FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

ARTICLES FOR FACULTY MEMBERS

Title/Author	Advances on polysaccharides from cactus: Analysis and review based on bibliometrics / Zheng, Y., Zhang, P., & Fu, L.
Source	*Journal of the Professional Association for Cactus Development* Volume 25 (2023) Pages 1–22 https://doi.org/10.56890/JPACD.V25I.513 (Database: JPACD.ORG)

Title/Author	Analysis of hydrocolloid excipients for controlled delivery of high-value microencapsulated prickly pear extracts / Fernández-Repetto, A., Gómez-Maqueo, A., García-Cayuela, T., Guajardo-Flores, D., & Cano, M. P.
Source	*Food Hydrocolloids for Health* Volume 3 (2023) 100115 Pages 1-10 https://doi.org/10.1016/j.fhfh.2023.100115 (Database: ScienceDirect)

Title/Author	An analysis of the plant- and animal-based hydrocolloids as byproducts of the food industry / Waraczewski, R., Muszyński, S., & Sołowiej, B. G.
Source	*Molecules* Volume 27 Issue 24 (2022) 8686 Pages 1-25 https://doi.org/10.3390/molecules27248686 (Database: MDPI)

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Title/Author	Application of fermentation for the valorization of residues from Cactaceae family / Carpena, M., Cassani, L., Gomez-Zavaglia, A., Garcia-Perez, P., Seyyedi-Mansour, S., Cao, H., Simal-Gandara, J., & Prieto, M. A.
Source	*Food Chemistry* Volume 410 (2023) 135369 Pages 1-15 https://doi.org/10.1016/j.foodchem.2022.135369 (Database: ScienceDirect)

Title/Author	Bioactive compounds of Barbados Gooseberry (Pereskia aculeata Mill.) / Egea, M. B., & Pierce, G.
Source	*Reference Series in Phytochemistry* (2021) Pages 225–237 https://doi.org/10.1007/978-3-030-57415-4_13 (Database: Springer, Cham)

Title/Author	Opuntia monacantha: Validation of the anti-inflammatory and anti-arthritic activity of its polyphenolic rich extract in silico and in vivo via assessment of pro- and anti-inflammatory cytokines / Abid, F., Saleem, M., Jamshaid, T., Jamshaid, U., Youssef, F. S., Diri, R. M., Elhady, S. S., & Ashour, M. L.
Source	*Journal of Ethnopharmacology* Volume 326 (2024) 117884 Pages 1-11 https://doi.org/10.1016/J.JEP.2024.117884 (Database: ScienceDirect)

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Source	*Foods* Volume 12 Issue 11 (2023) 2092 Pages 1-12 https://doi.org/10.3390/foods12112092 (Database: MDPI)

Title/Author	Physicochemical, nutritional, and medicinal properties of Opuntia Ficus-Indica (l.) Mill. and its main agro-industrial use: A review / Martins, M., Ribeiro, M. H., & Almeida, C. M. M.
Source	*Plants* Volume 12 Issue 7 (2023) 1512 Pages 1-45 https://doi.org/10.3390/PLANTS12071512 (Database: MDPI)

Title/Author	Trends in research on cacti: the food of the future / Coqueiro, J. M., Costa, L. D., Silva, L. C. e., dos Santos Conceição, L., da Silva Cardoso, P., Ferreira Ribeiro, C. D., & Otero, D. M.
Source	*Journal of the Science of Food and Agriculture* Volume 104 Issue 9 (2024) Pages 4939–4949 https://doi.org/10.1002/jsfa.13306 (Database: Wiley online Library)

Title/Author	Underutilized plants of the Cactaceae family: Nutritional aspects and technological applications / de Araújo, F. F., de Paulo Farias, D., Neri-Numa, I. A., & Pastore, G. M.
Source	*Food Chemistry* Volume 362 (2021) 130196 Pages 1-17 https://doi.org/10.1016/j.foodchem.2021.130196 (Database: ScienceDirect)

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Journal of the Professional Association for Cactus Development

Advances on polysaccharides from cactus: analysis and review based on bibliometrics

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Abstract. Plant polysaccharides are beneficial for developing new drugs, nutraceuticals and functional foods. Cactus is of interest to researchers in agronomy, medicine and food chemistry because of its long history of medicinal use, its simple growing requirements and biological basis for becoming a green vegetable. This review provides the first summary and analysis of the research history of cactus polysaccharides through a bibliometric approach. Bibliometrics was used to investigate the focus of different stages of development of the topic, with contributions from different countries and institutions. In addition, keyword analysis and keyword clustering were used to understand the different research directions of this topic. The analysis showed that: (1) considerable work on plant polysaccharides in the Cactaceae family has focused on *Opuntia spp.* (2) The study of cactus plant polysaccharides is a long-established topic but did not attract much attention in its early stages. (2) In 2018, research on cactus polysaccharides has received more attention than ever before. (3) Mexican institutions and scholars constituted the most important contributions to this topic. (4) This theme has only formed one complex network of cooperation, mainly composed of Mexican institutions and scholars. (5) Early studies on cactus polysaccharides focused on the detection, extraction and purification of polysaccharide content. (6) The biological activities of plant polysaccharides have gradually become the focus of research in recent years. (7) The biological activity of plant polysaccharides has been verified from *in vitro* and *in vivo* experiments with positive results.

Keywords: *Opuntia spp.; Statistical analysis; CiteSpace; Extraction; Biological activity*

Citation: Zheng, Y., Zhang, P., and Fu, L. 2023. Advances on polysaccharides from cactus: analysis and review based on bibliometrics. *Journal of the. Professional Association for Cactus Development. 25: 1-22*. https://doi.org/10.56890/jpacd.v25i.513.

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Introduction

Plant carbohydrates account for more than 80% of the dry matter and include monosaccharides, disaccharides, oligosaccharides and polysaccharides. Plant polysaccharide is a polysaccharide produced by plant cell metabolism (Warnakulasuriya *et al.,* 2018). Generally, plant polysaccharides are composed of hundreds of monosaccharides, and their properties differ from those of monosaccharides. Plant polysaccharides are widely used in food, pharmaceutical and daily chemical fields and have become a hot spot in food science, natural medicine and life science (Shao *et al.,* 2020). Numerous studies have shown that plant polysaccharides have biological activities such as immunomodulation, antioxidant, anti-fatigue, hypoglycemic, hypolipidemic, anti-tumour, anti-radiation, anti-bacterial, anti-virus, and liver protection (Li *et al.,* 2018). Several hundred polysaccharides have been isolated from natural products, among which water

soluble polysaccharides extracted from plants have been studied most extensively. Cactus is the common name of the species belong to family Cactaceae, comprising about 127 genera with 1750 known species. Cactus is native to tropical and subtropical arid regions of North and South America and is now found in temperate regions of the world, especially in Mexico, where it is most abundant and commonly cultivated as a nutritious food (Delgado-Ramírez *et al.,* 2021; Mitchell, 2021; Pensamiento-Niño *et al.,* 2021; Matus *et al.,* 2022).

Cactus contains energy substances such as fats, proteins and carbohydrates, as well as active ingredients such as alkaloids, polysaccharides and flavonoids (Bouaouine *et al.,* 2018; Hailu, 2020; Sigwela *et al.,* 2021). Polysaccharides are currently recognized as the main active component in cactus and have also become a hot topic of research on cactus in recent years. The composition and structure of cactus polysaccharides are complex, and many researchers have extracted and isolated different polysaccharides from different species and genera of cactus. Cactus polysaccharides contain homopolysaccharides, heteropolysaccharides and glycoproteins with different polysaccharide contents, relative molecular masses and monosaccharide composition ratios (Huang *et al.,* 2009; Riaz *et al.,* 2021). Cactus polysaccharide extraction methods mainly include the hot water method, acid method, enzyme method, ultrasonic method, microwave-assisted method, microbial method, etc. The extracted crude polysaccharide of cactus often contains protein, pigment, small molecules, and other impurities that need to be removed (Cai *et al.,* 2008; de Andrade Vieira *et al.,* 2021).

So far, several reviews have described the progress of research on cactus components (Goycoolea *et al.,* 2003; Stintzing *et al.,* 2005; Moßhammer *et al.,* 2006; Shedbalkar *et al.,* 2010; Shetty *et al.,* 2012). Some other reviews on plant polysaccharides include cactus polysaccharides (Xie *et al.,* 2016; Yuan *et al.,* 2021; Felicia *et al.,* 2022). However, the research advances in these reviews on cactus polysaccharides' extraction, purification, structural characterization and bioactivity need to be updated. In addition, this series of reviews used traditional methods for interpreting the highlighted literature. The development of bibliometrics in recent years has allowed for a more statistically dependent presentation of the reviews. Bibliometric analysis is a literature and information mining method based on mathematical statistics. It can reflect research trends and hotspots through clustering relationships of keywords in the literature and has become an important tool for global analysis in various scientific fields (Fu *et al.,* 2022; Jin *et al.,* 2022; Li *et al.,* 2022; Shen *et al.,* 2022; Zheng *et al.,* 2022 a; b; c). This review summarizes the research progress of polysaccharides from the cactus.

Material and Methods

Two bibliometrics software have been used in this systematic literature review. The first is CiteSpace, developed by Dr. Chaomei Chen, a professor at the Drexel University School of Information Science and Technology (Börner *et al.,* 2003; Chen, 2004, 2006; Chen *et al.,* 2010). CiteSpace 6.1R2 was used to calculate and analyze all documents. COOC is another emerging bibliometrics software (Xueshu *et al.,* 2022). COOC12.6 was used to analysis of annual publications. We used the core collection on Web of Science as a database to assure the integrity and academic quality of the studied material. "cactus polysaccharide", have been used as a "Topic." The retrieval period was indefinite, and the date of retrieval was July 30, 2022. 112 articles were retrieved, spanning the years 1991 to 2021.

Results

Literature Development Trends

The paper on polysaccharides from cactus in the core collection on Web of Science dates back to 1991. Nerd and Nobel (Nerd *et al.,* 1991) analyzed the composition of *Opuntia ficus-indica* (L.) Miller under well-watered and drought-stricken conditions for 15 weeks. The analysis of polysaccharides is one of the indicators. They found an increase of polysaccharides (probably starch) in the water-storage thin-walled tissue of *Opuntia ficus-indica* (L.) under drought conditions. The composition in *Cerus peruvianus* was investigated by Alvarez et al. (Alvarez *et al.,* 1992) in 1992. They found that the main component of gum fraction was an uronylated rhamnoarabinogalactan with a viscosity that may exceed 1000 mL g^{-1}. This composition was precipitated and washed by ethanol to obtain an almost protein-free soluble mucopolysaccharide that can be used as a flocculant for water impurities and as an adjuvant in cosmetic formulations. Figure 1 shows the annual and cumulative number of publications related to cactus polysaccharides. Although this topic has received attention as early as 1991 (this does not mean that the topic has only been investigated since 1991, as the papers used here only consider the core collection on Web of Science), not every year has been published. Since 2002, this topic has entered a stable phase, with papers published yearly. Until 2017, this topic did not show significant growth, and only some of these years had more annual publications, such as 2008, 2012 and 2016. Starting from 2017, this topic entered a period of significant growth. More than 5 articles were published each year, reaching a peak of 12 articles in 2019. While the number of papers published worldwide has increased significantly over the past five years, an increase in annual publications on a topic still means it is starting to attract more people to it. Although the survey of cactus plant polysaccharides can be published in a range of non-English speaking indigenous academic journals due to the geographical distribution of cactus, the increase in the number of papers on this topic in the core collection on the Web of Science database reflects that it is gaining more popularity among international scholars.

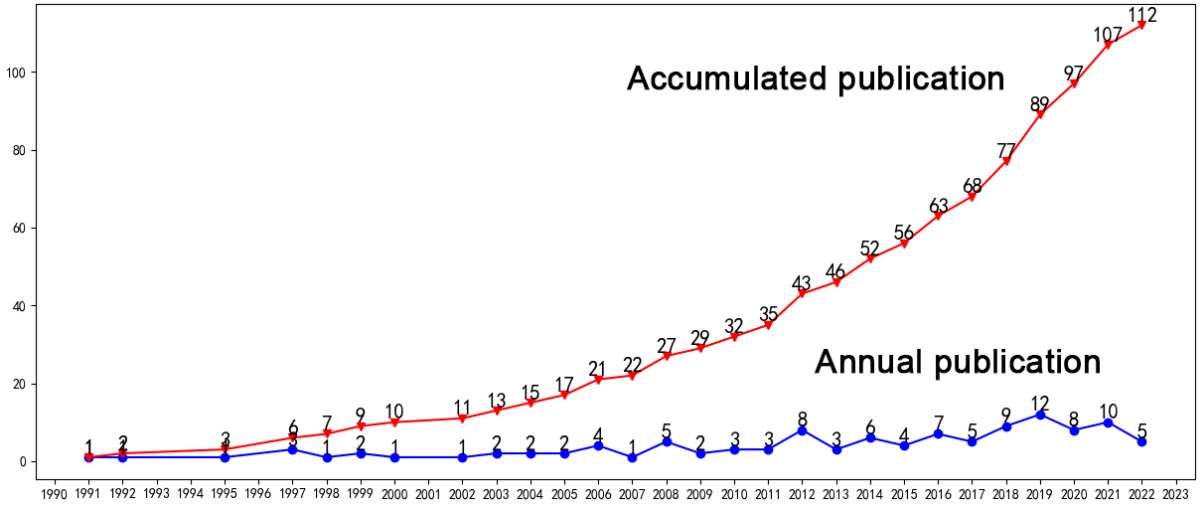

Figure 1. Annual and accumulated publications from 1991 to 2022 searched in the Web of Science about polysaccharides from cactus.

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Journals, Cited Journals and Research Subjects

Figure 2 shows a tree diagram of the top 6 journals publishing the number of polysaccharides from the cactus. Polysaccharides are carbohydrates; therefore, Carbohydrate Polymers has published the most significant number of papers on this topic. The papers in this journal focus on the investigation of the properties of cactus polysaccharides. For example, Manhivi et al. (Manhivi *et al.,* 2018) investigated the composition, rheology and thermal properties of *Opuntia spp.* mucilage. The Journal of the Professional Association for Cactus Development publishes scientific articles about the Cactaceaes and near-family species communities worldwide. Therefore, it has also made a remarkable contribution to this topic. For example, Armenta *et al.* (2009) investigated the structure of polysaccharides in *Opuntia matudae* fruits of different ripening stages. Cardenas *et al.* (1997) investigated the rheological behaviour of polysaccharides isolated from *Opuntia ficus-indica*. The journals in Figure 2 are all in the food field except for one botanically related. Because cactus mucilage has potential applications in many fields. Therefore, a series of works focused on mucilage extraction, purification and compositional analysis. For example, De Andrade Vieira *et al.* (2021) recently published a study on the physicochemical properties, structure and technological properties of the mucus extracted from seven cladodes of cacti (*Opuntia fícus-indica*, *Opuntia cochenillifera*, *Cereus jamacaru*, *Cereus hildmannianus*, *Pilosocereus gounellei*, *Tacinga inamoena* and *Pilosocereus pachycladus*) and found that arabinose is the main polysaccharide.

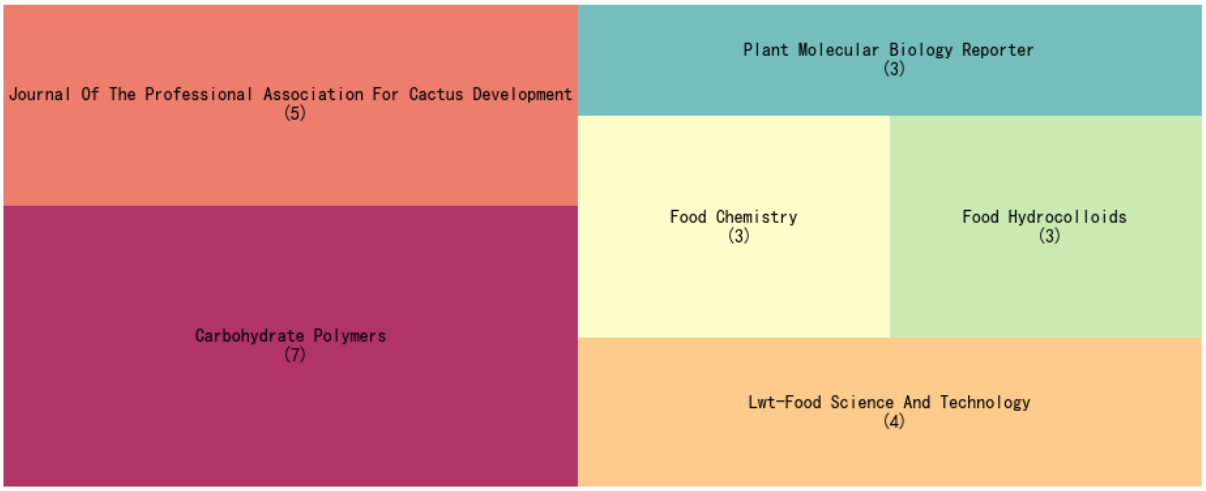

Figure 2. Top 6 journals that published articles on the polysaccharides from cactus.

In addition to the number of papers published by the journal on the topic, the frequency with which the journal is cited by papers related to the topic is also an important indicator. Table 1 shows the top 12 cited journals on the polysaccharides from the cactus. The table shows that Carbohydrate Polymers continue to rank first and are the most frequently cited journal. This represents not only a large number of papers on polysaccharides from cactus published in this journal, but the academic community has also recognized these papers. However, the rankings in Table 1 are not entirely objective, as whether a paper is widely cited receives influence from academic publishers. Therefore, we counted all journals with more than 20 citations in Table 1, representing that they all have an important contribution to the development of this topic. Most of the journals in Figure 2 appear in Table 1, except for Lwt-Food Science and Technology and Plant Molecular Biology Reporter. This means that the papers on polysaccharides from cactus published in these two journals have not attracted much attention. In

addition, some additional information is given in Table 1. For example, journals related to phytochemistry and analytical chemistry are included in Table 1. This represents polysaccharides from the cactus involved in the analysis of phytoconstituents. The appearance of Journal of Arid Environments also makes sense since the cactus is a very important group of plants in arid ecosystems.

Table 1. Top 12 cited journals on the polysaccharides from cactus.

No.	Citation	Cited Journal
1	63	Carbohydrate Polymers
2	47	Food Chemistry
3	40	Journal of Agricultural and Food Chemistry
4	35	Carbohydrate Research
5	31	Journal of the Professional Association for Cactus Development
6	29	Journal of Arid Environments
7	26	International Journal of Biological Macromolecules
8	26	Food Hydrocolloids
9	25	Phytochemistry
10	23	Analytical Biochemistry
11	22	Journal of Food Science
12	21	Food Research International

To further explore the information that journals provide, we constructed a co-occurrence network of cited journals related to polysaccharides from the cactus (Figure 3). Journals highlighted in Figure 2 and Table 1 are not labelled. Analytical Biochemistry, ranked #10 in Table 2, has a very strong betweenness centrality in Figure 3, representing that the content of this journal connects two different fields. Another journal of analytical chemistry, Analytical Chemistry, plays a similar role. After interpreting the journals connected around them, the information related to cactus polysaccharides in these journals is about the extraction, isolation and analysis of plant components. Another cluster is included in the lower part of Figure 3, where the journal with the strongest intermediary centrality is Journal of Arid Environments. In addition, Acta Horticulturae is also an important journal in this cluster. They link ecology and a range of food-related journals. In the upper left corner of the entire network is another journal with high intermediary centrality, Annals of the New York Academy of Sciences. It links to journals related to the analysis of cactus phytoconstituents and the exploration of molecular mechanisms (e.g. Cell Biology International, Journal of Clinical Neuroscience, Cellular and Molecular Neurobiology, etc.).

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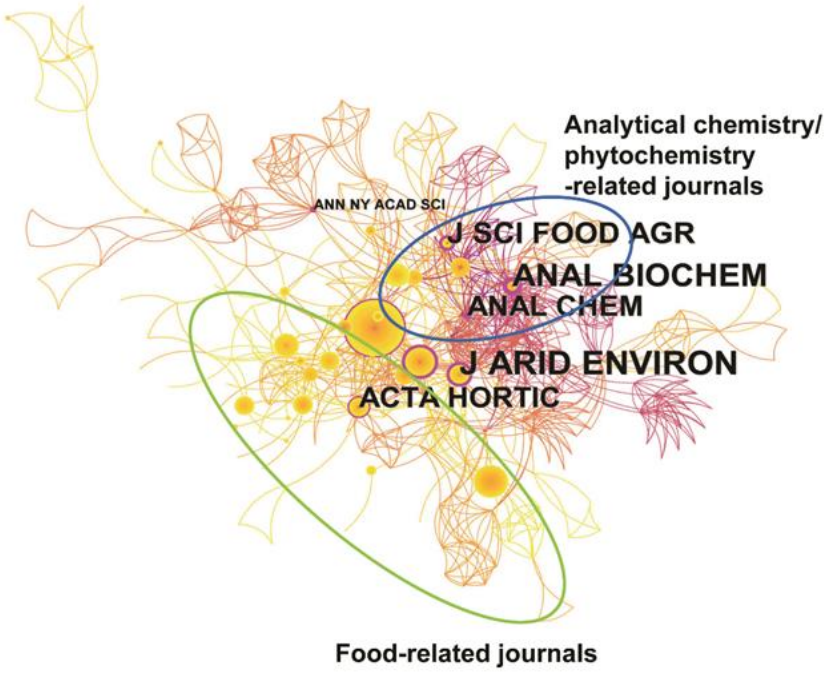

Figure 3. Co-occurrence network of cited journals for polysaccharides from cactus.

Table 2 shows which cited journals this theme was extended to for the first time between 2020-2022. Analyzing journals that have published papers related to a topic for the first time in recent years provides an insight into the most current developments. As shown in Table 2, the journals that published papers on cactus polysaccharides for the first time in the last three years were still mainly concentrated in food, biological and comprehensive journals. However, it is worth noting that materials science-related journals, especially polymer-related journals, appear in 2020 and 2022. Shanmugavel *et al.* (2020) tried the incorporation of bio-additives prepared from cactus extracts into cement concrete mixtures and tested the fresh and hardened state of the modified concrete. In addition, there are the first publications on this topic in water treatment/water environment-related journals in 2021 and 2022. Hussain and Haydar (Hussain *et al.,* 2021) prepared a novel plant flocculant using *Opuntia stricta* (Adjeroud *et al.,* 2015, 2018; Djerroud *et al.,* 2018; Adjeroud-Abdellatif *et al.,* 2020). The results showed that the main component in the flocculant was polysaccharide. Asnam *et al.* (2022) prepared porous composites from the cactus extract and sodium alginate and tested the potential of this material to replace PVA as an adsorbent gel.

Table 2. List of journals has appeared in the co-occurrence network in the last three years.

Year	Journal
2022	Biomolecules; Journal of Water Process Engineering; Natural Product Research; Polymer Bulletin
2021	Clean-Soil Air Water; Journal of The Serbian Chemical Society; Phyton-International Journal of Experimental Botany
2020	Agrociencia; Construction and Building Materials; International Journal of Dairy Technology; International Journal of Polymer Analysis and Characterization; Journal of Food Quality; Metabolites; Polymers; Scientific Reports

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The category of the published paper can reflect the evolution of the topic. Figure 4 shows the evolution of the category of polysaccharides from the cactus over time. The topic began in Plant Sciences and covered a range of plant and biology-related areas through 2000. It is noteworthy that this topic had already started to venture into the field of material science in this period. There are two possible interpretations here. As mentioned before, the cactus extract may be used as a green additive to prepare new types of concrete. Another possibility is that the cactus extract was used as a raw material for a green preservative for metal preservation. The preparation of green preservatives from plant extracts is a subject that has a long history and has been attracting the attention of materials scientists (Alrefaee *et al.,* 2021). Since 2003, cactus polysaccharides have entered another important field, Food Science & Technology. After that, this topic started to be cross-researched with chemistry and pharmacology. At the same time, the application of cactus polysaccharides began to be investigated, so some papers were published in journals related to Immunology, Neurosciences, Cell Bilogy. In 2018, the application of cactus polysaccharides was extended to Water Resources. In recent years, this topic started to enter some completely different fields than before, such as Physics (Civil), Energy & Fuels, Materials Science (Coatings & Films).

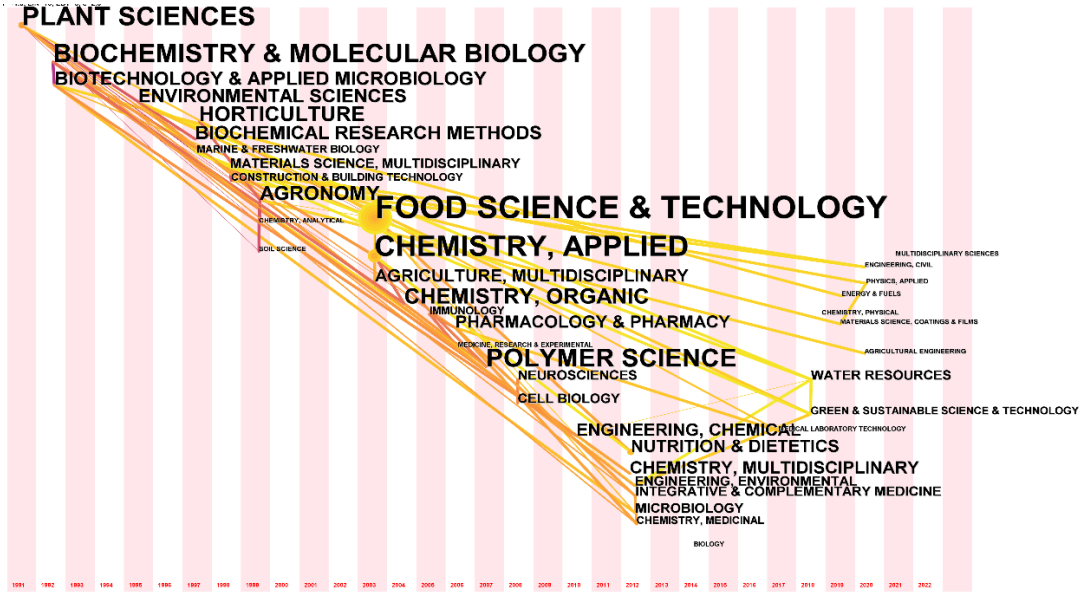

Figure 4. Time-zone view of research categories for polysaccharides from cactus.

Geographic Distribution

Figure 5 shows the pie chart of papers related to polysaccharides from cactus contributed by different countries. Mexico significantly contributed to this topic, contributing 18.1% of the papers. Both China and USA contributed >7% of the papers. France and Brazil contributed >5% of the papers. As seen in Figure 5, although the growth of the cactus is geographically limited, the investigation of its polysaccharides has not been restricted to a few countries. On the contrary, scholars from all geographical regions have demonstrated interest in this topic. As of 2022, a total of 46 countries are investigating this topic, with U.A.E, Poland and Romania being the first to publish on this topic after 2020.

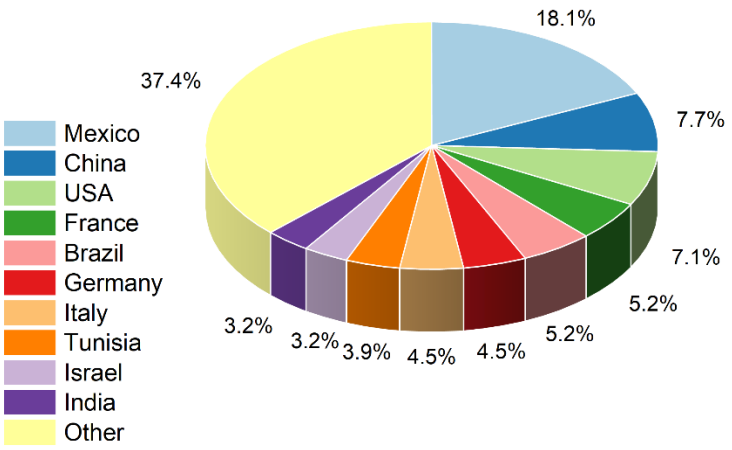

Figure 5. Pie chart of papers related to polysaccharides from cactus contributed by different countries.

Figure 6 illustrates the cooperation network between the different institutions on this topic. Although the topic has attracted the participation of different countries around the globe, only one network of cooperation has been formed. This collaborative network occupies a very important place in this topic. This collaborative network includes a series of Mexican universities and research institutions, mainly led by Colegio de Postgraduados, Universidad Autónoma del Estado de Hidalgo, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Universidad Autónoma de Sinaloa. In addition to this significant collaborative network, only a scattering of other institutions has taken to the collaborative investigation of this topic. It is worth noting that these collaborations are limited to domestic cooperation rather than international cooperation. This may be due to differences in the distribution and introduction of cacti in different countries. On the other hand, this topic may not have a challenge that needs to be urgently tackled, so different researchers focus on different perspectives.

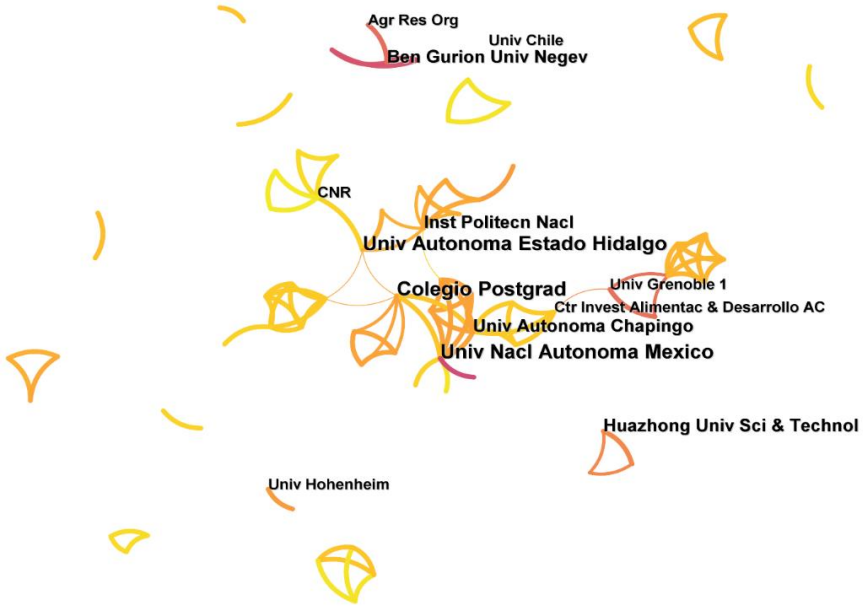

Figure 6. Institution cooperation network for polysaccharides from cactus.

Keyword Analysis and Evolution of The Field

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The most effective way to understand the direction of investigating concerns in a topic is the analysis of keywords. Table 3 lists the top 17 keywords in this topic. Not surprisingly, Polysaccharide was the most frequently occurring keyword in this theme. The second most frequent keyword was *Opuntia ficus-indica*, representing it as the most popular specie among the many cacti. *Opuntia ficus-indica* is a species of cactus that has long been a domesticated crop plant grown in agricultural economies throughout arid and semiarid parts of the world. It has assumed the role of "model plant" in a series of scholarly studies on cacti. Research on plant polysaccharides cannot be done without extraction techniques. Appropriate extraction techniques are necessary to obtain extracts containing as many polysaccharides as possible. The high purity of plant polysaccharides can be obtained by separating the extracts. Therefore, extraction is also a keyword that appears frequently. In the extraction of cactus, besides polysaccharides, other substances such as Mucilage, Dietary fibre and Pectin have also received attention (Cárdenas *et al.,* 2008; Lira-Ortiz *et al.,* 2014; Gheribi *et al.,* 2018). The plant organs of the cactus used to extract polysaccharides were also investigated. Table 3 shows that Cactus pear fruit (Montoya-Arroyo *et al.,* 2014; Robert *et al.,* 2015) and Cladode (Petera *et al.,* 2015) were the most frequently investigated organs. When the properties of cactus polysaccharides are investigated, the antioxidant properties are the ones that are most often focused on.

Table 3. List of top 17 keywords for polysaccharides from cactus.

No.	Freq	Centrality	Keywords
1	48	0.29	Polysaccharide
2	40	0.15	*Opuntia ficus-indica*
3	19	0.23	Extraction
4	17	0.33	Antioxidant activity
5	15	0.13	Cactus pear
6	13	0.13	Mucilage
7	12	0.09	Fruit
8	8	0.08	Dietary fiber
9	7	0.19	Acid
10	6	0.28	Cactus
11	6	0.13	Cladode
12	5	0.06	Chemical characterization
13	5	0.18	Extract
14	5	0.14	Film
15	5	0.00	Optimization

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16	5	0.03	Food
17	5	0.17	Pectin

Cluster analysis can further understand the different directions of investigation in this topic. Figure 7 shows that 9 clusters were formed after clustering the keywords. Some of these clusters have overlapping sections, representing a strong similarity between the papers. However, it is also possible to see that some of these clusters are more independent from others, representing that they focus on a specific topic. From the clustering results, the study of polysaccharides from cactus has shown different directions in the course of history. This situation is generally because the investigation of a topic has undergone different stages. After a series of problems have been overcome or investigated, a topic begins to move into a deeper investigation. Table 4 describes the clusters and their ID, size (number of papers), silhouette, and respective keywords. The following is a short explanation of each cluster:

Table 4. Knowledge clusters in the topic polysaccharides from cactus on keyword co-occurrences for each cluster.

Cluster ID	Size	Silhouette	Keywords	References
0	44	0.929	Polysaccharide; Apoptosis; Extract; Aloe vera; Nitric oxide;	(Huang *et al.,* 2008 a, 2009; Chen *et al.,* 2011, 2021; Deters *et al.,* 2012; Ejaz *et al.,* 2014; Li *et al.,* 2014; Ben Saad *et al.,* 2017; da Silva Brito *et al.,* 2020; Otálora *et al.,* 2021; Zhang *et al.,* 2022)
1	33	0.877	Cactus pear; Chemical characterization; Color; Betalain; Quality	(Mattagajasingh *et al.,* 2006; Moßhammer *et al.,* 2006; Luna-Paez *et al.,* 2007; Ramírez-Truque *et al.,* 2011; Di Cagno *et al.,* 2016; Giglio *et al.,* 2020)
2	30	0.879	Extraction; Acid; Optimization; Aroma; Rheological property; Cellulose	(Ninio *et al.,* 2003 a; Cárdenas *et al.,* 2008; Felkai-Haddache *et al.,* 2016; López-Mercado *et al.,* 2018; Camelo Caballero *et al.,* 2019; Silva *et al.,* 2019; Nagarajan *et al.,* 2020; Santagata *et al.,* 2022)
3	25	0.945	Fruit; Behavior	(Fox *et al.,* 2012; Nharingo *et al.,* 2015; Madera-Santana *et al.,* 2018; Asnam *et al.,* 2022)
4	24	0.872	Dietary fiber; Chemical composition; Bioactive compound; Functional property	(Armenta *et al.,* 2009; Nuñez-López *et al.,* 2013; de Campo *et al.,* 2018; El-Shahat *et al.,* 2019)
5	24	0.979	Antioxidant activity; Stroke; Polyphenol; Brain slice; Food; Biological activity	(Huang *et al.,* 2008 b; a; Kim *et al.,* 2013, 2014; Xie *et al.*, 2016)
6	23	0.896	Ficus indica; Mucilage; C13 NMR; Water relation	(Nobel *et al.,* 1995; Mondragon-Jacobo *et al.*, 2000; Vignon *et al.,* 2004; Ramírez-Truque *et al.,* 2011; Lira-Ortiz *et al.,* 2014; Montoya-Arroyo *et al.,* 2014; Rivera-Corona *et al.,* 2014; Manhivi *et al.,* 2018;

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				Raimundo *et al.,* 2018; Hussain *et al.,* 2021; Makhloufi *et al.,* 2022)
7	22	0.964	Cactus; Cladode; Cactaceae; Plant	(Nerd et al., 1991; Nobel *et al.,* 1995; Peña-Valdivia *et al.,* 2012; Ciriminna *et al.,* 2019)
8	16	0.905	Prickly pear; Component; Constituent	(Ninio *et al.,* 2003 b; Habibi *et al.,* 2004)

0 (Polysaccharide) The papers on this topic contain many papers focusing on the determination of a range of properties of cactus extracts and polysaccharides. For example, Huang *et al.* (2009) determined cactus polysaccharides' neuroprotective and antioxidant effects *in vivo* and *in vitro*. Rats were used in this work for the study. The study found that cactus polysaccharides reduced neurological deficit scores after cerebral ischemia-reperfusion. It reduces cerebral infarct volume and cortical neuronal loss by decreasing inducible nitric oxide synthase protein synthesis. Saad *et al.* (2017) discovered the protective effect of lithium cactus polysaccharide-induced liver injury in rats. Li *et al.* (2014) investigated the antitumor effect of cactus polysaccharides on lung squamous carcinoma cells SK-MES-1.

1 (Cactus pear) The study of cactus pear is often separated from the study of cladode. This cluster contains a series of papers investigating cactus pear. Ramírez-Truque *et al.* (2011)investigated the composition of cell wall polysaccharides in dragon fruit pulp. Di Cagno *et al.* (2016) investigated how to improve the shelf life, rheological, organoleptic and functional properties of cactus pear puree. Moßhammer et al. (Moßhammer *et al.,* 2006) summarized the physical and chemical properties of cactus pear and their potential applications.

2 (Extraction) The active ingredients in cactus require rational extraction techniques to obtain them. This cluster contains a series of extractions and characterizations of the active/valuable components of the cactus. The process of polysaccharide research generally includes extraction, separation, purification and purity determination. Among them, extraction is related to polysaccharide yield, structure and conformation. Separation and purification further improve the purity of polysaccharides, and their success and effectiveness will be directly related to the feasibility and credibility of subsequent structural studies. Cactus polysaccharides are polar macromolecules. Extraction is usually done by degreasing and decolorizing the raw material and then extracting it with water, salt or diluting alkali at different temperatures. The extracts are concentrated and then precipitated by centrifugation with precipitating agents (e.g. acetone, ethanol, etc.). The precipitated part is usually concentrated under reduced pressure or processed by membrane separation or ultrafiltration, etc., and then cold dried to obtain crude polysaccharide. According to the nature of the polysaccharide to be extracted and the purpose of the study, the extraction agent and extraction conditions are selected. A water-based solution extraction method is often used, or extraction with hot or cold dilute acid or dilute alkali. Avoid using strong acids and bases for extraction to prevent the breakage of glycosidic bonds.

3 # 3 (Water treatment) This cluster contains mainly a series of papers on using cactus raw materials for water treatment. Nharingo *et al.* (2015) investigated the potential use of cactus powder for coagulation-flocculation processes, especially for the adsorption and removal of lead ions. Fox et al. (Fox *et al.,* 2012) also attempted the removal of As (V) from water with cactus mucilage. Asnam et al.

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(Asnam *et al.,* 2022) attempted a composite of the cactus extract with sodium alginate. This composite has promise as an adsorbent material for anhydrous treatment.

4 (Bioactive compound) There is some overlap between the papers in this cluster and #1 and #6, which mainly contain a series of investigations on the active ingredients of cactus. For example, Nuñez-López *et al.* (2013) investigated the physicochemical, nutritional and antidiabetic properties of *Opuntia ficus-indica* at different stages of maturation. Armenta and Peña-Valdivia (Armenta *et al.,* 2009) investigated the variation of polysaccharides in *Opuntia matudae* fruits at different stages of ripening.

5 (Enzyme) This cluster consists of two main aspects. The first is the use of enzymes to enhance the extraction. For example, Kim et al. proposed an enzyme-assisted extraction method for improving the extraction and recovery of bioactive materials from cactus. The other category is the potential of cactus extracts in neurological applications. Huang *et al.* (2008 b) investigated the protective effect of cactus polysaccharides on H_2O_2-induced cortical and hippocampal damage in rats.

6 (Mucilage) This cluster contains mainly a series of papers on cactus mucilage. For example, cactus mucus is used to prepare bio-packaging films (Makhloufi *et al.,* 2022). Cactus mucilage can also modify the physicochemical properties of sorghum starch (Rivera-Corona *et al.,* 2014).

7 (Cladode) In addition to the fruit of the cactus, the cactus cladode is the most commonly investigated organ. In early investigations, the relationship between cactus cladode growth and the environment was brought into focus (Nerd *et al.,* 1991; Nobel *et al.,* 1995). Later, how to extract polysaccharides in cactus cladode became the main direction of interest (Sánchez-Hernández *et al.,* 2006; Ciriminna *et al.,* 2019).

8 (Prickly pear) This topic contains only two papers. Ninio *et al.* (2003 b) monitored changes in sugar, acid and volatile components in *Cereus peruvianus* (L.) Miller fruit at different stages of ripening. Habibi *et al.* (2004) detected arabinogalactan in the pericarp of *Opuntia ficus-indica*.

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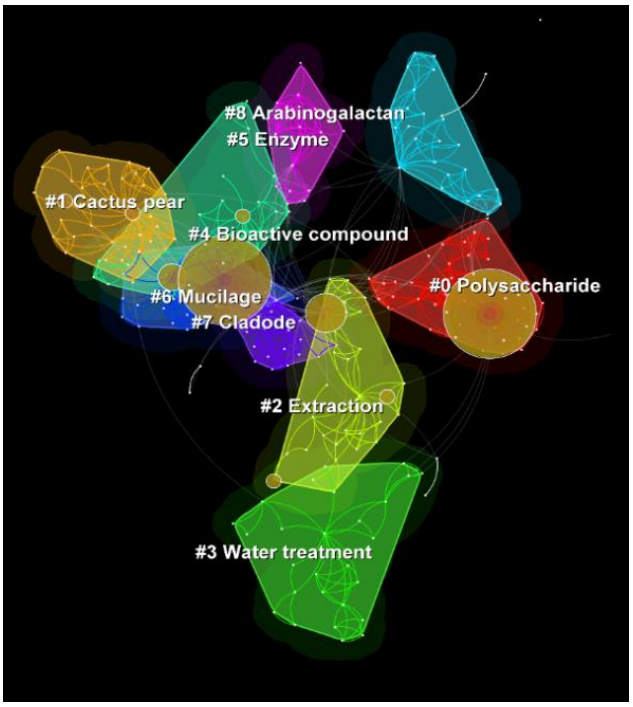

Figure 7. Grouping of keywords for polysaccharides from cactus.

Conclusions

In recent years, polysaccharides have shown significant promise in developing food, pharmaceutical, agricultural and cosmetic industries. As an edible and medicinal plant, cactus polysaccharides have a variety of important medicinal and edible values. Currently, the focus of cactus polysaccharides is mainly on immunomodulatory, antioxidant, hypoglycemic, antitumor and hepatoprotective properties. We analyzed the research process on cactus polysaccharides using a bibliometric approach and obtained the following main analytical conclusions:

(1) The paper on polysaccharides from cactus in the core collection on Web of Science dates back to 1991. Since 2002, this topic has entered a stable phase, with papers published yearly. Starting from 2017, this topic entered a period of significant growth.
(2) Not only have a large number of papers on polysaccharides from cactus published in Carbohydrate Polymers, but the academic community has also recognized these papers.
(3) Materials science-related journals, especially polymer-related journals, started publishing papers on polysaccharides from cactus recently.
(4) Cactus polysaccharides and mucilage can be potentially used as materials for water treatment, so this topic has also been published in water environment/water treatment-related journals in the last two years.
(5) The topic began in Plant Sciences and covered a range of plant and biology-related areas through 2000. Since 2003, cactus polysaccharides have entered Food Science & Technology. After that, this topic started to be cross-researched with chemistry and pharmacology.
(6) Mexico significantly contributed to this topic, contributing 18.1% of the papers. Both China and USA contributed >7% of the papers.
(7) Only one network of cooperation occupies this topic. This collaborative network includes a series of Mexican universities and research institutions.

(8) Cladode and the cactus fruit are the most commonly used organs to be extracted for polysaccharides.
(9) The extraction principle of the hot water method is similar to that of the acid method, in which the cell wall is ruptured by hot water or acidic solution to release polysaccharides.
(10) Enzymatic, ultrasonic and microwave-assisted methods have been used for extract cactus polysaccharides via breaking the cell wall.

The above analysis shows that bibliometrics is a powerful statistical technique for the analysis of research trends on a topic. We believe that this analytical technique can be used to further analyze topics related to cactus, such as the progress of cactus extraction process.

ETHICS STATEMENT

Not applicable

CONSENT FOR PUBLICATION

Not applicable

AVAILABILITY OF SUPPORTING DATA

Data sharing is not applicable to this article as no datasets were generated or analyzed during the current study.

COMPETING INTERESTS

The authors declare that they have no competing interests

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AUTHOR CONTRIBUTIONS

Conceptualization, Y.Z. and L.F.; methodology, L.F; software, P.Z.; validation, Y.Z. and P.Z.; formal analysis, Y.Z. and P.Z.; investigation, Y.Z.; resources, L.F.; data curation, Y.Z.; writing—original draft preparation, Y.Z. and P.Z.; writing—review and editing, L.F.; visualization, L.F.; supervision, L.F.; project administration, L.F.

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ARTICLES FOR FACULTY MEMBERS

FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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Analysis of hydrocolloid excipients for controlled delivery of high-value microencapsulated prickly pear extracts

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ABSTRACT

Prickly pears (*Opuntia ficus-indica*) are potential sources of functional ingredients because they are rich in betalains and phenolic compounds. However, mentioned bioactives may degrade during storage when exposed to air, light, and heat which could limit their application. To increase the stability and bioaccessibility of prickly pear extracts, we compared the ultrasound-assisted freeze-dried microencapsulation of seven excipient mixtures. The physical and physico-chemical properties (humidity, hygroscopicity, thermal analysis and morphology) and the qualitative and quantitative analysis of betalains and phenolic compounds (measured by high performance liquid chromatography) were analysed in each microparticle formulation. Stability-improving factors such as low humidity and hygroscopicity were observed in all microparticles. However, microparticle morphology was influenced by the excipient formulation. Encapsulation efficiency was higher than 60% for betalains and phenolic acids, however, flavonoids encapsulation efficiency was 14–35%. Based on the previous, the three best microparticles were selected: 100% maltodextrin (E2); 50% maltodextrin, 25% microcrystalline cellulose, 15% hydroxyl-propyl-methyl cellulose, and 10% xanthan gum (E5); and 100% β-cyclodextrin (E7). A static in vitro gastrointestinal digestion (INFOGEST method) was performed with these microparticles where the quantitative analysis of the bioactive compounds (HPLC) and their bioaccessibility was assessed. The bioaccessibility of bioactive compounds in encapsulated prickly pear extracts was improved when compared to the control. Microparticles containing maltodextrin and microcrystalline cellulose (E2) had the highest bioaccessibility and showed potential for the future formulation of functional foods.

1. Introduction

Prickly pears (*Opuntia ficus-indica* L. Mill.) are the most abundant species of the Cactaceae family. Although native to Mexico, they can be found in many arid and semi-arid climates of tropical and subtropical regions in the world. In Spain, prickly pears grow in the Canary Islands, Extremadura, Andalusia, and the Balearic Islands, where they occupy around 6000 hectares of cultivated area. Due to their Crassulacean Acid Metabolism (CAM) and anatomical structures, prickly pears are tolerant to drought and can reach a high biomass with great efficiency in the use of water (Feugang, Konarski, Zou, Stintzing & Zou, 2006).

Prickly pear fruits are rich in health-promoting compounds such as betalains and phenolic compounds which contribute to their health potential (Gómez-Maqueo, García-Cayuela, Fernández-López, Welti-Chanes & Cano, 2019, 2021). Red-colored betanin and yellow-colored indicaxanthin are the most abundant betalains in these fruits and are responsible for their free radical scavenging and antioxidant activity (Kanner, Harel & Granit, 2001). Prickly pear fruits are also abundant in phenolic acids such as piscidic acid, also found in other members of the Cactaceae family. Piscidic acid has shown anti-hypercholesterolemia effects by inhibiting cholesterol permeation in vitro as well as anti-inflammatory activity (Ressaissi, Attia, Pacheco, Falé & Serralheiro, 2017). Prickly pear fruits are rich sources of flavonoids such as isorhamnetin glycosides, which possess significant antioxidant and anti-inflammatory activities (Antunes-Ricardo, Gutiérrez-Uribe, López-Pacheco, Alvarez & Serna-Saldívar, 2015).

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From a nutritional point of view, the concentration of bioactive compounds that are potentially absorbed by our bodies is more important than the concentration in the food (Rodríguez-Roque et al., 2015). Bioaccessibility refers to the fraction of compounds that are released from the food matrix during digestion and that can be absorbed in the small intestine. The composition of the food matrix has a strong influence on the bioaccessibility and bioavailability of bioactive compounds. Consuming foods with a higher concentration of bioactives does not necessarily mean that a higher concentration of these will reach the target tissues in order to promote health.

In vivo digestion models are the best to determine the bioaccessibility and bioavailability of bioactive compounds. However, their implementation is a complicated and expensive process that normally limits the number of samples that can be studied. In comparison, in vitro gastrointestinal digestion models have the advantage that they are faster, simpler and less expensive. The INFOGEST consortium has published an in vitro static gastrointestinal digestion protocol as a standardized proposal to allow results to be comparable between different research groups (Minekus et al., 2014).

The degradation of betalains occurs in the stomach (gastric stage) and small intestine (intestinal stage), thus reducing their absorption and fecal excretion (Tesoriere et al., 2013). Meanwhile, the bioaccessibility of phenolic compounds is strongly influenced by their association with other molecules. For example, the bioaccessibility of phenolic glycosides is high since they may be hydrolysed during digestion to release the aglycones.

Microencapsulation is a technique where one or several bioactive compounds are covered by a biopolymer, thus protecting them from contact with oxygen, water and other conditions, and improving their stability (Saénz, Tapia, Chávez & Robert, 2009). Microencapsulation also prevents the formation of conglomerates, improves flow capacity, compression and mixing properties, and modifies the density of the particles (Ravichandran et al., 2014). There are different types of encapsulating agents, which can be polysaccharides, lipids or proteins. The choice of encapsulating material depends on the component to be encapsulated, as well as other characteristics such as electrical charge, molecular weight, viscosity, surface tension, and digestibility. The type of encapsulant used will considerably influence the morphology and surface of the generated extract, in addition to encapsulation efficiency (da Silva, Barreira & Oliveira, 2016; Đorđević et al., 2015; Sun-Waterhouse, Wadhwa & Waterhouse, 2013).

Among the different encapsulating materials that exist, semi-hydrolysed starch polymers such as maltodextrin (MD), cellulose derivatives such as microcrystalline cellulose (MCC) and hydroxymethylcellulose (HPMC), gums and cyclodextrins are good encapsulating materials for hydrophilic polymers (Dias, Ferreira & Barreiro, 2015; Otálora, Carriazo, Iturriaga, Nazareno & Osorio, 2015).

The use of encapsulation methodologies, such as ultrasound-assisted (conventional method), is suitable for the protection of these compounds against degradation to enable their use as functional ingredients in the food and pharmaceutical industry. The aim of this study was to compare different excipients using a conventional microencapsulation of extracts rich in betalains and phenolic compounds from *O. ficus-indica* fruits to improving its physicochemical properties and bioaccessibility.

2. Materials and methods

2.1. Encapsulating agents

The encapsulating agents used in this work were maltodextrin 10 - 10MD (Cargill, Minnesota, United States), microcrystalline cellulose-MCC (DFE, Goch, Germany), hydroxy-propyl-methyl-cellulose Pharmacoat 603 – HPMC (DVA Mexicanas S.A de C.V, Granada, Mexico), guar gum (Central de Drogas S.A de C.V, Naucalpan de Juárez, Mexico), Xanthan gum (Central de Drogas S.A de C.V, Naucalpan de Juárez, Mexico), α-cyclodextrin – α-CD (Cyclodex, Trappsol, Florida, United States), and β-cyclodextrin– β-CD (Sigma Alimentos, San Pedro Garza García, Mexico).

2.2. Plant material

Purple prickly pear (*Opuntia ficus-indica* L. Mill.) fruits from the Morada variety were obtained from Archena (Murcia, Spain, 38°7′N, 1°18′W; 121 m over sea level). Ripe fruits were selected according to uniform color, size, and absence of bruising (Supplementary Fig. S1). The physical and physicochemical characteristics (Supplementary Table S1), such as total weight (g), titratable acidity (g citric acid/100 g f.w.), pH, soluble solids (°brix), firmness (N/g f.w.), and CIE*LAB external color parameters (L*, a*, b*) were determined according to Gómez-Maqueo, Antunes-Ricardo, Welti-Chanes and Cano (2020). Whole fruits were cut into small pieces (20 × 20 mm), vacuum-sealed in polyethylene bags, frozen with liquid nitrogen, and freeze-dried (Telstar Lyo Beta 15). They were pulverized (Grindomix GM200, Retsch, Germany) to a particle size lower than 2 mm and seeds were removed. Pulverized samples were vacuum-sealed and stored at −20 °C.

2.3. Extraction of betalains and phenolic compounds

The extraction of betalains and phenolic compounds was performed simultaneously according to the methodology reported by García-Cayuela, Gómez-Maqueo, Guajardo-Flores, Welti-Chanes and Cano (2019). 1 g of freeze-dried whole prickly pear fruit was mixed with a 5 mL methanol: water (1:1, v/v) solution in a vortex for 1 min. Afterwards, samples were placed in an ultrasound bath (Ultrasons, JP Selecta, Barcelona, Spain) for 4 min and then centrifuged at 10,000 rpm. The supernatant was collected, and the pellet was re-extracted two more times with 3 ml methanol: water (1:1, v/v), and one more time with 3 mL of pure methanol. Combined supernatants were concentrated in a rotavapor and made up to a final volume of 5 mL with ultrapure water.

2.4. Microencapsulation

Prickly pear extracts were encapsulated according to the methodology by Kalogeropoulos, Yannakopoulou, Gioxari, Chiou and Makris (2010) with modifications. 0.8 g of microencapsulating agent was mixed with 4 mL aqueous prickly pear fruit extract. Seven different formulations were prepared by ultrasound and freeze-drying following the design shown on Table 1. Encapsulating ingredients were weighed and dissolved in 100 mL of ultrapure water, where 4 mL of aqueous whole fruit prickly pear extract were added. The suspension was sonicated (Ultrasons, JP Selecta, Barcelona, Spain) at 45 kHz frequency and 200 W power in an ice bath at 15 min intervals for a total of one hour. The solution was frozen at −20 °C and then freeze dried for 5 days at −45 °C and 1.3×10^{-3} MPa (Lyo Beta 15, Azbil Telstar, S.L., Terrasa, Spain). The same process was followed for the control (E0) but without the addition of encapsulating agents.

2.5. Moisture content and hygroscopicity

To determine moisture, 40 mg of sample were placed in a drying oven at 70 °C for 24 h until constant weight and weight loss was recorded (AOAC, 1998). Moisture content is expressed as percentage (%). To analyze hygroscopicity, a saturated sodium chloride solution (NaCl) was added to the base of the desiccator to create relative humidity of 75.5% inside the desiccator (Tonon, Brabet & Hubinger, 2008). 20 mg of sample were placed in the interior and the weight difference was recorded after 5 days. Hygroscopicity was calculated according to Eq. (1) and is expressed as percentage (%) or 1 g of absorbed moisture per 100 g dry solids (g/100 g) (Caparino et al., 2012).

$$HG = \frac{\Delta m/(M + M_i)}{1 + \Delta m/M} \quad (1)$$

Table 1
Materials used as encapsulating agents. Encapsulating matrixes (E1-E7) represent (or not) a combination of some individual encapsulating agents.

Encapsulating matrix	Individual encapsulating agents[1]						
	MD	MCC	HPMC	Guar	Xanthan	α-CD	β-CD
E0 (control)	–	–	–	–	–	–	–
E1	0.8 g	–	–	–	–	–	–
E2	0.4 g	0.4 g	–	–	–	–	–
E3	0.4 g	0.2 g	0.2 g	–	–	–	–
E4	0.4 g	0.2 g	0.12 g	0.08 g	–	–	–
E5	0.4 g	0.2 g	0.12 g	–	0.08 g	–	–
E6	–	–	–	–	–	0.8 g	–
E7	–	–	–	–	–	–	0.8 g

[1] Individual encapsulating agents: MD, maltodextrin; MCC, microcrystalline cellulose; HPMC, hydroxyl-propyl-methyl cellulose; Guar, guar gum; Xanthan, xanthan gum; α-CD, α-cyclodextrin; and β-CD, β-cyclodextrin.

Where Δm (g) is the increase in weight of powder after equilibrium, M is the initial mass of powder and M_i (%wb) is the free water contents of the powder before exposing to the humid air environment.

2.6. Differential scanning calorimetry (DSC)

Calorimetry was performed in a Discovery Ta DSC (Brand and country). Samples with no previous treatment were analysed between 20 and 350 °C at 10 °C/min in TzeroTM standard particles made from aluminum. Nitrogen (N_2) was used to purge with a flow of 50 mL/min. The equipment was calibrated (156,60 °C; ΔHf= 28,71 J/g) prior to analysis.

2.7. Scanning electron microscopy (SEM)

Scanning electron microscopy was used to analyze the morphology of the generated microstructures. Freeze-dried samples were used for this analysis. Samples were first metalized using a sputter Queorum (Q150-t) by covering them in gold. Once metalized, samples were introduced into the electronic scanning electron microscope (Philips XL 30 S-FEG). The design and nanolithography were analysed with Raith Elphy Quantum.

2.8. Encapsulation efficiency

Total encapsulated bioactive compounds were quantified according to Saénz et al. (2009). 100 mg of each microparticle was dissolved in 1 mL of ethanol-acetic acid-water (50: 8: 42) solution. Mixtures were homogenized for 1 min and sonicated two times for 20 min each time. Ice was added continuously to prevent heating. Then samples were centrifuged at 10,000 rpm. The supernatant was collected and filtered with a 0.45 μm filter (Analisis Vínicos S.L, Tomelloso, Spain). Afterwards, the pellet was re-extracted with 0.5 mL of ethanol-acetic acid-water (50: 8: 42). The filtered supernatants were combined and betalains and phenolic compounds were quantified by liquid chromatography.

Non-encapsulated surface bioactive compounds were quantified to establish encapsulation performance according to the procedure by Saénz et al. (2009). 100 mg of microparticle were mixed with 1 mL of ethanol: methanol (1: 1) solution for 1 min and filtered with a 0.45 μm filter. Betalains and phenolic compounds in the filtered solution were quantified by liquid chromatography.

The encapsulation efficiency was calculated as the net bioactive compounds (total-surface) of each encapsulation divided by the content of those same compounds in the control (E0) (Eq. (1)):

$$\text{Encapsulation efficiency}(\%) = \frac{\text{Net bioactive compounds}}{\text{Total bioactive compounds in control (E0)}} x\ 100 = \frac{(\text{Total bioactive compounds} - \text{superficial})}{\text{Total bioactive compounds in control (E0)}} x\ 100$$

Equation 1. Calculation of encapsulation efficiency.

2.9. Quantification of bioactive compounds by HPLC-DAD-ES-MS

Betalains and phenolic compounds were determined simultaneously by high-performance liquid chromatography (García-Cayuela et al., 2019). Chromatographic analyses were performed in a 1200 Series Agilent HPLC System (Agilent Technologies, Santa Clara, CA, USA) with a reverse phase C18 column (Zorbax SB-C18, 250 × 4.6 mm i.d., S-5 μm; Agilent) maintained at 25 °C. Elution solvent A consisted of 1% formic acid (v/v) in water, while solvent B was a mixture of methanol and formic acid (1%, v/v). Separation was achieved using an initial solvent composition of 15% (B) during 15 min, increased to 25% (B) within 10 min, and subsequently ramped to 50% (B) within 10 min, increased to 75% (B) in 15 min, followed by a decreased period of 15% (B) in 5 min prior to isocratic re-equilibration at 15% (B) for 10 min. The Flow rate was fixed at 0.8 mL/min and the injection volume was 20 μL. The UV–vis photodiode array detector was set at 4 wavelengths for monitoring simultaneously different compound families: a) 280 nm for phenolic acids, b) 370 nm for flavonoids, c) 480 nm for betaxanthins, and d) 535 nm for betacyanins. Additional UV/Vis spectra of each compound were recorded between 200 and 700 nm.

The HPLC-DAD was coupled on-line to a mass spectrometry detector (LCMS SQ 6120, Agilent) with an electrospray ionization (ESI) source operating in positive ion mode. Nitrogen was used both as the drying gas at a flow rate of 3 L/min and as nebulizing gas at a pressure of 20 psi. The nebulizer temperature was set at 300 °C and a potential of 3500 V was used on the capillary. Helium was used as coliseum gas and the fragmentation amplitude was 70 V. The spectra were recorded in the range *m/z* 100–1000.

The identification of the bioactive compounds was carried out by comparing the retention times, mass spectra and UV / Vis absorption with commercial, purified or semi-synthesized standards (García-Cayuela et al., 2019) and may be consulted in Supplementary Table S2. A betanin-rich extract was obtained from commercial beetroot and purified in a Sephadex L20 resin to obtain the betanin. Indicaxanthin was semi-synthesized from purified betalain by raising the pH with ammonia to obtain betalamic acid and by reacting with proline. Piscidic acid was purified by semi-preparative high-performance liquid chromatography (HPLC) from extracts of *Opuntia ficus-indica* peels. Standards for rutin, isorhamnetin glycosides, and kaempferol glycoside were provided by Sergio Serna-Saldivar's laboratory in Centro de Biotecnología FEMSA (Mexico), where these compounds were previously isolated from *Opuntia* cladodes. Hydroxybenzoic acid and quercetin were purchased from Sigma-Aldrich. To carry out the quantification, calibration curves were previously made for each of the standards, with five points in a range of 0–300 μg / mL. The content of each compound was expressed in μg / g dry weight.

2.10. HPLC-DAD-ESI-QTOF

Further mass spectrometry analyses were carried out using a maXis II LC-QTOF equipment (Bruker Daltonics, Bremen, Germany) with an ESI source and the same chromatographic conditions described above (Supplementary Table S2). The ESI-QTOF detector worked in positive ion mode, recording spectra in the range m/z 50–3000. The operating conditions were set as follows: temperature 300 °C, capillary voltage 3500 V, charging voltage 2000 V, nebulizer 2.0 bar and dry gas at 6 L/min. MS/MS study was performed by using the bbCID (Broad Banding Collision Induced Dissociation) method at 30 eV.

2.11. Simulated gastrointestinal digestion

The in vitro digestion assay of encapsulated prickly pear extracts was performed according to the standardized INFOGEST protocol (Brodkorb et al., 2019; Minekus et al., 2014). The solutions for mouth (Simulated Saliva Fluid, SSF), stomach (Simulated Gastric Fluid, SGF), and small intestinal (Simulated Duodenal Fluid, SDF) compartments were prepared according to a previous article (Eriksen, Luu, Dragsted & Arrigoni, 2017). The addition of enzymes in the preparation of digestive fluids was performed daily and moments prior to the digestive assay. After each phase (oral, gastric, and intestinal) of the simulated digestion, betalains and phenolic compounds were extracted as described previously (Section 2.3) to by quantified by liquid chromatography.

The bioaccessibility of antioxidants such as betalains and phenolic compounds were calculated as the ratio between their concentration in the intestinal fraction and their initial concentration in the fruit tissue (Equation 2).

$$\text{Bioaccessibility } (\%) = \frac{\text{antioxidant compounds intestinal phase}}{\text{Antioxidant compounds fruit tissue}} x\ 100$$

Equation 2. Bioaccessibility of antioxidants.

2.12. Statistical analysis

The statistical analysis of the results obtained was performed by analysis of variance (ANOVA) and the minimum significant differences were calculated at a significance level of $p < 0.05$. For this, the statistical program IBM SPSS Statistics version 25.0 was used. All the analyses were carried out in duplicate thanks to the biological replications carried out in each of the tests.

3. Results and discussion

3.1. Physical and physicochemical properties of microparticles

3.1.1. Moisture content

Moisture content refers to the percentage (%) of water that is bound to the microparticles after being freeze dried. Moisture content in the non-encapsulated control extract (E0) was significantly higher (12.01 ± 0.30% on a wet basis) than in the microparticles (Supplementary Fig. S2). On the other hand, no significant differences in moisture content were observed between the different microparticles, which presented an average of 7.76 ± 0.63% on a wet basis. The lower moisture content in the microparticles (when compared to the control; E0) is an indication of encapsulation efficiency. The moisture content of our microparticles (by high-speed homogenization and ultrasound) was higher than the 2.2–5.2% moisture content observed by Saénz et al. (2009) by using maltodextrin (6–30%) or inulin (3–15%) and spray drying at inlet temperature ranging from 120 to 160 °C. Otálora et al. (2015) observed similar moisture content (2.9–4.8%) by homogenizing and spray-drying prickly pear fruit extracts with maltodextrin at 170 °C inlet and 98 °C outlet air temperatures. High moisture in microparticles reduces the retention of the materials to betalains, thus decreasing the encapsulation performance over time. Low moisture content limits the ability of water to act as a plasticizer, prevents a reduction in the glass transition temperature and provides long-term stability to the product, preventing microbiological contamination.

3.1.2. Hygroscopicity

Hygroscopicity is the ability of materials to absorb atmospheric moisture. A low value is desirable to facilitate industrial scalability. In powdered fruit extracts, glucose and fructose are mainly responsible for their high hygroscopicity due to the strong interactions of the water molecule with the polar ends of these disaccharides. Encapsulated extracts showed a lower hygroscopicity of 5–14% compared to the 25% of the non-encapsulated control. Similarly, anthocyanin microparticles prepared with maltodextrin, β-cyclodextrin, whey protein isolate, and gum Arabic materials by Tao et al. (2017) showed 15% hygroscopicity by the same microencapsulation method (high-speed homogenization and ultrasound) as used in this study.

3.1.3. Stability of microparticles

Another relevant parameter for microparticles is the stability of the vitreous system and the association of vitreous behavior and chemical stability. Molecular mobility above the glass transition temperature (Tg) makes recrystallization more likely. Identifying it's Tg is important to guarantee that the system remains below this value to minimize the risk of physical instability (Craig, 2006). Hancock, Shamblin and Zografi (1995) suggest that the product be stored at least 50 °C below the Tg to ensure its stability. As shown in Fig. 1, the unencapsulated material (E0) has a glass transition temperature (Tg) of 49.5 °C, higher than those found in the maltodextrin microparticles E1 (47.90 °C) and maltodextrin and microcrystalline cellulose microparticles E2 (48.71 °C) which would be the most thermally stable. In other terms, the Tg of the microparticles additionally containing hydroxyl-propyl-methyl cellulose (E3), guar gum (E4), and xanthan gum (E5), were 43.2, 42.9, and 43.9 °C, respectively. These values are similar to those reported by Otálora et al. (2015) where the prickly pear extract encapsulated with maltodextrin showed a Tg of 40 °C. Microparticles with α and β cyclodextrin had the lowest Tg which was 40 °C and 30 °C for E6 and E7, respectively.. The Tg is related to the molecular weight of the excipients, but also to the length and structure of the polymer chain. Based on the results, the microparticles should be stored at freezing temperatures (−20 °C) to ensure their stability.

3.1.4. Microparticle morphology

Scanning electron microscopy (SEM) images (Fig. 2) reveal that the morphology of the microparticles vary as a function of the encapsulating agents. The freeze-dried whole fruit extract (E0) has an amorphous matrix with internal cavities, possibly due to the freeze-drying process (Habibi, Mahrouz & Vignon, 2009). Contrarily, all the microparticles showed a homogeneous appearance with defined spherical or filamentary shapes. Microparticles E1, E2 and E3 had a spherical morphology due to the presence of maltodextrin in their formulations. Low molecular weight sugars such as maltodextrin can act as plasticizers which reduce the contacts of the polymer chain, thus reducing the stiffness of the three-dimensional layered structure. Additionally, some glass-like components can be observed in these formulations. Several authors using homogenization and freeze-drying to create maltodextrin microparticles have described these as flake-like or resembling broken glass (Papoutsis et al., 2018; Yamashita et al., 2017) arguing that due to the low temperature involved, there is a lack of forces for breaking up the frozen liquid intro droplets (Chen, Chi & Xu, 2012). Meanwhile, microparticles E6 and E7 (whose encapsulating agents are α-cyclodextrin and β-cyclodextrin, respectively) had a larger particle size than the rest of the microparticles. This larger size could bring benefits in the encapsulation process, such as higher throughput. The least uniform microparticles were E4 and E5, whose formulation contained hydroxy-propyl-methyl-cellulose in addition to guar gum and xanthan gum, respectively. The presence of gums makes its degree of compaction more prominent due to its higher molecular weight. Furthermore, spherical structures

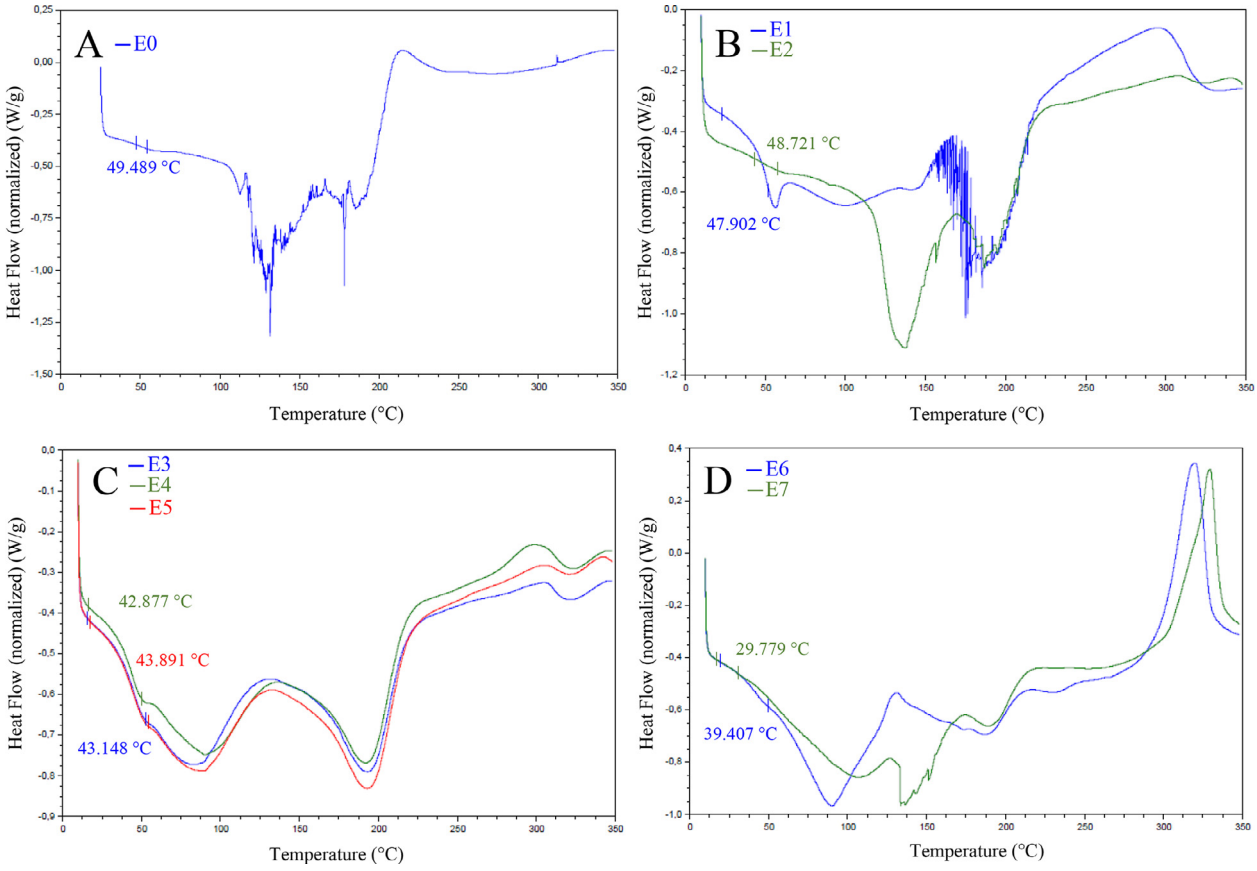

Fig. 1. Thermograms for encapsulated extracts of Spanish Morada prickly pear (*O. ficus-indica*) whole fruit. Glass transition temperature is indicated for each sample. E0, control sample without encapsulating agent; E1, with MD; E2, with MD+MCC; E3, with MD+MCC+HPMC; E4, with MD+MCC+HPMC+Guar; E5, with MD+MCC+HPMC+Xanthan; E6, with α-CD; and E7, with β-CD. Details for encapsulating agent composition are shown in Table 1.

are more difficult to distinguish in already encapsulated extracts due to the formation of a continuous matrix. We used ultrasound to create our microparticles as it positively affects drop size. A higher uniformity and smoothness are desirable to facilitate an industrial level handling of the extracts. Spherical particles can flow freely due to the absence of roughness and do not form agglomerates.

3.2. Identification of betalains and phenolic compounds by HPLC

The profile of betalains and phenolic compounds was analyzed using HPLC-DAD-ESI/MS (with ESI and QTof detectors) with absorbance detection at 280, 370, 480, and 535 nm, to identify phenolic acids, flavonoids, betaxanthin, and betacyanins, respectively. Supplementary Fig. S3 shows the simultaneous determination of these bioactive compounds. The HPLC retention times, UV/Vis spectra, and MS spectral data of the principal peaks are shown in Supplementary Table S2.

The HPLC analysis monitored at 480 and 535 nm allowed the identification of 3 betalains (1 betaxanthin and 2 betacyanins). Peak 1 (Rt = 10.47; λ_{max} at 478 nm) was identified as bx-proline (indicaxanthin), the major betaxanthin in orange and yellow prickly pear fruits. Peak 3 (Rt = 13.71; λ_{max} at 534 nm) was identified as betanin, the major betacyanin in red and purple prickly pear fruits. Peak 3 (Rt = 20.67; λ_{max} at 534 nm) was identified as isobetanin. The HPLC retention times, UV/Vis spectra, and MS spectral data matched our reference standards.

We identified 11 phenolic compounds, which included 2 phenolic acids (λ_{max} near 280 nm) and 9 flavonoids (λ_{max} near 370 nm). Peak 2 (Rt = 12.08; λ_{max} at 232 and 275 nm) was a common phenolic acid found in cactus called piscidic acid. Another phenolic acid, 4-hydroxybenzoic acid glycoside (peak 5; Rt = 32.34; λ_{max} at 274 nm), was also identified. Regarding flavonoids, 3 quercetin glycosides were identified as quercetin glycoside (QG1) (peak 6; Rt = 37.49; λ_{max} at 266 and 351 nm), quercetin glycoside (QG2) (Rt = 38.07; λ_{max} at 269 and 350 nm), and quercetin-3-rutinoside (rutin) (Rt = 37.49; λ_{max} at 266 and 351 nm). In addition, kaempferol-gluosyl-rhamnoside (KG) (peak 13; Rt = 42.65; λ_{max} at 352 nm) flavonoid was also identified. Isorhamnetin glycosides found were isorhamnetin glucosyl-rhamnosyl-rhamnoside (IG1) (peak 8; Rt = 39.06; λ_{max} at 352 nm), isorhamnetin glucosyl-rhamnosyl-pentoside (IG2) (peak 9; Rt = 39.43; λ_{max} at 352 nm), isorhamnetin glucosyl-hexosyl-hexosyl-pentoside (IG3) (peak 10; Rt = 39.71; λ_{max} at 353 nm), isorhamnetin glucosyl-pentoside (IG4) (peak 11; Rt = 40.12; λ_{max} at 299 and 352 nm), and isorhamnetin glucosyl-rhamnoside (IG5) (peak 12; Rt = 40.43; λ_{max} at 293 and 352 nm). These flavonoids were first characterized in cladodes (Antunes-Ricardo et al., 2015), while they have also been reported in *O. ficus-indica* fruits or juices (García-Cayuela et al., 2019; Gómez-Maqueo et al., 2020).

3.3. Encapsulation efficiency

The characterization of betalains, phenolic acids and individual flavonoids in the microparticles (E1-E7) and the control sample without encapsulating agents (E0) was carried out by HPLC with UV–Visible detection (diodes) and mass spectrophotometry. The encapsulation efficiency based on the content of bioactives measured by HPLC in the microparticles is shown in Table 2. The encapsulation performance provides information on the percentage of compounds bound to the excipients in each of the microparticles (E1-E7). It has been calculated by comparing the content of net encapsulated compounds with those found in the control without encapsulating materials (E0) (Supplementary Table S3).

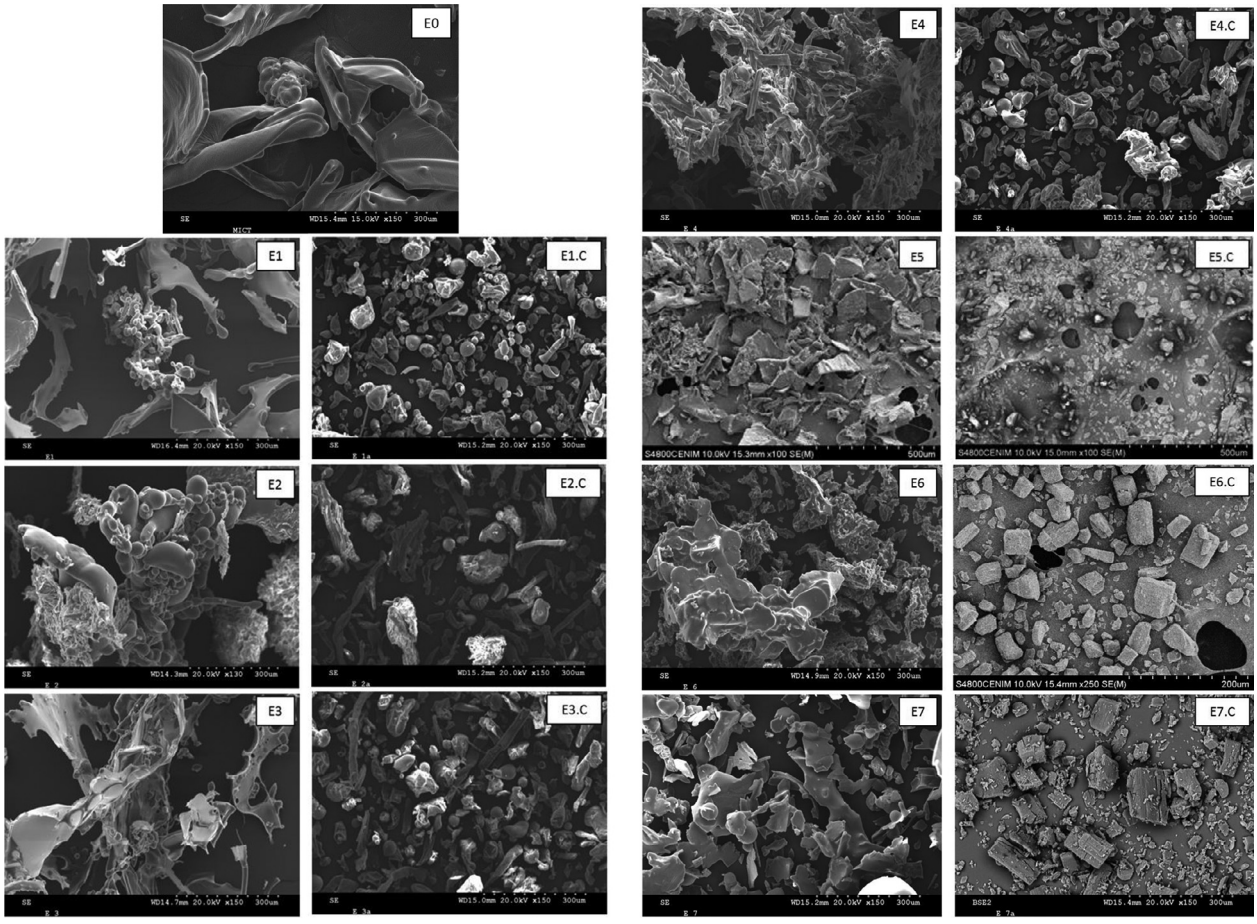

Fig. 2. Scanning electron micrographs for encapsulated extracts of Spanish Morada prickly pear (*O. ficus-indica*) whole fruit. E0, control sample without encapsulating agent; E1-E7, encapsulated extracts; E1.C-E7.C, encapsulating agents without extract used as controls. Details for encapsulating agent composition are shown in Table 1.

Table 2
Encapsulation efficiency based on the content of bioactives measured by HPLC in the microparticles (E1-E7)[1].

Bioactives	Encapsulated extracts[4]						
	E1	E2	E3	E4	E5	E6	E7
Betalains							
Indicaxanthin	68.6 ± 2.1^{a}	66.5 ± 7.8^{a}	74.9 ± 1.1^{a}	77.5 ± 1.6^{a}	76.6 ± 5.6^{a}	71.5 ± 2.6^{a}	72.3 ± 7.8^{a}
Betanin	61.8 ± 1.9^{ab}	56.8 ± 1.5^{b}	62.8 ± 0.2^{ab}	68.5 ± 4.0^{a}	64.4 ± 6.0^{a}	62.2 ± 2.9^{ab}	60.4 ± 6.5^{ab}
Isobetatin	61.5 ± 8.9^{ab}	68.6 ± 7.7^{a}	64.3 ± 3.5^{ab}	52.3 ± 0.9^{b}	63.7 ± 5.6^{ab}	69.2 ± 6.2^{a}	73.5 ± 2.4^{a}
Total betaxanthins[2]	68.6 ± 2.1^{a}	66.5 ± 7.8^{a}	74.9 ± 1.1^{a}	77.5 ± 1.6^{a}	76.6 ± 5.6^{a}	71.5 ± 2.6^{a}	72.3 ± 7.8 ^{a}
Total betacyanins[2]	61.8 ± 1.1^{a}	57.6 ± 3.7^{a}	62.9 ± 0.1^{a}	66.3 ± 4.8^{a}	64.3 ± 5.9^{a}	62.7 ± 3.1^{a}	61.4 ± 5.9^{a}
Phenolic acids							
Piscidic acid	72.6 ± 3.1^{a}	79.6 ± 7.5^{a}	79.8 ± 0.1^{a}	82.7 ± 2.3^{a}	80.6 ± 7.1^{a}	80.3 ± 0.3^{a}	80.7 ± 8.6^{a}
HBAG[3]	48.0 ± 3.9^{a}	51.2 ± 3.9^{a}	52.0 ± 0.2^{a}	53.1 ± 1.0^{a}	47.9 ± 4.2^{a}	51.6 ± 0.2^{a}	52.2 ± 5.1^{a}
Total phenolic acids[2]	71.7 ± 4.2^{a}	78.6 ± 7.3^{a}	78.9 ± 2.1^{a}	81.6 ± 2.2^{a}	79.4 ± 7.0^{a}	79.2 ± 1.3^{a}	79.7 ± 3.5^{a}
Flavonoids							
IG1[3]	63.6 ± 3.9^{a}	62.4 ± 1.9^{a}	62.9 ± 3.2^{a}	66.1 ± 3.0^{a}	64.3 ± 2.9^{a}	65.3 ± 1.9^{a}	66.5 ± 5.0^{a}
IG2[3]	69.1 ± 6.9^{a}	64.6 ± 2.1^{a}	68.8 ± 3.8^{a}	71.8 ± 1.0^{a}	67.4 ± 6.6^{a}	71.4 ± 4.3^{a}	63.7 ± 2.3^{a}
IG3[3]	46.2 ± 1.2^{f}	52.1 ± 2.3^{e}	75.2 ± 5.5^{b}	53.0 ± 2.2^{e}	84.8 ± 1.5^{a}	66.2 ± 2.8^{c}	60.0 ± 1.3^{d}
IG4[3]	68.3 ± 1.0^{ab}	65.8 ± 2.7^{ab}	64.5 ± 1.7^{b}	70.0 ± 3.3^{ab}	74.2 ± 5.3^{a}	66.7 ± 2.9^{ab}	81.7 ± 4.8^{a}
IG5[3]	71.9 ± 5.9^{a}	67.4 ± 6.6^{a}	75.3 ± 0.9^{a}	70.2 ± 1.4^{a}	74.0 ± 2.1^{a}	71.1 ± 0.3^{a}	68.5 ± 5.8^{a}
QG1[3]	40.9 ± 2.3^{a}	40.5 ± 5.1^{a}	43.4 ± 4.9^{a}	50.4 ± 7.5^{a}	46.2 ± 2.0^{a}	47.3 ± 1.7^{a}	46.5 ± 1.6^{a}
QG2[3]	53.9 ± 4.3^{ab}	59.6 ± 5.0^{ab}	55.7 ± 3.0^{ab}	55.7 ± 2.9^{ab}	49.4 ± 4.1^{b}	55.5 ± 1.3^{ab}	61.2 ± 3.4^{a}
Rutin	51.6 ± 2.6^{c}	56.3 ± 5.9^{c}	71.8 ± 1.6^{a}	52.0 ± 1.3^{c}	57.8 ± 1.7^{c}	66.7 ± 1.6^{b}	34.6 ± 1.5^{d}
KG[3]	72.1 ± 0.9^{b}	82.6 ± 1.0^{a}	77.5 ± 1.1^{b}	82.1 ± 2.6^{a}	86.9 ± 2.0^{a}	84.4 ± 1.5^{a}	86.2 ± 0.9^{a}
Total flavonoids[2]	63.0 ± 2.6^{ab}	62.4 ± 3.3^{ab}	67.7 ± 1.6^{a}	65.2 ± 1.9^{a}	67.4 ± 1.9^{a}	67.3 ± 0.5^{a}	63.2 ± 0.7^{b}

[1] Values are shown as percentage ± standard deviation respect to the bioactive content in control sample (E0). Lowercase letters indicate statistically significant differences ($p \leq 0.05$) between microspheres.

[2] Represents the percentage from the algebraic sum of the quantified bioactives by HPLC.

[3] Bioactive identities are shown in figure 4.

[4] Microspheres: E1, with MD; E2, with MD+MCC; E3, with MD+MCC+HPMC; E4, with MD+MCC+HPMC+Guar; E5, with MD+MCC+HPMC+Xanthan; E6, with α-CD; and E7, with β-CD. Details for encapsulating agent composition are shown in Table 1.

No significant differences were observed between the encapsulation performance of indicaxanthin, betacyanins and phenolic acids in the different microparticles. Indicaxanthin has an average efficiency of 72.57%, while that of betacyanins is 62.57% and 77.24% in the case of phenolic acids. On the other hand, the encapsulation performance of the flavonoids is significantly lower in the E3, E4 and E5 encapsulations (25.02% on average) than the encapsulation with α-cyclodextrin (E6) whose performance is 49%. Saénz et al. (2009) encapsulated prickly pear extracts by spray-drying with maltodextrin, and the encapsulation efficiency of betacyanins and indicaxanthin was greater than 90%. The previous information differs from that found in this study, most likely due to the effect of the encapsulation method. Meanwhile, Saénz et al. (2009) and Romano, Masi, Pucci, Oliviero and Ferranti (2017) observed that polyphenols, encapsulated with maltodextrin by spray-drying, presented encapsulation efficiency in the range of 39 to 79%. This is in line with our results for flavonoids and phenolic acids, with a mean of 31% and 77%, respectively.

3.4. *In vitro simulated gastrointestinal digestion*

The simulated gastrointestinal digestion assay was carried out with three microparticles (E2, E5 and E7) and the control (E0). The choice of microparticles was based on the characteristics observed in the thermal and morphological analysis, and on the encapsulation performance. E0 was selected as a control without encapsulating materials. E2 (encapsulated with maltodextrin and microcrystalline cellulose) was selected as it has a higher glass transition temperature than E1 as well as higher encapsulation performance and total content of bioactive compounds. Meanwhile, E5 (maltodextrin, microcrystalline cellulose, and hydroxyl-propyl-methyl-cellulose and xanthan gum) was selected as it has a higher glass transition temperature as well as higher efficiency than E3 and E4. Finally, E7 (with β-cyclodextrin) was included in the comparison as it presents similar characteristics to the previous encapsulated and had a more homogeneous particle appearance than E6 (α-cyclodextrin).

The bioactive content in encapsulated Morada *Opuntia ficus-indica* extracts after each phase of in vitro digestion is presented in Table 3. Meanwhile, Table 4 shows the bioaccessibility of individual betalains and phenolic compounds present in the different microparticles.

In the control (E0), indicaxanthin content decreased throughout the digestion and its bioaccessibility reached a value of 71%. A similar tendency was reported by Tesoriere, Fazzari, Angileri, Gentile and Livrea (2008). Encapsulated E2 (with maltodextrin, MD; and microcrystalline cellulose, MCC) and E5 (MD; MCC; HPMC, hydroxy propyl -methyl-cellulose; and xanthan gum) had the highest bioaccessibility of 99% and 84%, respectively. This demonstrated that these two microparticles may adequately protect indicaxanthin from degradation through the intestinal tract. However, encapsulation with β-cyclodextrin (E7) showed 78% indicaxanthin bioaccessibility which was close to that of the control.

Similarly, betanin in the control (E0) was also degraded when passing through the intestinal tract due to the low pH of the gastric phase. The bioaccessibility of betanin was 57% in agreement with a study by Tesoriere et al. (2008). Meanwhile, microparticles E2 and E5 had a higher bioaccessibility than the control and E7, there values were 89 and 82%, respectively. Although the encapsulation with cyclodextrin (E7) showed a lower bioaccessibility (76%), it was still higher than the control (E0). None of the microparticles increased the bioaccessibility of isobetanin, and its bioaccessibility remained around 50% in all samples. However, betanin's bioaccessibility did increase up to 92% in E2, 84% in E5 and 79% in E7 compared to 58% in the control (E0). These results could indicate that the chemical excipients used in the study have a higher binding force for betanin than for isobetanin, thus allowing greater protection against degradation during digestion. During digestion betalains can be susceptible to degradation due to the strongly acidic pH of the gastric-like environment as well as by attack of α-amylase activity of pancreatin in the intestinal-like milieu (Tesoriere et al., 2008).

Total phenolic acids (measured as the sum of the content of piscidic acid and the 4-hydroxybenzoic acid derivative) after digestion in the control (E0) had 77% bioaccessibility. Following a similar trend as for betalains, the E2 and E5 microparticles had a greater bioaccessibility than the phenolic acids, being close to 93% in both cases. In encapsulation with β-cyclodextrin (E7) the bioaccessibility of phenolic acids was slightly lower, 85%, although it is also higher than the control (E0).

Piscidic acid in the control (E0) decreased during digestion (Table 3) until reaching a bioaccessibility of 77% (Table 4). This is in accordance with a previous study by Gómez-Maqueo et al. (2020) where the bioaccessibility of piscidic acid in colored prickly pear varieties was 27–47% in pulps and 52–80% in peels. Microparticles E7 showed a slightly higher result of 85%. Meanwhile, the encapsulated E2 and E5 both had a high bioaccessibility of at least 90%, indicating greater protection of this compound. In the case of the 4-hydroxybenzoic acid derivative, in the control (E0) its bioaccessibility was 70% in agreement with previous studies (Gómez-Maqueo et al., 2020). In all microparticles the bioaccessibility was higher, standing out in the E2 and E5 encapsulations with vales of 111% and 134%, respectively. As a general rule, polymeric or glycosylated phenolic compounds need to be transformed in the intestine before they can be absorbed. This is the case for most flavonoids, which are naturally glycosylated. Flavonoid glycosides are too hydrophilic to be passively absorbed by diffusion in the small intestine. In fact, if they are deglycosylated, they can enter enterocytes by passive diffusion (Ribas-Agustí, Martín-Belloso, Soliva-Fortuny & Elez-Martínez, 2018). In other terms, the bioaccessibility of the flavonoids in the control (E0) was 51%. This is slightly lower to the bioaccessibility (73–81%) that has been reported for prickly pear mucilage (Antunes-Ricardo et al., 2015). Within the different microparticles, E5 presented a lower flavonoid encapsulation performance of approximately 65%. Its bioaccessibility of total flavonoids was the lowest due to the unstable union of these compounds with the excipients.

On the other hand, flavonoids in extracts encapsulated with maltodextrin and microcrystalline cellulose (E2) and with β-cyclodextrin (E7) showed a bioaccessibility greater than 80%. In both cases, the bioaccessibility of quercetin glycosides (QG1) increased from 25% to more than 70%, and from 93% to more than 140% for QG2.

IG1 (isoramnetin glucosyl-rhamnosyl-rhamnoside) showed a bioaccessibility of 60% in the control (E0), which increased to 85% in E2, 82% in E5 and 77% in E7. In the case of IG2 (isoramnetin glucosyl-rhamnosyl-pentoside), the bioaccessibility in the control is 62%, which increased to more than 80% in the three microparticles, standing out in E7, which increased up to 90%. IG5 (isoramnetin glucosyl-rhamnoside) shows 47% bioaccessibility and increased the same in the E2 (82%) and E7 (73%) packages, while in the E5 package it maintained similar values to the control.

4. Conclusions

The microencapsulation of prickly pear fruit extracts with different excipients positively affected their moisture content and hygroscopicity which could result in shelf-life extension. Microparticles encapsulated with cyclodextrin had the lowest moisture and hygroscopicity as well as high glass transition temperature. Microparticles which contained maltodextrin and microcrystalline cellulose in their formulation were the most thermally stable and showed spherical morphology. Indicaxanthin, phenolic acids, and betanin were the compounds with the highest encapsulation efficiency. Meanwhile, flavonoids found in prickly pear fruit extracts such as isorhamnetin, quercetin, and kaempferol glycosides showed a lower encapsulation efficiency. Microencapsulation enhanced the bioaccessibility of antioxidant compounds found in the extracts, especially in E2 (MD and MCC) and in E5 (MD, MCC, HPMC, and xanthan gum). E2 microparticle had the highest increase in the bioaccessibility of all bioactive compounds found in the extracts. Mentioned

Table 3
Bioactive content[2] in encapsulated Morada *Opuntia ficus-indica* extracts after each phase of in vitro digestion.

	E0 (control without encapsulating agents)				E2 (with MD+MCC)				E5 (with MD+MCC+HPMC+Xanthan)				E7 (with β-CD)			
	Initial	Oral	Gastric	Digesta	Initial	Oral	Gastric	Digesta	Initial	Oral	Gastric	Digesta	Initial	Oral	Gastric	Digesta
Betalains																
Indicaxanthin	179.6	166.4	143.5	127.3	122.1	121.7	121.1	120.7	137.6	122.9	113.7	115.6	129.9	119.8	106.9	102.3
	±3.3	±3.3	±3.7	±5.7	±3.5	±2.5	±9.4	±4.0	±9.9	±6.3	±5.3	±9.4	±4.0	±1.3	±3.0	±4.7
Betanin	665.3	521.9	456.9	384.0	377.6	376.2	374.7	346.6	428.4	406.1	385.1	361.9	402.0	390.1	386.4	317.4
	±10.3	±8.3	±11.0	±9.7	±10.1	±5.7	±7.9	±5.5	±9.6	±8.3	±12.8	±9.8	±9.2	±7.5	±2.2	±6.4
Isobetatin	50.6	29.4	27.3	26.1	34.8	24.9	18.5	18.8	32.2	29.5	19.7	17.1	37.2	19.6	18.3	17.7
	±8.2	±9.2	±3.3	±1.8	±3.9	±7.9	±2.1	±1.3	±2.9	±4.0	±6.3	±1.9	±1.2	±2.9	±1.3	±0.3
Total betaxanthins[2]	179.6	166.4	143.5	127.3	122.1	121.7	121.1	120.7	137.6	122.9	113.7	115.6	129.9	119.8	106.9	102.3
	±3.3	±3.3	±3.7	±5.7	±3.5	±2.5	±9.4	±4.0	±9.9	±6.3	±5.3	±9.4	±4.0	±1.3	±2.9	±4.7
Total betacyanins[2]	715.9	551.3	484.2	410.1	412.3	401.3	393.2	365.4	460.6	465.3	404.8	379.0	439.2	409.7	404.7	335.1
	±13.9	±13.1	±12.1	±10.5	±11.4	±10.4	±8.8	±6.2	±10.6	±9.9	±14.9	±10.6	±9.8	±8.6	±3.2	±6.9
Phenolic acids																
Piscidic acid (mg/g)	19.2	17.2	17.7	14.9	15.3	14.8	14.3	14.1	15.5	15.4	13.2	14.5	15.5	15.4	14.3	13.2
	±2.8	±1.2	±0.5	±1.5	±1.4	±0.4	±2.2	±1.5	±1.4	±0.1	±0.9	±1.0	±1.7	±0.2	±1.3	±1.4
HBAG[4]	743.8	552.8	529.0	518.4	381.1	382.7	355.1	357.9	558.5	556.1	537.9	533.3	388.5	403.5	365.7	383.1
	±24.1	±21.0	±8.7	±1.5	±9.1	±7.7	±8.9	±4.6	±3.3	±4.9	±10.2	±6.2	±9.7	±8.8	±17.8	±13.5
Total phenolic acids[2] (mg/g)	20.0	17.8	18.2	15.4	15.7	15.2	14.6	14.4	16.1	15.9	13.5	15.1	15.9	15.8	14.7	13.6
	±2.8	±1.3	±0.5	±1.5	±1.5	±0.4	±2.3	±1.5	±1.4	±0.1	±0.9	±0.9	±1.7	±0.2	±1.2	±1.4
Flavonoids[3]																
IG1	173.6	142.9	131.7	102.9	108.2	107.9	105.0	92.5	111.7	109.6	100.4	92.1	115.5	115.9	103.8	88.6
	±5.1	±2.7	±7.6	±5.1	±3.2	±2.1	±13.0	±11.5	±5.1	±3.0	±3.1	±4.4	±8.6	±1.6	±4.2	±5.6
IG2	135.9	123.7	121.8	84.81	87.8	88.3	89.2	75.2	91.7	94.2	89.4	76.5	86.8	89.9	86.3	77.4
	±14.4	±8.4	±3.8	±0.7	±2.8	±9.0	±8.7	±4.7	±8.9	±7.2	±5.2	±5.2	±3.1	±5.9	±1.9	±5.4
IG3	46.2	26.4	21.6	16.4	25.0	25.9	24.7	24.9	39.2	37.6	27.3	19.9	27.7	25.5	20.1	15.4
	±5.4	±1.5	±1.9	±3.1	±1.1	±1.5	±2.5	3 ± 8.3	±6.9	±4.4	±4.5	±1.6	±5.8	±3.1	±2.7	±1.9
IG4	81.5	78.3	63.3	36.2	53.7	57.9	54.9	44.4	60.5	56.8	49.1	31.9	66.6	65.1	60.9	43.3
	±8.7	±6.1	±5.8	±0.2	±2.2	±4.9	±7.0	±4.2	±4.3	±3.8	±8.7	±2.1	±3.9	±2.9	±0.1	±5.2
IG5	200.3	190.8	150.1	94.3	135.0	127.2	119.2	110.6	148.3	146.8	124.5	76.5	137.3	139.3	120.9	96.8
	±2.8	±5.0	±6.5	±1.5	±3.2	±4.3	±5.7	±2.0	±4.3	±9.7	±7.9	±7.4	±1.5	±7.9	±7.7	±3.2
QG1	48.5	24.4	29.6	12.6	19.7	19.9	19.5	19.1	22.4	17.7	16.0	10.6	22.6	16.6	16.2	15.8
	±2.1	±1.2	±5.4	±1.6	±2.5	±0.5	±2.2	±5.3	±0.9	±0.3	±1.5	±2.0	±0.8	±1.2	±3.7	±0.6
QG2	47.8	37.1	33.3	24.8	28.5	26.8	28.0	23.8	23.6	24.3	21.1	21.4	29.3	27.3	23.0	17.4
	±2.7	±0.5	±2.3	±2.2	±1.4	±0.2	±7.2	±3.6	±3.9	±0.3	±1.5	±5.5	±1.6	±1.5	±5.9	±5.5
Rutin	107.6	77.9	58.6	32.3	60.6	44.2	39.4	38.8	62.2	58.4	57.3	29.8	37.2	37.9	36.3	35.4
	±2.5	±3.7	±1.5	±2.2	±6.4	±5.1	±3.4	±5.9	±2.1	±3.9	±7.1	±0.9	±.1.6	±1.8	±2.8	±2.6
KG	31.5	28.6	22.8	19.4	32.6	31.1	29.3	28.7	42.3	42.6	26.7	24.1	34.1	34.0	29.2	25.8
	±0.1	±4.0	±1.3	±1.8	±0.4	±0.8	±3.9	±5.8	±3.6	±0.6	±0.3	±4.7	±0.4	±6.6	±3.1	±2.4
Total flavonoids[2]	841.5	701.4	609.9	404.0	518.4	498.2	479.8	429.4	559.5	545.2	485.1	358.7	523.0	517.6	467.5	390.1
	±19.1	±13.2	±13.8	±7.3	±9.2	±12.5	±20.4	±30.8	±15.0	±14.3	±15.8	±12.9	±11.7	±13.0	±12.4	±12.0

[1]Results are expressed as the mean ± standard deviation in μg/g dry weight (dw), except for piscidic acid and total phenolic acids (mg/g dw).
[2] Represents the algebraic sum of the quantified bioactives by HPLC.
[3] Bioactive identities are shown in Table S2 (Supplementary material).Details for encapsulating agent composition are shown in Table 1.

Table 4
Bioaccessibility[1] (%) of encapsulated Morada *Opuntia ficus-indica* extracts.

	E0 (control without encapsulating agents)	E2 (with MD+MCC)	E5 (with MD+MCC+HPMC+Xanthan)	E7 (with β-CD)
Betalains				
Indicaxanthin	70.7 ± 3.2^{c}	98.9 ± 1.6^{a}	84.0 ± 6.8^{b}	78.7 ± 3.6^{b}
Betanin	57.7 ± 4.6^{c}	91.8 ± 6.8^{a}	84.5 ± 9.3^{a}	79.0 ± 4.1^{b}
Isobetatin	51.6 ± 3.5^{a}	54.2 ± 3.7^{a}	50.0 ± 5.8^{a}	47.5 ± 0.7^{a}
Total betaxanthins	70.9 ± 3.2^{c}	98.9 ± 1.6^{a}	84.0 ± 6.8^{b}	78.7 ± 3.6^{b}
Total betacyanins	57.3 ± 4.0^{c}	88.6 ± 6.5^{a}	82.3 ± 9.1^{a}	76.3 ± 3.8^{b}
Phenolic acids				
Piscidic acid	77.5 ± 3.7^{b}	91.9 ± 3.7^{a}	93.8 ± 3.1^{a}	84.9 ± 4.0^{ab}
HBAG[2]	69.7 ± 0.2^{b}	93.9 ± 10.6^{a}	95.5 ± 7.2^{a}	98.6 ± 3.5^{a}
Total phenolic acids	77.2 ± 3.4^{b}	92.4 ± 3.4^{a}	94.7 ± 3.1^{a}	85.2 ± 3.8^{ab}
Flavonoids[2]				
IG1	59.2 ± 2.9^{b}	85.5 ± 10.6^{a}	82.4 ± 3.9^{a}	76.7 ± 4.8^{a}
IG2	62.4 ± 0.5^{b}	85.7 ± 5.8^{a}	83.5 ± 5.6^{a}	89.4 ± 7.7^{a}
IG3	35.4 ± 6.7^{c}	99.7 ± 9.3^{a}	50.9 ± 4.0^{b}	55.6 ± 6.7^{b}
IG4	45.0 ± 0.3^{d}	82.8 ± 6.8^{a}	52.8 ± 3.5^{c}	65.0 ± 8.7^{b}
IG5	47.1 ± 0.7^{c}	82.0 ± 3.5^{a}	51.6 ± 5.0^{c}	73.0 ± 3.5^{b}
QG1	25.7 ± 3.2^{d}	97.3 ± 7.8^{a}	47.2 ± 8.8^{c}	70.2 ± 2.5^{b}
QG2	51.8 ± 4.7^{b}	83.6 ± 5.5^{a}	90.5 ± 8.3^{a}	59.4 ± 7.1^{b}
Rutin	30.0 ± 2.0^{d}	64.0 ± 9.2^{b}	47.9 ± 3.4^{c}	95.2 ± 6.9^{a}
KG	61.7 ± 2.6^{c}	88.1 ± 5.8^{a}	56.9 ± 6.2^{c}	75.8 ± 3.7^{b}
Total flavonoids	48.0 ± 1.6^{d}	82.8 ± 3.3^{a}	64.1 ± 5.2^{c}	74.6 ± 3.0^{b}

[1] Bioaccessibility as percentage ± standard deviation. Lowercase letters indicate statistically significant differences ($p \leq 0.05$) between encapsulates and control.
[2] Bioactive identities are shown in Table S2 (Supplementary Material).Details for encapsulating agent composition are shown in Table 1.

microparticle has a homogeneous structure and higher glass transition temperature which makes it a promising formulation to provide stability during its shelf life and subsequent use. Microencapsulation with maltodextrin and microcrystalline cellulose (E2) can be considered the best formulation to prolong the stability of the bioactive compounds of the extracts of prickly pear and to increase it their bioaccessibility. Therefore, it may have great potential for the design of health-promoting foods. Future studies on the behavior of encapsulation in food matrices and storage stability are recommended.

Ethical statement

The research conducted for the manuscript "Analysis of Hydrocolloid Excipients for Controlled Delivery of High-Value Microencapsulated Prickly Pear Extracts" did not contemplate any study in humans and animals

Supplementary Material

Figure S1. Photographs of Spanish *Morada* prickly pear (*Opuntia ficus-indica*) variety.

Figure S2. Water content (A) and hygroscopicity (B) of encapsulated extracts from Spanish Morada prickly pear (*O. ficus-indica*) whole fruit.

Figure S3. C18 HPLC chromatograms obtained from Spanish Morada prickly pear (*O. ficus-indica*) whole fruit extract at 480 nm (betaxanthins; orange color), 535 nm (betacyanins; red color), 370 nm (flavonoids; green color), and 280 nm (phenolic acids; blue color).

Table S1. Physical and physico-chemical characteristics in Spanish Morada prickly pear (*Opuntia ficus-indica*) tissues.

Table S2. HPLC retention times, UV/Vis spectra, and MS spectral data of betalains and phenolics from Spanish Morada prickly pear (*Opuntia ficus-indica*).

Table S3. Total, superficial, and net content of bioactive compounds (μg/g dw) in micromicroparticles measured by HPLC.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRediT authorship contribution statement

Ana Fernández-Repetto: Data curation, Formal analysis, Methodology, Validation, Writing – review & editing. **Andrea Gómez-Maqueo:** Formal analysis, Visualization, Writing – original draft, Writing – review & editing. **Tomás García-Cayuela:** Formal analysis, Supervision, Validation, Writing – review & editing. **Daniel Guajardo-Flores:** Methodology, Writing – review & editing. **M. Pilar Cano:** Conceptualization, Funding acquisition, Project administration, Resources, Supervision, Visualization, Writing – review & editing.

Data Availability

No data was used for the research described in the article.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.fhfh.2023.100115.

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FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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Review

An Analysis of the Plant- and Animal-Based Hydrocolloids as Byproducts of the Food Industry

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Abstract: Hydrocolloids are naturally occurring polysaccharides or proteins, which are used to gelatinize, modify texture, and thicken food products, and are also utilized in edible films and drug capsule production. Moreover, several hydrocolloids are known to have a positive impact on human health, including prebiotics rich in bioactive compounds. In this paper, plant-derived hydrocolloids from arrowroot (*Maranta arundinacea*), kuzu (*Pueraria montana var lobata*), Sassafras tree (*Sassafras albidum*) leaves, sugarcane, acorn, and animal-derived gelatin have been reviewed. Hydrocolloid processing, utilization, physicochemical activities, composition, and health benefits have been described. The food industry generates waste such as plant parts, fibers, residue, scales, bones, fins, feathers, or skin, which are often discarded back into the environment, polluting it or into landfills, where they provide no use and generate transport and storage costs. Food industry waste frequently contains useful compounds, which can yield additional income if acquired, thus decreasing the environmental pollution. Despite conventional manufacturing, the aforementioned hydrocolloids can be recycled as byproducts, which not only minimizes waste, lowers transportation and storage expenses, and boosts revenue, but also enables the production of novel, functional, and healthy food additives for the food industry worldwide.

Keywords: food byproducts; food processing; waste material; novel hydrocolloids; waste management

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1. Introduction

The food industry produces many by-products worldwide. Approximately 38% of waste comes from food processing [1] specifically approximately 20% from meat, fish, and poultry, approximately 4% from dairy (mainly whey—50 million m^3 yearly), 33% from oil crops, and 35% from fruits, vegetables, and tubers industries [2]. Torres-León et al., [3] claim that waste from fruit processing exceeds 50%—namely bagasse, peels, trimmings, stems, shells, bran, and seeds. A lot of seemingly useless solids and liquids come from plant-based food manufacturers. Food waste can cause environmental issues and generate additional management, storage, and processing costs. Food byproducts are often processed into fodder. Such fodder consists of cereal industry waste, such as rice bran, maize and wheat seeds, husks, hull, banana peels, or feathers. Byproducts not suitable for animal feed, such as onion peels and roots or excess banana peels, are being disposed of [4]. However, many food byproducts can be used instead of discarded. They contain valuable polysaccharides—dietary fiber fractions pectin, chitosan, cellulose, hemicellulose, lignin, and gums; proteins, e.g., single-cell protein of yeast, proteins obtained from de-oiled sunflower press cake, for example, β-lactoglobulin, α-lactalbumin, immunoglobulin, bovine serum albumin, lactoferrin, and lactoperoxidase; lipids with high levels of unsaturated fatty acids; ω-3 PUFAs; natural colorants—apple pigments, anthocyanin-based pigments; bioactive compounds such as citric and linoleic acids, tocopherols, δ-Tocotrienolfunctions,

dihydrochalcones, flavanols, polyphenols, ascorbic and phenolic acids, isorhamnetin-O-(di-deoxyhexosyl-hexoside); or hydrocolloids, e.g., starches, glucosides, proteins, gums, and fiber [1,5]. The addition of these compounds may modify the structure of other food products desirably, contribute to functional value, and provide additional income for the industry. Some of the most valuable byproducts, considering texture modification, are starches and gelatin [6].

Starches are renewable polysaccharides that naturally form in most plants, serving a nutritional backup purpose. These biopolymers may be found in plants' rhizomes, branches, fruits, seeds, and tubers. Starch's two separate components are amylose and amylopectin [7]. Both compounds contain D-glucose chains; however, they are connected in various ways. Amylose consists of unbranched glucose units linked with α (1–4) glycosidic bonds. Amylopectin also consists of glucose, but it is heavily branched, and the units are linked with α (1–4) glycosidic and α (1–6) glycosidic bonds. Native starches consist of about 10–40% amylose and 70–90% amylopectin. The ratio of those two polysaccharides is unique to individual starches and therefore is responsible for starch's properties [8]. The weight, shape, and scale of amylose and amylopectin molecules define differences in pasting, retrogradation, rheology, and viscoelastic properties [9]. Exposing starch granules to a specific temperature and moisture makes its structure undergo several changes such as swelling by absorbing water, decreasing the level of crystallinity by amylopectin double helix dissociation, elution of amylose into the aqueous phase, and fracturing starch granules. These changes are known as starch gelatinization [10]. The reverse process, where amylose and amylopectin partially regain their ordered structure, is termed starch retrogradation [11]. Starches derived from tubers and roots require a relatively low temperature to gelatinize, the process is quick, and granules swell uniformly [12]. Moreover, root and tuber starches show a higher viscosity profile and paste clarity than grain starches, yet they tend to retrograde easily. Amylose to amylopectin ratio is the reason for these unique physicochemical properties. Almost all of these starches show B-type X-ray patterns [13].

Gelatin is a protein primarily obtained from the animal industry byproducts—pig skin, bovine and porcine cartilage, bones, and hides during partial hydrolysis of collagen. Collagen is the most common structural protein in animals' bodies, making up about 30% of all proteins. Animal species majorly influence gelatin properties and tissue types. They are obtained from [14,15]. There are two main uses of gelatin in the food industry: to modify the texture by water binding, providing creaminess and foam, fining [16], gelling, stabilizing, emulsifying, altering the viscosity, or to produce packaging films and coatings, which inhibit the environmental impact on food, prolonging the shelf life. Moreover, the addition of gelatin affects the aroma of food products. An increase in viscosity impedes volatile aroma compound penetration from the inside to the outside of a food product [15]. In addition, the firmer the gelatin gel, the harder it is for aroma substances to be released [17].

This work focuses on the novel utilization of plant and animal food industry byproducts. The byproducts' physicochemical characteristics, health-promoting benefits, and use as hydrocolloids have been discussed. It is important to establish how many valuable compounds are present in the seemingly useless waste. This way, such ingredients can be identified and extracted or obtained, providing additional income, reducing storage and transportation costs, and mitigating harmful impacts on the environment. The information gathered in this work aims to clarify the topic of novel hydrocolloids from byproducts and show their use in food technology.

2. Description of Arrowroot, Kuzu, Sassafras, Sugarcane, Acorns, and Gelatin Byproducts Utilization

2.1. Arrowroot

Arrowroot *(Maranta arundinacea)*, also known as sago banban, sago rare, sago andrawa, sagu, Patat, arut, jelarut, irut, larut, labia walanta, or hudasula [18], is a native plant to the West Indies [19], Indonesia [20], or tropical regions of South America [21]. The physiochemical properties of arrowroot from various sources are quite similar [22]. Arrowroot can reach

0.9–1.5 m in height [23]. Its flowers are white, and its leaves are big, green, and 10–20 cm long. Rhizomes are fleshy, cylindrical, and tuberous with a width of 2.5–3 cm and 20–40 cm long [24]. The plant's roots are long and abundant in fibers [25]. Its promising properties were introduced worldwide over colonization times, then the export of starch from tubers and rhizomes outside India began. Arrowroot is commonly cultivated in the Philippines as a perennial crop and used in various bakery products, e.g., Spanish shortbread *polvoron* or pancake topped with grated, young coconut flesh-*saludsod*. The direct consumption of arrowroot rhizome by humans is unclear, because of its very fibrous texture [26]. Arrowroot starch is used instead of many grain flours because of no gluten content [19] and is safe for people with celiac disease, gluten intolerance, and FODMAP (fermentable, oligosaccharides, disaccharides, monosaccharides, and polyols) sensitivity [21]. Three suitable cultivars are cultivated in Brazil: common, creole, and banana [27]. Arrowroot is mainly used for starch extraction, shown in Figure 1, because of its high content in plant rhizomes [28]; however, plant fibers also found utilization as packaging and tissue paper [29]. Arrowroot starch may also produce edible films [30]. Considering various purposes of arrowroot, in some fields such as fiber gathering or pro-health substances extraction, the starch is a byproduct that can be reused.

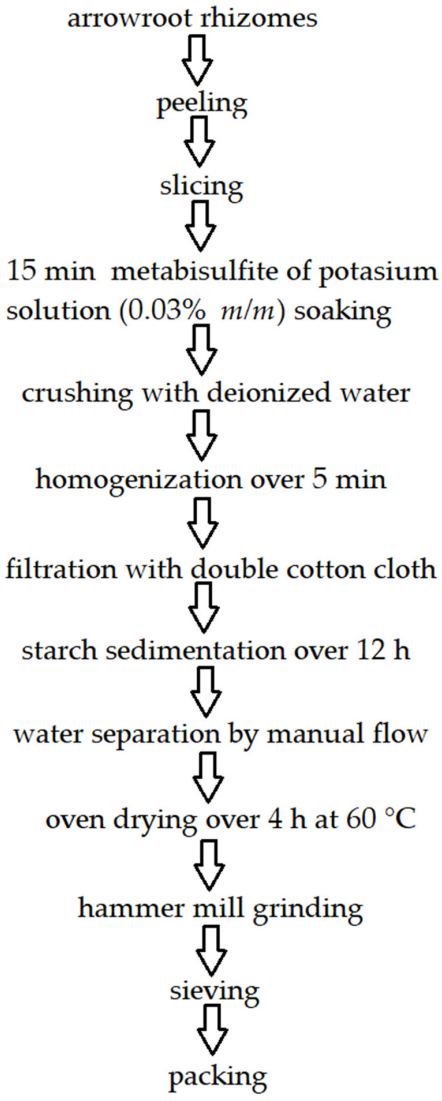

Figure 1. Arrowroot starch extraction. Own figure based on Ref. [29]. 2022, Tarique et al.

The plant's rhizome starch composition is presented in Tables 1 and 2.

Table 1. Arrowroot rhizome starch composition. N/E—not evaluated.

Compound	Moisture (%)	Fat (%)	Protein (%)	Carbohydrates (%)	Ash (%)	Soluble Fiber (%)	Insoluble Fiber (%)	Reference
Starch	11.90	0.84	0.14	rest	0.58	5.00	8.70	[31]
	15.24	0.01	0.40	83.91	0.33	N/E	N/E	[30]
	10.2	N/E	0.6	84.2	N/E	N/E	N/E	[32]
	7.06	1.43	3.75	80.77	3.60	3.96		[33]

Table 2. Amylose and amylopectin composition of arrowroot rhizome starch. N/E—not evaluated.

Polysaccharide	Amylose (%)	Amylopectin (%)	Reference
Starch	21.9	62.3	[32]
	22	N/E	[34]
	19.0–19.9	N/E	[35]
	15.21	84.79	[36]
	24.8	N/E	[37]
	>40	N/E	[38]
	20	80	[18]

The likely reason for the differences in the starch composition is the plant's age. Extraction of amylaceous fractions carried out on 12- and 14-month plants indicated an increase in amylose content from 17.9 to 20.0%, respectively. Moreover, starch granule size increased with the plant's development. On the other hand, the values of viscosity (peak, breakdown, final, and the tendency of retrogradation) decreased as the plant got older [39]. A large amount of amylose, such as 20–30%, is beneficial in gelatinization, lowering the energy required to start the process. Starches with higher amylose content have fewer crystalline regions and lower gelatinization temperatures [21].

The granules' size is 7–16 µm. Arrowroot starch exhibits a high purity of over 99% [12]. However, Guilherme et al. [26] indicated that arrowroot starch has a high amount of carboxylic acid, suggesting contamination problems and possible unwanted fermentation [40,41]. Microscopic analysis of arrowroot starch showed that granules are circular, ellipsoid, and oval, and their sizes vary [30]. Starch gelatinizes at 63.94 °C and has a B-type crystalline structure [38].

Arrowroot starch exhibits significant thickening, stabilizing [29], and shear thinning properties and may be used as a fat replacer in food [42]. Cassava and potato starches may be fortified with arrowroot starch, to increase the final gel's stability [12]. Since arrowroot starch is tuber-derived, its granules swell fast and evenly and have a high viscosity profile, surpassing grain starches [12,43]. Numerous physicochemical properties suggest that arrowroot can be mixed with other starches, e.g., to improve resistance to retrogradation and thermal and freeze-thaw stability of the whole composite [12]. Arrowroot starch is found in biscuit, cake, pudding, oatmeal, pie filling [29], soup, candy, condiments, pudding, and ice cream production. Moreover, it can replace wheat flour being a safe alternative for people with celiac disease [18]. Arrowroot starch appears to be a good ingredient for the extrusion process, exhibiting a high expansion ratio and low bulk density in the final product. The extrusion process makes arrowroot starch absorb more water and oil, which is a common phenomenon among starches. Products with extruded arrowroot starch exhibit desirable texture and color. Moreover, the extrusion process lowers the digestibility of the arrowroot starch, by making it more resistant, compared to native starch, which is sought after by people on low-calorie diets [44]. Arrowroot starch is a novel material for the walls of microcapsules. Arrowroot-enriched microcapsules indicated sufficient oxidative stability, shelf life, encapsulation efficiency, low water activity (0.05–0.23), and were hermetic.

Moreover, arrowroot starch acted as a cryoprotectant during freeze-drying [45]. Since Arrowroot starch exhibits antioxidative properties [21], it is expected to inhibit oxidation of lipids, thus may be used to prolong the shelf life of products that contain fats such as biscuits, pastries, margarine, a plant-based mayonnaise, and so on [46].

Despite various advantages, native arrowroot starch has some industrial limitations. Due to significant viscosity and discord with some hydrophobic polymers, it has finite solubility and unsatisfactory processability. Due to the immensely low phosphorus content (4.6 nmol mg^{-1}), its impact on arrowroot starch is insignificant, limiting some gel and paste behavior modifications [22]. To use the full potential of arrowroot starch, it needs modifications to improve its hydrophobicity, crystallinity, and stability to enzymatic and thermal degradation [47]. Gamma radiation treatment on arrowroot starch resulted in increased breakdown value, pointing to the low stability of the starch granules and low setback value, indicating high resistance to retrogradation thus, suggesting the arrowroot starch utilization for cold and frozen food products [48].

2.2. *Kuzu*

Kuzu, also known as kudzu, kudzuvine, kudsu, wa yaka, aka, nepalem, Japanese arrowroot, kudzu comun (Spanish), vigne japonaise (French) and kopoubohne (German) [49] is a bulbous, climbing shrub in the *Fabaceae* family of *Pueraria* genus native to Asia (China, Japan, Korea, Thailand, Vietnam, and Taiwan) and Malesia (Indonesia, Malaysia, Papua New Guinea, and the Philippines). The most popular kuzu variety is *Pueraria montana var lobata* [50]. Eastern Asians have been using kuzu for several years to create functional properties of food. Starch extracted from kuzu can form a clear, colorless, high-strength gel [8]. Kuzu starch is used as a food stabilizer, microencapsulated wall material, raw material for edible films, texture modifier, and emulsifier [51]. However, emulsifying properties were proven poor compared to protein or surfactants [52]. This plant has also been used as fodder and has various medical purposes [53,54]. Kuzu starch is also used in bioplastic production, constituting nontoxic, biodegradable, transparent, slightly reddish/yellowish material [55]. Kuzu root contains oleanene-type triterpenes and triterpenoid glucosides, including kudzusaponin, kudzusapogenol, and soyasapogenol [56–58] fragrant components namely methyl palmitate, methyl stearate, 2-methoxyethyl acetate, acetyl carbinol, and butanoic acid responsible for gently sweet and fruity-wine aroma [59] and a minimal amount of minor constituents such as 5-methylhydrantoin, tuberosin, choline chloride, acetylcholine chloride, D-mannitol, glycerol 1-monotetracosanoate [60], eicosanoic acid, hexadecanoic acid, tetracosanoid acid-2,3-dihydroxypropyl ester [61], diacetonamine, and D-(+)-pinitol [62].

A common method of isolating kuzu starch is the standard precipitation method, shown in Figure 2. Since starch and isoflavones have low water solubility, they coexist after precipitation [63] (C_2H_5OH).

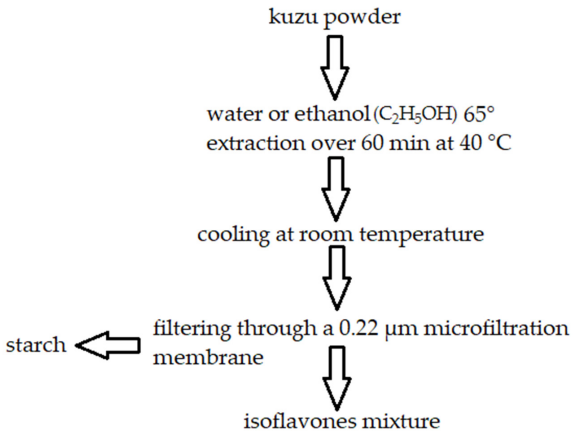

Figure 2. Extraction of kuzu starch and isoflavones. In this method, starch is a by-product. Own figure based on Ref. [64]. 2022, Mocan et al.

The kuzu starch composition has been presented in Table 3.

Table 3. Composition of kuzu starch isolated from its roots.

Ingredient	Starch as Dry Basis (%) *w*/*w*	Amylose (%)	Amylopectin (%)	Reference
root	51.6	N/E	N/E	[65]
	15–35	19–24	20.5	[50]
	N/E	22.2–23.34	N/E	[66]

Studies indicate that kuzu has C-type starch [67–69]. However, kuzu starch obtained in Vietnam was of A-type and in Korea—B-type [70]. The differences might be due to the genotype and growing conditions [66]. The average degree of polymerization (DPn) of kuzu starch amylose is 1905 and of amylopectin—2017. Amylopectin's average chain length (CL) is 21, shorter than that of amylose, 151. However, the mean number of chains per molecule (NC) of amylose is 12.6, while that of amylopectin is 96.4 [68]. According to Van Hung and Morita [71], the region where kuzu is cultivated influences DPn, CL, and NC. The largest DPn and NC of amylose and amylopectin is established for kuzu from Vietnam. The longest CL of amylopectin molecules (30) was found in kuzu starch from Japan. The highest CL of amylose (236) exhibited starch from Korea. The composition of isoflavones varies depending on the cultivars, growing regions, isolating techniques, plant's growth phase [72], where starch was obtained, and if the process was carried out commercially or at home. The highest concentration of daidzein and daidzin was found in Korea—16.41 mg/100 g starch, whereas starch from Japan had 2.18 mg of daidzein per 100 g of starch [71]. Starch is located in the plant's roots [73]. Starch manufactured commercially has no daidzin, genistein, genistin, or puerarin. Daidzein was present with a concentration of 0.011 mg/g dry basis. In the homemade sample, all the mentioned isoflavones were present with a total concentration of 8.277 mg/g dry basis [74]. Given the medical use of the mentioned bioactive compounds, effective purification and separation from the starch are essential. In the aftermath, starch is considered a byproduct, which should not be discarded due to its significant value and may be used in the food industry.

Kuzu starch exhibits a high lightness value (L*) of 93.34, which makes it a desirable product [67,75]. Granules are irregular, polygonal, spherical, and hemispherical and have a smooth surface without cracks [76] and a diameter of 3 to 23 µm. On average, the amylopectin molecule weight is 2.05 × 108 Da, and of amylose, it is 1.89 × 106 Da [69]. The starch has a polysaccharide structure with a high pasting temperature of 70–76 °C. It is rich in micronutrients such as phosphorus, iron, and calcium, providing a valuable alternative to thickening and binding agents such as gelatin [77]. Kuzu starch is used to manufacture edible films, stabilize emulsions, encapsulate oxidizable functional substances, modify the texture of food, and develop functional foods [78–80]. Native kuzu starch is used worldwide as a pro-health ingredient in foods such as nutritional powders, beverages, noodles, or vermicelli [81]. The functional properties include transparency, solubility, swelling power, freeze-thaw stability, gelatinization, retrogradation, pasting property, dynamic rheological property, and in vitro digestion [66]. Kuzu starch exhibits a transparency of 50.6 [82], indicating high phosphorus content [83]. The solubility is 8.55%, and swelling power is 3.95% at 50 °C. The increase in temperature to 90 °C caused an increase in solubility and swelling power—the reason being the loosening of chemical bonds in starch granules. Native kuzu starch has poor syneresis resistance. After five freeze-thaw cycles, the percentage of syneresis increased from 12% to 52.56% [84]. Starch content (*w*/*v*) affects gel structure. 0.1% concentration makes the gel resemble homogeneous, stubby, and curved strands, while 2%—thick masses and entangled aggregates. Storing gels at 4 °C for a week caused the gels to be retrograde, creating an opalescent surface and, at a concentration of 2%, fibrous clusters [85]. To establish kuzu starch thermal properties, differential scanning calorimetry (DSC) is mainly used [66]. Native kuzu starch shows higher gelatinization temperatures and enthalpy than retrograded kuzu starch due to fewer

crystalline regions [69]. Inappropriate chain length hinders the retrogradation of starch molecules. The best length for starch to retrograde is 14–24 degrees of polymerization [86]. Most molecules of kuzu starch have a degree of polymerization ranging from 13 to 24, making kuzu starch a starch of a high retrogradation degree (RD)—44.4% [69]. However, kuzu starch RD is higher the lower the temperature is. The reason is starch molecules diffusion and lower nucleation of the molecules. Additionally, incorporating saccharides or sodium chloride may alter kuzu starch RD based on its storage temperature [87]. To prevent starch retrogradation, tea polyphenols and catechins might be applied [88]. Compared to potato starch, canna starch, fern starch, and adzuki bean starch, kuzu starch exhibits the greatest pasting temperature because of its little starch granules, limiting swelling capacity at high temperatures [67,69]. Kuzu starch pasting abilities might be modified by adding other hydrocolloids, e.g., xanthan gum and soluble soybean polysaccharide. The reason is interactions with leached amylose or amylopectin from starch granules [66]. Kuzu starch exhibits shear thinning behavior when the content is not less than its Ce [85]. As kuzu starch concentration increases, so do the storage modulus (G′), loss modulus (G″), and shear viscosity. Additionally, adding sodium chloride, sucrose, and maltodextrin can modify the rheological properties [89]. Regarding the application, kuzu starch should be treated differently. The best conditions to prepare kuzu starch pastes for application in the food industry are temperatures of 80 °C and 15 min time; for the pharmaceutical industry—95 °C and 75 min; for the cosmetic industry—80 °C and 30 min [90].

Raw kuzu starch shows moderate in vitro digestibility. After 120 min of exposure to digestive enzymes, many eyelets were noticed on the surface of kuzu starch granules. Kuzu starch exhibits a rapidly digestible starch (RDS) content of 5.66%, slowly digestible starch (SDS)—of 25.88%, and 68.46% of resistant starch (RS). Such properties suggest that kuzu starch might be used as a functional ingredient for lowering the glycemic index in food. However, after gelatinization, RS lowered to 11.38% because of heat-damaging crystalline regions, making starch chains easier for amylase to interact with [91,92]. The percentages of SDS and RS can be raised by heating them for 1–24 h at the temperature of 50 °C causing amylose-amylose and amylose-amylopectin interactions to amplify, probably making starch chains "temper" and more resistant to enzymes [15,93]. Another method of limiting kuzu starch digestibility is a fortification with xanthan gum (1–2% w/w). Xanthan gum tends to adsorb on the starch granules, providing a defense against digestive enzymes [94].

Natural extracts present in kuzu flour limit the increase in crystallinity and recrystallization of starch [8]. There is inaccuracy in the case of the kuzu starch level of crystallinity. Starch from Vietnam exhibits a degree of crystallinity of 38.6%, Japan—35.9%, and Korea—35.7% [70]. According to Wang et al. [68], relative crystallinity is at a level of 35.25%. Reddy et al. [67] suggest 23.45% confirmed by X-ray diffraction analysis. The most likely reasons for these contractions are the kinds of cultivars, sample growing conditions, or quantification methods [95].

Most isoflavones are destroyed while starch is extracted from the roots. Thus, another isolation method is advised [66].

Native kuzu starch has some industrial limitations, such as low solubility or low stability; thus, some structural modifications might be needed [66]. Kuzu starch may be modified physically, including annealing or extrusion treatment [96,97]. Annealing involves heating the starch granules to a temperature between the glass transition and the initial gelatinization for a given time in an aqueous environment [98,99]. Heating at 50 °C for 1–9 days does not change the C-type of kuzu starch, but the ratio of B-type polymorphs grows compared to non-modified starch. What is more, annealing caused an increase in gelatinization temperatures, enthalpy, pasting temperature, and prior stability and a reduction in pasting viscosities, granular swelling power, and solubility because of internal rearrangement and interplay of starch particles [97]. Furthermore, annealed kuzu starch might have health benefits for individuals who require lowered digestibility of food—the SDS and RS percentage in tempered kuzu starch is 10% higher compared to native kuzu starch. The reason is amplifying interplay between amylose and amylose or

amylopectin after tempering [93]. Annealed kuzu starch might find use in manufacturing canned and frozen foods because of lowered swelling power, solubility and increased paste stability and crystallinity. Moreover, lowered granular swelling and amylose leaching and increased heat and shear stability suggest utilization in noodle production [97]. Enzymatic modification utilizing α-amylase and transglucosidase may be used to address the inferior pasting qualities and propensity for retrogradation of kuzu starch. This way, modified kuzu starch exhibits higher solubility, paste clarity, gelatinization temperature, and lower viscosity due to slower retrogradation [81]. Another method of kuzu starch modification is esterification with octenyl succinic anhydride. Such treatment improves emulsification properties, viscosity, and granule swelling compared to native kuzu starch [52]. According to Chen et al., [98], it is possible to cross-link kuzu starch using sodium trimetaphosphate, which grants the starch more desirable thermal, freeze-thaw, and retrogradation stability, and higher viscosity. This may prove useful in jelly, jam, gummy candy, mayonnaise, preserves, sauces, instant meals, and pastry production. Kuzu starch modified with dodecenyl succinic anhydride exhibits larger granule size, higher viscosity, lower gelatinization temperature and enthalpy value. Starch modified this way shows better emulsification properties compared to native starch and may be used as a wall stabilizer in the production of microcapsules filled with oil or bioactive compounds [99].

2.3. Filé Powder

Sassafras albidum is a deciduous tree species native to North America. The tree's root was used in folk medicine and as a spice in soft drinks such as root beer but was prohibited in 1960 because of an unsafe amount of carcinogenic alkaloid safrole (4-allyl-1,2-methylenedioxybenzene). In the present day, safrole-free extracts are allowed to be used as flavorings. Many other alkaloids were found in the roots, but none in the leaves [100].

Filé powder is a spice and a thickening agent made of young, dried, and ground Sassafras tree (*Sassafras albidum*) leaves; however, Parekh [101] refers to filé powder as ground sassafras root. Filé powder is a crucial ingredient of Creole gumbo [102]. Originally gumbo was made using okra instead of filé powder by l Choctaw Native Americans. Filé powder was introduced later and proven useful when okra was out of season [103]. At high temperatures, filé powder thickens unusually, forming unappetizing gelatinous strings [104].

Parts of the sassafras tree have many uses. Orange-wood is used to make barrels, buckets, posts, and furniture; oil is used in perfume and soap production; the drinkable brew is made of roots' bark [105]. However, sassafras bark hot water infusions are not recommended due to the harmful safrole content and may interfere with medicine intake [106]. Considering the processing of sassafras trees, the leaves are a byproduct, which may be used in the production of food additives instead of being discarded.

Given the thickening properties of filé powder and its herby taste, it is expected to find use in instant soups and meals, salty sauces, meat pies, vegetable pastes, loaves, bread spreads, cocktail mixers etc. To the authors' best knowledge, there is very little information about filé powder utilization in food technology.

2.4. Sugarcane

Sugarcane places among the most valuable crops in food and energy industries. Sugarcane production in 2020 reached 1.9 billion tons [107]. Its notable trait is to accumulate large content of sucrose in its stems and a very high yield of 80 tons/ha but in theory, it is possible to achieve over 380 tons/ha due to the breeding programs or gene engineering. Sugarcane is mainly cultivated for sugar and, following, ethanol production [108]. The sugarcane industry is responsible for a lot of waste. For every 1 ton of sugar produced, 9 tons of byproducts are generated—3–3.4 tons of bagasse, 4.5 tons of molasses, 0.3 tons of filter (press) mud, and, consequently during the manufacturing process, 12 tons of fumes. These byproducts are abundant in carbon compounds and minerals, thus may be used for extraction, physicochemical transformation, or fermentation to fortify products such as construction materials, drugs, substrates for enzymes in the production of chemicals, food

and fodder, pesticides or to obtain fiber, low-calorie sweeteners, vitamin acids, beverages, oils, protein, fodder, fertilizer (press mud), and fuel. However, there are byproducts of lesser commercial value which are trash, green tops, wax, fly ash, and spent wash. Numerous organizations in leading sugar-producing nations such as Australia, Brazil, Cuba, Mauritius, Taiwan, South Africa, China, and India have been revolutionized into "Sugar Complexes" which supplied not only sugar but also waste-derived products due to the abundance of economic opportunities in the production of sugarcane byproducts [109,110].

Bagasse is a fibrous waste generated after sugarcane is crushed during sugar production. Fresh bagasse has a high moisture content of about 50% and is later dried to a composition of 45% cellulose, 28% pentosans, 20% lignin, 5% sugar, 1% minerals, and 2% ash [110], however, Paturau, [111] reports 55–58% cellulose, 26–32% hemicellulose, 19–22% lignin, and Sangark and Noomhorm [112], claim 45% cellulose, 26% hemicellulose, and 19% lignin. Bagasse is a complex carbohydrate biopolymer structure. Monomers of these biopolymers are connected by four bonds, namely ether, ester, hydrogen, and carbon-carbon bonds [113]. Functional groups are connected via ether bonds, hemicelluloses, cellulose, and lignin are connected via ester bonds, hydrogen bonds are present in carbon-carbon structure in the aromatic rings and the cellulose polymer chains, β-1,4 glycosidic bonds are present between long, linear homopolysaccharide of anhydroglucose and the cellulose fractions. Bagasse cellulose molecular mass is 157,800 to 168,400 g/mol and the cellulose fibers are 1–1.5 mm in size. Bagasse is resistant to enzymatic and chemical hydrolysis due to cross-linkage between hemicellulose matrix and micro- and macrofibrils of cellulose and due to the degree of polymerization, which depends on glucose units in a polymer. The crystallinity index of bagasse is established as 56.7% [114]. Bagasse hemicellulose consists of β-1,4 xylopyranose backbone, β glucans, xyloglucans, glucomannans, galactomannans, and scarcely of uronic acids. Bagasse lignin's weight averages from 507 to 3973 mol/g. Syringic acid, ferulic acid, vanillic acid, p-coumaric acid, xylose, glucose, arabinose, galactose, acetosyringone and syringaldehyde are present in bagasse lignin fractions [115]. Dried bagasse composition is similar to that of wood. Approximately half the generated dried bagasse is sufficient to provide the sugarcane processing unit with energy and ethanol–fuel. The leftovers are often stockpiled, threatening the environment with spontaneous combustion [116]. Powdered sugarcane bagasse has been found to have a mean particle size of 105.30 μm, a surface area of 4.105 m^2/g, pore diameter of 2.23 nm, and pore volume of 0.005 cc/g [117]. Due to the significant content of cellulose in bagasse, it is used in various paper types, construction boards, panels, insulating boards, and particleboard production and due to a high content of pentosans it is utilized to obtain chemicals, e.g., lactic acid. Moreover, by fermenting the bagasse or adding manure the biogas of approximate caloric value of 5500 $kcal/m^3$ can be produced, which may be used to power petrol or diesel engines. Bagasse is also used to manufacture biodegradable plastic (PHB), agriculture mulch, mushroom subsoil, and ethanol via simultaneous saccharification-cum-fermentation by enzymatic or acid hydrolysis [110]. Bagasse may be hydrolyzed to obtain 85–95% xylose and small percentages of arabinose and glucose. In China and Brazil xylitol is made of bagasse using the reduction process [118]. Due to bagasse's significant polysaccharide content, it may be processed into insoluble, neutral in taste and odor dietary fiber if processed using alkaline hydrogen peroxide. This process is accompanied by stirring which reduces the content of lignin by about 50% and increases water holding capacity by about 50%, the reason being mechanical shear, which opens fiber structure and makes cellulose hydroxyl groups bind with water [112]. Another method of dietary fiber production is by using sodium hydroxide (NaOH), shown in Figure 3 [119]. Furthermore, sugarcane dietary fiber has been found to be efficient as a gelling agent, generating gels of high strength and displaying a remarkable capacity to hold water [120]. This is especially true when the fiber is present in high concentrations (6 percent). According to Zhuang et al. [121], sugarcane insoluble dietary fiber increased the quality of myofibrillar protein gels by strengthening its structure, increasing the stability of the gel network and reducing its syneresis. Sugarcane dietary fiber may be used to fortify bread [122].

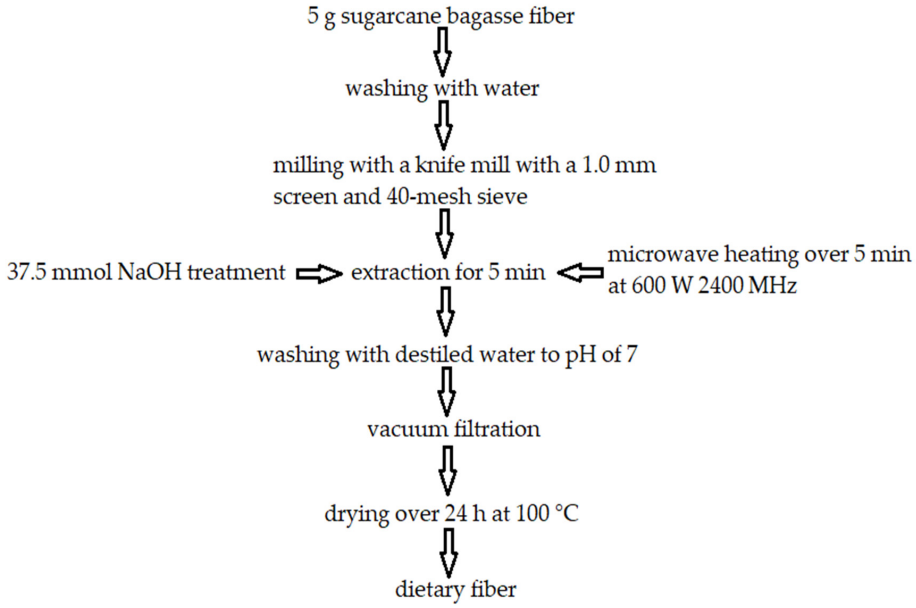

Figure 3. Production of dietary fiber from raw sugarcane bagasse fibers. Own figure based on Ref. [119]. 2022, Gil-López et al.

Another byproduct of the sugarcane industry is black strap molasses—a thick liquid of significant viscosity. Molasses is rich in sugar, which further crystallization is not profitable and is not usually meant for direct human consumption due to its chemical composition and unappealing, dark color. About 23–28 L of molasses is generated for every ton of crushed sugarcane. Molasses consists of 45–55% fermentable sugars—30–35% sucrose, 10–25% glucose and fructose, 2–3% non-sugar compounds, water, and minerals. Molasses is used as an ethyl alcohol production substrate during yeast fermentation, then ethyl alcohol is utilized in other chemicals' production, e.g., ethyl benzene, ethylene oxide, propionic acid, mono chloroacetic acid, their salts, Acetic acid, Beta picoline (3-Methyl Pyridine), Styrene, or Dibutyl phthalate. Ethanol is also used as a fuel oxygenator with 5% concentration—5–20% as a blend with gasoline (Gasohol), or F-95% as fuel extender/replacement. During yeast fermentation for alcohol, carbon dioxide is produced as well, which can be utilized as a cooling agent or to carbonate beverages. As a component of the subsoil for *Aspergillus niger*, a producer of citric acid, baker's and food yeast, monosodium glutamate, and substitute for coffee, molasses is implicitly utilized in the food industry. Molasses can be used as a compound of fodder, which benefits the microflora of ruminants' stomachs and helps them digest fibrous feed such as straw. Moreover, fodder enriched with molasses inhibits the development of bronchial disease. It is also used to obtain itaconic acid—a plasticizer and a chemical intermediate; acetone and butanol; dextran—a blood plasma expander and a toothpaste, paint, glue, iron-dextran complex (a medicine for anemia), sulfate dextran (anticoagulant) ingredient; ephedrine—a cough syrup ingredient; biocides; Nitromiel—an explosive; potassium salts; denaturants; activated carbon; asphalt; cement; drawing lubricants; dehydrating agent in mineral clarifying processes; sealing agents [109].

Press mud cake (or press mud) is a leftover residue produced during the filtration of sugarcane juice. It consists of 50–70% moisture, 5–14% crude was and fat, 15–30% fiber, 5–15% sugar, 5–15% crude protein, and a notable deal of Si, Ca, P_2O_5, MgO, Fe, and Mn. Press mud is rich in phosphorus, and thus is used as a fertilizer, increasing yield of sugarcane. The mixture of molasses and press mud is utilized as a fertilizer and is useful in animal feed production. Press mud is also used to produce n-triacontanol—a plant growth regulator; policosanol—higher aliphatic alcohol; cement; distemper paints; foaming agents; activated carbon; filter aids; proteins; carnauba wax replacement [109].

During the processing of sugarcane, a significant amount of foliage is generated. Furthermore, almost all of the leftover sugarcane green tips are converted into cattle feed,

which is an inexpensive and wholesome source of feed. Dried leaves are used as a fertilizer or may be ground and utilized as a filler in plastics and linoleum production [109].

2.5. Acorn

"Acorn" is a common name for the fruit of plants of the genus *Quercus* (oak trees), belonging to the family Fagaceae. The genus *Quercus* grows in the USA, temperate Europe, Asia, and subtropical Africa [123–125]. Acorns have been present in the human diet for ages being used, e.g., as flour in bread craft [126]. Nowadays, acorns are sometimes used in Mediterranean countries in times of food scarcity or as an ingredient of traditional beverages such as Raccahout (Turkish drink resembling hot chocolate), Eichel Kaffee (acorn coffee), or Licor de Bolota (Portuguese alcoholic drink) [127]. These fruits grow in the wild, most often being unused and their valuable functional ingredients such as proteins, carbohydrates, and lipids are wasted. By gathering and processing, such compounds can be of use in the food industry [128]. Acorns may also be used as hog feed because of their notable content of macronutrients. Oils isolated from acorns exhibit resemble olive oil in terms of color, iodine value, UV extinction, coefficient, fatty acid composition, and refractive index [129,130]. Acorns contain a significant amount of unsaturated fatty acids (60% of oleic acid, ω9, and 16% of linoleic acid, ω6), fiber, chlorophylls, carotenoids, phenolic compounds [127], typically 2–5% proteins, vitamins A and E, minerals—P, K, Ca, and Mg, high amount of glycine, lysine, and proline thus, being more nutritious than many cereals. Acorns may be exploited as a source of new hydrocolloids in the food sector due to their high starch content (approximately 50% greater than cereals) and fiber content. Moreover, acorn starch exhibits high paste consistency thus, may be utilized to thicken food and as a stabilizing agent. Acorn protein emulsifying properties come from lysine, which linear structure is believed to act like a potent surfactant at its isoelectric point [128]. Utilization of plant proteins, including acorn protein, as emulsifiers, stabilizers, or foaming agents is a novel approach to food texture modifications [131,132]. Acorn protein has been proven effective as an emulsifier in oil/water environments with a protein concentration of 0.5–2% (*w*/*v*). Acorn protein reduces the viscosity of o/w emulsions [128] Suggested acorn taxa for protein extraction is *Q. infectoria* spp. *boissieri*, because it contains the most protein (8.44%) among other acorn taxa. Acorn protein extraction was presented in Figure 4.

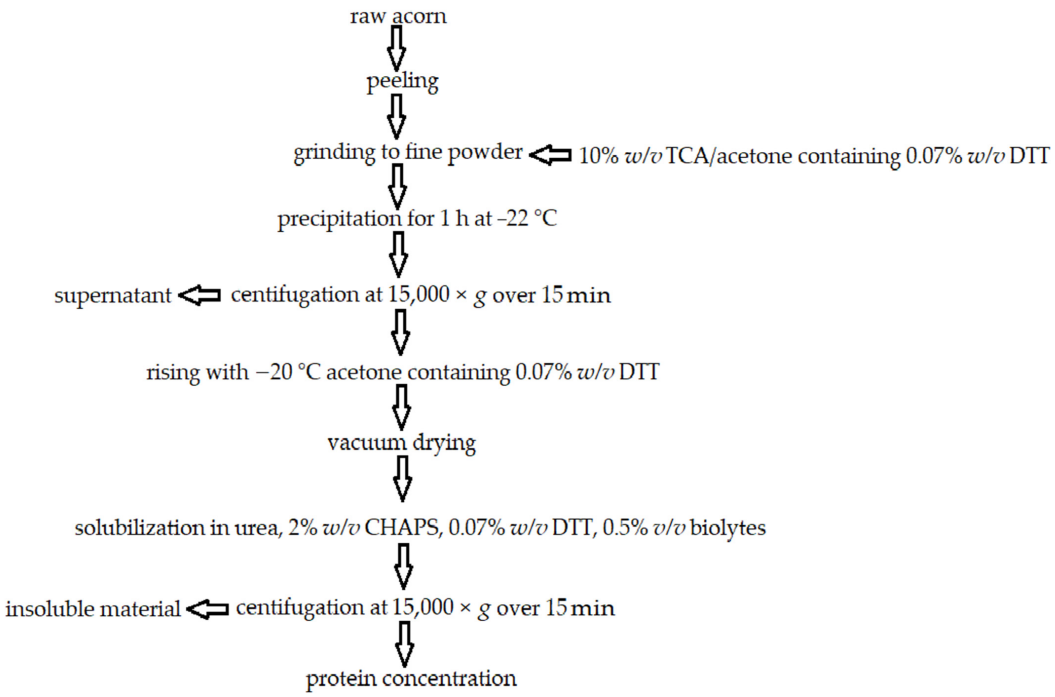

Figure 4. Flow chart of acorn protein extraction. Own figure based on Ref. [133]. 2022, Galván et al.

2.6. A New Approach to Gelatin

Gelatin is made by breaking cross-linkages between the polypeptide chains and bonds in the parent protein collagen, obtaining a heterogeneous mixture. Even further treatment of enzymes yields gelatin hydrolysates, which show pro-health benefits [134–136] and can be used as functional ingredients to provide cryoprotective effects helpful with food exposed to freeze-thaw cycles [137]. According to FAOSTAT [138], poultry meat production increased by about 35% between 2010 and 2020, resulting in corresponding byproduct production. The yearly manufacture of gelatin is about 375,000–400,000 tons [139], of which only 2% is not from mammals [140].

Despite its wide range of applications, gelatin concerns some consumers because of religious (haram or not kosher food) and health (possibility of prion disease in bovine gelatin) reasons. To overcome those issues and make use of meat and fish byproducts, it is suggested to produce gelatin from poultry skins, feet, heads, and bones and process waste of the fishery industry [141–143]. The gelatin yield depends on the amount of collagen present in a byproduct, the least abundant being the poultry head and feet (28% wet basis) and the most abundant fish skins, bones, and fins (33% wet basis) [144]. The most common fish used for gelatin production are Atlantic salmon, cod, sin croaker, short fin scad, Alaska pollock, big eye snapper, brown stripe red snapper, yellow-fin tuna, Nile perch, black and red tilapia, grass, and silver carp [145]. When gelatins from several aquatic animal species are merged, new properties emerge, allowing for a wider range of applications in the food industry [146]. Compared to traditional bovine gelatin, gelatin from cold-water fish, e.g., cod, megrim, tuna, and tilapia exhibits lower gelling and melting temperatures and similar gel strengths because of lower hydroxyproline and proline, the content of the amino acids. Amino acid content decreases as the environment of the fish is colder. Cold-water fish gelatin requires chemical or enzymatic modifications to be effective in commercial use, or its utilization would be limited to refrigerated products. The gelatin obtained from warm fish byproducts exhibits similar physicochemical properties to porcine or bovine gelatin, and thus, may replace them without significant modifications [147]. Another drawback of fish gelatin is its unpleasant, fishy odor [148]; however, it can be almost entirely neutralized by sulfuric acid, citric acid, and sodium hydroxide treatment. Moreover, such a procedure dramatically increases the gel's clarity [149]. Porcine skin gelatin exhibits higher foam capacity and foam stability compared to shark cartilage and precooked tuna fin gelatin [150,151]. In the case of protein films, gelatin from channel catfish and Nile perch exhibit film strength, tensile strength, percentage of strain, and water vapor permeability comparable with mammal-derived gelatin [152,153]. A comparison between various aquatic animals-derived gelatin has been provided in Table 4. Although gelatin is not a novel hydrocolloid, its production from poultry and fishery byproducts is innovative.

Table 4. Aquatic animals derived gelatin compared.

Source	Yield (Wet Basis)	Bloom/gel Strength	Reference
Atlantic salmon skin	4–11.3%	80–108 g	[154]
Atlantic cod skin	44.8% [c]	71 g	[154]
Bigeye snapper skin	6.5%	105.7 g	[155]
Bigeye snapper skin	40.3% [a]	138.6 g	[156]
Brownbanded bamboo shark	19.06–22.81%	56.53–217.26 g	[157]
Blacktip shark	21.17–24.76%	10.43–207.83 g	[157]
Black tilapia skin	5.39%	181 g	[158]
Bigeye snapper skin	6.5%	105.7 g	[155]
Channel catfish	19.2% [b]	252 g	[159]
Cod skin	17%	180 g	[149]
Cuttlefish skin	36.82% [c] (dorsal skin) and 59.69% [c] (ventral skin)	126 g (dorsal skin) and 137 g (ventral skin)	[160]
Giant catfish skin	20.1%	153 g	[161]

Table 4. *Cont.*

Source	Yield (Wet Basis)	Bloom/gel Strength	Reference
Giant squid inner and outer tunics	12%	147 g	[162]
Grass carp	11.3% [a]	N/E	[163]
Lumpfish skin	14.3%	N/E	[164]
Megrim skin	10%	360 g	[165]
Nile perch bone	2.4%	134–160 g	[152]
Nile perch skin	16%	134–229 g	[152]
Pollock skin	18% [b]	460 g	[136]
Red tilapia skin	7.81%	128 g	[158]
Shark cartilage	17.34%	111.9 kPa	[150]
Shortfin scad skin	7.25%	177 g	[14]
Sin croaker skin	14.3%	125 g	[14]
Silver carp skin	11% [a]	600 g	[166]
Tilapia skin	N/E	263 g	[149]
Tuna fin	1.25%	126 g	[151]
Yellowfin tuna skin	89.7%	426 kPa	[167]

[a]—based on the hydroxyproline content of the gelatin in comparison with that in the skin. [b]—based on the protein content of the gelatin in comparison with the wet weight of raw material. [c]—dry weight. N/E—not evaluated.

The process of acid and alkali gelatin extraction from fishery byproducts has been shown in Figure 5.

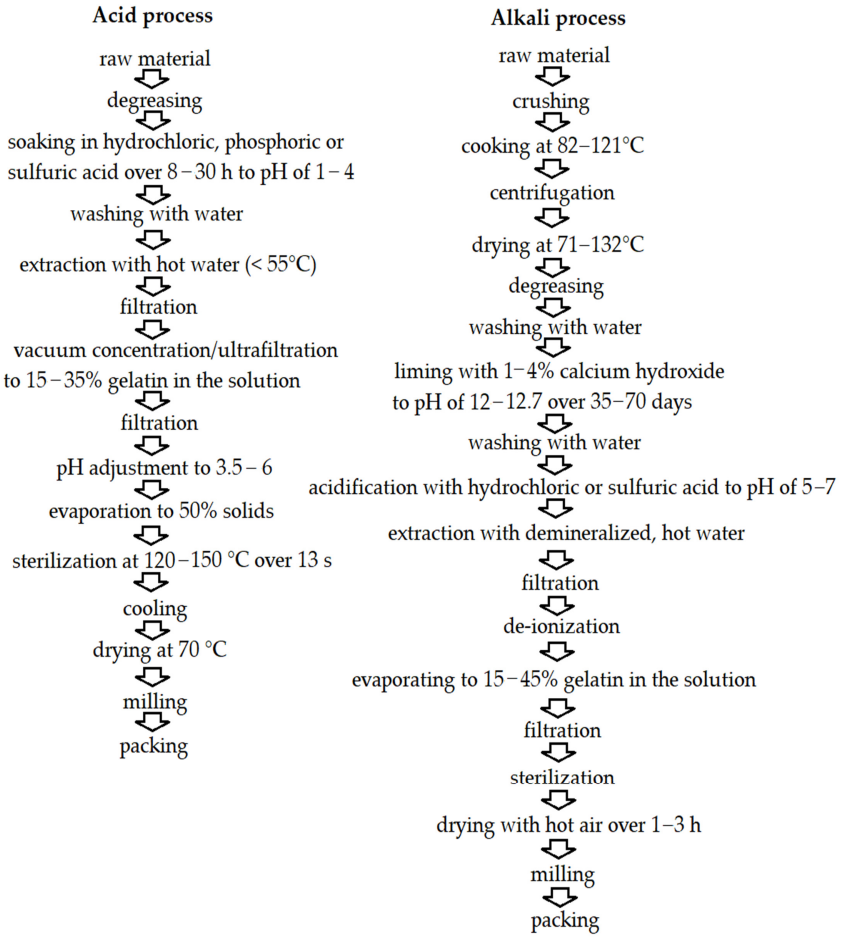

Figure 5. Flow diagram of acid and alkali process of gelatin production. Own figure based on Ref. [146]. 2022, Wasswa et al.

3. Health-Promoting Properties of Waste Gelatin from the Fish and Poultry Industry and Byproducts from Arrowroot, Kuzu, Sassafras Tree, Sugarcane, and Acorn

It is important to note the health advantages of arrowroot starch. It is characterized by high digestibility and high content of dietary fibers; contains raffinose, lactulose, stachyose, and fructooligosaccharides which might serve the purpose of prebiotics [19], meaning they promote the growth of favorable bacteria such as *Bifidobacterium* and *Lactobacillus* in the large intestine without stimulating the harmful bacteria such as *Clostridium perfringens*. As a result, humans absorb microelements including Ca, Mg, and Fe more readily and are less likely to develop diseases such as large intestine cancer and disorders brought on by excessive cholesterol levels. Presently, in the large intestine, the prebiotics ferment, producing short-chain fatty acids, which can limit the development of pathogens [168]. As a result of the starch's anti-inflammatory and anti-irritating effects, it is used to alleviate tissue and bowel disease [169]. This seems especially meaningful regarding ecology and human safety since traditional drugs cause a variety of side effects and may interfere with the environment. Arrowroot starch nanocrystals do not cause such harmful effects [170]. The rhizome of arrowroot is safe for those with phenylketonuria since it is high in alkaloids, glucosides, phenolic compounds, terpenoids, saponins, flavones, tannins, [171] phosphorus, sodium, potassium, magnesium, iron, calcium, and zinc, and has a medium level of phenylalanine. A rhizome is also known for immunostimulatory—significantly increased IgG, IgM, and IgA levels in mouse serum [31]. Arrowroot starch shows antioxidant properties [21] most likely by trapping peroxyl and hydroxyl radicals [172]. This leads to the mitigation of diabetes, cardiovascular disease, high blood pressure, and cancer control [173]. Its short fibers are easy to digest, making it useful for baby diets and children with autism or down syndrome [174]. Arrowroot is also a source of type III resistant starch, proven beneficial for health as a dietary fiber fraction and a valuable food processing element [175]. Another benefit of arrowroot starch is a very low glycemic index of 14 [24]. Foods with a low glycemic index (GI) are more favorable compared to those with a high GI, considering health issues. A diet composed of low-GI foods help maintain proper body weight, body fat, manage hyperlipidemia, and diabetes [176]. Although arrowroot is mainly used for starch production, many works point out its medical applications [22]. Arrowroot is abundant in a good deal of healthy substances, e.g., alkaloids, steroids, phenolic compounds, and flavonoids. Solvents and found compounds are presented in Table 5. Moreover, arrowroot leaves may extract antidiarrheal compounds [177].

Table 5. Solvents and soluble compounds of arrowroot extracts.

Compound	Reference					
	[178]			[179]		
	Methanol	Water	Ether	Chloroform	Methanol	Water
alkaloids	P	P	N	N	P	N
steroids	P	N	P	P	N	N
phenolic compounds	P	P	N	N	P	P
flavones	N/E	N/E	N	N	P	N
flavonoids	P	N	N	N	N	N
flavonones	N/E	N/E	N	N	P	N
glycosides	P	P	P	P	P	P
saponins	P	P	P	P	P	N
terpenoids	P	P	P	P	P	N
tannins	P	N	N	N	P	N

P—present, N—not present, N/E—not evaluated.

Kuzu is known for its use in diabetes effects mitigation due to the significant content of isoflavonoids, especially puerarin, known for its ability to restore glucose balance. [116]. Pueranin is used against migraines as well because it regulates cerebral blood circulation [180]. Kuzu is also used in the treatment of flu, fever, nausea, allergies, and

diseases of the upper respiratory tract. Moreover, kuzu has a significant alkaline effect that is useful in deacidifying, detoxifying, and regulating the body's metabolism. Intake of kuzu increases the content of happiness hormones–serotonin and dopamine, which are responsible for maintaining a positive mental state and preventing stress. It also lowers blood pressure, increasing the economization of heartbeat and reducing the risk of a heart attack. The consumption of kuzu may also help stimulate the immune system by penetrating the human intestines to cope with bacterial infections and suppress smooth muscle contractions [77]. Furthermore, kuzu root contains bioactive isoflavones: isoflavonoid glucosides, coumarins, puerarols, but-2-enolides and their derivatives [181], daidzein, daidzin, puerarin, formononetin-7-O-glucoside (ononin), 3-methoxypuerarin, 6-O-D-xylosylpuerarin, 3-methoxydaidzein, genistein, biochanin A, formononetin and isoflavone glucosides, e.g., daidzein 8-C-apiosyl-(1→6) glucoside. These compounds exhibit hepatoprotective [182], antioxidant [183], anti-diabetes [184], neuroprotective [185], cardiovascular protective [186], anti-inflammatory [187], estrogenic [188], antineoplastic, antiatherogenic, antiarrhythmic, antihypertensive, detoxifying, and diuretic activities. However, the mentioned substances exhibited different antioxidant properties than expected, suggesting that more research is needed [189]. Puerarin can dilate blood vessels, which decreases blood pressure [180]. Daidzein, also known as phytoestrogen, helps with alcoholism prevention, reducing the urge to alcohol consumption by up to 80%. Daidzein is also known for its antioxidant properties, alleviating the consequences of alcohol intoxication, and helping heal organs already damaged by alcohol. Intake of kuzu also helps with coping with nicotine addiction. The great advantage of kuzu therapy is the lack of side effects [81]; however, Wong et al. [180] state that there are no regulations regarding contaminants in kuzu root, such as excessive or banned pesticides, microbial contaminants, heavy metals, and chemical toxins.

Sassafras leaves contain many essential oils, including geranial, neral, limonene, caryophyllene, α-pinene, (Z)-3-hexenol, linalool, the caryophyllene oxide [190]. In traditional medicine, sassafras infusions are used to treat colds, high blood pressure, heart troubles, swelling, worms, fever [191], stomach ache, urinary retention, scurvy, jaundice, pregnancy difficulties, cancer, typhus, dropsy [192], diarrhea, rheumatism, measles, scarlet fever, burns, lower chest pain, nausea, vomiting, indigestion, constipation, loss of appetite, gallstones, bladder pain, or as a blood purifier [193]. Formerly, boiled sassafras leaves were used as an abortifacient [194].

Sugarcane bagasse and sugarcane tops show promising pro-health benefits if processed, to dietary fiber using alkaline (NaOH or H_2O_2) treatment. Dietary fibers from both bagasse and sugarcane used in the food industry demonstrated significant nutritional value. However, H_2O_2 treatment promoted oxidation and free radical occurrence thus, being threatening for some human food macromolecules. In terms of chapatti-style bread and pasta noodles, an inclusion of no more than 8% of these fibers has been deemed agreeable [119]. The main pro-health characteristics of dietary fibers are inhibition of carbohydrate and fat digestion, which helps deal with diabetes [195]; hyperglycemia control [196]; diabesity prevention; inflammation control; Alzheimer's disease and vascular dementia prevention; depression and anxiety mitigation; hypocholesterolemic effect; lowering the blood pressure; cardiovascular disease prevention; colon cancer prevention [197]. Dietary fiber is known to have even more health-promoting properties; however, dietary fiber benefits are not the main topic of this work.

Acorn protein may provide some health-promoting benefits. Acorn proteins consist, among others, of legumin, legumin precursors, which show antioxidant activities, and arterial pressure regulating properties, by inhibiting I-converting enzyme [198]; 2-Cys peroxiredoxin and peroxiredoxin-2b proteins, both responsible for mitigating the oxidative stress in plants thus, most likely serving an antioxidative purpose; chitinase being a protein responsible for defensive mechanism against pathogens [140] thereby, inhibits the growth of fungi and helps with cancer prevention [199]. Acorn protein has a significant amount of leucine, isoleucine, and threonine presented in Table 6. However, acorn protein alone

cannot provide a sufficient number of amino acids [200]. Mentioned compounds are not synthesized in the human body and need to be supplied with food for proper health. Lack of indispensable amino acids in the human diet results in several health conditions such as depression, anxiety, insomnia, fatigue, weakness, and growth stunting in the young. The more severe consequences of indispensable amino acids deficiency are kwashiorkor–a state of malnutrition manifesting as peripheral edema, dry peeling skin with hyperkeratosis and hyperpigmentation, ascites, liver malfunction, immune deficits, anemia, and relatively unchanged muscle protein composition; and marasmus–severe physical wasting [201].

Table 6. Aminogram of acorn *(Quercus rotundifolia)* kernel protein. According to [202].

Amino Acid (AA)	Protein Content (g AA/ kg Protein)
Essential amino acids	
arginine	65
lysine	43
histidine	18
isoleucine	47
leucine	62
methionine	22
Methionine + cystine	45
phenylalanine	45
Phenylalanine + tyrosine	64
threonine	32
valine	58
Non-essential amino acids	
Aspartic acid	205
Glutamic acid	143
serine	42
glycine	43
alanine	46
proline	65
tyrosine	26
cystine	23

It is possible to obtain fish and aquatic animal gelatin hydrolysates using various proteolytic enzymes. Gelatin hydrolysates and gelatin-derived peptides show health-promoting properties [15], shown in Table 7.

Table 7. Health benefits of aquatic animal gelatin hydrolysates and gelatin-derived peptides with used enzymes.

Fish or Aquatic Animals	Enzyme Used	Pro-Health Benefits	Reference
Alaska pollock skin	Pronase E	Antioxidant	[203]
Atlantic salmon skin	Flavourzyme	Dipeptidyl-peptidase IV enzyme inhibitory activity–type 2 diabetes, symptoms mitigation	[204]
Amur sturgeon skin	Alcalase	Antioxidant, cryoprotective benefit	[205]
Brownstripe red snapper skin	Trypsin-like proteases from pyloric caeca	Antioxidant	[206]
Blacktip shark skin	Papain, papaya latex crude enzymes	Antioxidants, hypertension prevention, human LDL cholesterol inhibition, DNA oxidation inhibition, metal ion chelation	[207–210]
Chum salmon skin	Papain, Alcalase	Cell proliferation, cycle progression, apoptosis	[211]

Table 7. *Cont.*

Fish or Aquatic Animals	Enzyme Used	Pro-Health Benefits	Reference
Hoki skin gelatin	Trypsin	Antioxidant	[212]
Japanese flounder skin	Pepsin	Antioxidant	[213]
Jumbo squid skin	Trypsin	Antioxidant	[214]
Nile tilapia scale	Alcalase	Antioxidant	[215]
Pacific cod scale	Pepsin, trypsin, α–chymotrypsin	Antioxidant, antihypertensive benefit	[216]
Pacific cod skin	Papain	Antioxidant, ACE-inhibition (hypertension prevention)	[208]
Squid inner and outer tunics	Protamex, trypsin, neutrase, savinase, NS37005, esperase, alcalase	Antioxidant, hypertension prevention, anticancer benefit against lines MCF-7 and U87	[217]
Squid skin	Pepsin	Hypertension prevention	[218]
Tilapia skin	Properase E, multifactor neutral	Antioxidant, photoaging prevention	[219,220]

4. Conclusions

Many food industries focus on manufacturing products, which are allegedly the most profitable and easiest to produce, discarding the byproducts into landfills or directly to the environment polluting it. This causes additional transportation and storage costs. Moreover, pollution poses a threat to wild animals and humans as well. Fortunately, production waste has a lot to offer to manufacturers, since it is most often loaded with useful materials, functional substances, and health-promoting compounds. Byproducts such as poultry feet, beaks, feathers, skin or fish scales, fins, heads, and bones are typically discarded or processed into fodder. However, such a common ingredient might be used in a novel way to produce safe gelatin and a substrate for gelatin hydrolysates that are beneficial for health. Many plants are cultivated for their medical application. After processing, leftovers are discarded. These byproducts frequently contain valuable, functional compounds such as foaming agents, surfactants, gelling and thickening agents, cryoprotectants, or syneresis inhibitors. Moreover, the byproducts often pose medical applications too. They may exhibit antioxidative, nutritional, anti-cancer, diabetes regulating, or prebiotic activity. It is worth considering the economic strategy used by sugarcane manufacturers, who not only produce sugar but also utilize the waste to generate fuel for their units, fodder for locals, and fertilizer for their plants. Such technology may be introduced to more food processing industries such as starch, meat, grain, or plant-derived bio-compounds industries. Doing so can reduce the cost of production, transit, and storage, therefore, providing consumers with lower prices without losses for the manufacturers.

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FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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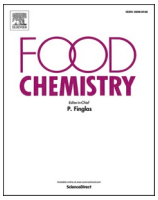

Application of fermentation for the valorization of residues from Cactaceae family

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ABSTRACT

Cactaceae family is well-known for their adaptations to drought and arid environments. This family, formed by four subfamilies (Cactoideae, Opuntioideae, Pereskioideae, and Maihuenioideae) are known for being leafless stem succulent plants with numerous spines, and their commercial fruits, distinguished by their bright colors and their skin covered with bracts. Some of these species have been traditionally used in the food industry (*e.g.,* pitaya, cactus, or prickly pear) or as pharmaceuticals to treat specific diseases due to their active properties. The processing of these fruits leads to different residues, namely pomace, skin, spines, and residues from cladodes; besides from others such as fruits, roots, flowers, mucilage, and seeds. In general, Cactaceae species produce large amounts of mucilage and fiber, although they can be also considered as a source of phenolic compounds (phenolic acids, flavonols and their glycosides), alkaloids (phenethylamines derived betalains), and triterpenoids. Therefore, considering their high content in fiber and fermentable carbohydrates, together with other target bioactive compounds, fermentation is a potential valorization strategy for certain applications such as enzymes and bioactive compounds production or aroma enhancement. This review will comprise the latest information about Cactaceae family, its potential residues, and its potential as a substrate for fermentation to obtain active molecules with application in the food industry.

1. Cactaceae family

1.1. Main genera, target species and current use

Cactaceae Juss. constitutes a wide plant family containing 1,922 species distributed into 130 genera (Ana Novoa et al., 2015). Due to the taxonomic heterogeneity of this plant family, no generic consensus on its phylogenetic classification has been achieved, but it is collectively assumed that Cactaceae species can be classified into 4 major subfamilies, namely: Cactoideae, Opuntioideae, Pereskioideae, and Maihuenioideae (Bárcenas et al., 2011). These species are considered xerophytic, presenting several common characteristics, such as high resistance to drought and heat as a consequence of their adaptation to the arid conditions found in their natural habitat (Nuzhyna et al., 2018), located in the American continent, specially Mexico, and other South American countries in a lesser extent, such as Brazil, Argentina, and Chile, among others (Bravo-Avilez et al., 2019; Ortega-Baes & Godínez-Alvarez, 2006). Among the adaptative anatomical and physiological traits attributed to Cactaceae species, the performance of crassulacean acid metabolism together with a spinous succulent body and extended root systems are some of the major characteristics devoted to the improvement of their water management systems (Santos-Díaz &

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Camarena-Rangel, 2019). Moreover, most species also contain edible fruits with sweet pulp, low acidity, and different attracting colors, ranging from yellow to red–purple (Bakar et al., 2020).

Concerning the human exploitation of Cactaceae, multiple applications have been largely established with different industrial purposes, including horticultural, food and livestock feed, and medicinal uses (Ana Novoa et al., 2015), being *Opuntia ficus-indica*, *Hylocereus undatus* and *Hylocereus polyrhizus* the most commercially employed species, commonly known as prickly pear, white pitahaya, and red pitahaya, respectively, whereas other species are still poorly underexploited, as it is the case of *Pilosocereus gounellei*, also named as xiquexique, and *Cereus jamacaru*, commonly known as mandacaru (de Araújo et al., 2021). With respect to industrial applications, cacti have been classically exploited in the food industry for the development and production of a wide range of products. Thus, *P. gounellei* flour has been satisfactorily used for cake and bread production, obtaining dough formulations with higher consumer acceptability and better color and flavor than those derived from wheat (Da Silva et al., 2018). The pulp from *C. jamacaru* was used to develop functional ice creams with enhanced physicochemical attributes for their industrialization, showing a greater sensory acceptance (de Fidelis, 2015). In parallel, the jelly from *C. jamacaru* pulp was incorporated to goat yogurt, promoting a longer storage with improved acidity, and lactose content profiles (Nobrega et al., 2020). Furthermore, *O. ficus-indica* fruits were also used for the elaboration of juice, retaining a high content of bioactive compounds after clarification (Cassano et al., 2010), whereas its red–purple betalains were shown to enhance the color stability of gummy candies (Otálora et al., 2019). In addition, several authors have reported the production of different active packaging systems for food purposes based on film structures derived from *O. ficus-indica* (Aparicio-Fernández et al., 2018) and *H. polyrhizus* (Qin et al., 2020).

More recently, besides their food applications, Cactaceae species have been exploited in other industrial sectors, including technological, pharmaceutical, and chemical applications (Fig. 1). For instance, cacti have been used for the production of nanomaterials, including gold, lithium, and zinc oxide nanoparticles with improved therapeutical performance (Alvarez-Bayona et al., 2019; Vishnupriya et al., 2020), cosmetic nanoemulsions with moisturizing properties (De Azevedo Ribeiro et al., 2015), cellulose nanowhiskers (Nepomuceno et al., 2017) and nitrogen-doped carbon dots (Arul et al., 2017). In the field of livestock feeding, different cladodes from Cactaceae species have been use for such purpose, showing no negative effects on meat quality with the exception of *O. ficus- indica*, which prompted an increase in polyunsaturated fatty acids on lamb and goat meats (Mahouachi et al., 2012). Another chemical application of cacti derived products has awaken much interest in the field of water treatment, since solid cactus materials from *O. ficus-indica* provoked the removal of turbid artifacts in water, thanks to their flocculating properties due to starch and quercetin (Bouaouine et al., 2019). In addition, the production of natural dyes has also enabled the exploitation of cacti in the chemical and cosmetic industry, mainly due to the isolation of pigments from *O. ficus-indica* (Guesmi et al., 2013) and *H. polyrhizus* pulp (Utpott et al., 2020). Finally, the pharmaceutical applications related to Cactaceae are a consequence of the biosynthesis of secondary metabolites with health-promoting properties, in response to the climatic and biological threats to which these species are subjected in their natural habitat (P García-Pérez et al., 2020).

1.2. Residues from Cactaceae family

1.2.1. Invasive species: A source of potential residues

Cactaceae family is native from American continent, from Southern Argentina to Northern Canada, except from *Rhipsalis baccifera* naturally distributed in Africa. However, in the last years, these species have been spread by seed companies and botanical gardens worldwide (A. Novoa et al., 2016). The easiness of this family for spreading has been linked to their rapid vegetative propagation and adaptation to arid conditions (Podda et al., 2017). This is the case of *Opuntia* spp., which has been frequently categorized as a highly invasive alien species specially in the Mediterranean Basin and South Africa (Erre et al., 2009; Podda et al., 2017). Different studies have researched the main factors that influence *O. stricta* distribution and proposed strategies to control these

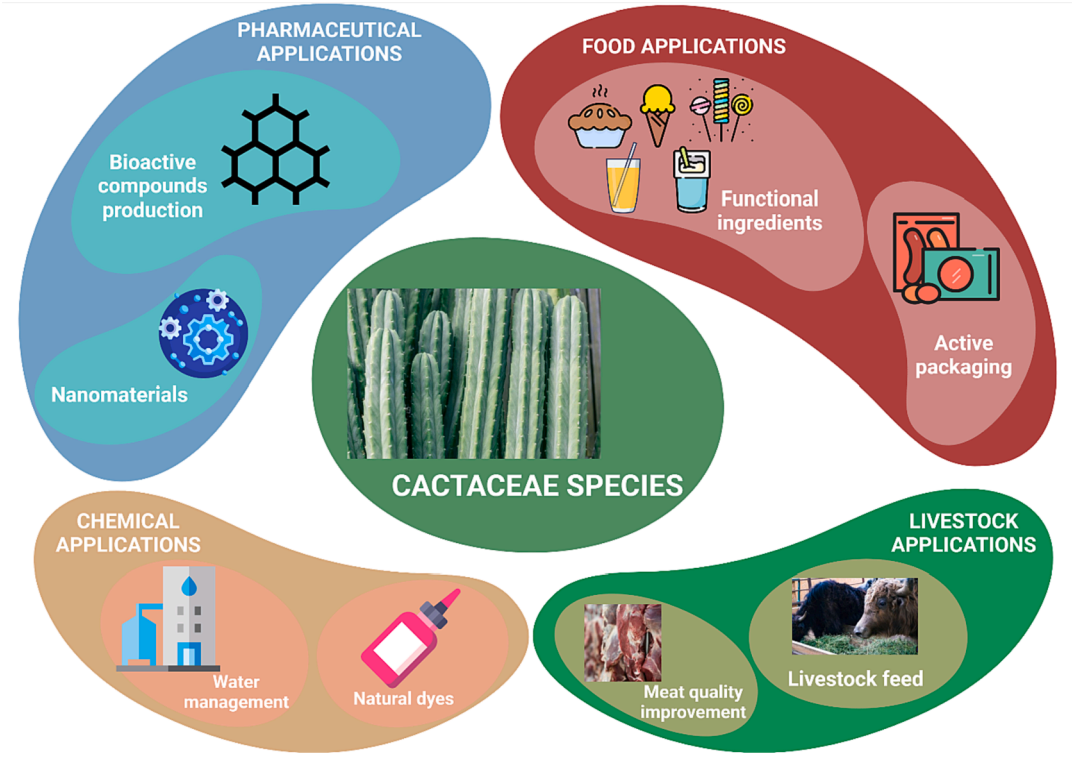

Fig. 1. Industrial applications of Cactaceae and derived by-products.

populations and protect natural biodiversity (Foxcroft et al., 2007; Rule & Hoffmann, 2018). From this perspective and to avoid the perturbation of ecosystems, some approaches have been considered, *i.e.*, phytoremediation, bioenergy production or development of added-value products with industrial applications (Kumar Rai & Singh, 2020). Removing these species from invaded ecosystems is a potential valorization strategy to use this underutilized discarded biomass to produce bioactive compounds or as a substrate for fermentation while taking advantage of the rapid growth of these species (Ahmed et al., 2020). However, even though it is critical to managing the spread of Cactaceae species, it should be considered that valorization strategies could have a counter-productive effect and promote the growth of these species so risks assessment should be deemed.

1.2.2. Industrial waste derived from food and beverage production

Generally, Cactaceae species are formed by leaves or spines, areoles (unique structure of cacti), stems, fine roots, fruits, and flowers. Among them, stems and fruits are the parts that have been mostly used for food applications. Their fruits are also known as prickly or cactus pear (*Opuntia* spp.) or pitahaya or dragon fruit (*Hylocereus* spp.) and are composed of a thick peel with small thorns or bracts that protects a juicy pulp with a lot of seeds (the edible part) (Jiménez-Aguilar et al., 2015). The fruit is usually consumed as fresh fruit, juice, or jam. In turn, the stems, also called cladode or '*nopal*', are leafless and succulent covered with spines and multicellular hairs and are frequently used for animal feed or discarded (Marin-Bustamante et al., 2018). Likewise, some cultures have included cladodes in diverse food preparations, such as salads, jam, chutney, or pickles and candied *nopales* (Ginestra et al., 2009).

Processing Cactaceae plants for food and non-food applications leads to different residues, namely pomace, skin, spines, and residues from cladodes. Several industries can profit of such by-products for the development of sustainable products, including food and beverages (*e.g.*, functional foods, alcoholic and non-alcoholic beverages), livestock feed products (*e.g.*, supplements, feed from cladodes and waste from fruit processing, including skin and seeds), nutraceuticals (*e.g.*, fiber and flours from cladodes), pharmaceuticals (*e.g.*, gastric mucosal protectants from mucilage extracts, tablets and capsules of cladode powder and flower extracts), binding compounds for the construction industry (*e.g.*, binding compounds from mucilage/cladodes), biogas for the energy sector (*e.g.*, biogas from digestion of cladodes and factory waste streams), and agricultural inputs (*e.g.*, soils, organic materials, and improved drainage from the use of cactus pear plant products). However, not all the valorization strategies have the same importance; namely, those by-products recycled into new food products are the best option for managing residues in the context of the circular economy. On the contrary, those residues considered as "unfitted for human consumption" consist of degraded products that will be used for other purposes, such as animal feed or fuel production (Lavelli, 2021). In this context, this article will focus on the recovery strategies of active compounds for residues from the Cactaceae family for food applications.

Cactaceae family is cultivated around the world. According to their human exploitation, since *Opuntia* and *Hylocereus* are the most commercially employed species in the food industry, and their residues have been the most studied. *Opuntia* spp. is grown in more than 20 countries and up to 2.6 million ha are dedicated to this crop, mainly used for forage. In terms of production, more than 600,000 tons are produced annually and 96 % of the world's production is represented by 3 countries, namely Mexico (80 %), Italy (12,2%) and South Africa (3.7 %). Yields per country also vary between 6.5 t/ha in Mexico to 26 t/ha in US and Israel (FAO and ICARDA, 2017). *O. ficus-indica* is the most economically important species worldwide, being Mexico the main producer (*ca.* 44 % of the world production), followed by Italy, which is the main world exporter. However, during fruit processing (peeling, squeezing, clarification, etc.) waste and by-products are generated from the prickly pear (skin, seeds, and part of the pulp, depending on processing) and the cladodes (spines, glochids and the outer edible coating). These by-products account for 45 % of the whole fruit and are mainly peels (30 %) and endocarp with seeds (Melgar et al., 2017; Morales et al., 2015). *O. ficus-indica* by-products are valuable sources of beneficial nutrients, such as minerals and fatty acids and fiber (M. A. Silva et al., 2021). *Hylocereus* species are also cultivated worldwide in tropical or sub-tropical areas. The last data indicate a total production of *Hylocereus* spp. of more than 1 million tons. Vietnam is the top producer and exporter with more than 60 % of the production followed by China (20 %) and Indonesia, Taiwan, Malaysia, and Nicaragua. Yields per country vary between 4 and 6 t/ha in Mexico to 40–45 t/ha in Vietnam (Mercado-Silva, 2018). Fruit processing also generates by-products such as peels, seeds, and pulp. Around 50 % of *Hylocereus* spp. is the pericarp, majorly formed by peel by-products (30–35 %) (Montoya-Arroyo et al., 2014; Roriz et al., 2022). These by-products contain natural antioxidants like tocopherols, sterols or phenols that can be used in the food industry (Lim et al., 2010). For example, their seed oil displays fatty acid and phenolic compositions compared to canola, flaxseed, and grape seed oils.

A deeper insight into these bioactive compounds, mainly phenolic compounds, alkaloids, betalains, and terpenes, and their associated bioactivities are reported later in this review. Moreover, the high fiber and fermentable carbohydrates content of Cactaceae species and their potential as substrates for fermentation purposes and their applications in the food industry will be addressed.

2. Target bioactive compounds of Cactaceae residues

Cactaceae species should cope with harsh environmental conditions in their natural habitats, including intense heat, drought, radiation, and poor nutrient availability from the soil, together with biotic stresses, caused by insect and herbivore attacks. Consequently, to deal with this paradigm, cacti synthesize a wide range of secondary metabolites with bioactive properties, as part of their chemical defensive system, *i.e.*: phenolic compounds, alkaloids, and terpenoids, among others (Harlev et al., 2013). In addition to the environmental dependence, the phytochemical composition of cacti is also influenced by other factors, such as the cultivar, geographical distribution, and the plant tissue to be analyzed. Among the different species belonging to this family, those from the genus *Opuntia* have been largely defined in terms of bioactive compounds production, in particular the species *O. ficus-indica*, including both edible parts and derived by-products, like cladodes and fruits peels and seeds (Aruwa et al., 2018). Other cacti genera poorly characterized by means of bioactive compounds are *Hylocereus*, *Pereskia*, *Mamillaria*, and *Coryphantha* (Das et al., 2021). In this section, a detailed description of the phytochemicals present in Cactaceae by-products is provided in Table 1 and summarized in Fig. 2.

2.1. Phenolic compounds

Phenolic compounds (PC) constitute the largest family of secondary metabolites, with more than 10,000 individual compounds identified to date, being ubiquitously found in the plant kingdom (García-Pérez et al., 2021a), including cacti. Considering by-products, much attention has been paid to the phenolic composition of fruit peels, mainly, together with seeds, cladodes, and flowers (Table 1). Due to their role as antioxidant compounds, and UV-radiation scavengers, is it assumed that such compounds should be accumulated in the aerial parts of cacti, which are the tissues presenting a higher exposure to environmental threats (García-Pérez et al., 2019). PC have been identified in Cactaceae, ranging from simple phenolics, such as phenolic acids, including hydroxycinnamic and hydroxybenzoic acids, to more complex polyphenols, as it is the case of flavonoid glycosides, mostly represented by flavonol and flavone glycosides, tannins, coumarins, anthocyanins, and stilbenes.

A wide variety of common phenolic acids has been reported to cacti

Table 1
Relevant bioactive compounds found on Cactaceae species by-products.

Species	Residue	Extraction	Analysis	Compounds	Quantification*	Ref.
Opuntia joconostle	Fruit peel	SLE, 80 % MeOH and 50 % acetone	HPLC-PDA-EI/MS	Phenolic acids: protocatechuic, 4-hydroxybenzoic, caffeic, vanillic and syringic acids / Flavonoids: rutin, and quercetin, anthocyanins / Alkaloids: betacyanins	PA: 259.9 μg/g	(Osorio-Esquivel et al., 2011)
Opuntia spp.	Cladodes	SLE, 75:25 60 % MeOH: 6 M HCl	HPLC-DAD	Phenolic acids: caffeic, chlorogenic, *p*-coumaric, ferulic, gallic, *p*-hydroxybenzoic, syringic, and vanillic acids / Flavonoids: apigenin, isorhamnetin, quercetin, rutin, and luteolin glycosides	PA: 99.6 μg/g F: 31.2 μg/g	(López-Palacios & Peña-Valdivia, 2020)
		SLE, hexane	HPLC-UV/Vis	Terpenoids: β-amyrin, oleanolic acid, and peniocerol	67.2 μg/g	
	Fruit peel	SLE, 70 % EtOH	Spectro-photometry	Total phenolic compounds, total flavonoids, total tannins	3,760 μg GAE/g	(Cardador-Martínez et al., 2011)
	Ground seeds	SLE, 99:1 MeOH:HCl			4,600 μg GAE/g	
Opuntia ficus-indica	Cladodes	SLE, 99.9:0.1 80 % MeOH:HCOOH	UPLC-ESI-QTOF/MS	Phenolic compounds: hydroxycinnamic and hydroxybenzoic acids, lignans, tyrosols, anthocyanin, flavone, and flavonol glycosides	PA: 1453.8 mg FAE/kg	(Rocchetti et al., 2018)
		Extrusion and precipitation, EtOH	n.d.	Mucilage	n.d.	(Otálora et al., 2019)
	Seeds	Soxhlet, hexane	GC	Fatty acids: palmitic, stearic, oleic, vaccenic, and linoleic acids	9.3 % oil rate	(Chougui et al., 2013)
		SLE, 80 % MeOH	HPLC-DAD-LTQ/MS	Phenolic acids: ferulic acid and sinapic acid derivatives	890 μg GAE g	
		SLE, hexane	HPLC-refractometer detector	Tocopherols: α-, β-, γ-, δ-tocopherols	117.6 mg/kg	(El Kharrassi et al., 2018)
		SLE, 1:2 MeOH: $CHCl_3$ saponification	HRGC-FID	Fatty acids: myristic, palmitic, stearic, palmitoleic, oleic, linoleic, and linolenic acids / Phytosterols: ergosterol, campesterol, stigmasterol, lanosterol, β-sitosterol, and avenasterol	FA: 98.8 g/kg P: 9.3 g/kg	(Ramadan & Mörsel, 2003)
	Fruit peel	SLE, 80 % EtOH	HPLC-DAD-ESI/MS	Betalains: indicaxanthin, betanidin / Flavonoids: isorhamnetin, quercetin, and kaempferol glycosides	B: 3.9 mg/g F: 3.7 mg/g	(Melgar et al., 2017)
		SLE, MeOH	HPLC-PDA-MS/MS	Phenolic acids: quinic, malic, ferulic, syringic, *p*-coumaric, and sinapic acids and glycosides / Flavonoids: quercetin, kaempferol, rhamnetin, isorhamnetin glycosides / Fatty acids: eicosanoic acid, behenic acid	TPC: 165.2 mg GAE/g	(El-Hawary et al., 2020)
		SLE, THF, saponification with 30:70 MeOH:KOH	HPLC-PDA-APCI/MS	Xhantophylls: violaxanthin, neoxanthin, anteraxanthin, lutein, zeaxanthin, cryptoxanthin / Carotenes: α- and β-carotenes, and lycopene	TC: 1,693 μg/100 g	(Cano et al., 2017)
	Flowers	Maceration, 99:1 80 % acetone:HCl	HPLC-MS-ESI/MS	Phenolic acids: ferulic acid, chlorogenic acid, syringic acid, coumaric acid, caffeic acid / Flavonoids: isorhamnetin and quercetin glycosides	PA: 3.2 mg GAE/g F: 1.6 mg CAE/g	(Benayad et al., 2014)
Hylocereus polyrhizus and *Hylocereus undatus*	Fruit peel	SLE, 99:1 80 % MeOH: HCOOH, acid, and alkaline hydrolisis	UPLC-TOF/MS	Phenolic acids: gallic, syringic, sinapic, chlorogenic, *p*-hydroxycinnamic, caffeic, *p*-coumaric, ferulic, quinic, and isoferulic acids / Flavonoids: epicatechin, rutin, isoquercetin, kaempferol, isorhamnetin, quercetin, diosmetin, baicalein, tectorigenin	0.05–131.67 mg/kg	(Tang et al., 2021)
		Precipitation with acetone	n.d.	Mucilage	n.d.	(Le et al., 2020)
	Seeds	Soxhlet, pethroleum ether	GC	Fatty acids: myristic, palmitic, stearic, arachidic, palmitoleic, oleic, linoleic, linolenic acids	28.37 % oil rate	(Lim et al., 2010)
			HPLC-UV	Tocopherols: α- and γ-tocopherols	43.5 mg/100 g	
		SLE, 50:50 MeOH: hexane	HPLC-PDA	Phenolic acids: gallic, protocatechuic, *p*-hydroxybenzoic, vanillic, caffeic, syringic, and *p*-coumaric acids	4.26 mg/100 g	
		Saponification, 94:6 EtOH:KOH	GC-FID	Phytosterols: campesterol, stigmasterol, β-sitosterol	1,040 mg/100 g	
Hylocereus megalanthus	Fruit peel and seed mixture	Fungal fermentation & maceration, 50 % EtOH	HPLC-UV/Vis	Phenolic acids: gallic, *p*-hydroxybenzoic, vanillic, syringic, *p*-coumaric, and cinnamic acids / Flavonoids: catechin, epicatechin, quercetin / Stilbenes: resveratrol	102.32 mg/100 g	(Zambrano et al., 2018)
Hylocereus spp.	Fruit peel and flowers	SPE and SLE, 95:5 MeOH:TFA	HPLC-ESI-MS/MS	Betalains: betanidin, betaxanthin, indicaxanthin, betanin, isobetanin, phyllocactin, hylocerenin, isophyllocactin, and isohylocerenin glycosides	n.d.	(Wybraniec et al., 2007)
Pereskia aculeata	Leaves	SLE, 70 % EtOH	HPLC-DAD-ESI/MS	Phenolic acids: caftaric acid, caffeic acid derivatives / Flavonoids: quercetin, isorhamnetin, and kaempferol glycosides	PA: 12.32 mg/g F: 11.4 mg/g	(Garcia et al., 2019)
	Fruit peel	SLE, acetone	HPLC-DAD	Carotenoids: α- and β-carotenes	76.2 μg/g	(Agostini-Costa et al., 2014)

(*continued on next page*)

Table 1 (*continued*)

Species	Residue	Extraction	Analysis	Compounds	Quantification*	Ref.
Pereskia spp.	Leaves	SLE, acetone	HPLC-DAD	Carotenoids: α- and β-carotenes, zeaxanthin	210 μg/g	(Agostini-Costa et al., 2014)
Mammillaria herrerae	Callus	SLE, 80 % MeOH	UPLC-MS/MS	Citric acid, tyramine, sinapic, ferulic, and *p*-coumaric acids, quercetine, flavone, rutin, kaempferol, and isoflavone glycosides, bruguierol A	TPC: 15.22 mg GAE/g	(Song et al., 2020)
Mammillaria spp.	Stem	UAE, MeOH	HPLC-DAD	Phenolic acids: protocatechuic, gentisic, chlorogenic, *p*-hydroxybenzoic, caffeic, and sinapic acids	74.3 mg/100 g	(Elansary et al., 2020)
Coryphantha macromeris	Callus	UAE, MeOH	UHPLC-PDA-HESI-Orbitrap-MS/MS	Phenolic acids: protocatechuic, caffeic, syringic, ferulic, piscidic, benzoic, and sinapic acid glycosides / Chalcones: aspalathin / Flavonoids: glycitin, scutellarein, apigenin, and isoflavone glycosides	n.d.	(Cabañas-García et al., 2021)
Coryphantha spp.	Stems and roots	SLE, 80 % EtOH	Qualitative determination	Alkaloids, sterols, flavonoids, saponins	n.d.	(Sánchez-Herrera et al., 2011)

Abbreviations: APCI, atmospheric-pressure chemical ionization; DAD, diode-array detector; EI, electron-impact ionization; ESI, electrospray ionization; EtOH, ethanol; FID, flame ionization detector; GC, gas chromatography; HCl, hydrochloric acid; HCOOH, formic acid; HESI, heat-assisted electrospray ionization; HPLC, high performance liquid chromatography; HRGC, high-resolution gas chromatography; KOH: potassium hydroxide; LTQ, linear trap quadrupole; MeOH, methanol; MS, mass spectrometry; MS/MS, tandem mass spectrometry; n.d., not defined; PDA, photodiode array detector; QTOF, quadrupole coupled to time-of-flight; SLE, solid–liquid extraction; SPE, solid phase-assisted extraction; TFA: trifluoroacetic acid; THF, tetrahydrofuran; TOF, time-of-flight; UAE, ultrasound-assisted extraction; UPLC: ultra-high performance liquid chromatography; UV/Vis, ultraviolet/visible spectrophotometric detector; CE: cyanidin equivalents, GAE: gallic acid equivalents, FAE: ferulic acid equivalents; CAE: catechin equivalents. TC: Total carotenoids; TPC: total phenolic content. * Quantitative data correspond to the maximum total values obtained in each study.

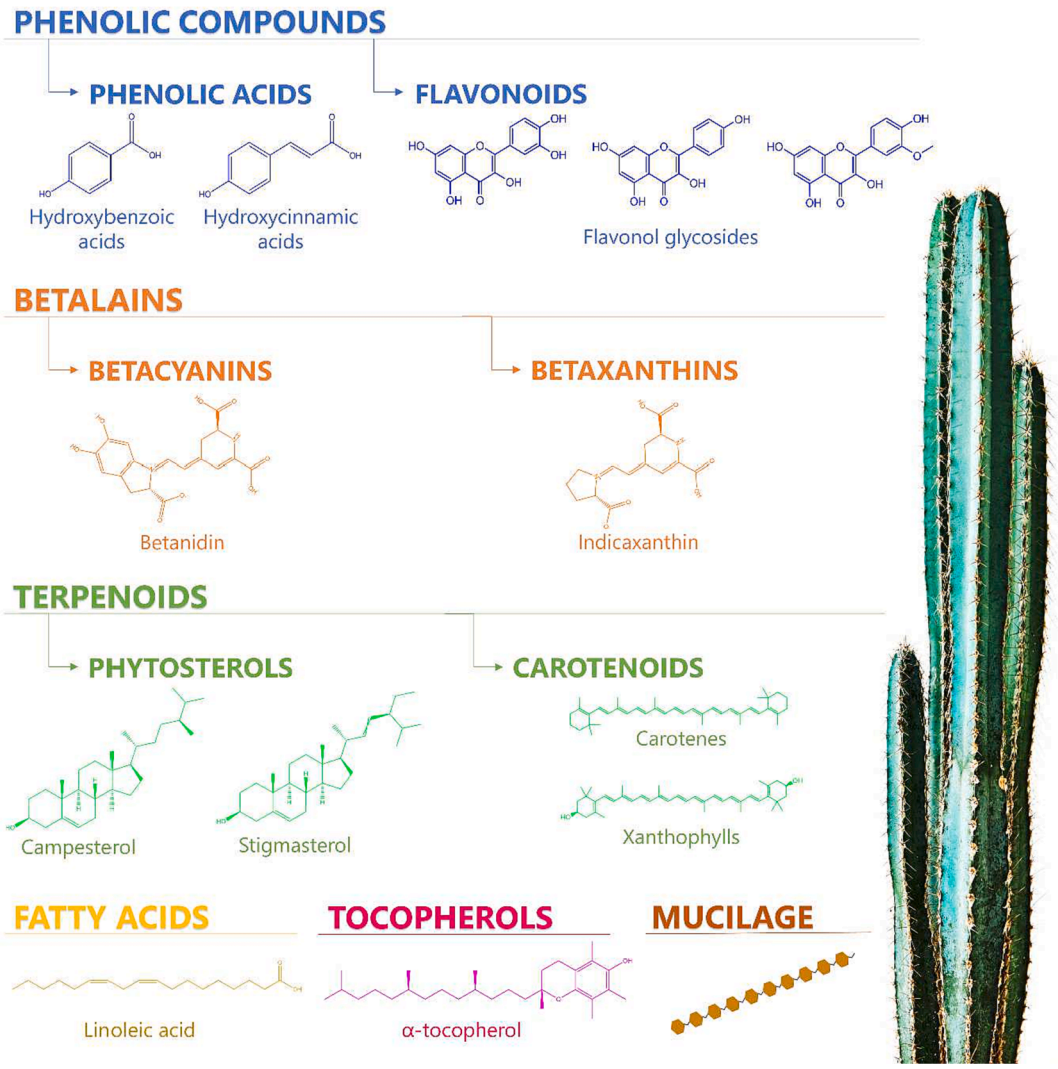

Fig. 2. Representative bioactive compounds from Cactaceae species and by-products.

by-products, whereas the flavonoid family was essentially formed by quercetin, kaempferol, and isorhamnetin glycosides (Cardador-Martínez et al., 2011; López-Palacios & Peña-Valdivia, 2020; Osorio-Esquivel et al., 2011). On the other hand, other subfamilies of PC were more restricted to some species, as it the case of anthocyanins in the cladodes of *O. ficus-indica,* with contents up to 1,444 mg/kg cyanidin equivalents (Rocchetti et al., 2018), or catechins (368.8 mg/kg) and resveratrol (28.2 mg/kg) in the mixture of fruit peels and seeds from *Hylocereus magalanthus* (Zambrano et al., 2018). Thus, *Opuntia* by-products have been revealed as promising sources of PC, especially, cladodes, seeds, fruit peels, and flowers (Benayad et al., 2014; Chougui et al., 2013; El-Hawary et al., 2020; Melgar et al., 2017; Rocchetti et al., 2018). Similar phytochemical composition has been reported to *Hylocereus* by-products, particularly those derived from *H. polyrhizus* and *H. undatus*, whose fruit peels, flowers and seeds have been thoroughly characterized in terms of phenolic profiling (phenolic acids concentration between 0.05 and 131.67 mg/kg) (Le et al., 2020; Lim et al., 2010; Tang et al., 2021). In a lesser extent, several species belonging to *Pereskia*, *Mammillaria*, and *Coryphantha* have been also subjected to the phytochemical analysis of their by-products, including leaves, calli, stems, and roots, showing not only the widely distributed phenolic acids and flavonol glycosides, but also more specialized compounds, such as caftaric acid, bruguierol A or aspalathin, probably due to existence of endangered species on these genera (Cabañas-García et al., 2021; Elansary et al., 2020; Song et al., 2020). Regarding the total phenolic content (TPC), it ranges between 15 and 165 mg GAE/g depending on the species (El-Hawary et al., 2020; Song et al., 2020).

2.2. Alkaloids

Alkaloids constitute a heterogeneous family of plant secondary metabolites characterized by the presence of nitrogen-containing heterocycles within their basic structure (García-Pérez et al., 2021b). Owing to biosynthetic properties, two major alkaloid classes are reported in Cactaceae: isoquinoline and phenethylamine derivatives, accounting for up to 50 and 80 individual compounds, respectively, in the whole family (Das et al., 2021). Between them, phenethylamines have focused the research on cacti alkaloids since many decades, firstly reported in the genus *Opuntia* (Meyer et al., 1980), although isoquinoline alkaloids, like tryptamine, have been also detected in the callus cultures of *Mammillaria* spp., and mescaline and pellotine in *Lophophora williamsii* and *Lophophora diffusa*, both with hallucinogenic effects (Santos-Díaz & Camarena-Rangel, 2019).

The most important phenetylamine derivatives found in cactus species and their by-products are betalains, considered as the major pigments of these species (Rahimi et al., 2019). Depending on the substitution degree of the basic betalain structure, known as betalamic acid, two different groups are described: red-violet betacyanins and yellow-orange betaxanthins (Slimen et al., 2017). Because of their pigment nature, betalains have been mostly reported as phytoconstituents of cacti fruits peels, causing their coloration (Table 1). Thus, betacyanins were reported in the peels of *Opuntia* fruits, like *Opuntia joconostle*, with maximum concentrations of 230.3 mg betanin/kg in the pericarp (Osorio-Esquivel et al., 2011) and *O. ficus-indica*, mostly represented by betanidin and its glycosides, isobetanin and gomphrenin I, with total betacyanin compounds up to 3.97 mg/g of extract (Melgar et al., 2017). These compounds are also found in the fruit peels of some *Hylocereus* species (pitahayas) in the form of apiosyl, feruloyl and sinapoyl esters, together with other specific compounds, such as phyllocactin and hylocerenin and their isomers and esterified derivatives (Herbach et al., 2006; Wybraniec et al., 2007). On the hand, betaxanthins have been reported in a lesser extent, being essentially reported in the fruit peels of *O. ficus-indica*, where up to 25 different compounds were isolated, including indicaxanthin as the most prevalent compound followed by vulgaxanthin I-IV, dopaxanthin, portulacaxanthin II-III and miraxanthin III (Kugler et al., 2007; Melgar et al., 2017).

2.2.1. Terpenoids

Terpenoids also make part of the secondary metabolites found on cacti, deriving from the condensation of different isoprene subunits, giving rise to different subfamilies depending on the condensation degree (García-Pérez et al., 2021b). In the case of Cactaceae family, highly condensed terpenoids are predominantly found, represented by triterpenoids and triterpenoid acids, phytosterols, and carotenoids. Due to the lipophilic nature of these compounds, they are mainly found as constituents of seed oils, although they can be secondarily isolated from other tissues, such as fruit peels and cladodes, where they constitute another source of pigmentation together with betalains (Table 1).

Thus, López-Palacios et al. isolated some triterpenoids from the cladodes of *Opuntia* spp., such as β-amyrin (1.33–4.39 μg/g), oleanolic acid (13.92–63.26 ng/g), and the phytosterol peniocerol (0.73–0.83 μg/g) (López-Palacios & Peña-Valdivia, 2020). Considering *O. ficus-indica*, both the seeds and derived oil are rich in a wide range of phytosterols, *i.e.*, ergosterol, cholesterol, campesterol, stigmasterol, lanosterol, avenasterol, and β-sitosterol (Ramadan & Mörsel, 2003), whereas the fruit peels contains relevant amount of carotenoids (total 16.93 mg/kg), including xanthophylls, such as violaxanthin, neoxanthin, anteroxanthin, lutein, zeaxanthin, and cryptoxanthin, as well as carotenoids: α- and β-carotenes, and lycopene (Cano et al., 2017). Accordingly to *Opuntia* seeds, those from *Hylocereus* species were also reported to contain different phytosterols (up to 10.4 mg/kg), like cholesterol, campesterol, stigmasterol, and β-sitosterol (Lim et al., 2010). Moreover, the leaves and fruit peels of different *Pereskia* species were shown to present α- and β-carotenes, together with zeaxanthin, only in the case of leaves (210 μg/g) (Agostini-Costa et al., 2014). Finally, other authors demonstrated the presence of sterols in the stems and roots of *Coryphantha* spp. (Sánchez-Herrera et al., 2011).

2.3. Other compounds

Together with the most relevant bioactive compounds found on cacti and their by-products, additional compounds with associated biological properties are present in a lesser extent (Table 1). For instance, fatty acids constitute the major constituents of cacti seeds and oil. Depending on the cacti species, different oil rates have been obtained ranging from 9.3 % to 28.37 % (Chougui et al., 2013; Lim et al., 2010) Thus, they present a combination of saturated, monounsaturated, and polyunsaturated fatty acids (SFA, MUFA, and PUFA, respectively). Cacti SFAs are mostly represented by palmitic, stearic, myristic acids, found in the seeds of *O. ficus-indica* (Chougui et al., 2013), *H. polyrhizus*, and *H. undatus* (Lim et al., 2010), whereas behenic acid has been also isolated in the fruit peels of *Opuntia* species (El-Hawary et al., 2020). In the case of unsaturated fatty acids, they were found in lower proportions in the seeds of the same species, containing oleic and palmitoleic acids as the principal MUFAs, whereas both omega-3 and omega-6 PUFAs have been also reported, namely linoleic and linolenic acids, respectively (Chougui et al., 2013; Lim et al., 2010). In addition to fatty acids, tocopherols were also reported in the seeds of cactus species, representing bioactive antioxidants of lipophilic nature. In this sense, Lim et al. identified α- and γ-tocopherol in the seeds of *Hylocereus* spp. (435 mg/kg) (Lim et al., 2010). More recently, other authors spotted the presence of multiple tocopherol isoforms, including α-, β-, γ, and δ- isomers in both oil seed and cladode essential oils of *Opuntia megacantha* (117.6 mg/kg) (El Kharrassi et al., 2018).

On the other hand, concerning polysaccharides, mucilage is considered a functional biopolymer, mostly isolated from cactus cladodes, and largely used with industrial purposes in both food sector, as a gelling, stabilizing and encapsulating agent, and in pharmacy, due to its antioxidant and other bioactivities, together with its properties as an efficient natural drug delivery system (Gheribi & Khwaldia, 2019; Messina et al., 2021). As previously observed for other bioactive compounds, the cladodes of *O. ficus-indica* represents an important source of cactus mucilage (Otálora et al., 2019), although it can also be isolated from the

fruit peels of *Hylocereus* spp., being incorporated in different food-related applications (Le et al., 2020).

3. Fibers and fermentable carbohydrates of Cactaceae residues

Cactaceae species have gained attention not only because of its attractive nutritional composition but also for being a rich source of bioactive compounds with promising health-related benefits (M. A. Silva et al., 2021). Among the different species belonging to this family, those from the genus *Opuntia* is the most extensively explored and distributed genus within the Cactaceae family and includes nearly 1,500 species (Chahdoura et al., 2015). Therefore, waste and by-products are mostly generated from the prickly pear and cladodes processing. It has been estimated that about 20 % of cladodes and 45 % of fruits fresh weight are discarded, and such by-products have a higher content of dietary fiber than their corresponding commercial edible parts (Bensadón et al., 2010). Therefore, the valorization of *Opuntia* wastes and by-products by recovering their bioactive compounds and dietary fiber is a potential strategy to sustainably manage these residues. In this context, the composition of dietary fiber present in *Opuntia* spp. by-products is described in this section. The physicochemical characteristics, as well as their health-related properties, are also presented.

3.1. Composition and types of fiber

Opuntia spp. waste and by-products are rich sources of dietary fiber. The insoluble fiber is composed of structural polysaccharides, such as cellulose, and hemicellulose and acts as protecting barrier against pathogenic microorganisms. On the other hand, the soluble fiber is composed of pectin, mucilage, and loosely bound hemicelluloses whose main function in plants is to avoid tissue dehydration by forming complexes with water (Ventura-Aguilar et al., 2017). The content of dietary fiber strongly depends on the part of the plant, cultivar, geographical location, maturity stage, etc. Table 2 depicts the dietary fiber composition of the by-products of *Opuntia* spp. cultivated in different geographical regions.

Regarding prickly pears by-products, El Kossori et al., 1998 found that *O. ficus-indica* sp. cultivated in Morocco had the highest total fiber content in the seeds (54.2 % w/w DW, dry matter), followed by the skin (40.8 % w/w DW) and the pulp (20.5 % w/w DW) (El Kossori et al., 1998). These authors observed that seed's fiber was mostly composed of cellulose (83.2 % of total fiber), while skin's fiber was rich in hemicellulose and cellulose and the pulp had the highest pectin content (70.3 % of total fiber). None of these by-products showed a significant content of lignin in their composition (El Kossori et al., 1998). Similar results were found by Jiménez-Aguilar et al., 2015 who reported a higher content of total fiber in the seeds (81.5–93.8 % w/w, DM) of four varieties of *O. ficus-indica* from Mexico, followed by skin (43.2–58.1 % w/w DM) and pulp (14–16.6 % w/w DM) (Jiménez-Aguilar et al., 2015). These authors also reported that seeds were mainly composed of insoluble fiber (98 % of total fiber), whereas a higher content of soluble fiber was found in skin (22–38 % of total fiber). In addition, the highest content of soluble fiber was reported in pulp (50–70 % of total fiber). On the other hand, in a recent study, it was reported that the total fiber content in the skin and pulp of *O. ficus-indica* cultivated in Egypt (El-Beltagi et al., 2019) was significantly lower than that previously reported (El Kossori et al., 1998), obtaining values of 5.83 and 4.65 % w/w DW, respectively. Similarly, Medina et al., 2007 found that the total fiber content in green and orange pulp of *O. ficus-indica* from the Canary Islands was 5.65 and 4.86 % w/w DW, respectively (Medina et al., 2007). Thus, these differences can be attributed to geographical variation, harvesting time, growth conditions, cultivar, etc. In a deep study, it was determined the composition of total dietary fiber of sweet and acid fruits of *Opuntia* spp. (Peña-Valdivia et al., 2012). These authors observed that the total fiber content of the whole sweet fruits was in the range of 6.17–10.07 % w/w DW, being the soluble fiber the predominant fraction (59–75 % of total fiber). The soluble fiber of sweet fruits of *Opuntia* spp. was composed of mucilage (0.79–1.03 %), pectin (1.45–2.14 %), and loosely bound hemicelluloses (1.58–2.11 %). These authors highlighted that the content of these three soluble non-starch polysaccharides was significantly lower than that found in the acid cultivar which in turn, was similar to those values found in cladodes (Peña-Valdivia et al., 2012).

Concerning cladodes' by-products, Ramírez-Moreno et al., 2013 found that the total fiber content in cladodes of *O. ficus-indica* ranged

Table 2
Dietary fiber of the *Opuntia ficus-indica* by-products cultivated in different geographical regions.

PRICKLY PEAR			CLADODE		Reference
Pulp	**Seed**	**Skin**	**Spine**	**Whole cladode**	
Total dietary fiber (% w/w DM)					
20.5	54.2	40.8	–	–	(El Kossori et al., 1998)
14–16.6	81.5–93.8	43.2–58.1	–	–	(Jiménez-Aguilar et al., 2015)
4.65	–	5.83	–	–	(El-Beltagi et al., 2019)
4.86–5.65	–	–	–	–	(Medina et al., 2007)
–	–	–	46.12	–	(Marin-Bustamante et al., 2018)
–	–	–	–	30.93	(Ayadi et al., 2009)
–	–	–	–	62.05–64.25	(Bensadón et al., 2010)
–	–	–	–	23–45	(Peña-Valdivia et al., 2012)
–	–	–	–	20.86–29.43	(López-Palacios et al., 2012)
–	–	–	–	47.48–51.14	(Ramírez-Moreno et al., 2013)
Soluble fiber (% w/w DM)					
14.41	3.74	3.14	–	–	(El Kossori et al., 1998)
7.6–12.4	2.2–2.9	9.8–19.3	–	–	(Jiménez-Aguilar et al., 2015)
–	–	–	–	13.38–21.19	(López-Palacios et al., 2012)
–	–	–	–	16–30	(Peña-Valdivia et al., 2012)
–	–	–	–	7.83	(Ayadi et al., 2009)
–	–	–	–	5.68–7.07	(Ramírez-Moreno et al., 2013)
–	–	–	–	8.92–9.8	(Bensadón et al., 2010)
Insoluble fiber (% w/w DM)					
6.08	50.48	37.62			(El Kossori et al., 1998)
4.2–7.6	78.6–91.3	28.2–40.9			(Jiménez-Aguilar et al., 2015)
–	–	–	–	6.62–8.47	(López-Palacios et al., 2012)
–	–	–	–	7.8–16.2	(Peña-Valdivia et al., 2012)
–	–	–	–	20.87	(Ayadi et al., 2009)
–	–	–	–	41.80–44.07	(Ramírez-Moreno et al., 2013)
–	–	–	–	53.13–54.45	(Bensadón et al., 2010)

Abbreviations: DM, dry matter.

47–51 % w/w DM, being the insoluble fiber the predominant fraction (41.80–44.07 % of total fiber) (Ramírez-Moreno et al., 2013). Similarly, (Bensadón et al., 2010) reported that the insoluble fraction in the cladodes' by-products (*O. ficus-indica*) ranged between 53.13 and 54.45 % w/w DW whereas the soluble one was within 8.92 and 9.8 % w/w DW. These authors highlighted that the by-products' composition was comparable to that observed in the edible parts of cladodes, suggesting that by-products of cladodes are an underexploited source of health-promoting compounds. Regarding the composition of dietary fiber, López-Palacios et al. (2012) found that *nopalitos* (edible young cladodes) were a rich source of soluble fiber mainly composed of mucilage (11.72 % w/w DM), followed by loosely bound hemicellulose (4.59 % w/w DM) and pectin (1.83 % w/w DM) with the lowest content (López-Palacios et al., 2012). In contrast, the insoluble fraction of *nopalitos* was composed of cellulose (5.49 % w/w DM) and tightly bound hemicellulose (2.30 % w/w DM). In this line, it was found that *nopalitos* of *O. ficus-indica* were composed of pectins (6.1–14.2 % w/w DM), mucilages (3.8–8.6 % w/w DM) and loosely bound hemicelluloses (4.9–10.7 % w/w DM), representing the soluble fiber (Peña-Valdivia et al., 2012). These authors agreed that mucilages and pectins can protect *Opuntia* spp. from drought through a hydrating mechanism allowing them suitably grow in the arid or semiarid areas. Regarding the insoluble structural polysaccharides, Peña-Valdivia et al. (2012) reported that low content of tightly bound hemicellulose (2.2–4.6 % w/w DM) were found in *nopalitos* while the cellulose content ranged 5–15 % w/w DM, representing the most abundant structural polysaccharide (Peña-Valdivia et al., 2012). These authors also reported the absence of lignin in *nopalitos* of *Opuntia* cultivars. On the other hand, spines represent 3.7 % of the total waste from *O. ficus-indica* processing in Mexico per year. In addition, other authors found that spines were mostly composed of cellulose (39.7 % w/w DM), hemicellulose (32.8 % w/w DM) and lignin (24.6 % w/w DM) (Marin-Bustamante et al., 2018). In this regard, the insoluble structural polysaccharides profile makes spines a cheap and underexploited lignocellulosic biomass to produce bioethanol (de Souza Filho et al., 2016).

3.2. Physicochemical properties

The functional properties of *Opuntia* spp. by-products, such as water retention capacity, swelling capacity, glucose retention index, fat adsorption capacity and rheological properties should be considered when dietary fiber is used as a functional ingredient during the development of novel foods. In this regard, purple cactus pear waste showed similar functional properties (water retention capacity, swelling capacity and emulsion capacity) than those observed in commercial fiber (Monter-Arciniega et al., 2019). These authors highlighted that cactus waste exhibited the highest fat adsorption capacity, suggesting that dietary fiber from cactus waste could be used for delaying lipid digestion and thus controlling cholesterol levels (Monter-Arciniega et al., 2019). In addition, dietary fiber obtained from cactus pear waste showed a non-Newtonian pseudoplastic behavior and high viscosity at a concentration of 5 %, which was comparable with xanthan gum at the same concentration (Monter-Arciniega et al., 2019). Such properties may support the use of cactus pear waste as sustainable food ingredients according to the circular economy concepts. In this way, Ramírez-Moreno et al., 2013 observed that water retention and swelling capacity of raw cladodes (*O. ficus-indica*) were unaltered after boiling (as it is commonly processing), suggesting that the high content of insoluble fiber and mucilage with high molecular weight could form a complex network with water capable of producing an increase in viscosity of the liquid phase (Ramírez-Moreno et al., 2013). However, boiled cladodes also reported a thixotropic behavior with an irreversible structural breakdown of the gel. This fact was attributed to the hydrolysis of mucilage's structure by cooking, leading to the release of soluble fiber components into the boiling water, thus reducing the gel's consistency and viscosity (Ramírez-Moreno et al., 2013). These changes in rheological properties of boiled cladodes negatively affected their glucose retention capacity into the food matrix (57–75 % lower than untreated cladodes) and thus reducing the physiological benefits of cladodes on the control of post-prandial glucose levels.

Likewise, Ayadi et al., 2009 found that cladodes from *O. ficus-indica* obtained similar values of swelling capacity than other vegetable sources (wheat and carrot). In addition, the dietary fiber of cladodes showed high water retention capacity, demonstrating their ability to interact with water. In turn, their fat adsorption capacity was comparable with that found in other vegetable sources, suggesting their technological potential. These authors also enriched wheat flour with cladode powder at different concentrations (5, 10, 15 and 20 %) obtaining significant effects on dough properties. In fact, they observed that increasing the cladode proportion in the flour led to an increase in dough's tenacity, energy, adhesion, stickiness, and hardness. However, sensory quality limited the amount of dietary fiber from cladodes that could be added to wheat flour since incorporating more than 5 % of cladodes led to unacceptable scores of overall qualities (Ayadi et al., 2009).

3.3. Associated health benefits

Opuntia spp. by-products have shown health-related properties associated to dietary fiber, such as antioxidant and antimicrobial activities *in vitro* (Sánchez et al., 2014) and anti-inflammatory, and anti-diabetic properties *in vivo* (Fernandez et al., 1994). Controlling the postprandial serum glucose level (Frati et al., 1990) and reducing the blood cholesterol levels (Fernandez et al., 1994), as well as retarding gastric emptying have been attributed to soluble fiber. In turn, the increase in fecal bulk, reduction of constipation's symptoms and the stimulation of colonic fermentation (resulting in the production of short chain fatty acids) have been ascribed to the insoluble fraction (Sáenz et al., 2004). These physiological effects were associated with previously mentioned functional properties (water retention capacity, fat adsorption capacity, emulsion activity, and swelling). In this regard, several diseases (*e.g.*, cardiovascular, type II diabetes mellitus and obesity) could be prevented by daily consuming fiber-enriched foods (recommended dietary fiber intake 25–30 g/day) (Abirami et al., 2014).

Bensadón et al., 2010 found that cladodes and fruits' by-products of *O. ficus-indica* cultivars have high total antioxidant activity measured through ABTS (52.37–66.33 μmol trolox equivalents/g DM) and FRAP (40.39–65.33 μmol trolox equivalents/g DM) assays, and these values were comparable to those of other foods (*e.g.*, nuts and fruits) (Bensadón et al., 2010). This was attributed to their high content of phenolic (1.54–3.71 GAE/100 g DM) and carotenoid (15.16–22.84 mg β-carotene equivalents/g DM) compounds. These authors reported that polyphenols and some carotenoids could be bound to insoluble dietary fiber and the resulting complex may resist the gastrointestinal digestion and reach the gut almost intact. Therefore, by-products obtained from cladodes and fruits may combine the beneficial effect of both dietary fiber and antioxidant compounds in a single source, providing a product with multiple properties suitable as dietary supplement or food ingredient. However, considering the high content of dietary fiber in *Opuntia* spp. by-products (53.13–54.45 % w/w DW), comprehensive knowledge about the bioavailability of these bioactive compounds to ensure their physiological role is needed. Similarly, Melgar et al., 2017 identified twelve PC in the *Opuntia* spp. skin, which showed high correlation with all antioxidant activity assays (DPPH radical scavenging activity, reducing power and β-carotene bleaching), indicating that these bioactive compounds contributed to the total antioxidant activity (Melgar et al., 2017). As it can be observed from Table 2, *Opuntia* spp. skin is also a good source of dietary fiber, and thus the insoluble fraction could bind to the PC affecting their release during the passage through the gastrointestinal tract.

Sánchez et al., 2014 studied the antibacterial activity of cladodes from eight cultivars of cactus pear against *Campylobacter jejuni, Vibrio cholera, and Clostridium perfringens* (Sánchez et al., 2014). The minimum

bactericidal concentrations (MBC) of cladodes were in the range of 1.1–1.25 mg/mL (*C. jejuni*), 4.4–30 mg/mL (*V. cholera*), and 0.8–16 mg/mL (*C. perfringens*). These authors also reported that total phenolic content (TPC) in cladodes ranged 1.49–3.80 mg GAE/100 g DM and the total flavonoids content in cactus cultivars were within 15.4 and 36.6 mg QE/g DW. From these results, (Sánchez et al., 2014) hypothesized that PC in cladodes composition with demonstrated antimicrobial activity were responsible for such effect. Likewise, the antibacterial and antifungal activity of *Opuntia* spp. skin was studied against eight pathogenic strains. It was found that *Opuntia* extracts showed a higher inhibition capacity against the pathogenic microorganisms in comparison to ampicillin and ketoconazole or bifonazole drugs. This was also attributed to the phenolic composition of *Opuntia* spp. by-products whose antimicrobial effect could be explained by their ability to be adsorbed to cell membranes, interact with enzymes, or deprive of substrate and metal ions (Melgar et al., 2017).

4. Fermentation of residues as a valorization strategy

Cactaceae species have been processed for food and non-food applications, including diverse fields such as construction, energy, or agriculture. Such potential opens several possibilities for innovation, in which fermentation might play a key role because of both the occurrence of a variety of fermentable carbohydrates and derivatives in the by-products derived from these plants (*e.g.*, poly and oligosaccharides, mucilage) and the diversity of microorganisms able to use them as carbon sources (*e.g.*, lactic acid bacteria (LAB), fungus, yeasts). Among the different species belonging to this family, *Opuntia* has been the most studied genus for fermentation applications due to its high content in dietary fiber although some examples in other species can be found.

4.1. Microorganisms used in fermentation and fermentation design

Opuntia fruit seeds, skin, and spines are the most common residues arising from the use of pulps to prepare juices and other food products. Skin and spines are rich in mucilages, hydrocolloids that can be employed as thickeners and be fermented by microorganisms. In addition, the fragility and ease deterioration of *Opuntia* fruits (*e.g.*, almost neutral pH and high availability of carbon sources) may limit the large-scale marketing. Therefore, their fermentation with appropriate microorganisms appears as an appropriate strategy to add them value. Besides contributing to environmental sustainability, fermentation of *Opuntia* residues enables the release of many nutritionally relevant compounds, making them more bioaccessible and thus, bioavailable. In this regard, different species of lactobacilli and bifidobacteria can release polyphenols. Likewise, dehydrated skin from *Opuntia* has been used as substrate of fermentation, leading to products employed in animal feed. Tripodo et al., 2002 used skin and juices as substrates to produce food grade yeasts (*Saccharomyces cerevisiae*) and animal feed. Using *Geotrichum candidum* on these substrates allowed increasing the protein content of the feedstuffs up to 8.1 %. In addition, the digestibility of skin also increased as result of yeasts' fermentation (Tripodo et al., 2002). Skin and spines enable the production of the feeding of ruminants, stable for up to 21 days and with high content of fiber and non-fiber carbohydrates, soluble sugars, able to be fermented by LAB (cocci and bacilli) (T. C. do Santos et al., 2015; Todaro et al., 2020).

O. ficus-indica skin and spines were successfully used as carbohydrate feedstock to improve the production of baker's yeasts (Diboune et al., 2019). (Diaz-Vela et al., 2013) reported that skins from *O. indica* were suitable sources of carbon for fermentation using probiotic strains (*Pediococcus pentosaceus*, *Aerococcus viridans, Lacticaseibacillus rhamnosus*) that led to the increase of total fiber, which is an appropriate carbon source for LAB, also having prebiotic properties. Fermenting *Opuntia* spp. cladodes with strains of *Lactiplantibacillus plantarum* (CIL6, POM1 and 1MR20), *Levilactobacillus brevis* (POM2 and POM4), *Lactobacillus rossiae* 2LC8 and *Pediococcus pentosaceus* CILSWE5 enhanced the production of γ-amino butyric acid, the antioxidant, and immunomodulatory properties, also retaining the levels of ascorbic acid and carotenoids (Filannino et al., 2016).

On the other hand, the richness of cladodes in lignocellulosic residues and proteins convert them in substrates of great potential for the fermentation by aspergilli fungi without supplementation with nitrogen, minerals, and vitamins. Growing *Aspergillus niger* in cladodes led to an increase in the protein content and a reduction of cellulose, hemicellulose, and lignin, which facilitated their degradability and the production of bioethanol (de Souza Filho et al., 2016). Growing *Lactobacillus diolivorans* in cladode hydrolysates (inoculum 5 %) was comparable with that obtained in MRS at industrial level and using cladode hydrolysates as the only sugar source showed a 1,3-propanediol production close to that of a medium having only glucose as the sugar source. This supports using cladode hydrolysates to produce 1,3 propanediol in a sustainable and cost-effective manner (J. S. de Santana et al., 2021). In turn, growing *Candida utilis* in cladodes' hydrolysates from *O. ficus-indica* also improved the total protein content of the biomass product as result of the addition of yeasts proteins to cladodes (Akanni et al., 2015).

Moreover, fermentation has been also used as a valorization strategy in other Cactaceae species. Growing *L. acidophilus* LA-05 or *Bifidobacterium animalis* ssp. *lactis* BB-12 in red pitaya pulp increased the content and bioaccessibility and antioxidant activity of phenolics and flavonoids (*e.g.*, catechin, epigallocatechin gallate, procyanidin B2) as well as that of organic acids resulting from bacterial metabolism (Mora-Cura et al., 2017; Morais et al., 2019). In addition, supernatants of red pitaya pulps fermented with *L. plantarum, P. pentosaceus and L. pentosus* strains demonstrated strong antifungal properties against *A. niger* and *Cladosporium sphaerospermum*. These properties were ascribed to a higher content of PC, released in supernatants after fermentation (Omedi et al., 2019). Similarly, red pitaya pulps fermented with *L. plantarum* FBS05 showed inhibitory effect on *Escherichia coli, Salmonella Typhimurium, Pseudomonas aeruginosa* and *Staphylococcus aureus* (Muhialdin et al., 2020).

4.2. Application of fermentation

4.2.1. Production of enzymes

Enzymes are mostly produced when Cactaceae species are fermented with different genera and species of fungi. Solid-state fermentation of *O. ficus-indica* with *A. niger* potentiated the release of industrially relevant and stable xylanases, β-glucosidases (Tamires Carvalho dos Santos et al., 2018), amyloglucosidases (R. S. M. de Santana et al., 2012) and cellulases (Oliveira et al., 2001) in a cost-effective manner. In turn, the growth of *Acremonium* sp. from Antarctica in *O. ficus-indica* led to the production of extracellular proteases with promising biotechnological applications (Nascimento et al., 2015).

4.2.2. Aroma enhancement

Aroma compounds mostly result from the fermentation of *Opuntia* spp. with LAB and yeasts, especially when producing alcoholic beverages. The composition of volatiles is strongly determined by the species within the genus *Opuntia* employed and from the color, size, seed content, sugars, proteins, fats, pectins and non-volatile organic acid present in the fruits, which change during ripening. Such composition is different from that of other fruits because of the higher content of nonen-1-ols, which produce a melon-like flavor (Arrizon et al., 2006). Fermentation of *O. humifusa* with LAB led to an increase in the content of isorhamnetin, quercetin, total polyphenols, and total flavonoids. In addition, fermented extracts of *O. humifusa* increased the α-glucosidase inhibition activity, increased the overall sensory acceptability, the DPPH radical scavenging activity and the glucose tolerance (Park et al., 2021). Solid-state fermentation of *O. ficus-indica* with *Klyuveromyces marxianus* produced nine fruity aroma compounds, including alcohols, esters and aldehydes, ethyl acetate (responsible for the fruity aroma), ethanol and acetaldehyde being the major compounds produced (Medeiros et al.,

2000).

Alcoholic fermentation of prickly pear juice from *O. hemifusa* with *Pichia fermentans* and *S. cerevisiae*, followed by malolactic fermentation with *Oenococcus oeni* led to the production of many aroma compounds, including ethyl acetate (etherial, fruity, sweet, grape), 2-methylpropan-1-ol (bitter), 3-methylbutyl acetate (fruity, particularly banana), ethyl hexanoate (fruity, strawberry, anise), isopentanol 3-methylbutan-1-ol (alcoholic, pungent, etherial, cognac, fruity, banana and molasse), ethyl octanoate (fruity, floral, green leafy, menthol, anise), 3,4-dimethylpentan-1-ol, ethyl decanoate (grape, fruity, particularly apple), ethyl dec-9-enoate (fruity, sweet), 2-phenylethyl acetate (sweet, honey, floral rosy, cocoa, and balsamic nuance) ethyl dodecanoate (fatty and fruity), 2-phenylethanol (sweet, honey-like, yeast-like, floral, spicy), octanoic acid and decanoic acid (Navarrete-Bolaños et al., 2013; Rodríguez-Lerma et al., 2011). Other authors fermented *O. hemifusa* with *Citrus junos* and *Psidium guajava* L, observing changes in the content of isoharmnetin, quercetin and total polyphenols and flavonoids, and a decrease in the concentration of hesperidin and naringin (Park et al., 2021).

4.2.3. Bioactive compounds production

The most studied bioactive properties of fermented *Opuntia* wastes (pulp, skin, cladodes, spines) are those associated with antioxidant and immunomodulatory properties through modulation of cytokine secretion, which opens several perspectives for innovation in the development of diverse products with health benefits (Barba et al., 2020). Lactic acid fermentation can increase the content of bioactive peptides, short chain fatty acids or polysaccharides, whereas the contents of sugar or antinutritional compound can decrease. During lactic acid fermentation, PC are converted into substances with increased biological value, and the fermented products are also sources of pre and probiotics, with well-known beneficial effects (Barba et al., 2020).

Fermentation of cladodes with *Lactiplantibacillus plantarum* and *Bacillus subtilis* was reported to enhance the production of nitric oxide, cytokines secretion, nuclear factor-κB (NF-κB) activity, and mitogen-activated protein kinase (MAPK) phosphorylation in RAW 264.7 cells, thus supporting a promising use as immunostimulatory therapeutics (Filannino et al., 2016; Hwang & Lim, 2017). In the same line, fermentation with *Leuconosctoc mesenteroides* markedly inhibited the inflammatory status of Caco-2/TC7 cells, maintaining the integrity of tight junctions and showed antioxidant properties (Di Cagno et al., 2016). Administration of *O. ficus-indica* fermented with *L. plantarum* to obese mice decreased the body weight gain, improved the insulin resistance, and reduced the hyperglycemia and hyperlipemia associated to obesity (Verón et al., 2019).

5. Trends and applications of Cactaceae family residues in the food industry

In recent years, technological applications of some species of cacti have been noted, as they constitute a good source of healthy food and natural food ingredients. As mentioned before, and due to its diversity and economic importance, one of the most studied genera of the Cactaceae family is *O. ficus-indica* which is abundant in dietary fiber and natural antioxidants. Their by-products, such as cladodes and fruits, may be suitable for inclusion in functional foods (Bensadón et al., 2010; El Kossori et al., 1998; Jiménez-Aguilar et al., 2015). Seeds of the *O. ficus-indica* varieties contained a significant amount of oil and unsaturated fatty acids (Chougui et al., 2013). Their peels can be incorporated into food products such as beverages, capsules, and colorants (Jiménez-Aguilar et al., 2015). However, by-porducts from other species of belonging to Cactaceae family have been tested for industrial applications in the food industry. Among them, *H. polyrhizus, H, undatus, Cereus jamacar* and *Pilosocereus gounellei* can be highlighted (Table 3).

Bakery is one of the food industries where cacti by-products have been used. Fortifying wheat flour with cladodes powder from *O. ficus-*

Table 3
Applications of Cactaceae family residues in the food industry.

Species	Product	Purpose	Result	Reference
Opuntia ficus-indica	Bakery	Increase dietary fiber and increase water absorbency in flour	Dough showed better functional properties	(Ayadi et al., 2009)
	Rice- and corn-based snacks	To fortify extruded products	Increase in the content of flavanols	(Moussa-Ayoub et al., 2015)
	Wine	To obtain high-quality fermented beverages	Produce a fermented product with a unique flavor and taste	(Rodríguez-Lerma et al., 2011)
	Edible films	To produce carboxymethyl cellulose (CMC) edible films	High concentration of aqueous extract and peel powder in CMC films increased the bioactive compounds content, antioxidant capacity	(Aparicio-Fernández et al., 2018)
	Kiwifruit	Packaging	The use of mucilage coating improved the quality of products such as firmness, ascorbic acid, pectin contents and flavor	(Allegra et al., 2016)
Hylocereus polyrhizus	Bread	Use of flour as a fat replacer	The formulated bread showed acceptable physical characteristics and sensory response	(M Utpott et al., 2018)
	Ice cream	To enhance the antioxidant properties of ice cream	Increased in color acceptability and functional properties	(Gengatharan et al., 2021)
	Ice cream	Use as a fat replacer	Reduced the fat content of ice cream and improved acceptability and viscosity	(Utpott et al., 2020)
	Beer	Intelligent packaging by incorporating betalains	Monitored the freshness of beverage	(Qin et al., 2020)
Cereus jamacaru	Bread	Fruit skin flour for bread formulation	More favorable taste and color	(Nascimento et al., 2015)
Pilosocereus gounellei	Cake	Partial replacement of wheat flour	The cake observed that formulations containing 20, 40 and 60 % of the flour of this species had greater acceptability	(da Silva, 2019)
Cereus jamacaru and *Opuntia ficus-indica*	Ice cream and yogurt	Better physical properties and development of by-products	The formulation showed desirable physicochemical properties and high content of vitamin C	(de Fidelis, 2015)

(*continued on next page*)

Table 3 (*continued*)

Species	Product	Purpose	Result	Reference
Hylocereus undatus	Fruit juice	To produce lacto-fermented dragon fruit juice with high biological activity	Improved the functional properties, consumer acceptability and shelf life	(Muhialdin et al., 2020)

indica increased dietary fiber content, resulting in increased water absorbency in flour and altered properties of the dough. The cladodes powder increased dough and cake tenacity and considerably decreased their elasticity. According to the baking test, cladodes incorporation resulted in a significant change in cake quality and aspect and it also influenced the cake's crust color and tone (Ayadi et al., 2009). Cakes made with cladodes flour scored poorly on sensory evaluation compared to control cakes except when it was 5 % of total flour content (Ayadi et al., 2009; de Waal et al., 2015). The same results were obtained for bread using cladode flour mixed with whole wheat flour (Msaddak et al., 2017). In another study, 10 % flour from the *Cereus jamacaru* fruit skin was used for producing a bread product more appealing, flavorful, and with improved color (M. A. G. do Nascimento, 2014). The loaf bread flour from *Hylocereus polyrhizus* skin resulted in low specific volume and lightness value and high redness and yellowness values. It showed higher crumb and crust firmness, integrity, plasticity, and gumminess with excellent sensory response (M Utpott et al., 2018). Cookies and carrot cakes made with cladode flour replaced with respectively 10 % and 25 % of flour resulted in acceptable taste and texture (de Waal et al., 2015). Cake formulated with specific percentages of *Pilosocereus gounellei* powder showed high acceptance (C. E. da Silva, 2019).

Cacti have been also used in ice cream and yogurt production. Some studies have been conducted in line with this concept. Studying yogurt formulations based on adding the pulp or peel of *C. jamacaru* and *O. ficus-indica* showed the highest percentage of soluble solids, pH, total acidity, and content of vitamin C (de Fidelis, 2015). *O. ficus-indica* is an excellent alternative to produce ice cream due to its state of acidity, sugary taste, aspects of nutrition, and colors. Adding the pulp to ice cream produced a very suitable product, so it would be feasible to produce *O. ficus-indica* ice cream on a large industrial scale (El-Samahy et al., 2009). The ice creams made from *C. jamacaru* and *P. gounellei* pulp are technically suitable for manufacturing. Overall, all formulations demonstrated good sensory acceptability. Gengatharan et al., 2021 reported that betacyanins from *H. polyrhizus* act as a functional natural colorant that can be used in ice cream, increasing the color acceptability of the product (Gengatharan et al., 2021). Using *H. polyrhizus* skin flour in another study reduced the fat content of ice cream and improved acceptability and viscosity (Utpott et al., 2020). Juice concentrates from *O. ficus-indica* had the potential for using in ice cream or yogurt preparations as a proper coloring aliment (Moβhammer et al., 2006). *O. ficus-indica* juice is a convenient way to consume the fruit. However, because of its high pH value, juice requires a stabilization treatment to preserve its microbiological quality (Barba et al., 2017). A study has recently assessed clarifying effects on *O. ficus-indica* fruit juice composition. The results showed that the microfiltration (MF) and ultrafiltration (UF) processes did not impact soluble solids, pH, or acidity (Cassano et al., 2010). Fermentation of *H. undatus* juice with *Lactobacillus plantarum* showed high potential for being marketed as a functional drink. Its antibacterial activity was significantly improved by fermentation. Also, the antioxidant activity of fermented juice was higher than fresh juice. This study showed that consumers highly accepted fermented juice with fresh fruit and had a low microbial load while retained a steady shelf life (Muhialdin et al., 2020).

Sarkar et al., 2011 reported that the utilization of prickly pear fruit solids mixed with rice flour can improve the quality of extruded products (Sarkar et al., 2011). Incorporating *O. ficus-indica* peel acquired during processing with rice or maize flour could be used to produce cereal-based snacks high in flavanols (Moussa-Ayoub et al., 2015). It was found that the improved *O. ficus-indica* snacks had a more significant nutritional effects than either the rice-based snacks or the corn-based snacks. Moreover, the snacks obtained by this process were well received by consumers. There was also a significant increase in the amount of β-carotene, improving both polyphenolic content and antioxidant activity (Namir et al., 2017). Cacti also have the potential to use them in the wine and beer industry. A traditional method of fermenting *O. ficus-indica* juice provides low-alcohol beverages, which can be further processed into spirits and vinegar by a subsequent acetic fermentation (FAO, 2013). In a different study, a selected mixed culture (*P. fermentans and S. cerevisiae*) for the fruit of prickly pears fermentation produced a wine with distinct flavor and aroma characteristics. This study showed the existence of major volatile compounds essential for a delicate wine flavor (Rodríguez-Lerma et al., 2011). de Waal et al. (2015) reported that fermented beer made from maize or sorghum was produced to replace 25 % of the flour with cladode powder of *O. ficus-indica* (de Waal et al., 2015).

Food Packaging is one of the essential activities in food industry where cacti can play an important role. Novel food packaging films were manufactured by adding betacyanin-rich *H. polyrhizus* peel extract into starch/polyvinyl alcohol. These films are also used as intelligent packaging to monitor the freshness of protein-rich animal foods (Qin et al., 2020). Biodegradable food packaging materials combined with beneficial antioxidant combinations can be produced from *O. ficus-indica* peel. The addition of red prickly pear peel powder and its aqueous extracts significantly impacted the physical and antioxidant features of carboxymethyl cellulose (CMC) palatable films. The developed palatable films showed the existence of betalains and PC (Aparicio-Fernández et al., 2018). Mucilage from *O. ficus-indica* and *O. elatior* has been used for several industrial applications. Using this biopolymer as packaging to ensure food safety and quality will extend new opportunities and bring new trends to the food packaging market. Whether used as palatable film and coating, the use of cacti mucilage could also be economically profitable due to its low price, accessibility, and usefulness when used as primary packaging for food products and because of its advantageous effects could be used for packaging applications in the future (Gheribi & Khwaldia, 2019).

6. Conclusions

Cactaceae family comprises different genus of succulent plants adapted to cope with severe environmental conditions, including high temperatures, drought, and poor nutrient availability. Among its four subfamilies, Cactoideae and Opuntioideae are the most representative with *Hylocereus* and *Opuntia* as the most known genera. The residues from processing Cactaceae species can be considered as a source of bioactive compounds, namely phenolic compounds (hydroxycinnamic and hydroxybenzoic acids, flavonoid glycosides, tannins, coumarins, anthocyanins, and stilbenes), alkaloids (isoquinoline and phenethylamine derivatives), terpenoids (triterpenoids, phytosterols, and carotenoids) and other compounds such as fatty acids, tocopherols, or polysaccharides (mucilage). However, it has been estimated that this discarded biomass has a higher content of dietary fiber than their corresponding commercial edible parts so they can be considered as excellent substrate for fermentation. The content of dietary fiber depends on different factors such as the part of the plant, cultivar, geographical location, or maturity stage, but their health benefits have been associated to both soluble and insoluble dietary fiber. Different microorganisms can be used for fermentation of residues, but lactic acid bacteria should be highlighted. At last, Cactaceae residues have been exploited for the formulation of new products of the food industry mainly bakery and dairy products, but also fermented drinks and food packaging systems. Taking all together, Cactaceae species could be exploited for different industrial sectors including food, technological,

pharmaceutical, and chemical applications because of their high content in fiber and active compounds. Nevertheless, studies are limited to few species and more research is still needed in terms of using fermentation as a valorization strategy.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

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ARTICLES FOR FACULTY MEMBERS

FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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Bioactive Compounds in Underutilized Vegetables and Legumes

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Preface

Vegetables and legumes are the major components of a balanced human diet and they provide essential nutrients such as carbohydrates, proteins, fat, vitamins, and minerals. Researches are in the exploration of various plants other than traditional vegetables and legumes to meet the global food demands and promote the plants which have been neglected and underutilized. Nutritional and phytochemical analysis of many neglected and underutilized vegetables and legumes has revealed that they are source nutrients, proteins, and vitamins. Further, underutilized vegetables and legumes are also proved to be rich in valuable secondary metabolites which account for bioactive principles.

This book encompasses research work on bioactive compounds of underutilized vegetables and legumes across the globe to present the latest research on these plants for the enhanced appreciation of this topic. The chapters presented in this volume throw light on several research subjects that have provided critical information on the synthesis of plant secondary metabolites and their bioactive principles specifically from underutilized/neglected vegetables and legumes. Each chapter also provides background information of the plant, parts used, and their nutritional composition, chemical compounds, and their biological activities. Self-explanatory illustrations and tables have been incorporated in each chapter complementary to the main text. The topics included in this volume are not intended to be comprehensive, but our approach has been wide-ranging, presenting the most exciting aspects of bioactive compounds from underutilized vegetables and legumes.

We would like to thank and express our deepest gratitude to all contributors who helped us to complete this book. We also thank Professor Jean-Michel Merillon and Professor Kishan Gopal Ramawat, Series Editors, for their constant encouragement. We thank Dr. Sylvia Blago, Dr. Sofia Costa, and Dr. Johanna Klute for their support. Finally, we also express indebtedness and thankfulness to the Springer team for completing this assignment successfully.

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Bioactive Compounds of Barbados Gooseberry (*Pereskia aculeata* Mill.)

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Mariana Buranelo Egea and Gavin Pierce

Contents

Abstract

Pereskia aculeata Mill., a species of the family Cactaceae, is considered a non-conventional leafy vegetable. This species has been consumed mainly for its protein and mineral (iron and calcium) content, and represents a strategy for improving the nutritional value of diets in rural communities. *P. aculeata* protein is of high quality (with an abundance of the essential amino acids lysine and tryptophan) and high digestibility. Additionally, *P. aculeata* seems to be a promising thickening ingredient as it is rich in a mucilage, which has potential to serve as a functional food ingredient that contributes to favorable sensory properties and to dietary soluble fiber requirements. Further, *P. aculeata* leaves are an abundant source of vitamin C, the provitamin A carotenoids α- and β-carotene, and several xanthophylls, including lutein and violaxanthin. Additionally, the *P. aculeata* essential oil contains several terpenes and terpenoids commonly found

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in herbs and spices used as culinary seasonings and in traditional medicinal practices. The objective of this chapter was to gather information on the active compounds present in *P. aculeata* leaves that can demonstrate beneficial health effects.

Keywords

Cactus · Carotenoids · α-carotene · Hydrocolloid · Leafy vegetable · Lutein · Mucilage · Tryptophan

Abbreviations

AI	Atherogenic index
ABTS	2,2′-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid)
DPPH	2,2-diphenyl-1-picrylhydrazyl
FM	Fresh matter
GAE	Gallic acid equivalent
MRSA	Methicillin-resistant *Staphylococcus aureus*
PI	Protection index
RAE	Retinol activity equivalents
RDA	Recommended daily allowance
TBARS	Thiobarbituric acid reactive substances

1 Introduction

Nonconventional vegetables are valuable alternatives to traditional food crops due to their abundant content of macro- and micronutrients. The use of non-conventional vegetables has been increasing, mainly due to the increase in adherence to vegan or vegetarian diets for health reasons, and increasing consumer awareness and concern with climate change [1, 2]. *Pereskia aculeata* Mill., a species of the family *Cactaceae*, grows naturally in the American continent, South and Southeast Africa, Northeast and Southeast Australia. It is popularly known as "Barbados Gooseberry" and in Brazil such as "ora-pro-nobis" (Latin, pray-for-us) [3–5]. The flower is perigynous and presents a hypanthium with bracteoles and aculeus. The fruit is pomaceous, type cactídio, with succulent hypanthium, pericarp, and seeds immersed in a yellowish gelatinous mass. The seed is exotestal and develops from an amphitropous, bitegmic, and crassinucelate ovule [6, 7] (Fig. 1).

The common edible part is the leaves that has been used in local cuisine as a source of digestible vegetable protein, vitamins, and minerals. In general, the production and commercialization of *P. aculeata* leaves is still rudimentary, which hinders its establishment as an agricultural crop and value-added commercial product [8–11]. Among the bioactive compounds reported for *P. aculeata* leaves are phenolic compounds such as caftaric acid, rutin, and narcissin; carotenoids including pro-vitamin A carotenoids and bio-compatible xanthophylls, and

Fig. 1 Flowers and leaves (**a**) and ripe fruits (**b**) of *Pereskia aculeata*

betalains [12–14], among others, which contribute to the biological activity of this plant.

P. aculeata leaves have been used in the interior states of Brazil, mainly in Minas Gerais, as a traditional medicinal treatment for iron deficiency anemia, as a therapeutic agent for cancer, for the prevention or treatment of osteoporosis, and for the treatment of intestinal constipation [15]. However, few studies have yet demonstrated the biochemical components of *P. aculeata* that contribute to its healing action [16], apoptosis in breast carcinoma [17], antibacterial activity [13], anti-inflammatory activity [18], and antinociceptive potential [19].

As this plant has been reported as an important source of nutrients, it can be utilized as a practical vegetable for economically disadvantaged populations of Brazil, both in urban and rural environments to increase their nutrient intake [15, 20]. Several researches have been developing methods to include the leaves of *P. aculeata* as a functional ingredient in food products such as bread rolls [21], noodles [22], pasta dough [23–25], juices [26], ice cream [27], cake [28, 29], emulsified cooked sausages [30], and milk beverages [31], among others. The objective of this chapter was to gather information on the active compounds present in the leaves of *P. aculeata* that confer health effects.

2 Nutritional Composition and Antinutritional Factors

Table 1 shows the proximal composition *P. aculeata* leaves on a dry basis. Fresh leaves have an average of ~90% moisture [32], which can hinder its shelf life and utility as a fresh vegetable. The available literature highlights the large amount of proteins (~24%), lipids (4%), and carbohydrates (~40%), with emphasis on the high content of dietary protein (~24%) present in the dried matter.

As it is a vegetable source of essential amino acids, *P. aculeata* leaves may be an important contributor to the protein content of the human diet moving forward, to reflect changes in consumer preferences. A major concern when considering potential plant-based protein sources is the quality of the protein sources provided. Dietary protein quality is rated based on the essential amino acid composition of a protein, as it relates to human needs, in addition to the ability of the protein to be digested, absorbed, and retained by the body [39]. *P. aculeata* leaves have shown less true protein digestibility (~75%) [40, 41] than casein (96%) [40], however, this value is comparable to other hallmark plant sources of protein such as soy, oat, and quinoa [42]. Additionally, protein digestibility can be modified with different methods of

Table 1 Chemical composition of *P. aculeata* leaves

	Average	Median	Range	References
Macronutrients				
Moisture (g 100 g^{-1})	8.3	7.6	5.9–12.5	[21, 24, 25, 30, 33, 34]
Protein (g 100 g^{-1})	23.8	22.9	12.4–40.7	[21, 22, 24, 25, 30, 32–35]
Lipid (g 100 g^{-1})	4.0	3.7	2.4–5.2	[21, 24, 25, 30, 32–34]
Ash (g 100 g^{-1})	16.6	16.1	14.8–18.8	[21, 24, 25, 30, 32–34]
Carbohydrate (g 100 g^{-1})	43.7	45.5	29.5–59.0	[21, 24, 25, 33, 34]
Total dietary fiber (g 100 g^{-1})	30.2	39.1	8.7–41.8	[21, 25, 32, 34, 36]
Soluble fiber (g 100 g^{-1})	4.3	5.2	2.4–5.2	[32, 34, 36]
Insoluble fiber (g 100 g^{-1})	29.0	33.9	19.2–33.9	[32, 34, 36]
Energy (g 100 g^{-1})	288.2	271.8	269.2–323.6	[21, 24, 33]
Total sugar (g 100 g^{-1})	48.0			[35]
Uronic acid (g 100 g^{-1})	26.0			[35]
Micronutrients				
Iron (mg 100 g^{-1})	19.8	15.6	9.4–38.7	[23, 25, 37]
Zinc (mg 100 g^{-1})	5.7	5.9	4.0–7.3	[23, 37]
Calcium (mg 100 g^{-1})	2679.3	2880.0	1346.7–3660.0	[23, 25, 37, 38]
Magnesium (mg 100 g^{-1})	1065.3	680.0	450.0–2560.0	[23, 25, 37, 38]
Phosphorus (mg 100 g^{-1})	447.4	320.0	150.0–1130.0	[23, 25, 37, 38]
Potassium (mg 100 g^{-1})	3266.0	3380.0	2420.0–3910.0	[23, 25, 37, 38]
Copper (mg 100 g^{-1})	0.7	0.9	0.1–1.2	[23, 37]
Boron (mg 100 g^{-1})	4.1	4.1	2.8–5.5	[23, 37]
Manganese (mg 100 g^{-1})	23.6	24.5	2.8–43.5	[23, 37, 38]
Sulphur (mg 100 g^{-1})	640.8	716.7	150.0–980.0	[23, 25]

food preparation, such as cooking time or drying [32, 43], and may be underestimated since fermentable fibers increase the activity of intestine microbiome and may indirectly increase protein digestibility [40].

The amino acid profile (mg g^{-1} of protein) of *P. aculeata* leaves can contain different amounts of phenylanine + tyrosine (53.7 and 84.4), leucine (69.0 and 66.3), valine (50.1), lysine (53.4 and 41.7), isoleucine (36.9 and 40.87), threonine (30 and 36.64), methionine + cystine (17.18 and 22.26), histidine (24.0 and 16.23), and tryptophan (21.1 and 5.10), as reported by Zem et al. [40] and Silveira et al. [41], respectively. Silveira et al. [41] highlight the presence of leucine (6.96%), lysine (5.37%), and phenylalanine (5.02%), while Takeiti et al. [32] and Zem et al. [40] highlight tryptophan in *P. aculeata* leaves. This amino acid profile demonstrates that this leaf has similar essential amino acids to legumes, mainly for lysine (high in beans) and tryptophan (high in garbanzo). Still, proteins from *Pereskia* are classified as incomplete, as they are insufficient sources of some essential amino acids [41]. This highlights the importance of a diverse diet in order to obtain the full spectrum of essential amino acids.

For adults between 19 and 50 years old, the contribution of 40 grams of *P. aculeata* dried leaves to the RDA for micronutrients for females and males [44], respectively is 524 and 410% of manganese, 107% of calcium (for both genders), 137% and 107% of magnesium, 50% and 38% of potassium, 44% and 99% of iron, 26% of phosphorus (for both genders), and 29% and 21% of zinc. *P. aculeata* leaves appear to be an excellent source of minerals and may even serve as a food supplement in areas where human malnutrition is a problem. Vegetarian and vegan diets often contain low amounts of calcium that can be resolved with ingestion of 40 grams (~2 spoons) of *P. aculeata* dried leaves. Dietary calcium optimizes bone density and protect against bone resorption reducing risk of osteoporosis [45, 46].

P. aculeata leaves showed ~20 mg 100 g^{-1} FM of folic acid, ~186 mg 100 g^{-1} FM of vitamin C (determined using titration method) [32], and 439 μg 100 g^{-1} FM of vitamin E including α-tocopherol was the major 91%, followed by γ-tocopherol (5%), α-tocotrienol (3%), and β-tocopherol (1%) [47].

The antinutritional factors studied for *P. aculeata* are oxalic acid, saponins, trypsin-inhibiting tannins, and hemagglutination activity. Oxalic acid has been reported between 41 and 99 mg 100 g^{-1}, total tannins between 1813 and 3804 mg 100 g^{-1}, trypsin inhibitory activity between 0.2 and 1.8 UTI mg^{-1}, saponins of 0.3 mg 100 g^{-1}, and no hemagglutination activity [34, 41, 43]. Although it has become better known and introduced in food recently and there is a concern with the presence of antinutrients and alkaloids, the toxicity of *P. aculeata* extracts against the liver primary culture PLP2 [13] or growth and development of Wistar rats has not been reported [37], demonstrating that it is safe for human consumption.

2.1 Polysaccharides in *P. aculeata* Leaves

P. aculeata leaves and fruits have been reported as a plant source of hydrocolloids that are commonly referred to as mucilage. This mucilage mainly contains type I

arabinogalactan with partially esterified galacturonic acid and fucose, and to a lesser extent galactose, rhamnose, and galacturonic acid, which are highly ramified [35, 48–50]. The composition of the mucilage from *P. aculeata* leaves has been linked to wound healing properties [16, 51, 52].

The *P. aculeata* mucilage generally contains the saccharides arabinose, galactose, rhamnose, and galacturonic acid in a mole ratio of 5.1: 8.2: 1.8: 10 [48]. The mucilage contains abundant carboxyl groups, which may serve as ion binding sites that contribute to its gel-forming ability (ability to interact with water to form a proper gel) [53]. Under standard conditions, *P. aculeata* hydrocolloids obtained by hot water extraction exhibits apparent viscosity values that decrease with the shear rate until stabilization (pseudoplastic fluid) and thixotropic behavior that increases with relation to mucilage concentration. The addition of salts to *P. aculeata* mucilage decreases viscosity by the presence of positive ions that reduce repulsion and molecule expansion. Conversely, viscosity and thermal stability are increased by the presence of sucrose [54].

The mucilage of *P. aculeata* possesses characteristics similar to Arabic gum and cashew gum, which are commonly utilized in the food industry as a beverage thickener and to replace fat in fermented milk products [31]. Addition of this arabinose- and galactose-rich mucilage to films provides increased tolerance to salinity, but the surface of the films the demonstrated the low capacity of the binding of Arabic gum over the fibers of cellulose [55, 56].

The dietary fiber content in the leaves of *P. aculeata* is ~23 g 100 g^{-1}. *P. aculeata* has been extensively utilized in traditional medicine for its beneficial health effects. Among the applications of this plant, the literature has highlighted it as a source of bioactive fiber, with the potential to increase intestinal motility, reduce total body weight and visceral fat gain, increase the protection index (PI), and reduce the Atherogenic index (AI) (per Lee index) to confer heart protection, as has been demonstrated in mice [33]. The beneficial properties of the fiber were confirmed by Vieira et al. [57], in their in vivo study (men, 20–50 years old) which found the improvement of gastrointestinal symptom rating scores, flatulence reduction, and increase of satiety when cookies with *P. aculeata* flour (36 g of cookie providing 6.42 g fiber day^{-1}) were consumed during a14-week period.

2.2 Lipids in *P. aculeata*

Lipids comprise 0.4% of fresh *P. aculeata* leaves, and ~4.0% of the dried leaf matter (Table 1), as determined using the Soxhlet method [32]. These levels are comparable to other leafy green vegetables, as lipids represent 0.4% of the weight of fresh spinach leaves (*Spinacia oleaceae*). Hydrodistillation of the dried leaves yielded 0.02% essential oil, indicating that *P. aculeata* leaves primarily contain non-volatile lipids. Modest quantities of tocopherols and tocotrienols have been found in fresh *P. aculeata* leaves (436.68 μg 100 g^{-1}). While the quantities of tocopherols and tocotrienols are unlikely to represent a significant contribution to the human diet,

the primary vitamin E compound found in tested samples was α tocopherol, the most bioactive vitamin E molecule, at 400.34 μg 100 g^{-1} [18].

2.2.1 *P. aculeata* Essential Oils

The composition of the essential oil on *P. aculeata* leaf lipids consists of monoterpenes, diterpenes, hydrocarbons, and select fatty acids. Dietary fatty acids found in *P. aculeata* leaves are primarily palmitic acid (16:0) and linoleic acid (18:1, ω-6) [58, 59]. The essential oil of *P. aculeata* leaves consist primarily of terpenes and terpenoids. Many of the terpenes and terpenoids present within *P. aculeata* are found in common medicinal plants, herbs, and spices.

Essential oils constitute a minor portion of *P. aculeata,* leaves, but they may represent a contributor to the bioactive properties of the vegetable in vivo. Hydrodistillation of dried *P. aculeata* yields 0.02–0.03% of essential oils, comprised of 30 compounds. The composition of the *P. aculeata* leaves appear to be highly variable, as samples collected from the same campus (Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil) at the same time of year (October), from the same lab had dramatically different composition when harvested two years apart. Whether these differences are due to differences in the annual weather or inter or intra-plant differences remains to be determined. Two dietary fatty acids, linoleate and palmitate, are found in the essential oil. The remaining compounds in the essential oil include oxygenated monoterpenes, oxygenated sesquiterpenes, oxygenated diterpenes, diterpene hydrocarbons, hydrocarbons, and phytol. Aside from fatty acids, the most abundant compounds in samples analyzed are phytol (29.4%) or acorone (30%) depending on the study used for reference. As both the samples in Table 2 are from the same lab, it is possible that the acute differences in lipid composition are due to annual changes in weather. The essential oil has been demonstrated to act as a bactericidal to *Bacillus cereus, Bacillus subtilis, and Staphylococcus epidermidis* ATCC 25923 at 100 μg/mL, although it does not appear to act as a bactericidal agent towards Gram-negative bacteria [59].

2.2.2 Carotenoids

The leaves and fruit of *P. aculeata* are both exceptionally rich in carotenoids, rivaling other common dietary sources (Table 3). The leaves of *P. aculeata* are especially rich in the xanthophylls, lutein, and zeaxanthin, while the fruits are rich in both α- and β-carotene, thus providing pro-vitamin A activity. A useful tool for determining the vitamin A activity of foods and supplements is retinol activity equivalents (RAE), where the standard is 1 μg of retinol. Absorption of carotenes is critical for vitamin A activity to be obtained, with dietary fats facilitating the absorption of carotenes. β-carotene is absorbed more readily than α-carotene, and thus confers greater provitamin A activity, Supplemental carotenes are not bound in a food matrix, and are absorbed six times higher than dietary carotenoids, and so dietary β-carotene has a RAE ration of 12:1, and α-carotene has a RAE of 24:1. The presence of abundant α-carotene is relatively uncommon in popular edible fruits. Leaf carotenoids appear to fluctuate in response to sunlight, with shady conditions favoring increased production of all

Table 2 Lipids in essential oil of *Pereskia aculeata* obtained via hydrodistillation (mg 100 g^{-1})

Essential oil compounds	[59]	[58]
(5*E*,9*E*)-Farnesyl acetone		5.70
(*E*)-β-Ionone	0.1	0.75
(*Z*)-3-Hexenyl salicylate		0.17
(*Z*,*Z*)-Methyl-4,6-hexadecanolide		16.34
14-Hydroxy-(*Z*)-caryophyllene	0.6	0.29
14-Hydroxy-4,5-dihydro-caryophylle	1.6	
14-Hydroxy-9-*epi*-(*E*)-caryophyllene	0.6	0.28
1-Hexadecene	0.3	
1-Nonadecen-ol	6.18	
1-Octadecene		0.62
1-Tetradecene	0.2	
2-Hexyl-(*E*)-cinnamaldehyde		0.60
2-Ethylhexyl salicylate		1.73
6-Methyl-α-ionone	7.2	
Acorone		30.0
ar-Tumerone		1.10
Caryophyllene oxide	0.3	0.51
cis-Dihydro-mayurone	Trace	0.17
cis-Thujopsenal	0.9	
Citronellyl butyrate	0.3	
Cyclopentadecanolide	Trace	5.48
Dihydro-β-agarofurann		0.57
Ethyl hexadecanoate	0.6	
Eudesma-4(15),7-dien-1β-ol	0.3	
Heptadecane	1.9	
Hexadecanoic acid	17.4	
Isopropyl hexadecanoate	0.7	0.42
Linoleic acid	12.7	4.74
Methyl hexadecanoate	2.6	4.92
Methyl isovalerate	0.4	
Methyl linoleate	3.0	4.44
Methyl octadecenoate		0.69
n-Eicosane	2.9	
n-Hexadecene	1.3	
n-Octadecane	1.0	
Nonadecane	2.9	
n-Pentadecane	0.3	
Phytol	29.4	5.11
α-Cadinol	0.9	
α-Muurolol	0.5	0.22

carotenoids measured. Total carotenoids shifted from 119 μg g^{-1} in the leaves when grown in full sun to 210 μg g^{-1} when grown in half shade. The profound difference in leaf carotenoid content suggests that growing methods could be

Table 3 Carotenoids in *P. aculeata* leaves and berries (μg g^{-1})

Compounds	Leaves, sun [14]	Leaves, shade [14]	Berries [14]	Berries [61]
Lutein	57.4	102	2.3	6.5
α-carotene	3.8	11.9	18.9	22.7
α-cryptoxanthin/ zeaxanthin				2.7
(allE)β-carotene	18.6	35.1	29.2	34.3
(9Z)β-carotene	3.0	5.1		
(13Z)β-carotene	0.7	2.9	3.0	
Cis-β-carotene				2.8
Neoxanthin	9.3	16.1		
Violaxanthin	20.0	27.3		
Lutein-like	2.5	5.0		
Zeaxanthin-like	3.3	5.3		
Total	186	375	53.3	71.7

manipulated to yield exceptionally high-carotenoid leaves, for use as a vegetable or dietary supplement [14].

The quantity of xanthophylls in the leaves is substantial. Lutein and zeaxanthin are known to aculeate in the macula to protect the eye from blue light [60]. Although xanthophylls do not have any provitamin-A activity, their biocompatibility and protective properties in vivo have prompted the production of lutein + zeaxanthin supplements, which generally contain up to 20 mg of lutein, approximately the same amount as is supplied by 100 g of cooked kale (18 mg). Raw shade-grown *P. aculeata* leaves contain over five times as much lutein as cooked kale leaves, making this vegetable a richer source of lutein than any commonly consumed leafy vegetables on a w/w basis.

2.3 Phenolic Compounds and Antioxidant Activity in *P. aculeata*

Phenolic compounds vary from 5 to 118 mg GAE g^{-1} in *P. aculeata* leaves, according to the solvent used in the extraction (petroleum ether, chloroform, methanol, water, ethanol, or acetone) [13, 34, 62]. In the *P. aculeata* berries, phenolic compounds were ~65 mg GAE g^{-1} [61]. Phenolic compounds are known for their high antioxidant activity. In the extract of *P. aculeata* leaves the antioxidant activity has ranged from 7 to 107 mg mL^{-1} to sequestre 50% of DPPH radicals [13, 34, 58, 62], IC_{50} of 40 μg mL^{-1} using ABTS method, IC_{50} of 39 μg mL^{-1} using TBARS method, and 63–82% of inhibition of β-carotene oxidation [62].

Among the compounds found in the leaves are (in ascending order of amount) (mg g^{-1}): *cis* caftaric acid (9.5), quercetin-3-*O*-rutinoside (3.56), isorhamnetin-*O*-pentoside-*O*-rutinoside (2.27), *trans* caftaric acid (2.22), quercetin-*O*-pentoside-*O*-rutinoside (2.11), isorhamnetin-3-O-rutinoside (1.30), kaempferol-3-*O*-rutinoside

(0.81), quercetin-*O*-pentoside-*O*-hexoside (0.74), isorhamnetin-*O*-pentoside-*O*-hexoside (0.63), and caffeic acid derivative (0.57) [13].

Caftaric acid, an ester form of caffeic acid, was the most abundant in *P. aculeata* leaves, and this phenolic acid has demonstrated to have excellent antioxidant and anti-inflammatory activity [63]. Rutin was the second most abundant phenolic compound reported for *P. aculeata*. This compound has been linked to its antibacterial activity more active against Gram positive bacteria (*Enterococcus faecalis*, *Listeria monocytogenes*, and MRSA – Methicillin-resistant *Staphylococcus aureus*) than against Gram-negative bacteria (*Escherichia coli*, *Klebsiella pneumoniae*, *Morganella morganii*, *Proteus mirabilis*, and *Pseudomonas aeruginosa*) [13].

3 Conclusions

P. aculeata has a potential as a vegetable, functional food, and source of bioactive compounds including protein, mucilage, and carotenoids. While the protein in *P. aculeata* is not considered complete, the abundance of lysine and tryptophan make it an excellent complimentary protein, as many plant proteins are deficient in these amino acids. Further, *P. aculeata* leaves serve as a rather unusual delivery method for plant proteins, providing the potential for enrichment of dishes that are not traditionally protein rich.

There are several other components of *P. aculeata* that position the plant as a unique health-promoting vegetable. In addition to being high in protein, this vegetable is rich in mucilage that serves as a source of a unique soluble fiber. While no studies have yet investigated the effects of *P. aculeata* on the gut microbiome, it is probable that this fiber contains unique prebiotic activity, as a consequence of its unique chemical composition. Further, the abundance of vitamin C, minerals, and carotenoids poise *P. aculeata* to become an important nutrient-dense food for the future.

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ARTICLES FOR FACULTY MEMBERS

FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

Title/Author	**Opuntia monacantha: Validation of the anti-inflammatory and anti-arthritic activity of its polyphenolic rich extract in silico and in vivo via assessment of pro- and anti-inflammatory cytokines / Abid, F., Saleem, M., Jamshaid, T., Jamshaid, U., Youssef, F. S., Diri, R. M., Elhady, S. S., & Ashour, M. L.**
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Opuntia monacantha: Validation of the anti-inflammatory and anti-arthritic activity of its polyphenolic rich extract in silico and in vivo via assessment of pro- and anti-inflammatory cytokines

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ABSTRACT

Ethnopharmacological relevance: Opuntia monacantha belongs to the cactus family Cactaceae and is also known by cochineal prickly pear, Barbary fig or drooping prickly pear. It was traditionally used to treat pain and inflammation. *O. monacantha* cladodes showed pharmacological effects such as antioxidant potential owing to the presence of certain polysaccharides, flavonoids, and phenols.

Aim of the study: This research aimed to evaluate the anti-inflammatory as well as the anti-arthritic potential of ethanol extract of *Opuntia monacantha* (E-OM).

Materials and methods: In vivo edema in rat paw was triggered by carrageenan and used to evaluate anti-inflammatory activity, while induction of arthritis by Complete Freund's Adjuvant (CFA) rat model was done to measure anti-arthritic potential. *In silico* studies of the previously High performance liquid chromatography (HPLC) characterized metabolites of ethanol extract was performed by using Discovery Studio 4.5 (Accelrys Inc., San Diego, CA, USA) within active pocket of glutaminase 1 (GLS1) (PDB code: 3VP1; 2.30 Å).

Results: EOM, particularly at 750 mg/kg, caused a reduction in the paw edema significantly and decreased arthritic score by 80.58% compared to the diseased group. It revealed significant results when histopathology of ankle joint was examined at 28th day as it reduced inflammation by 18.06%, bone erosion by 15.50%, and pannus formation by 24.65% with respect to the diseased group. It restored the altered blood parameters by 7.56%, 18.47%, and 3.37% for hemoglobin (Hb), white blood count (WBC), and platelets, respectively. It also reduced rheumatoid factor RF by 13.70% with concomitant amelioration in catalase (CAT) and superoxide dismutase (SOD) levels by 19%, and 34.16%, respectively, in comparison to the diseased group. It notably decreased mRNA expression levels of COX-2, IL-6, TNF-α, IL-1, NF-κβ and augmented the levels of IL-4 and IL-10 in real time PCR with respect to the diseased group and piroxicam. HPLC analysis previously performed showed that phenolic acids and flavonoids are present in E-OM. Molecular docking studies displayed pronounced inhibitory potential of these compounds towards glutaminase 1 (GLS1), approaching and even exceeding piroxicam.

Conclusions: Thus, *Opuntia monacantha* could be a promising agent to manage inflammation and arthritis and could be incorporated into pharmaceuticals.

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1. Introduction

Rheumatoid arthritis is type of arthritis which is also an autoimmune disorder, that is progressive, long-lasting, & disabling (Deyab et al., 2021). It is characterized by joint swelling, discomfort, and synovial joint rigidity. Etiology of arthritis is unknown; however, it was believed to be caused by an autoimmune reaction exaggerated by various factors related to genes and environment. According to an epidemiological analysis, arthritis affects 1% of the global population. Arthritis is excruciating as joints connect bones and assist the movement of different body parts (Abid et al., 2022a,b). Furthermore, it is defined by inflammation in synovium and the elevated generation of nuclear factor-κβ (NF-κβ), that makes formation of pro-inflammatory cytokines (Joung et al., 2020). Stimulation of cyclooxygenase-2 (COX-2) by NF-κB exaggerates inflammatory processes (González-Sarrías et al., 2010). COX-2 activation is linked with excessive formation of prostaglandin-E2 (PGE2) that consequently leads to erosion of bone and destruction of cartilage, and it is associated with heart complaints such as stroke and myocardial infarction (Mobashar et al., 2022).

Besides, various mechanistic approaches are now followed in rheumatoid arthritis progression, comprising two markers namely tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6) (Yokota et al., 2021), where former causes stimulation of latter (Lima et al., 2021). Meanwhile, IL-6 is significantly involved in immune cells activation, then events lead to inflammatory mechanism, which ends in erosion of bone and formation of pannus (Li et al., 2022). Increased levels of these parameters are primarily implicated in pathogenesis of disease (Arjumand et al., 2019). It is noteworthy to highlight that inflammation, which is characterized by swelling, discomfort, and heat, might nevertheless impair a patient's mobility (Muley et al., 2016; Talaat et al., 2018; Ayoub et al., 2021). Meanwhile, uncontrolled inflammation, can increase the creation of C-reactive protein (risk factor for cardiac ailments) (Koenig, 2013).

The current treatment approaches include nonsteroidal anti-inflammatory medicines (NSAIDs), corticosteroids, and disease modifying antirheumatic therapies (DMARDs), that helps to decrease course of rheumatoid arthritis-related inflammation. Although medications are efficient in reducing pain, they cannot deliver a complete cure. The disadvantage of using synthetic medications to treat arthritis is stomach ulcers produced by the long-term use of NSAIDs and steroids (Labib et al., 2017; Rahimi et al., 2021). Because of the increased susceptibility of patients to infections, disease-modifying anti-arthritic medicines (methotrexate & IL-1β & TNF-α antagonists are not in common use (Guo et al., 2018). Bleeding, bone erosion and gastric perforation are side effects of NSAIDs due to prostaglandins inhibition (Scheiman, 2016). Opiates are associated with dependence & tolerance (Deane and Holers, 2019). Peptic ulcers, diabetes precipitation, osteoporosis, and elevated risk of infection capturing are among the side effects of corticosteroids (Fernández et al., 2021). However, due to serious adverse effects, first-pass effect, very poor absorption, rapid metabolism and low bioavailability, administrating of medications through parenteral and oral routes is limited (Qindeel et al., 2020).

Because of the terrible morbidity, medicinal costs, fear of surgery, and unremitting quality of sickness, conventional medications are now being replaced with plant drugs for their relative safety and ease of availability compared to synthetic drugs (Ashour et al., 2018; Mamadalieva et al., 2018; Thabet et al., 2018). It was estimated that 60–90% of patients suffering from rheumatoid arthritis seek complementary and alternative medicine (Mobashar et al., 2019). *Opuntia monacantha* belongs to the cactus family Cactaceae and is also known by cochineal prickly pear, Barbary fig or drooping prickly pear (Majeed et al., 2021). It was traditionally used because of their pronounced effectiveness in diabetes, burns, indigestion, and bronchial asthma (Abid et al., 2020). *O. monacantha* cladodes showed pharmacological effects such as antimicrobial and antioxidant potential owing to the presence of certain polysaccharides, flavonoids, and phenols (Bari et al., 2012; Dick et al., 2019). Besides, *Opuntia monacantha,* is native to Brazil, and commonly known in the coastal plains of Rio Grande do Sul and other Brazilian states. It produces abundant fruits however, it is little consumed, except by indigenous populations, who also use the species as an anti-inflammatory (da Silva et al., 2023).

Hence, this research was done to evaluate anti-arthritic and the anti-inflammatory effect of ethanol extract of *Opuntia monacantha* cladodes. Carrageenan-triggered paw edema in animals was used as model of acute inflammation, whereas chronic arthritis model can be induced in animals via Complete Freund's Adjuvant (CFA). Furthermore, proinflammatory and anti-inflammatory cytokines such as NF- κβ, IL-10, TNF-α, IL-4, IL-1, IL-6, and Iκ-B levels in addition to COX-2 were measured, which could indicate a potential mechanism of *Opuntia monacantha* ethanol extract in treating rheumatoid arthritis patients that were further confirmed by histopathological examination. Furthermore, the levels of the markers of oxidative stress exemplified by catalase (CAT) superoxide dismutase (SOD) and glutathione peroxidase (GPx), hematological parameters including white blood cells count (WBC), platelets, and hemoglobin (Hb), and biochemical parameters represented by alanine transaminase (ALT), alkaline phosphatase (ALP), urea, aspartate transaminase (AST) & creatinine were also observed. Meanwhile, in silico studies of the previously characterized metabolites of ethanol extract was performed by using HPLC within active site of glutaminase 1 enzyme to further correlate the biological activity with the predominant secondary metabolites and to explore an additional mode of action of extract.

List of abbreviations

ALP	Alkaline phosphatase
ALT	Alanine transaminase
AST	Aspartate transaminase
CAT	Catalase
CFA	Complete Freund's Adjuvant
COX-2	Cyclooxygenase-2
DMARDs	Disease modifying antirheumatic therapies
E-OM	Ethanol extract of *Opuntia monacantha.*
GLS1	Glutaminase 1
GPx	Glutathione peroxidase
Hb	Hemoglobin
H&E	Haemotoxylin and eosin
HPLC	High performance liquid chromatography
IL-6	interleukin-6
NF-κβ	nuclear factor-κβ
NSAIDs	Non-steroidal anti-inflammatory drugs
PGE2	prostaglandin-E2
SOD	Superoxide dismutase
WBC	white blood cells count

2. Materials and methods

2.1. Plant material

Opuntia monacantha Haw. (Cactaceae) was gathered from the Government College University, Lahore (Botanical Garden). It was authenticated voucher # 3654 by Professor Dr. Zaheer-ud-Din, Department of Botany, Government Collage University Lahore.

2.2. Preparation of the plant extract

O. monacantha (3 kg) was washed, sun-dried for two weeks, ground into fine powder, and then steeped in 70% ethanol for a week at room

temperature. It was filtered using Whatman # 1 filter paper, and with help of rotary evaporator, solvent was evaporated and extract was made concentrated 40 °C under decreased pressure. The extracts were then dried to a semisolid or solid mass in hot air oven at 25 °C to give dried extract.

2.3. In vivo biological evaluation of O. monacantha ethanol extract

2.3.1. Animals

Wistar rats of 200–250g weight were used to conduct research. These were kept at the University of Lahore's Animal House, which was a part of the Pharmacy Department. The Institutional Ethical Committee at the University of Lahore, Pakistan, authorized the experimental with approval number IMBB/UOL/20/416. Procedures were achieved in conformity with the applicable regulations and rules. The rats were housed in steel cages under typical laboratory settings with 25 °C and 60% humidity. Rodents feed and water were in an adequate manner. The attending veterinarian assessed optimal condition of animals on routine basis. Animals were placed in laboratory for 14 days before conducting experiments. The current investigation used xylazine and ketamine together, as anaesthetic agent. Dose of ketamine was 100 mg/kg of body weight & dose of xylazine was 5 mg/kg of body weight.

2.3.2. Estimation of acute anti-inflammatory effect using carrageenan-induced paw edema model

Six wistar rats were added in every group and groups were designated as 1 to 5. Normal saline was given to first group and named as control group meanwhile piroxicam was administered to second group with dose of 10 mg/kg as pretreatment. E-OM was given to 3rd, 4th and 5th groups at different concentrations namely 250, 500 & 750 mg/kg as a pretreatment. After treating groups, induction of inflammation was done via the injection of 0.1 mL of 1% of carrageenan solution that is freshly prepared in sub-planter area of right hand paw of rats for five consecutive hours and digital water plethysomometer was used to measure edema size in paw (Mobashar et al., 2020).

2.3.3. Estimation of chronic arthritis inhibitory activity using arthritis induced Complete Freund's adjuvant (CFA) model

36 rats were split into 6 groups containing 6 animals in every group. They are vehicle control group that received normal saline, disease control, piroxicam group that orally administered with 10 mg/kg of piroxicam, treatment groups that orally received *O. monacantha* ethanol extract (E-OM) at 250, 500 and 750 mg/kg doses. Complete Freund's Adjuvant (CFA) (0.15 mL) was given to left paw of each animal except vehicle control group at day 0 to induce arthritis. Treatment was started on 8th and it was continued till 28th day. All rats were sacrificed on the 28th day using anesthesia (Mobashar et al., 2022). Paw edema and arthritic score were monitored using a digital water plethysmometer and macroscopic criteria (Akhtar and Shabbir, 2019), respectively, on day 8th 12th, 16th, 20th, 24th, and 28th. Macroscopic criteria protocol was followed using reference literature (Akhtar and Shabbir, 2019).

2.3.4. Determination of mRNA expression levels of NF-κβ, TNF-α, IL-6, IL-1β, COX-2, IL-10 and IL-4

Blood samples were treated with TRIzol reagent with 1 : 3 (200 : 600 μL), then chloroform was added for separation of phases, after that isopropanol was added to make precipitation of RNA, later washing was done with ethanol, then air dried followed by storage at −80 °C. Then, quantification was performed via nanodrop spectrophotometer. Then, performance of cDNA synthesis by using kit manufacturer's protocol (Thermo Scientific; Waltham, MA) was achieved. The template of RNA was treated with primer oligo dt_{18} followed by nuclease-free water (q.s). Incubation temperature was 65 °C for 5 min. After that, 4 μL of 5X reaction buffer (20 mM $MgCl_2$, 250 mM KCl (8.3 pH), 250 mM Tris-HCl and 50 mM DTT), 2 μl of 10 mM dNTPs mixture, 1 μL of Ribo Lock RNase inhibitor and 1 μL of 200U M-MuLV reverse transcriptase enzyme were mixed, followed by the incubation of the for 60 min at 42 °C. GAPDH was used as reference as internal reaction control gene. Manually designing of primers of GAPDH & TNF-α was done. Primers sequence for IL-6, IL-4, IL-1β, NF-κB, IL-10 & COX-2 were selected from previous researches and are as follows: IL-4; 5′-GTACCGGGAACGGTATCCAC-3' (forward), 5′-TGGTGTTCCTTGTTGCCGTA-3' (backward); IL-10, 5′-TTGAACCACCCGGCATCTAC-3' (forward), 5′-CCAAGGAGTTGCTCCCGTTA-3' (backward); TNF-α, 5′-ATGGGCTCCCTCTCATCAGT-3' (forward), 5′-GCTTGGTGGTTTGCTACGAC-3' (backward); IL-1β, 5′-GTCCTCTGCCAAGTCA GGTC-3' (forward), 5′-CAGGGAGGGAAACACACGTT-3' (backward); IL-6, 5′-CCCACCAGGAACGAAAGTCA-3' (forward), 5′-ACTGGCTGGAAGTCTCTTGC -3' (backward); GAPDH, 5′-AGTGCCAGCCTCGTCTCATA-3′, (forward), 5′-ACCAGCTTCCCATTCTCAGC-3' (backward); NF-kβ, 5′-CTGAGTCCCGCCCCTTCTAA-3' (forward), 5′-CTCCACCAGCTCTTTGATGGT-3' (backward); COX-II, 5′-ATGCTACCATCTGGCTTCGG-3' (forward), 5′-TGGAACAGTCGCTCGTCATC-3' (backward). Heating was done for 5 min at 70 °C, terminated reaction and cDNA (2 μL) was added with forward-reverse primer mix (1 μL), PCR Master Mix (6 μL) & nuclease-free water (3 μL). Denaturation was for 10 s at 95 °C, annealing was done for 20 s at 58 °C and 60 °C and extension was for 30 s at 72 °C, in thermal cycler (Mobashar et al., 2022).

2.3.5. Determination of oxidative stress markers

2.3.5.1. Determination of serum catalase (CAT). Material, reagents and instrumentation used in catalase assay are serum, 150 mM, phosphate buffer, H_2O_2, Spectrophotometer. Aebi's methodology for the determination of serum catalase was followed using UV spectrophotometer. The measurement was done at 240 nm. The rat's serum was homogenized in 150 mM phosphate buffer of pH 7 at 4 °C and centrifugation was done at 6000 rpm. The activity of catalase was expressed in unit per gram of serum sample. The absorbance was measured by UV spectrophotometer and values were compared with standard curve which was generated from known enzyme catalase (Goth, 1991).

2.3.5.2. Determination of superoxide dismutase (SOD). Estimation of SOD enzyme was measured by taking 100 μL of sample, 1200 μL of sodium orthophosphate buffer with pH 8.3 and 0.052 M concentration, 100 μL of phenazine methosulfate at 187 μm, 300 mL of NBT (300 μm), 200 μL of NADH (760 μmol) reaction was initiated by continuous adding of NADH. Then, it was incubated at 400 °C for a time span of 90 s, reaction was completed by the increment of 100 μl of glacial acetic acid. Afterwards, 4 mL of *n*-butanol were mixed in the reaction mixture with stirring. Afterwards, the mixture of the reaction was cooled for 10 min, followed by its centrifugation and the *n*-butanol layer was separated at the end. The color saturation of butanol layer was read at 560 nm against butanol and against SOD expressed as U/ml. The quantity measure depends on intensity of the enzyme to hinder the scavenging effect on superoxide anion radicals that was measured by NBT reduction method with slight modifications. The activity was determined as mentioned before (Nishikimi et al., 1972).

2.3.5.3. Determination of hematological and biochemical parameters. Hematological parameters such as while blood count (WBC), platelets, and hemoglobin (Hb) and biochemical parameters such as ALT, AST, creatinine and urea were determined as previously reported by Mobashar et al. (2022).

2.3.6. Histopathological investigation

After the animals were sacrificed on the 28th day, affected joints were used for histopathological examinations using Haemotoxylin and eosin (H&E) staining (Beziere et al., 2014). The histological scoring system was used to measure several factors such as pannus development, bone and cartilage deterioration (Mobashar et al., 2022).

2.4. In silico molecular docking studies

Molecular docking was done to identify flavonoids and phenolic metabolites existing in *O. monacantha* ethanol extract by using Discovery Studio 4.5 (Accelrys Inc., San Diego, CA, USA) within active pocket of glutaminase 1 (GLS1) (PDB code: 3VP1; 2.30 Å) (Thangavelu et al., 2012) as a novel target for counteracting rheumatoid arthritis that was taken from protein data bank, following C-Docker protocol where (ΔG) representing the binding energies were assessed from equation as described earlier (Labib et al., 2018; Talaat et al., 2018; Altyar et al., 2020; Sweilam et al., 2022).

2.5. Statistical analysis

Results were measured as Mean ± SEM. Graph pad prism 8.0.1 was used to interpret the data. Data were examined statistically using ANOVA followed by test of post Tukey's multiple comparison. The value $P \leq 0.05$ was taken as significant.

3. Results

3.1. Chemical characterization of O. monacantha ethanol extract using HPLC analysis

Quantitative determination of flavonoids and phenolic acids was previously reported by the authors for *O. monacantha* ethanol extract using HPLC analysis using reference standard of phenols and flavonoids i.e., quercetin, kaempferol, acids of ferulic, benzoic, vanillic, gallic, chlorogenic, caffeic, sinapic, syringic and coumaric in methanol (Abid et al., 2022a,b). Results showed that quercetin (2.14 μg/g), vanillic acid (4.87 μg/g), benzoic acid (1.24 μg/g), gallic acid (0.87 μg/g), sinapic acid (1.21 μg/g), *m*-coumaric acid (2.23 μg/g), *p*-coumaric acid (0.38 μg/g), syringic acid (0.11 μg/g), and caffeic acid (14.67 μg/g) were the predominant flavonoids and phenolic acids existing in *O. monacantha* ethanol extract (Table S1). A scheme showing the compounds identified in *O. monacantha* ethanol extract was illustrated in Fig. S1.

3.2. Acute anti-inflammatory activity using carrageenan induced paw edema model

Administration of carrageenan, a mucopolysaccharide, into the rat hind paw triggers a localized inflammatory response that is depicted by an elevated metabolism of arachidonic acid with concomitant elevation of vascular permeability that is accompanied by the occurrence of edema in addition to neutrophil extravasation (Gamache et al., 1986). Carrageenan induced paw edema model constitutes one of the most popular models used to screen the anti-inflammatory behavior of natural products (Peters et al., 1999). Edema was induced by Carrageenan in animals at the 1st, 2nd, 3rd, 4th, and 5th hours that was counteracted by oral administration of 10 mg/kg of piroxicam. Meanwhile, oral administration of *O. monacantha* ethanol extract (E-OM) at different doses namely 250, 500, and 750 mg/kg as a pretreatment slightly prohibited Carrageenan triggered paw edema at 1st, 2nd, 3rd, 4th, and 5th hours as shown in Fig. 1.

3.3. Chronic arthritis inhibitory activity using arthritis induced Complete Freund's adjuvant (CFA) model

3.3.1. Effect on body weight and arthritic score

Complete Freund's Adjuvant (CFA) injections in rat paw (sub-plantar

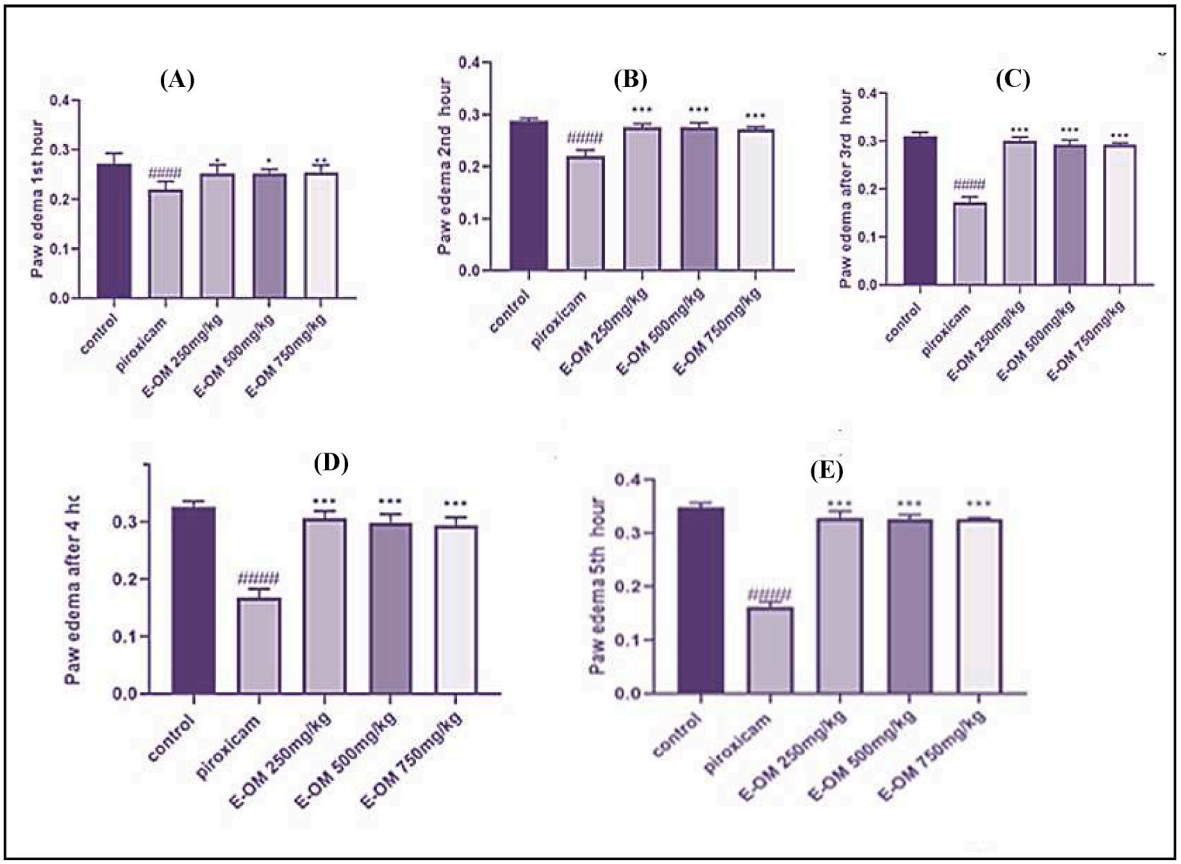

Fig. 1. Treatment at different time intervals where E-OM inhibited paw edema induced by Carrageenan, showing (A) treatment at 1st hour, (B) is at 2nd hour, (C) at 3rd hour, (D) at 4th hour and (E) 5th hour. *, **, *** showing $P \leq 0.05$, $P \leq 0.01$, $P \leq 0.001$; ###, pinpoints arthritic control bar to differentiate from among others, it shows all other groups were compared with arthritic control.

region) and enhanced inflammation there, culminating in a peak edema on the eighth day. No swelling was observed in normal control group in compared to arthritic group which showed an elevation in paw volume from the 8th to the 28th day. However, at the 28th day, the percentage inhibition exhibited with oral administration of different doses of E-OM was comparable to the reference treatment, piroxicam and even superior activity was exerted upon administration of E-OM at dose of 750 mg/kg. Arthritis formation changed the weight of the body of rats as inflammation developed. After one week to end of trial, the loss in weight was more noticeable in arthritic rats than in the normal group by 31.21%. From the 12th to the 28th day, however, with oral administration of different doses of E-OM namely 250, 500, 750 mg/kg and piroxicam improved body weight significantly in arthritic rats by 1.5%, 20.27%, 20.56% and 21.69%, respectively on the 28th day of experiment as shown in Fig. S2.

However, arthritic rats orally administered with doses of E-OM recovered body weight in a dose-dependent manner. The results recorded in Fig. 2 revealed a continual increase in the arthritic index in the arthritic group that received CFA only without any treatment. However, treatment with E-OM and piroxicam at a dose of 10 mg/kg decreased the arthritic index in compare to the control group through the 16th to the 28th day of the study. The maximum arthritic score observed at the 28th day was ameliorated by piroxicam by 74.30%, E-OM at 750 mg/kg by 80.58%, E-OM at 500 mg/kg by 77.95% and E-OM 250 mg/kg by 76.16% compared to arthritic control group. It is worthy to highlight that normal control group didn't exhibit any swelling during trial when it came to the arthritic index.

3.3.2. Effect on gross microscopic view and histopathology of rat paw

At the 28th day of the CFA model, gross microscopic and histopathological examination of rat paw examination revealed significant edema in arthritic rats whereas treatment with E-OM at different doses effectively ameliorated the arthritic condition in manner of dose dependent. E-OM (750 mg/kg) caused low bone erosion and less pannus development whereas E-OM (750 and 500 mg/kg) reduced vascular degeneration in treated rats. Piroxicam and E-OM (250, 500 and 750 mg/kg) reduced pannus formation score by 21.53%, 15.86%, 18.98% and 24.65%, respectively whereas they reduced inflammation score evidenced by reduced inflammatory cell infiltration by 19.44%, 15.83%, 16.11% and 18.06%, respectively in addition to decreasing bone erosion by 16.37%, 9.36%, 13.16% and 15.50%, respectively in compare to arthritic rats (Fig. S3). Besides, examination of histopathology of ankle joint at end of 28th day, it showed effective results where *O. monacantha* reduced inflammation, bone erosion and pannus formation. Significant results were observed $p \leq 0.001$ after treatment with piroxicam and E-OM (750 and 500 mg/kg) (Fig. 3).

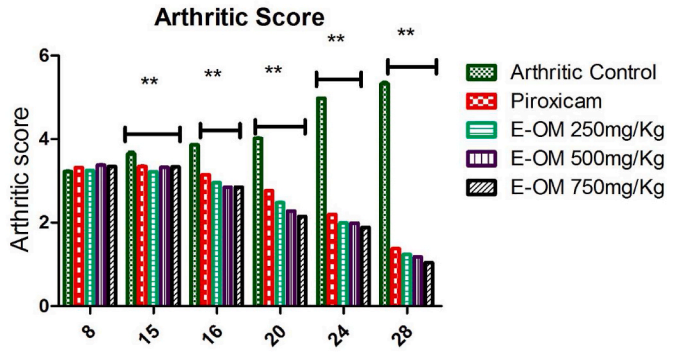

Fig. 2. Effect of oral administration of different doses of E-OM (250, 500 and 750 mg/kg) and piroxicam (10 mg/kg) on arthritic score in Complete Freund's Adjuvant (CFA) treated rats from 8th to 28th day.
Results were expressed as Mean ± SEM; (n = 6) and analysis was done by two-way ANOVA analysis, followed by Tukey test.; anti-arthritic effect of E-OM doses and piroxicam in a chronic CFA model showed level of significance of $^{*}p < 0.05$, ** = p < 0.01, ***p < 0.001 and ^{ns} $p > 0.05$ with respect to diseased group.

3.3.3. Effect on liver function tests

Arthritis induction in rats by CFA resulted in raised levels of ALP, ALT and AST by 53.47%, 57.14% and 1.23%, respectively in comparison with normal group. It was noted that in comparison with ALP and ALT, level of AST was not altered and results were insignificant compared with Piroxicam and E-OM treated group. Treatment with E-OM at different doses showed a substantial reduction in ALP, ALT and limited decrease in AST levels when compared to the disease control estimated by 4.52%, 9.09% and 2.44%, respectively at E-OM (250 mg/kg); 6.79%, 12.87% and 2.65%, respectively for E-OM (500 mg/kg) and by 8.14%, 18.36% and 1.95%, respectively for E-OM (750 mg/kg). These results are comparable to piroxicam that showed 10.86%, 14.55% and 1.93%, reduction in ALP, ALT and AST levels respectively (Fig. S4).

3.3.4. Effect on rheumatoid factor

The induction of rheumatoid arthritis increased level of rheumatoid factor (RF) in rats. However, treatment of E-OM at different doses considerably reduced the RF value in a dose dependent manner where the best activity was exerted by E-OM (750 mg/kg). Results showed that the arthritic control showed 277.47% elevation in RF with respect to normal vehicle group. On the contrary, administration of E-OM at 250, 500 and 750 mg/kg significantly reduced RF by 10.96%, 13.42% and 13.70%, respectively approaching piroxicam that resulted in 14.90% reduction in RF in comparison with arthritic group (Fig. S5).

3.3.5. Effect on hematological and kidney function parameters

The induction of rheumatoid arthritis in rats showed a pronounced decline in hemoglobin (Hb) estimated by 55.81% with concomitant elevation in white blood count (WBC) and platelets by 124.21% and 75.96%, respectively in comparison with normal control group (Fig. 4.). However, administration of E-OM at 250, 500 and 750 mg/kg displayed pronounced elevation in Hb by 2.59%, 7.45% and 7.56%, respectively whereas they reduced WBC by 6.78%, 18.48% and 18.47%, respectively in addition to a slight decreasing in the platelet count by 1.34%, 2.61% and 3.37%, respectively in comparison wi t h arthritic control group. E-OM treated groups particularly at 750 mg/kg showed activity approaching that of piroxicam that exhibited 6.91% elevation in Hb level and 18% and 3.66% reduction in both WBC and platelets, respectively. However, it was observed that induction of RA and treatment with E-OM at different doses as well as with the standard Piroxicam showed no significant effect on creatinine and urea that are kidney function tests.

3.3.6. Effect on oxidative stress markers

Rheumatoid arthritis in rats showed a pronounced decline in superoxide dismutase (SOD) and catalase (CAT) levels estimated by 57.02% and 52.7%, respectively as compared to the normal vehicle group. Besides, they showed a pronounced elevation in SOD level estimated by 13.18 %, 28.12% and 34.16%, respectively with concomitant increase in CAT by 8.07 %, 16.1% and 19%, respectively as compared to arthritic control group. Besides, piroxicam, standard drug, showed 32.12% and 24.18% elevation in SOD and CAT levels, respectively (Fig. 5).

3.3.7. Effect on mRNA gene expression of various inflammatory indicators

In the instance of gene expression research, various inflammatory indicators were assessed after a 28-day mRNA expression study in wistar rats. In the arthritic group, IL-4 and IL-10 expression were considerably decreased by 17.16% and 31.22%, respectively with respect to the control group. However, administration of E-OM at 250, 500 and 750 mg/kg, showed significant up-regulating and elevation in both interleukins estimated by 1.14 %, 7.88% and 11.18%, respectively for IL-4

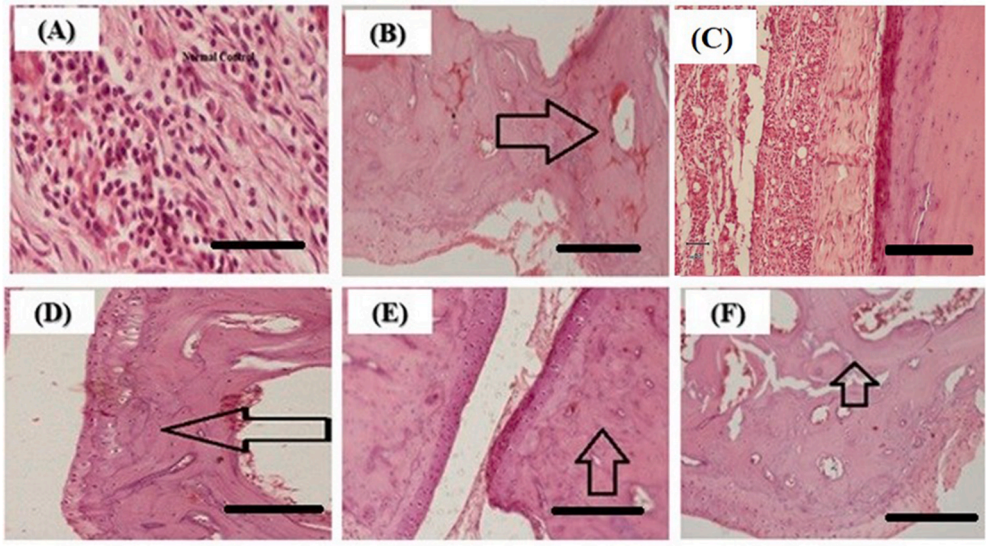

Fig. 3. Histopathology of ankle joint at 100x at the end of 28th day in Complete Freund's Adjuvant (CFA) induced arthritis model in rats. (A) Control group; (B) arthritic control group; (C) piroxicam treated group; (D) E-OM 250 mg/kg; (E) E-OM 500 mg/kg; (F) E-OM 750 mg/kg.

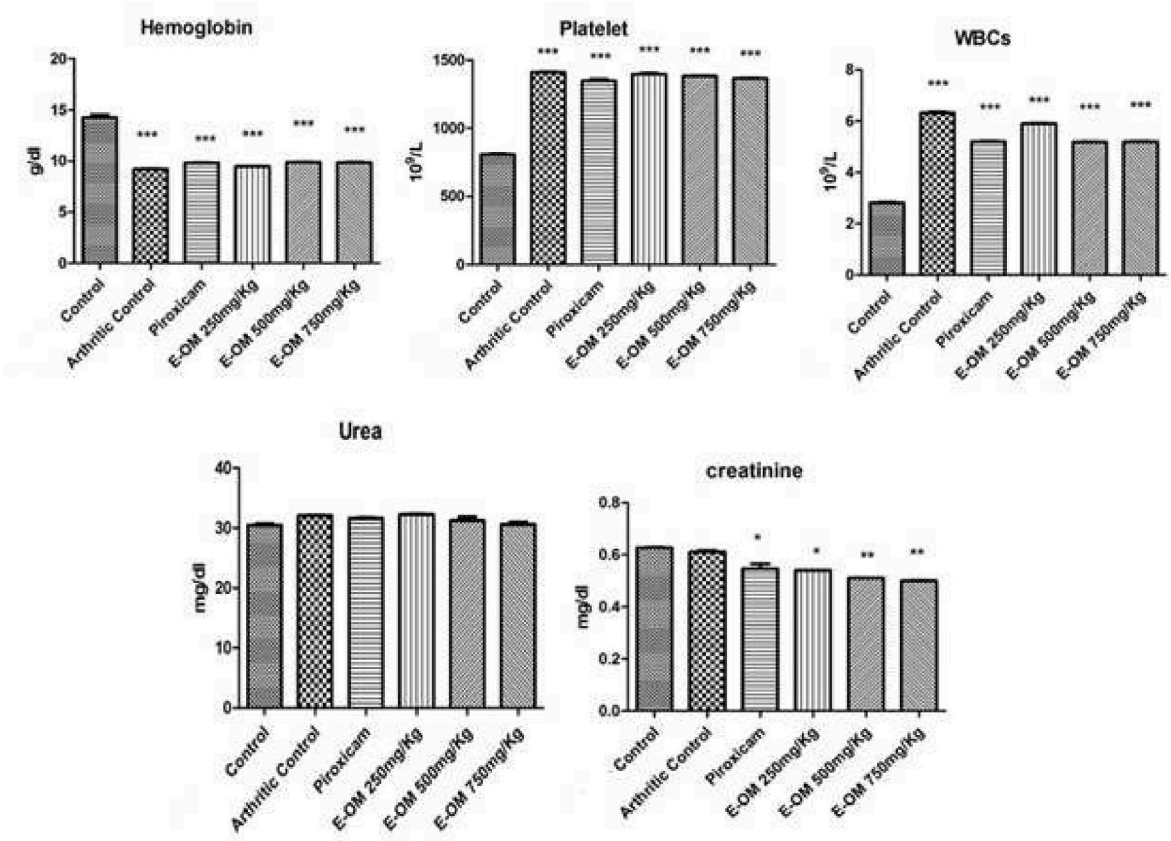

Fig. 4. Effects of oral administration of different doses of E-OM (250, 500 and 750 mg/kg) and piroxicam (10 mg/kg) on hematological and kidney function parameters in Complete Freund's Adjuvant (CFA) treated rats

Results were expressed as Mean ± SEM; (n = 6), and statistical analysis was done by two-way ANOVA analysis followed by Tukey test.; anti-arthritic effect of E-OM (250, 500 and 750 mg/kg) and piroxicam in a chronic CFA model showed level of significance of $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$ and $^{ns}p > 0.05$ with respect to diseased group.

and 16.09%, 18.41% and 22.47%, respectively for IL-10. Meanwhile, piroxicam elevated IL-4 and IL-10 by 11.98% and 20.77%, respectively with respect to the arthritic group illustrated in Fig. 6. In arthritic control group, IL-1β, IL-6, COX-2, TNF-α and NFκβ expression were considerably elevated by 32.54%, 29.86%, 26.36%, 41.25% and 31.51%, respectively with respect to the normal control. However, administration of E-OM at 250, 500 and 750 mg/kg showed a pronounced reduction in IL-1β expression estimated by 6.43%, 7.9% and

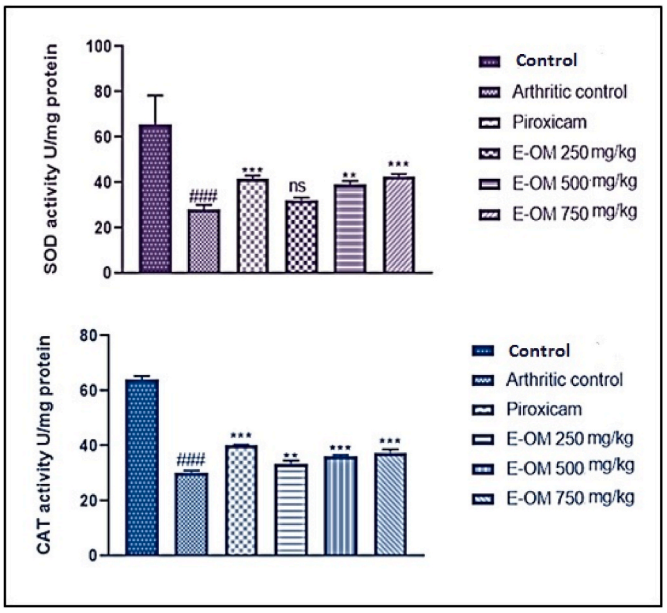

Fig. 5. Effect of oral administration of different doses of E-OM (250, 500 and 750 mg/kg) & piroxicam (10 mg/kg) on oxidative stress markers namely SOD (A) and CAT (B) in Complete Freund's Adjuvant (CFA) treated animals; *, **, *** indicate P ≤ 0.05, P ≤ 0.01, and P ≤ 0.001 respectively; ns: non-significant; ###, pinpoints arthritic control bar to differentiate from among others, it shows all other groups were compared with arthritic control.

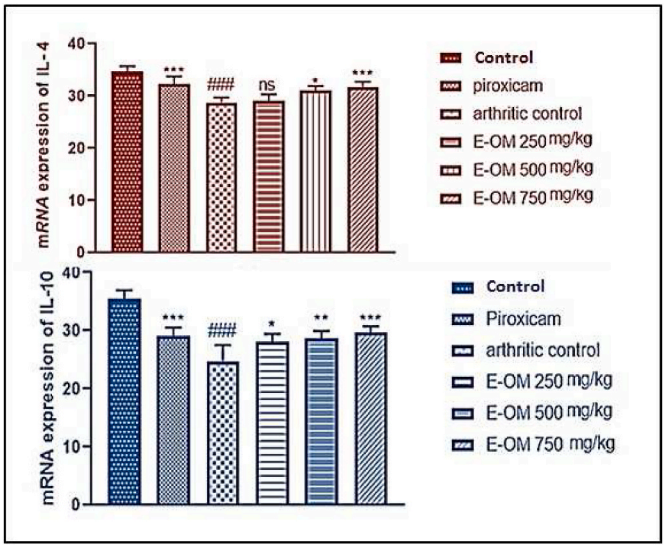

Fig. 6. Effects of oral administration of different doses of E-OM (250, 500 and 750 mg/kg) and piroxicam (10 mg/kg) on IL-4 and IL-10 expression in Complete Freund's Adjuvant (CFA) treated rats; *, **, *** indicate P ≤ 0.05, P ≤ 0.01, and P ≤ 0.001 respectively; ns: non-significant; ###, pinpoints arthritic control bar to differentiate from among others, it shows all other groups were compared with arthritic control.

7.17%, respectively; and 1.77%, 6.13% and 8.33% decrease in IL-6, respectively with concomitant decline in COX-2 by 0.1 %, 8.4% and 9.88%, respectively. Besides, they effectively reduced TNF-α by10.45 %, 10.87% and 11.88%, respectively and by 3.55 %, 5.82% and 6.86%, respectively for NFκβ expression Furthermore, piroxicam reduced IL-1β, COX-2, IL-6, TNF-α and NFκβ expression by 8.25%, 9.82%, 7.97%, 11.94% and 14.37%, respectively with respect to the arthritic diseased group (Figs. 7 and 8).

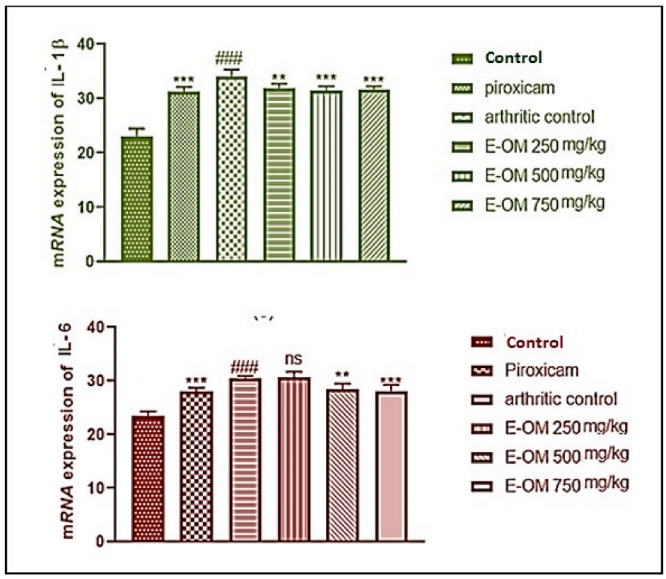

Fig. 7. Effects of oral administration of different doses of E-OM (250, 500 and 750 mg/kg) and piroxicam (10 mg/kg) on IL-1β and IL-6 expression in Complete Freund's Adjuvant (CFA) treated animals; *, **, *** indicate P ≤ 0.05, P ≤ 0.01, and P ≤ 0.001 respectively. ns: non-significant; ###, pinpoints arthritic control bar to differentiate from among others, it shows all other groups were compared with arthritic control.

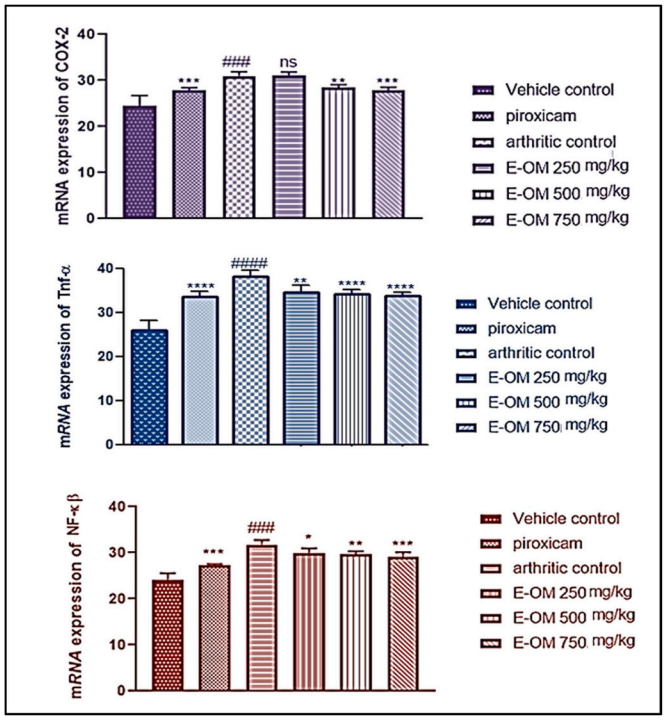

Fig. 8. Effect of oral administration of different doses of E-OM (250, 500 and 750 mg/kg) and piroxicam (10 mg/kg) on COX-2, TNF-α and NFκβ expression in Complete Freund's Adjuvant (CFA) treated rats; *, **, *** indicate P ≤ 0.05, P ≤ 0.01 and P ≤ 0.001 respectively. ns: non-significant; ###, pinpoints arthritic control bar to differentiate from among others, it shows all other groups were compared with arthritic control.

3.4. *In silico molecular docking studies*

Additionally, in an effort to discover an additional probable mode of action to the extract, molecular docking studies were done for the

previously identified flavonoids and phenolic metabolites existing in *O. monacantha* ethanol extract on the active sites of glutaminase 1 (GLS1). As illustrated in Table 1, all the tested compounds exhibited pronounced inhibitory potential towards GLS1 approaching piroxicam; however gallic acid, caffeic acid as well as quercetin and sinapic acid showed superior activity compared to piroxicam as revealed from their free binding energies (ΔG) which are −29.69, −28.27, −27.93, −24.41 kcal/mol, respectively whereas that of piroxicam is −23.64 kcal/mol (Table 1). Gallic acid showed the best fitting because of the formation of four hydrogen bonds with Asp327, Asn324, Tyr394; 1 Carbon–Hydrogen bond with Glu325 along with π-alkyl bond with Leu321at the binding site of glutaminase 1 (Fig. 9 A). Meanwhile, caffeic acid produced 3 Hydrogen-bonds with Asp327, Leu321, Phe322 amino acid existing at the active center (Fig. 9B). Piroxicam forms 1 hydrogen bond with Asp327; three Carbon–Hydrogen bonds with Asn324, Leu323, Glu325 in addition to 1 π-cation bond with Arg317 at glutaminase 1 active site (Fig. 9C).

4. Discussion

O. monacantha extract has been used frequently to treat arthritis in traditional medicine (Erdemoglu et al., 2003). Modern studies revealed that lesser side effects were observed when treatment of inflammation was done with natural products and acts as cost effective treatment in comparison with available conventional treatment (Bai et al., 2021). Complete Freund's Adjuvant (CFA)-induced arthritis model is much preferred as it possess much resemblance with arthritic disorders in humans, characterized by hyperplasia of synovium, formation of vessels, destruction of cartilage destruction and erosion of bone (Naz et al., 2020). *O. monacantha* ethanol extract was administered in current study at various doses, significantly reduced all pathological changes. Because of their pathophysiological similarities to human arthritis, CFA generated edema is a precise model to estimate anti-inflammatory effects of drugs during chronic inflammation in rats. In this study, rats were administered with *O. monacantha* ethanol extract at the different doses and showed substantial improvement in arthritic index after treatment of 28 days in comparison with injury group, which demonstrated a slowing of disease development.

Furthermore, serum biochemical and hematological indices, as well as inflammatory biomarkers i.e., CRP, NF-kB, COX-2, interleukins & TNF-α, are used to characterize the patient's inflammatory status (Uttra et al., 2018). Hypophosphatemia and hepatocellular injury are linked to elevated levels of AST, ALP and ALT. While levels of markers and other parameters such as platelets, RBCs and WBCs did not change in treated animals, hence in current study, it provided the bare minimum supportive environment for inflammation. However, the arthritic group had significant numbers of platelets and WBCs, which could indicate invoke immune responses against pathogens. Meanwhile, IL-10 and IL-4, inhibit Th-1 cell activity by suppressing interferon resulting in direct inhibitory effect on macrophage activity in arthritic patients' synovium. Results displayed herein showed an increase in IL-10 and IL-4 levels in both standard and extract-treated groups, which indicates potent anti-inflammatory properties.

Furthermore, when protease enzymes was released from neutrophils along with formation of reactive oxygen intermediates, quick and severe joint damage occurs, resulted in inflammation, hypercoagulability and hypoalbuminemia (Narazaki et al., 2017). IL-6 levels were found elevated in disease control group in comparison with treated groups, where they were not elevated and were within normal range, owing to anti-arthritic effects of the *O. monacantha* ethanol extract and piroxicam. Due to formation of proteolytic enzymes and reactive oxygen species, IL-1 plays important part in destruction of cartilage and activation of T-cells at inflammatory sites, whereas nuclear factor-kappa (NF-kβ) is responsible for production of inflammatory cytokines via toll-like receptor (TLR) pathway. The current study revealed that following conventional and extract treatments, the levels of IL-1 were reverted. This adds to the evidence that IL-1 has inhibitory effect and hinders in release of pro-inflammatory mediators. COX-2 increases PGE2 production, which enhanced TNF-α & IL-1β released from chondrocytes, causing discomfort and inflammatory processes in patients of rheumatoid arthritis (Lemos et al., 2009).

In current study, qPCR expression analysis revealed a re-establishment of COX-2 levels upon treatment with *O. monacantha* ethanol extract, resulting in reduced polygenetic arthritis symptoms. Moreover, various pathogenic stressors, such as growth factors, cytokines and oxidative stress, activate the inflammatory milieu by promoting NF-kB production. The synovial lining in rats after induction of arthritis by CFA was studied for a similar mechanism. In both the ethanol extract treated and control groups, normal levels of NF-kβ were restored. TNF-α is type of cytokine that is produced by many cells such as monocytes, macrophages and fibroblasts that promotes inflammation. It acts both as paracrine autocrine inducer of various mediators of inflammation (IL-1, IL-8, IL-6, GMCSF) and adhesion molecules (ICAM1), resulting in rheumatoid synovitis (Kaplanski et al., 2003). The arthritic control group showed a comparable reaction, while the ethanol extract and piroxicam treatment groups showed a significant decrease in TNF-α expression.

Besides, rheumatoid factor is considered as autoantibody that targets IgG antibodies. Detection of rheumatoid factor level is commonly adopted in classification of rheumatoid arthritis (Nielsen et al., 2012). Herein, rheumatoid arthritis resulted in elevation of rheumatoid factor (RF) in rats. However, treatment with E-OM at different doses as well as with piroxicam considerably reduced the RF value in a dose dependent manner, equivalent to anti-inflammatory properties of plant extract. Additionally, in the ethanol extract-treated groups, histological scoring as well as microscopical testing of rats' paws revealed less bone degradation and inflammation.

The potent anti-arthritic and anti-inflammatory properties which were exhibited by *O. monacantha* extract possible flavonoids and phenolic compounds presence in enormous amount as previously detected via HPLC analysis along with some other compounds such as that quercetin, ferulic acid, kaempferol, benzoic acid, vanillic acid, gallic acid, chlorogenic acid, sinapic acid, caffeic acid, coumaric acid and syringic acid in addition to eucalyptol, *β*-sitosterol and vitamin C (Abid et al., 2022a,b).

Secondary metabolites such as quercetin, benzoic acid, gallic acid, and coumaric acid are reported to possess notable anti-inflammatory, anti-oxidant as well as anticancer potential (Abid et al., 2022a,b). Linalyl acetate possess as anti-oxidant and anti-inflammatory agent (Veenstra and Johnson, 2019). Eucalyptol has also anti-oxidant and

Table 1
Free binding energies (kcal/mol) of major previously detected compounds of *O. monacantha* ethanol extract using HPLC in the active center of glutaminase 1 using molecular docking.

Compound	GLS1 (3VP1)	Number of bonds of Hydrogen & C–H	Number of formed π-πand π-alkyl & π-cation bonds
Benzoic acid	−17.45	3; Asp327, Lys398	–
Gallic acid	**−29.69**	5; Asp327, Asn324, Tyr394, Glu325	1; Leu321
m-Coumaric acid	−22.09	4; Asp327, Asn324, Phe322, Glu325	1; Leu321
p-Coumaric acid	−21.79	4; Asp327, Phe322, Glu325	–
Quercetin	**−27.93**	5; Asn324, Phe322, Glu325, Leu321	2; Leu321
Sinapic acid	−24.41	5; Asp327, Leu321, Glu325, Arg317	–
Caffeic acid.	**−28.27**	3; Asp327, Leu321, Phe322	–
Syringic acid	−21.11	1; Asp327	2; Leu321, Leu323
Vanillic acid	−21.53	1; Asp327	1; Leu321
Piroxicam	−23.64	4; Asp327, Asn324, Leu323, Glu325	1; Arg317

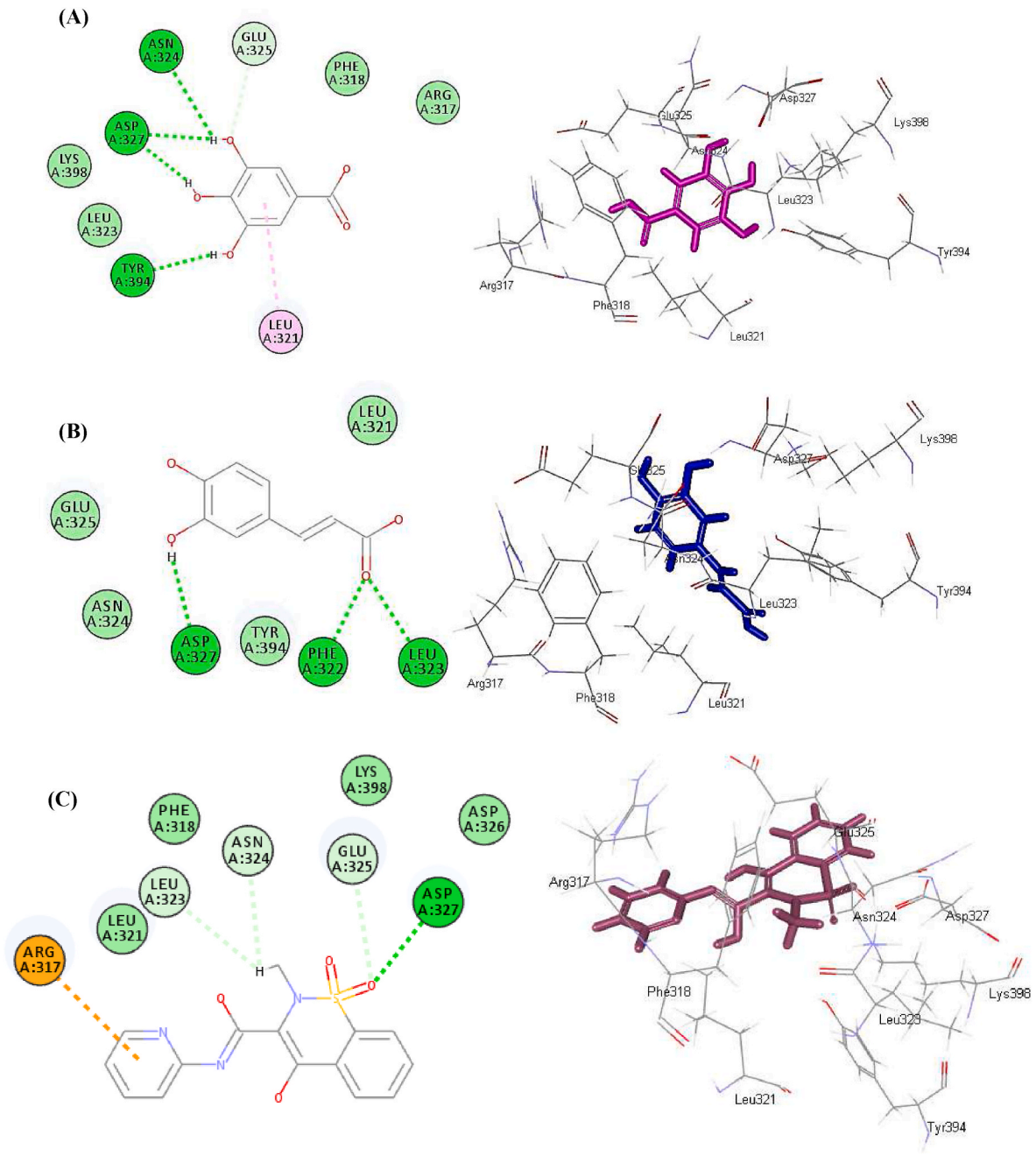

Fig. 9. 2D and 3D binding modes of gallic acid (A), caffeic acid (B) and piroxicam (C) in the active center of glutaminase 1 using molecular docking study.

anti-inflammatory capabilities by controlling MAPK and NF-κβ pathways in many diseases (Prasher et al., 2020) and is also incorporated in reduction of inflammation in lungs by declining the levels of IL-1β,TNF-α and NF-κβ (Dey et al., 2022). *p*-Coumaric acid and sinapic acid imparts this extract sufficient anti-inflammatory and anti-arthritic drug alternative (Hoda et al., 2019). Additionally, Sinapic acid exerts regulate COX-2 and other pro-inflammatory cytokines to display anti-inflammatory effect (Hwang et al., 2022). Gallic acid exerts its anti-inflammatory effect due to inhibition of pathway of NF-κβ, which is incorporated in inflammation and pathogenesis of rheumatoid arthritis (Abid et al., 2022a,b).

However, increased glycolysis levels allow the cells to fulfill their energy requirement, meanwhile glutaminolysis, offers the required biosynthetic precursors cope with the elevated metabolism. These elevations are coherent with the increased levels of elevated glutaminase 1 (GLS1) expressions in patients suffering RA (Ahmed et al., 2022). It was previously reported that GLS1 prohibition that controls the change of glutamine to glutamate, inhibits Fibroblast-like synoviocytes (FLS) from RA directly; RA-FLS proliferation, and hence improves the pathological intensity of rheumatoid arthritis (Takahashi et al., 2017). Hence, molecular docking studies were conducted on flavonoids and phenolic acids (identified earlier) of *Opuntia monacantha* ethanol extract in an effort to discover additional probable mode of action to the extract and examine their inhibitory potential *versus* glutaminase 1 active sites. All the tested compounds exhibited pronounced inhibitory potential towards GLS1 approaching piroxicam; however gallic acid, caffeic acid, quercetin and sinapic acid showed superior activity compared to piroxicam. This shed the light about the efficacy of *Opuntia monacantha* in the alleviation of inflammation and arthritis and thus could serve as promising agent to be incorporated in pharmaceutical preparations.

5. Conclusion

In summary, results illustrated in current research concluded that ethanol extract of *Opuntia monacantha* showed promising anti-arthritic and anti-inflammatory properties in animal models. This suggested that it could be a good candidate for treating inflammation and arthritis as well. Furthermore, *Opuntia monacantha* extract can reduce TNF-α, COX-2, IL-1, IL-6, NF-κβ levels, with concomitant elevation of the IL-10, IL-4, and Iκ-B. Besides, molecular docking studies displayed that all tested compounds exhibited pronounced inhibitory potential towards glutaminase 1 approaching piroxicam suggesting an additional probable mode of action to the extract. More research into the possible application of *O. monacantha* extracts in arthritic patients, as well as safety studies in addition to clinical trials, is required to determine the safety and feasibility of any *O. monacantha* extracts-based clinical therapies.

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Ethical approval

The Institutional Ethical Committee at the University of Lahore, Pakistan, authorized the experimental with approval number IMBB/UOL/20/416.

CRediT authorship contribution statement

Farah Abid: Conceptualization, Formal analysis, Investigation, Methodology, Writing – review & editing. **Mohammad Saleem:** Conceptualization, Formal analysis, Investigation, Methodology, Writing – review & editing. **Talha Jamshaid:** Conceptualization, Formal analysis, Investigation, Methodology, Writing – review & editing. **Usama Jamshaid:** Conceptualization, Formal analysis, Investigation, Methodology, Writing – review & editing. **Fadia S. Youssef:** Investigation, Software, Validation, Writing – original draft, Visualization. **Reem M. Diri:** Funding acquisition, Project administration, Resources, Validation, Writing – review & editing, Data curation. **Sameh S. Elhady:** Funding acquisition, Project administration, Resources, Validation, Writing – review & editing, Data curation. **Mohamed L. Ashour:** Data curation, Supervision, Validation, Writing – original draft, Project administration.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jep.2024.117884.

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FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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Review

Pereskia aculeata Miller as a Novel Food Source: A Review

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Abstract: *Pereskia aculeata* Miller is an edible plant species belonging to the Cactaceae family. It has the potential to be used in the food and pharmaceutical industries due to its nutritional characteristics, bioactive compounds, and mucilage content. *Pereskia aculeata* Miller is native to the Neotropical region, where it is traditionally employed as food in rural communities, being popularly known as 'ora-pro-nobis' (OPN) or the Barbados gooseberry. The leaves of OPN are distinguished by their nontoxicity and nutritional richness, including, on a dry basis, 23% proteins, 31% carbohydrates, 14% minerals, 8% lipids, and 4% soluble dietary fibers, besides vitamins A, C, and E, and phenolic, carotenoid, and flavonoid compounds. The OPN leaves and fruits also contain mucilage composed of arabinogalactan biopolymer that presents technofunctional properties such as thickener, gelling, and emulsifier agent. Moreover, OPN is generally used for pharmacological purposes in Brazilian folk medicine, which has been attributed to its bioactive molecules with metabolic, anti-inflammatory, antioxidant, and antimicrobial properties. Therefore, in the face of the growing research and industrial interests in OPN as a novel food source, the present work reviews its botanical, nutritional, bioactive, and technofunctional properties, which are relevant for the development of healthy and innovative food products and ingredients.

Keywords: plant proteins; mucilage; bioactive molecules; sustainable protein sources; functional properties; ora-pro-nobis

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1. Introduction

The continuous increase in the global population associated with agricultural expansion, climate changes, and awareness of the importance of ecological preservation has pressured researchers and industries to find more sustainable food sources [1–3]. In fact, animal source foods are related to increments of deforestation, greenhouse gas emissions, water consumption, and risks to human health [4,5]. As a consequence, there is a growing interest in plant-source foods as an answer to more sustainable agricultural practices [2,6]. Moreover, food consumption has also been influenced by ethnic, cultural, and religious beliefs, with an increasing number of people with dietary restrictions, such as vegans, vegetarians, and flexitarians. In this way, plant-source foods can satisfy the demand for edible proteins while providing essential nutrients to the human diet in a more sustainable manner [3,7].

Recently, many unconventional edible plants have been researched regarding their nutritional, biological, and industrial potentialities [8,9]. Among these alternative plant species, *Pereskia aculeata* Miller is a nutrient-rich cactus native to Latin American countries [10] that offers interesting characteristics for food and pharmacological applications. *P. aculeata* Miller is also known as 'ora-pro-nobis' (OPN), blade-apple cactus, Barbados

gooseberry, leaf cactus, lemon vine, and rose cactus. The leaves of OPN are distinguished by their nontoxicity and nutritional richness, including, on a dry basis, 23% proteins, 31% carbohydrates, 14% minerals, 8% lipids, and 4% soluble dietary fibers, besides significant content of vitamins A, C, and E and diverse phenolic, carotenoid and flavonoid compounds [11–14]. Furthermore, the OPN leaves and fruits contain mucilage composed of arabinogalactan biopolymer that presents technofunctional properties such as thickener, gelling agent, and emulsifier [15–17], as well as wound healing characteristics [18]. These assets demonstrate the potential use of OPN mucilage in the food and pharmaceutical industries.

In this context, this review focuses on the general aspects of OPN as a novel food source, including the botanical, nutritional, bioactive, and technofunctional properties of OPN leaves and fruits. We evaluated 241 scientific articles available on Web of Science and 34 on Scielo (Latin American scientific database) that contained the word "*Pereskia*" in any part of the text. It should be highlighted that this review was inspired by the potential of using OPN for the development of healthy, affordable, and innovative food products and ingredients, particularly in the case of Latin American countries where OPN is naturally found.

2. General Botanical Characteristics

According to an update of the angiosperm phylogeny group (APG) classification [19], the Cactaceae family is circumscribed to the eudicots group (or tricolpate), Superasterids clade, Caryophyllales order. Indeed, Cactaceae is one of the most interesting angiosperm botanical families, as it presents a photosynthetic mechanism that concentrates atmospheric inorganic carbon, described as crassulacean acid metabolism (CAM), specialized for survival in arid environments. CAM allows the opening of the stomata complex at night (saving water) to capture carbon dioxide (CO_2) that is stored in the form of malic acid (or malate) and used in photosynthesis during the next day [20]. Such a physiological mechanism permits their survival during prolonged drought while keeping a healthy tissue water status [21]. The Cactaceae family includes about 100 genera and approximately 1500 species that are mainly distributed in the Neotropical region (Mexico, Brazil, and Chile) except for the *Rhipsalis* genus that occurs in tropical Africa, besides many species of the *Opuntia* genus introduced in Africa, Australia, and India [22]. Among the main genera belonging to the Cactaceae family, *Pereskia* genus (subfamily Pereskioideae) is a leafy shrub and tree native to the American continent, comprising 17 species that are distributed from Argentina to Florida [10,21,23].

Phylogenetic analysis reveals that *Pereskia* is a paraphyletic genus that originated from all other cactus species. *Pereskia* retained numerous plesiomorphic character states, such as non-succulent stems, persistent leaves, cymose inflorescences, stylet (polyestemon), and, in some species, a superior ovary with basal placentation [22]. As these characteristics are absent in other Cactaceae family members, it is reported that *Pereskia* originated a clade characterized by the presence of complex stomata in the stem and by the late formation of the bark, which promoted photosynthesis in the stem [22]. In this sense, the *Pereskia* genus is considered an ancestral of the modern cacti. Although the species belonging to the *Pereskia* genus can grow under limited water availability (as a conventional cactus does), they differ from the other cacti mainly because they do not possess a photosynthetic stem and also because they present well-developed leaves, in which water and most nutrients are accumulated [14,21,24,25]. Among the *Pereskia* species, *Pereskia aculeata* Miller (OPN), in particular, is traditionally employed as food in Brazilian rural communities due to the sensorial acceptance and high nutritional content of its leaves, including significant amounts of proteins, fibers, minerals, and vitamins [14,26,27].

Regarding the OPN morphological characteristics, it can be described as a liana (woody vine) perennial and shrubby species (Figure 1), reaching 4 m in height or even more, that has become an invasive plant in the world [28]. The older well-lignified stems are endowed with prominent spines that demand care for handling [23,29]. It presents long branches

and simple elliptical leaves with short petioles and succulent texture, from 3–8 cm up to 15 cm long [23,29]. The inflorescences are short, numerous, with cream-yellow flowers, sometimes with a red center, arranged on the foliage and formed in spring or summer, visited by bees and carpenter bees. The OPN fruits, when ripe, are globular, yellowish, edible, and berry-like, with glochids and black seeds [30] (Figure 2).

Figure 1. (**A**,**B**) Flowers and (**C**,**D**) leaves of *Pereskia aculeata* Mill.

With respect to agronomic performance, OPN is a rustic species, tolerant to arid conditions, that presents vegetative growth all year long and that does not require high soil fertility; consequently, it is adapted to diverse soil types and edaphoclimatic conditions [23,29]. This species can be propagated with stem cuttings or by seeds and can also be used as a rootstock for other cacti [31]. When cultivated at high density (~10 plants per m^2), OPN can produce 5759 kg of proteins in the leaves (main edible part) per hectare per year [24]. For comparison purposes, considering only the grains, the average soybean (*Glicyne max* (L.) Merr.) and maize (*Zea mays* L.) protein productions per hectare per year are 1154 kg and 514.7 kg, respectively [24].

(**A**) (**B**)

Figure 2. (**A**) Green fruit and (**B**) mature fruit of *Pereskia aculeata* Mill.

3. Nutritional Characteristics of OPN Leaves

Several works have studied the chemical composition of OPN leaves, and the results are summarized in Table 1. As can be seen (Table 1), almost 90% *w*/*w* of OPN leaves are represented by water. The remaining dry matter is quite diverse and contains all classes of essential nutrients to the human diet.

Table 1. Nutritional composition of *Pereskia aculeata* Mill. (ora-pro-nobis) leaves. The values are presented on a dry matter basis.

Nutrient	Range	Average	Reference
Moisture fresh leaves (% *w*/*w*)	83.3–91.1	88.1	[13,24,32,33]
Proteins (% *w*/*w*)	14.3–29.0	23.3	[13,24,32–34]
Lipids (% *w*/*w*)	4.1–16.3	8.5	[13,32,33]
Carbohydrates (% *w*/*w*)	29.5–32.3	30.9	[32,33]
Soluble dietary fiber (% *w*/*w*)	2.4–5.2	3.8	[13,32]
Insoluble dietary fiber (% *w*/*w*)	19.2–33.9	26.6	[13,32]
Minerals (% *w*/*w*)	10.8–16.1	13.9	[13,32,33]
Calcium (% *w*/*w*)	1.3–4.6	3.4	[13,24,32,33]
Magnesium (% *w*/*w*)	0.6–1.9	1.0	[13,24,32,33]
Potassium (% *w*/*w*)	1.6–3.9	3.1	[13,24,32]
Phosphorous (mg/kg)	1560–5600	3770	[13,24,32,34]
Manganese (mg/kg)	88–464	327	[13,24,32]
Zinc (mg/kg)	37–267	106	[13,24,32,34]
Iron (mg/kg)	142–244	190	[13,24,32,34]
Copper (mg/kg)	12–14	13	[13,32]
Vitamin E (mg/kg)	14–49	32	[33,34]
Total carotenoids (mg/kg)	250–354	302	[33,34]
β-carotene (mg/kg)	78–430	250	[13,32–34]
Vitamin A (mg/kg)	23–25	24	[13,33]
Vitamin C (mg/kg)	430–1858	1144	[13,32]

Carbohydrates represent approximately one-third of the dry matter of OPN leaves. They are present as structural and highly ramified polysaccharides formed by galactopyranose, arabinofuranose, arabinopyranose, rhamnopyranose, uronic acid, and fucose [16,35]. These complex polysaccharides are known as mucilage (non-toxic) and can be used as hydrocolloids in food processing due to their high water absorption capacity [12,36,37]. In

fact, the first process for obtaining OPN mucilage was described in 1982 in Sierakowski's protocol [37] by using benzene, ethanol, water, and acetone. This study showed that OPN mucilage was composed of an arabinogalactan, formed mainly by arabinose and galactose, containing 3.5% protein in relation to the polysaccharide content. Lima-Junior et al. [35] optimized the OPN mucilage extraction process by using only ethanol as a solvent, and the obtained mucilage (as powder) presented 10.5% protein and 46.9% carbohydrates. OPN leaves have a high arabinogalactan content, characterized as a biopolymer primarily composed of a main backbone of (1→4) β-D-galactopyranose with branches of galactose, arabinose, rhamnose, and galacturonic acid at the C-3 position, in the proportion of 5.4:8.3:1.8:1.0, respectively [16,35]. Size exclusion chromatography analyses revealed that the arabinogalactan material is heterogeneous, presenting an average molar mass of 7.9×10^5 g/mol [16]. This arabinogalactan is considered 'type I' because proteins are associated with the polysaccharide chain by covalent bonds. This proteoglycan complex is typical of the cell walls of higher plants, and it is related to the physical–chemical properties of OPN mucilage [12,16].

Dietary fiber is a plant material resistant to enzymatic digestion, which is essential to human health because its consumption has been associated with a decreased incidence of numerous diseases [38]. Thus, another positive nutritional aspect of OPN leaves is their significant content of dietary fibers, including both soluble and insoluble fractions. Concerning the lipid fraction, which is also present in a considerable concentration in OPN leaves, to date, no work has evaluated their composition, i.e., the already published papers were limited to analyzing the total lipid concentration by Soxhlet methods [13,32,33,39]. Concerning minerals and vitamins, their contents in OPN leaves are impressive. Considering the nutrient reference value requirements (per day) for an adult, established by FAO [40], a portion of 30 g of OPN leaf flour (dried leaves) can provide, on average: 100% calcium, 100% magnesium, 160% phosphorus, 250–400% iron, 230–290% zinc, 430% copper, 3200% manganese, 900% vitamin A, 340% vitamin C, and 100% vitamin E.

Regarding the protein fraction, it is interesting to note that the protein content of OPN leaves, on a dry basis, is quite similar to whole milk powder from cows (~25% *w*/*w*) [41], which underlines the nutritional relevance of OPN leaves for human consumption. In this sense, their amino acid composition was analyzed by Takeiti et al., (2009) [13] and Silveira et al., (2020) [42]. Although these works diverged about the amino acid proportions, both reported that OPN leaves provide all essential amino acids in suitable proportions for child and adult nutrition. Only methionine and lysine were present slightly below the ideal levels recommended by the World Health Organization [43]. Furthermore, the protein fraction must be digestible by the human gastrointestinal tract to be considered a good protein source. According to Takeiti et al., (2009) [13], the in vitro digestibility of OPN leaf proteins was 75.9%, which is similar to the digestibility of beans, wheat, and rice [43].

Considering the physical–chemical characteristics, it was demonstrated by sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) that the OPN leaf proteins present molar masses ranging from 15 to 97 kDa, with significant bands found at 61 kDa, 53 kDa, 33 kDa, and 15 kDa [13]. After separation by cryogel chromatography, followed by Fourier-transform infrared (FTIR) spectroscopy, Neves et al., (2020) [44] showed that the secondary structures of OPN leaf proteins were mainly composed of β-sheet (46.5%) and α-helix (13.9%), indicating a stable conformation for these proteins. They also observed a zeta potential of 5.8 mV at pH 3.2, which suggests an isoelectric point slightly above this pH value. Finally, Morais et al., (2019) empirically evaluated the protein recovery of OPN leaf proteins by combining salting out, temperature, and isoelectric precipitation [45]. These authors noticed that the highest protein recovery in the presence of precipitating salts (about 69%) was achieved using ammonium sulfate (NH_2SO_4) at 0.5 M and 85 °C. In the absence of precipitating salts, the highest protein recovery was found between pH 3 (71%) and pH 4 (69%) at 85 °C. These results agree with Neves et al., (2020) [44] for an isoelectric point of OPN leaf proteins between pH 3 and 4. Ultimately, by accessing all scientific papers containing the word "*Pereskia*" in any part of the text (by using Web

of Knowledge and Scielo databases), it was verified that only three scientific works—Takeiti et al., (2009) [13], Morais et al., (2019) [45] and Neves et al., (2020) [44]—presented qualitative physical–chemical information about OPN leaf proteins.

4. Bioactive Properties of OPN Leaves

OPN is also popularly used for pharmacological purposes in Brazilian folk medicine, which is attributed to its metabolic, anti-inflammatory, antioxidant, and antimicrobial properties, among others, as reviewed by Agostini-Costa (2020) [46] and Porto et al., (2021) [14]. Concerning the metabolic aspects, Barbalho et al., (2016) [39] administrated OPN leaf flour to Wistar rats and observed significant health improvements, including a reduction in weight gain, visceral fat, levels of total cholesterol, triglycerides, low-density lipoprotein, very-low-density lipoprotein, and increased HDL-c and enhancement of intestinal motility. Moreover, in double-blind, randomized clinical studies with humans, Vieira et al., (2019, 2020) [47,48] noticed that the consumption of OPN leaf flour in biscuits and beverages improved intestinal health and reduced weight, waist circumference, and body fat and increased satiety. The authors suggested that these results are associated with the presence of dietary fiber and phytochemical compounds, such as polyphenols, in the OPN leaves.

Another important pharmacological use of OPN is related to its anti-inflammatory properties. Pinto et al., (2015) [49] studied the topical anti-inflammatory activity of a hexane extract of OPN leaves in acute and chronic ear dermatitis in mice. The authors verified that the OPN extracts significantly reduced inflammatory processes induced by various toxic agents. In addition, the OPN extracts did not exhibit toxicity for dermatological applications. The same research group [50] also verified an antinociceptive activity using a methanolic OPN extract in mice treated with acetic acid. The authors attributed the analgesic effect to alkaloids and quercetin in OPN extract. Carvalho et al., (2014) [18] produced ethanolic extracts of OPN cultivated in different soil types and evaluated their in vitro wound healing properties and cytotoxicity using mouse fibroblast cells. They observed that OPN extracts obtained from all tested soils were safe and effective as a wound healing agent, whose beneficial effects were attributed to the mucilage of the OPN leaves. The main health-related effects of OPN leaves are summarized in Table 2.

Table 2. Summary of the main health-related properties of OPN leaves.

Properties	Type of Study	Main Results	References
Metabolic	In vivo using Wistar rats	Improved metabolic profile Increased intestinal mobility	[39]
	In vivo randomized cross-over intervention with adult men	Improved gastrointestinal symptoms Increased satiety	[47]
	In vivo double-blinded randomized clinical trial with adult women	Reduced weight, waist circumference, body fat, eructation, and constipation Increased satiety Improved feces consistency	[48]
Anti-inflammatory	In vivo using Swiss and Wistar rats	Reduction of the inflammatory process of acute and chronic ear dermatitis Absence of dermal toxicity	[49]
	In vivo using Swiss rats	Antinociceptive activity (analgesic effect)	[50]
	In vitro using fibroblast cells L929	Wound healing capacity Absence of cytotoxicity	[18]

One of the main popular claims of OPN leaves concerns their antioxidant properties. In fact, OPN leaves contain high concentrations of several classes of antioxidants, including carotenoids (α and β-carotene, lutein, zeaxanthin, and violaxanthin) [51], phenolic compounds such as caffeic, chicoric, and coumaric acid derivatives, flavonoids (quercetin, kaempferol, and isorhamnetin glycoside derivatives) [52,53], and terpenoids

(phytol, γ-tocopherol, vitamin E, squalene, and lupeol) [54], among others. In general, the extraction procedures of antioxidant molecules from the OPN leaves are achieved by mixing them with organic solvents, such as ethanol, acetone, methanol, and hexane [51–53]. However, it is also possible to improve antioxidant extraction using supercritical fluid technology with CO_2 [54]. In a study focused on the antioxidant properties of OPN leaves [51], the authors produced a hydroethanolic extract that was effective in inhibiting the growth of Gram-positive and Gram-negative bacteria, including human pathogens such as *Listeria monocytogenes*, *Staphylococcus aureus*, and *Klebsiella pneumoniae*. Last, but not least, it is noteworthy that the works evaluating the toxicity of OPN leaves to humans showed that they are safe for food and therapeutic applications [52,55].

5. Technofunctional Properties of OPN Leaves

From a food processing perspective, technofunctional properties or technological functionalities can be defined as the effectiveness with which a particular food component or ingredient provides desirable attributes in a food or beverage [8,56]. To date, the technofunctional properties of OPN leaves have been studied in the formulation of food products in the forms of flour [48,57,58], mucilage [17,36], and fresh leaves [59].

In terms of flour, e.g., dried and grounded OPN leaves, Sobrinho et al., (2015) [57] evaluated the effects of OPN leaf flour on the texture, color, and sensory acceptance of cooked sausages. The authors observed that sausages with 1–2% of OPN leaf flour were darker and softer than the control ones, while sensory acceptance was kept unchanged. In this same direction, Sato et al., (2018) [58] studied the impact of adding 10–20% of OPN leaf flour on the functional and sensory characteristics of pasta. The authors noticed that, compared with the control, the enriched samples were darker, softer, gained more weight during cooking, lost less weight after cooking, and presented higher contents of proteins, fibers, and minerals. At the same time, the color, flavor, odor, texture, and overall appearance were equally appreciated by the consumers of both types of pasta. Finally, in a double-blind, randomized study, Vieira et al., (2020) [48] researched the effect of a dairy-based beverage containing 5% w/w OPN leaf flour on intestinal microbiota, gastrointestinal symptoms, and anthropometric parameters in women aged from 20 to 60 years old. The results demonstrated that the daily consumption of the beverage with OPN leaf flour for 6 weeks improved feces consistency, increased satiety, and reduced weight, waist circumference, and percent body fat. However, no effect was detected on the composition of the intestinal microbiota. Briefly, these works show that it is possible to use OPN leaf flour to produce various food products, improving their nutritional content while maintaining their technofunctional properties and sensory acceptance.

Recent studies have demonstrated the potential of OPN mucilage for industrial applications. It was shown that OPN mucilage could form gels and emulsions at different hydrocolloid concentrations and temperatures [35,60,61] in the presence or not of sodium chloride and sucrose and at different pH values [62]. Furthermore, OPN mucilage is able to form oil-in-water nanoemulsions through ultrasonication [63] and can be used to produce biodegradable films when associated with glycerol [64]. The technofunctional properties of OPN mucilage were also studied in fermented dairy drinks, leading to increased protein content and viscosity of the product, as well as reduced syneresis [65]. Neves et al., (2020) [66] produced microparticles of OPN mucilage and whey protein isolate to encapsulate α-tocopherol loaded in canola oil or coconut oil. After encapsulation, the bioactive activity of α-tocopherol was retained over 35 days, and its bioaccessibility was higher when using canola oil as a carrier. Lise et al., (2021) [36] analyzed the potential of OPN mucilage as an emulsifier and fat replacer in producing a mortadella-type meat product. The authors verified that it was possible to reduce the fat content of mortadella without impairing its texture and sensory characteristics.

Additionally, even as fresh leaves, OPN is technologically viable in food formulations, such as chocolate cake production, with a consequent increase in protein, fiber, and mineral contents and a reduction in total calories [59]. Neves et al., (2021) [67] evaluated the bioac-

tive compound levels, antioxidant activity, and bioaccessibility of carotenoids from OPN leaves submitted to different cooking methods. It was found that the cooking techniques (stir-frying, microwaving, and steaming) influenced the phytochemical composition of OPN leaves. In general, the cooking processes softened plant tissues, increased bioactive compounds' extraction, and enhanced their bioaccessibility. These results may be used to improve dietary intake recommendations for bioactive compounds and increase the nutritional value of food products.

Overall, the available data highlight the technofunctional and nutritional potential of using OPN leaves in food processing, whether as dried/fresh leaves or isolated extract (mucilage). However, to date, no paper has studied the effects of isolated OPN leaf proteins on the gelling, emulsifying, and foaming properties of food products. This shows the vast scientific potential to study these proteins and develop new food products from OPN leaves.

6. Characteristics of OPN Fruit

The OPN fruit originates from its flower (monocline and actinomorphic), where the ovary occurs within the hypanthium, being considered, therefore, as superior [68]. The flowers are arranged in terminal panicles, composed of four ovate green petals measuring 0.6 to 0.8 cm in length, and the corolla has 8 to 12 oblong white petals (longer than wide) of white color measuring 2 cm in length. This hypanthium presents a green color, fleshy consistency, thick cell walls, and green lanceolate bracteoles, besides spines at the bases of the leaves. At the beginning of its development, OPN fruit consists of a succulent hypanthium, pericarp, and seeds wrapped in gelatinous material [69]. In the young pericarp is observed a differentiation in mucilaginous cells, which occurs in large numbers in the innermost region of the mesocarp [69]. With the ripening process, the hypanthium acquires a yellow-orange color (Figure 2B) while it loses its bracteoles and aculeus; during this period, it develops a small opening in the apical region [68,69]. In the mature stage, the fruits are rounded, dark yellow, with a yellowish and sweet pulp involving two to four discoid seeds of brownish-black color. The inner part of the mesocarp and the entire endocarp change their structure, forming a gelatinous mass that surrounds the seeds [69,70].

Adult OPN plants produce their fruits between June and July, which are edible, pomaceous, cactidium-type, and small (1.8 to 2.0 g per fruit) [70]. The chemical composition of OPN ripe fruits was studied by Queiroz et al., (2009) [71] and Agostini-Costa et al., (2012) [70]. The average chemical content of OPN fresh fruits is shown in Table 3. In addition, Agostini-Costa et al., (2012) [70] characterized the total and phenolic carotenoid profile of mature OPN fruit, finding trans-β-carotene as the primary carotenoid, followed by α-carotene, lutein, and other minor carotenoids. Unlike the leaves, the use of OPN fruits for human nutrition is still very restricted due to the lack of studies on their properties and the difficulties in collecting and processing it. Consequently, research into the nutritional properties of OPN fruits can broaden its use, both in natura and after processing, as jam, syrup, or juices, for example.

Table 3. Chemical composition of OPN ripe fruits.

Characteristic	Range	Average	Reference
Moisture (% *w*/*w*)	87.4	87.4	[71]
Ashes (% *w*/*w*)	0.9	0.9	[71]
Protein (% *w*/*w*)	0.0–1.0	0.5	[70,71]
Lipids (% *w*/*w*)	0.2–0.7	0.5	[70,71]
Carbohydrates (% *w*/*w*)	6.3–11.5	8.9	[70,71]
Ascorbic acid (% *w*/*w*)	2.0–125.0	63.5	[70]
Niacin (mg/100 g)	0.9	0.9	[70]
Calcium (mg/100 g)	174.0–206.0	190.0	[70]
Phosphorus (mg/100 g)	26.0	26.0	[70]
pH at 22 °C	4.2	4.2	[71]

As observed for OPN leaves, the green fruits of OPN present mucilage with thickening and emulsifying properties. Silva et al., (2019) [72] extracted mucilage from OPN green fruits (Figure 2A) by cold extraction and lyophilization processes and investigated its physicochemical properties. The lyophilized mucilage from OPN green fruit exhibited, on a dry basis: 2.9% moisture, 67.2% carbohydrates, 19.8% proteins, 8.3% ashes, 0% lipids, 4.6% crude fiber, 0.2% phosphorus, 1.8% potassium, 2.7% calcium, 0.07% magnesium, 0.2% sulfur, 0.001% boron, 0.002% copper, 0.004% manganese, 0.005% zinc, 0.04% iron, and 0.02% sodium. FTIR spectroscopy of the mucilage obtained from OPN green fruit showed characteristic bands of polysaccharides and protein chains and an additional peak in 1722 cm^{-1}. This peak referred to the presence of carboxylic groups that serves as chemical sites for ionic bonds, which contribute to the ability to form gels [72,73]. Moreover, solutions of the mucilage from OPN green fruit with increasing concentrations (0.5, 1.0, 1.5, 2.0, and 2.5 g/100 mL of aqueous phase) presented pseudoplastic behavior [72]. It was also observed that increments of the mucilage concentration increased the apparent viscosity, emulsification capacity, and emulsion stability and decreased the mean size of the oil droplets.

7. Conclusions

OPN is a nonconventional edible plant adapted to diverse climate conditions that requires low soil fertility for satisfactory vegetative growth. As OPN is a cactus species that does not possess a photosynthetic stem, most of the water and nutrients are accumulated in its leaves, which are employed in traditional South American dishes. The OPN leaves are nontoxic and present all nutrient classes in significant amounts, including proteins, carbohydrates, fibers, lipids, minerals, vitamins, and diverse phenolic, carotenoid, and flavonoid compounds with metabolic, anti-inflammatory, antioxidant, and antimicrobial activity. Moreover, OPN leaves and fruits have been identified as an interesting source of mucilage composed of polysaccharides and proteins. This mucilage has a heterogeneous macromolecular profile with a polyelectrolyte behavior that can be employed as an emulsifying and stabilizing agent due to its interfacial adsorption properties. Regarding the protein fraction, the OPN leaf proteins are still little studied. However, when cultivated at high density, the protein productivity per hectare can be 500% higher than that of soybeans (*G. max*) and 1,100 % higher than that of maize (*Z. mays*), demonstrating the tremendous potential of OPN leaves as a perennial source of nutrients for human health and ingredients for the food industry. In general, the scientific literature demonstrates that it is possible to use OPN leaves in the form of flour, fresh leaves, or isolated extract (e.g., mucilage) to improve the nutritional content of food products while keeping their technofunctional properties. At the same time, there is still a broad field for future research and development focused on food applications of OPN leaves and fruits, comprising the optimization of their components' extraction as well as their use as gelling, emulsifying, and foaming agents in food formulation.

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ARTICLES FOR FACULTY MEMBERS

FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

Title/Author	**Physicochemical, nutritional, and medicinal properties of Opuntia Ficus-Indica (l.) Mill. and its main agro-industrial use: A review / Martins, M., Ribeiro, M. H., & Almeida, C. M. M.**
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Review

Physicochemical, Nutritional, and Medicinal Properties of *Opuntia ficus-indica* (L.) Mill. and Its Main Agro-Industrial Use: A Review

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Abstract: The cactus, *Opuntia ficus-indica* (L.) Mill. (OFI) belongs to the *Cactaceae* family, which contains about 130 genera and nearly 1600 species. This review aims to evaluate this plant from several perspectives, namely, botanic, physicochemical, nutritional, and medicinal properties, as well as agro-industrial use. The botanical aspects and morphological characteristics of OFI enable genetic variability, ecological adaptation, and broad geographic distribution. Due to its physicochemical and nutritional composition, it has several medicinal properties appropriate (or suitable) for several industries, such as pharmaceutical, food, and cosmetics. Its fruit, the prickly pear (PP), has potential agro-industrial expansion through the application of different conservation and transformation methods, making it possible to obtain a variety of products. The PP is a source of several nutrients and is an effective system to produce varied foods, which have several advantages from a nutritional, sensory, economic, and shelf-life point of view.

Keywords: *Opuntia ficus-indica*; prickly pear; food composition; medicinal properties; agro-industrial applications

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1. Introduction

Opuntia ficus-indica (L.) Mill. (OFI) belongs to the *Cactaceae* family, characterised by genetic variability, ecological adaptation, and broad geographic distribution (tropical and subtropical regions) [1–3]. This plant is originally from South America, namely Mexico, and can be found in the Middle East, South Africa, India, Australia, and some Mediterranean countries [1–3]. OFI adapts to extreme weather conditions, has rapid growth in poor soils, and has little need for water [4]. Thus, this species is the most cultivated in the world.

Before starting an OFI plantation, there are several aspects to consider, namely the climate of the place of cultivation, the physical and chemical characteristics of the soil and its preparation, the choice of the cultivar, the planting distances, the orientation crop rows, and the irrigation system [5].

The best harvest time depends on several factors, such as the size, weight, and firmness of the fruit, changes in peel colour, degree of receptacle depth of the flower, the total soluble solids (TSS) or sugar content, the minimum of 14 °Brix, the drop of glochids, the thickness of the peel, the ease of skin removal, and the pulp/peel ratio [6].

OFI consists of leaves, flowers, fruits (prickly pear), cladodes, and roots. OFI presents an ecological adaptation because of the photosynthetic metabolism in cactus cladodes, the crassulacean acid metabolism (CAM). The leaves are only visible on the tender cladodes. They are cylindrical and deciduous, and remain on the plant for over a month [7]. The flowers are large, showy, hermaphroditic, and pollinated by insects or wind; they have no aroma, but they have beautiful colours [4,7]. The prickly pear (PP) is a fleshy, oval, or

cylindrical berry 5 cm to 10 cm long and 4 cm to 8 cm wide, and weighs between 80 g and 200 g. The PP shows three components: peel (30 to 40%), pulp (60 to 70%), and seeds (2 to 10%) [7]. The edible part of the fruit has excellent organoleptic characteristics, such as a very aromatic pulp and hard integument seeds, which are excellent mineral sources [4,8].

The diversity of physicochemical and nutritional characteristics of OFI depends on the type of plant (genetic factors), its origin (climate) and agronomic factors (type of cultivation, fertilisation, and irrigation) [5]. The main constituent of cladodes is water, followed by carbohydrates, fibres, minerals (potassium, calcium, magnesium, phosphorus, sodium, manganese, iron, and zinc), proteins, reducing sugars, and lipids [2,9–11]. In addition, young cladodes contain ascorbic acid, carotenoids, and chlorophyll [5,12,13]. OFI flower extracts are a source of bioactive substances and have potential use as a food preservative [14]. The flowers predominantly accumulate yellow and red betalains and colourless phenolic compounds (gallic acid, 6-isorhamnetin 3-O-robinobioside, and 7-isorhamnetin 3-O-galactoside) [9,13,15]. The PP comprises water, sugars, ascorbic acid, fibres, amino acids, minerals, and antioxidant compounds such as phenols, flavonoids, betalains, and carotenoids [1,5,13,16]. The energetic value of this fruit varies between 31 and 50 kcal/100 g, comparable to other fruits such as apples, oranges, peaches, and pears [6].

OFI has several properties that allow its use in various applications, such as human and animal feed, in the pharmaceutical, cosmetic and food industries, in civil construction, in alternative fuels, in controlling soil erosion, protecting fauna, and as a source of nectar for bees [2,5,8,17,18].

Regarding the medicinal properties of OFI, several studies confirm that the fruits and cladodes can be used as a source of nutrients and phytochemicals. In this way, OFI is valued for contributing to a healthy diet and because it is rich in health-promoting substances [5,6]. Some medicinal properties of OFI include antioxidant, analgesic, anti-inflammatory, diuretic, anti-diabetic, anti-hypercholesterolaemic and anti-carcinogenic [3–5,9,19–27]. These properties allow the prevention of hangovers, lipid oxidation–reduction, prevention of some types of cancer, and reduction of the risk of diabetes (type 2) and cardiovascular diseases, two of the most common causes of death worldwide [5,28,29].

During storage, the PP can be contaminated or suffer changes in its physical, chemical, and biological characteristics, leading to losses of some food constituents. Among the most identified alterations are microbial contamination, enzymatic browning, reduced firmness, development of off-flavours, and physical deformations [8]. Furthermore, the PP has a short shelf life of about 3 to 4 weeks, is a low acidic fruit (0.05 to 0.18% citric acid equivalent) and has a pH value of 5.3 to 7.1, which compromises prolonged storage and distribution [9]. Thus, it becomes advantageous to apply conservation and transformation processes to PP: drying, freezing, concentration (physical methods), the addition of sugars, acidification, use of preservatives (chemical methods), and lactic or alcoholic fermentations (biochemical processes) [6].

Some of the products obtained from the various processing methods mentioned above are fresh preserved PP, minimally processed PP, dehydrated PP (dry or osmotically dehydrated), preserves, juices, fermented drinks, liquid sweetener, pulps (frozen or dehydrated), gums or gels, candies, jams or jellies, flours, seed oil, natural dyes, dietary fibres, and thickeners [7,8,30,31].

This review aims to approach the botanical aspects and morphological characteristics of OFI, its cultivation systems, worldwide distribution, physicochemical and nutritional composition, medicinal properties, applications, and agro-industrial uses, namely the conservation and transformation methods applied to the PP.

2. *Opuntia ficus-indica* (L.) Mill Species

2.1. Botanical Aspects

Cactus refers to the botanical family *Cactaceae*, which encompasses about 1600 species divided into 130 genera, divided into the three subfamilies *Pereskioideae*, *Opuntioideae* and *Cactoideae* [3,9]. In the order *Caryophyllales*, the botanical genus *Opuntia* is the most common,

grouping more than 300 species, namely the *Opuntia ficus-indica* (L.) Mill. (OFI) species [1]. The scientific name of this species was attributed in 1700 by the French botanist Joseph Pitton de Tournefort, due to the similarity to thorny plants that grew in the ancient Greek city of Opus. The OFI has various designations, depending on the country and the region where it is located. In Portugal, the common names of this species are prickly pear, agave, devil's fig tree, and tabaio or tabaibo (Madeira Archipelago) [7].

As a result of the long domestication to which it has been subjected, the OFI is characterised due to its genetic variability. Its taxonomy is complex, and its phenotypes vary according to the environmental conditions it is subjected to, increasing the diversity of adaptive responses. This plant has polyploidy, reproduction occurs sexually and asexually, and several hybrids are interspecific [5]. Thus, the taxonomic classification of this plant becomes difficult, and a detailed study is needed to recognise and identify each species, the varieties, and the adaptations reflected in its phenotype [6]. Table 1 describes the taxonomic classification of the OFI, according to Britton et al. (1919) and Bravo-Hollis et al. (1978) [32–34].

Table 1. Taxonomic classification of *Opuntia ficus-indica* (L.) Mill [32–34].

OFI Taxonomic	
Domain	*Eukaryota*
Kingdom	*Plantae*
Subkingdom	*Embryophyta*
Phylum	*Spermatophyta*
Division	*Magnoliophyta*
Class	*Magnoliopsida*
Subclass	*Caryophyllidae*
Order	*Caryophyllales*
Suborder	*Cactineae*
Family	*Cactaceae*
Subfamily	*Opuntioideae*
Tribe	*Opuntieae*
Genus	*Opuntia*
Subgenus	*Plantyopuntia*
Species	*Opuntia ficus-indica* (L.) Mill

OFI is derived from a diploid Mexican ancestry, but polyploidy is favoured by natural hybridisation [35]. Several studies have reported this species as octoploid, heptaploid, pentaploid, hexaploid, and diploid [36–39]. There is a variation in the number of chromosomes of this species, depending on the origin of the plant [5].

Cactaceae have a set of adaptive, evolutionary, and ecological strategies, which give them a great capacity for development in different habitats [8]. OFI adapts to extreme weather conditions and grows rapidly in poor soils with little need for water [4]. Thus, this species is the most cultivated in the world and the most economically important within the genus *Opuntia*, being a viable option in tropical and subtropical regions, where other plants cannot survive, mainly in arid areas (annual rainfall of less than 250 mm) and semi-arid regions (annual rainfall of 250 to 450 mm) [10].

2.2. Morphological Characteristics

OFI presents ecological adaptations due to the photosynthetic metabolism that occurs in cacti, namely, crassulacean acid metabolism (CAM). In these plants, the stomata open at night to fix carbon dioxide (CO_2) and accumulate oxygen (O_2), simultaneously losing water (H_2O), which leads to the gradual acidification of the stem. During the day, they keep the stomata closed and prevent the loss of H_2O through transpiration. The CO_2 is fixed overnight as malic acid ($C_4H_6O_5$), which is stored in vacuoles and used during the day [7]. In conditions of extreme water deficiency, stomata remain closed day and night, preventing transpiration and the entry of CO_2 [6]. In this way, CAM plants increase the efficiency of using H_2O and the ability to survive in arid and semi-arid environments [5].

Anatomically, the OFI consists of roots, cladodes, leaves, flowers, and fruits (Figure 1). OFI can reach up to 5 m in height and is very extensive, fleshy, and densely branched, with fine, absorbent surface roots. The root system develops horizontally, reaching laterally up to 10 to 15 m from the base of the plant [40,41]. The length of the roots is related to the environmental conditions, the type of soil, the availability of water, and the cultivation practices, mainly irrigation and fertilisation [6].

Figure 1. *Opuntia ficus-indica* (L.) Mill.

OFI stems, called cladodes, are fleshy and succulent, and are responsible for CAM (Figure 1). They have an ovoid or elongated shape, and their weight ranges from 40 to 100 g. The cladode length varies between 30 and 50 cm, the width varies between 20 and 30 cm, and the thickness varies between 2 and 4 cm. The outer part of the cladode is the chlorenchyma (green part), which is essential for its photosynthetic action, and the inner part of the cladode is the parenchyma, which corresponds to about 50 to 70% of the cladode, where H_2O and organic acids are stored [9,40,41]. Depending on environmental conditions, the cladodes have areolas, capable of developing new cladodes, flowers, or roots. Areolas have two types of spines in their cavity, serving as a means of defence against damage from prolonged exposure to sunlight and the attack of animals. Large thorns are modified leaves, and the glochids are sharp and grouped in large numbers [6,41]. As the plant ages, the base cladodes lignify and form a trunk-like structure [40].

The leaves develop on the areolas, only visible on the tender cladodes. The leaves are cylindrical and deciduous, remain on the plant for just over a month, and are widely used as animal feed [7,41].

The flowers are large and showy, develop in the upper margin of leaves, are hermaphroditic, and are pollinated by insects or wind [7]. They have no aroma, but beautiful colours, such as yellow, orange, pink, purple, red, or white. Due to the limited flowering duration (March to June), few studies have been carried out on this plant's flowers [4,41].

The prickly pear (PP) is the OFI fruit that is formed from an inferior ovary located in the stem tissues (Figure 2). Its maturation is completed about 110 to 120 days after flowering, and its final weight can vary between 80 and 200 g. PPs are ovoid or cylindrical, 5 to 10 cm long and 4 to 8 cm wide [7,40].

Figure 2. White, orange, and red prickly pear ecotypes and image of the respective cross-sections of the fruits.

The PP can be divided into peel, pulp, and seeds. Of the total weight of PP, 30 to 40% corresponds to the peel, 60 to 70% corresponds to the pulp, and 2 to 10% corresponds to the seeds [5,42].

The PP peel can be divided into pericarp and mesocarp. The pericarp is thin and presents the same morphology as the cladodes, including the glochids, and the mesocarp is edible and has nutritional value. However, the mesocarp is not customarily consumed because it is discarded when the fruit is peeled [3,7]. In the initial phase of fruit development, the peel is green, evolving with its maturation to other colours. Depending on the ecotype and variety of the plant, the peel may be greenish-white, yellow, orange, red, purple, or purplish [3,6]. The pulp is the edible portion of the fruit. It is soft, juicy, translucent, gelatinous, and velvety, with a sweet taste. Its colour corresponds to the colour of the peel and it has numerous tiny seeds with hard integuments [3,8]. The seeds, distributed regularly throughout the fruit, are dark, edible and have been extensively investigated [3,43]. The PP's short shelf life is 3 to 4 weeks, limiting its long-term storage and worldwide distribution [9].

2.3. Cultivation System

A successful PP crop must follow specific rules and procedures, always respecting good agricultural practices [44]. The main climatic factors affecting any culture's yield and quality are temperature, precipitation, sun exposure and wind. Therefore, before starting an OFI plantation, there are several aspects to consider, namely the climate of the place of cultivation, the physical and chemical characteristics of the soil, the choice of the cultivar, the preparation of the soil, the planting distances, the orientation crop rows, and the irrigation system, among other factors [5].

Regarding planting material, this can be obtained from simple asexual vegetative reproduction from whole cladodes or parts of cladodes or through in vitro micropropagation [6]. This last method provides high propagation rates, requires little space for cultivation, allows the production of healthy plants without the intervention of pathogens, selects specific genotypes, avoids physiological disturbances and morphological anomalies, and obtains plants with the desired characteristics [45]. Khalafalla et al. (2007) demonstrated that OFI micropropagation, through areolas, can combat desertification in arid and semi-arid areas [46].

The PP is a perennial crop with an annual cycle, which can occur naturally with a second flowering and subsequent fruiting when favourable conditions exist [40]. The physiological development of the PP occurs between 70 and 150 days after flowering, depending on the production season, variety/ecotype, climatic conditions, and post-harvest

treatments. This development begins with an intense cell multiplication, followed by an increase in volume, which gives rise to the appearance of vacuoles, which accumulate nutrients [8]. There are three phases to fruit growth: dry matter growth of the husk, the development of dry matter of the seeds, and the development of the pulp. During maturation, changes occur in the colour of the peel and the texture of the fruit, the drop of glochids occurs, and the total soluble solids (TSS) content increases [40].

The parameters that help define the best PP harvest time are size, weight, and firmness of the fruit, changes in peel colour, degree of receptacle depth of the flower, the SST content, the minimum of 14 °Brix, the drop of glochids, the thickness of the peel, the ease of skin removal, and the pulp/peel ratio [6].

PPs are difficult to harvest due to glochids and spines, which can pierce the peel and enter the eyes and respiratory system of the person performing the collection. Therefore, this is carried out manually, during the night and until the beginning of the morning, when there is greater ease of cutting, better resistance to damage, and higher maintenance of turgor of the fruit tissues, in which the glochids are more humid and attached to the fruit [5]. There are several harvesting techniques: turning or twisting, which is a procedure that causes damage to the fruit and is used when the product is intended for processing; cutting flush to the insert, through which fruits are obtained with little conservation time, used when the fruit is for immediate consumption or sale; and cutting a small piece of the cladode attached to the fruit, which is the most used technique for the commercialisation of PP, as it allows an increase in the time of conservation [40].

After harvesting, the glochids are removed mechanically without damaging the epidermis of the fruits. Then the fruits are selected according to the purpose for which they are envisioned (or "for the purposed application"). Subsequently, the fruits are washed with drinking water or chlorinated with sodium hypochlorite to reduce the microbial load. Then the fruits are waxed by immersion in, or sprinkling of, wax, to control the loss of water by transpiration, reduce the intensity of the fruits' gas exchange, improve the visual appearance, and prolong their conservation. The classification of PPs is performed manually or mechanically, according to colour and size [7,8]. Generally, the fruits are packaged on the day of harvest and transported to their destination in refrigerated conditions. If the fruits are handled a few days after harvest, they must be stored under certain conditions. For PP storage, low temperatures are recommended, between 5 °C and 8 °C, with a relative humidity between 85% and 95%, depending on various factors such as storage time, type of packaging, the season of harvest, and the variety of the fruit [8]. Under these conditions, the shelf life of the fruits varies between 3 and 8 weeks [40]. The *Codex Alimentarius* contains standards that describe the quality, presentation, packaging, and hygiene requirements relating to the PP [47].

Like other cultures, OFI can also suffer biotic diseases (fungal, bacterial, and viral infections) and abiotic diseases from frost, hail, herbicides, pesticides, and fruit splitting [5]. In Portugal, the PP species are relatively resistant to pests and diseases. Due to the resistance of this plant and the climatic conditions, which are not unfavourable, there are no stressful situations. Concerning diseases, these are mainly caused by fungi or bacteria and pests. The Mediterranean fly (*Ceratitis capitata*) is the insect that causes the most damage in the culture of PP, but damage can also be caused by ants, slugs, snails, mealybugs, and fruit flies [7,44].

2.4. Worldwide Distribution

The OFI originates from South America, namely Mexico. This species was introduced in Spain, by Christopher Columbus, in 1493, on one of his trips to America. Subsequently, the OFI was dispersed and naturalised in the Mediterranean area of Europe and northern Africa [35,41].

Due to its genetic variability, OFI has a high adaptability, ecological and, consequently, a wide geographic distribution, can be found in locations with diverse climatic conditions, including North, Central and South America, Northern, Central and Southern Africa, Middle East, Australia, India, and some Mediterranean countries [2,3].

The leading producers and consumers of OFI are Mexico and Italy, and it is in Mexico that this species has the highest degree of genetic diversity. Of the approximately 590,000 ha grown worldwide, Mexico and Italy contain 70% and 3.3%, respectively [10,29]. In Mexico, cladodes are the fifth most consumed vegetable, and PP is the third most consumed fruit [37].

In Europe, the economic and agricultural importance of the PP dates to the 16th century. The OFI began by decorating bourgeois-class gardens and properties, and then served as a hedge to delimit rural properties due to the thorns and, later, its fruits became a food source for the lower classes in times of food scarcity. Currently, PP presents itself in different forms, from fresh to processed, also being sold as a gourmet product [7,41].

In Portugal, mainly in the Alentejo and Algarve, the cultivation of OFI is allowed, being an introduced, naturalised, non-invasive species, which is in the phase of expansion, often being found on the edges of rural roads and paths or even to delimit private land [42,44]. Recently, the private sector has started focusing on this species' growth for fruit production in the semi-arid areas of the Alentejo and Algarve. Since 2009, with the help of a programme for unemployed young farmers, more than 200 ha have been planted, and a further 500 ha will be planted in the coming years [5].

3. Physicochemical and Nutritional Composition

3.1. Cladodes and Flowers

The physicochemical composition of OFI depends on the plant part, ecotype or variety, environmental factors, growing area, type of fertilisation, season, the state of maturation, and post-harvest procedures. Therefore, the nutritional values vary between species, varieties, and plant parts, and should not be taken as absolute values [3,5].

The cladodes are fleshy and adapt to arid environments, their primary function being water storage [7]. For this reason, the main constituent of cladodes is water (88–95%), and, consequently, cladodes are a low-calorie food (27 kcal/100 g) [2]. In their chemical composition, based on fresh weight, the water content is followed by carbohydrates (3–7%), fibre (1–2%), minerals (1–2%), proteins (0.5–1%), reducing sugars (0.64–0.88%), and lipids (0.2%) [2,9–11]. In addition to these constituents, young cladodes contain ascorbic acid (10–15 mg/100 g), carotenoids (30 µg/100 g), mainly β-carotene, and chlorophyll (12.5 mg/100 g) [5,12].

Regarding minerals, cladodes contain potassium (2.35–55.20 mg/100 g), calcium (5.64–17.95 mg/100 g), magnesium (8.8 mg/100 g), phosphorus (0.15–2.59 mg/100 g), sodium (0.3–0.4 mg/100 g), manganese (0.19–0.29 mg/100 g), iron (0.09 mg/100 g), and zinc (0.08 mg/100 g) [1,48].

Several studies with cladode juices have shown pH values of around 4.6, with 0.45% titratable acids (low-acid vegetable) and 6.9% total soluble solids (TSS) [11,15]. The cladodes are characterised by fluctuating levels of malic acid (95–985 mg/100 g) due to CAM. During the night, OFI fixes CO_2 as $C_4H_6O_5$, releasing O_2, and during the day $C_4H_6O_5$ is decarboxylated, and the released CO_2 is converted into glucose ($C_6H_{12}O_6$) through photosynthetic action [49]. Other organic acids are also present in cladodes, including citric acid (31–178 mg/100 g) [50], and oxalic acid, which occurs in a dissolved (0.61 mg/g dry weight) or crystalline form (34.5 mg/g dry weight) when calcium sequestration occurs, forming calcium oxalate crystals (11.5–14.3 mg/100 g) [1,51].

One of the main constituents of cladodes is dietary fibre, a class of compounds characterised by a mixture of polymeric carbohydrates of vegetable nature, and oligosaccharides or polysaccharides, such as cellulose, hemicellulose, pectic substances or resistant starch, which may be associated with lignin and other components such as polyphenols, waxes, saponins, cutin, phytates, or proteins. The benefits associated with dietary fibre intake include the decreased risk of coronary heart disease, diabetes, obesity, and cancer [6]. Total dietary fibre (TDF) is classified according to its solubility in water in two fractions: soluble dietary fibre (SDF) and insoluble dietary fibre (IDF) [52]. The SDF is composed of mucilage, gums, pectins, and some hemicelluloses, and is associated with the reduction of cholesterol

levels and the control of the absorption of glucose. IDF is composed of cellulose, lignin, and most hemicelluloses, and presents water-binding capacity, which increases stool weight, ion exchange, and absorption of bile acids, minerals, and vitamins, as well as interactions with the microbial flora [6]. Regarding IDF, cladodes contain cellulose (11%), hemicellulose (8%), and lignin (3.9%) [2].

Mucilage is a complex polysaccharide produced by specialised cells of the cell wall, and forms molecular networks capable of retaining large amounts of water [17]. In cladodes, mucilage is composed of arabinose (42%), xylose (22%), galactose (21%), galacturonic acid (8%), and rhamnose (7%) [53].

The age of cladodes, the environmental conditions, the soil type, and the climate may explain variations in the polyphenol content of cladodes: nicotiflorin (2.89–146.5 mg/100 g), narcissine (14.69–137.1 mg/100 g), isoquercetin (2.29–39.67 mg/100 g), ferulic acid (0.56–34.77 mg/100 g), isorhamnetin-3-*O*-glucoside (4.59–32.21 mg/100 g), rutin (2.36–26.17 mg/100 g), coumaric acid (14.08–16.18 mg/100 g), 3,4-dihydroxybenzoic acid (0.06–5.02 mg/100 g), 4-hydroxybenzoic acid (0.5–4.72 mg/100 g), salicylic acid (0.58–3.54 mg/100 g) and gallic acid (0.64–2.37 mg/100 g) [1,13,54]. The carbohydrates, carotenoids, and acidity increase during development, and the proteins and fibres decrease with cladode age [11].

Several studies show a great interest in OFI flower extracts, e.g., as a source of bioactive substances and potential use as a food preservative [14]. The flowers accumulate predominantly yellow and red betalains (betaxanthins and betacyanins, respectively), and colourless phenolic compounds [3,9,48]. Regarding the phenolic content, the main compounds present in the flowers are gallic acid (1630–4900 mg/100 g), 6-isorhamnetin 3-O-robinobioside (4269 mg/100 g), and 7-isorhamnetin 3-*O*-galactoside (979 mg/100 g) [1,3,55].

Bousbia et al. (2022) did a study to evaluate the phytochemicals and antioxidant activity of the peels, flowers, and seeds of OFI from Souk-Ahras, Algeria. They determined the content of total polyphenols, flavonoids, and betalains, and the antioxidant activities by DPPH, FRAP (ferric reducing antioxidant power) and TAC (total antioxidant capacity) tests, and concluded that flowers were the organ richest in polyphenols, and the flavonoids and peel had a high content of betalains. The flowers and seeds presented considerable DPPH free radical scavenging power, and the peel extracts showed an important total antioxidant capacity and a consequent ferric iron reducing power [56].

A study by Ammar et al. (2012) on the composition of OFI flowers during and after flowering indicates that the main compounds present in hexane extracts are carboxylic acid (28–97%), terpenes (0.2–57%), esters (0.2–27%), and alcohols (<1.8%). This study also demonstrates antibacterial activity against *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Escherichia coli*, and antifungal activity against *Aspergillus niger* and *Candida lipolytica*. It should be noted that the post-flowering phase corresponds to the accumulation maximum of phenolic compounds, and, consequently, there is an increase in the antioxidant and antibacterial properties [14]. Regarding the composition of vitamins in the OFI's flowers, the results are not very conclusive, given the small number of studies [1,3].

3.2. Prickly Pears

Due to the wide varieties/ecotypes of OFI and its wide geographic distribution, information on the physicochemical and nutritional composition of the PP is scarce and varied [57]. The type of plant (genetic factors), the origin of the plant (climate) and the agronomic characteristics (type of cultivation, fertilisation, and irrigation) are decisive in the diversity of physicochemical and nutritional parameters of the fruit [5,6]. Tables 2–4 [58–60] compile various data regarding the physical–chemical composition and nutritional status of various PP ecotypes in different regions.

Martins et al. (2022) completed a study on three PP ecotypes (white, orange, and red) cultivated in Portugal, mainly in the south of the country, in the Alentejo and Algarve. This team characterised these PPs according to the morphometric, physicochemical, nutritional, and toxic profiles of the fruit and mesocarp. The three ecotypes

showed similar physicochemical and nutritional profiles: 79.9 to 83.0% of water content in fruits and 82.6 to 84.8% of water content in mesocarps; 14 to 20% of total carbohydrate contents, with 8 to 14% corresponding to reducing sugars; 1.4% total dietary fibre in fruits and 4.8% total dietary fibre in mesocarps; protein values in the order of 0.5%; fat values between 0.12% and 0.15%; 21 to 32 mg/100 g of ascorbic acid; 18 to 43 mg/100 g of carotenoids; and 109 to 158 mg/100 g of total polyphenols. All ecotypes had a low caloric content, varying between 150.0 and 160.7 kcal/100 g. This study also included the analysis of 30 minerals (essential nutritive, non-nutritive, and toxic macrominerals and microminerals) of the PP fruits and mesocarps. The macromineral with the highest concentration was potassium (140–650 mg/100 g), followed by calcium (18–286 mg/100 g), magnesium (13–96 mg/100 g), phosphorus (3.7–25 mg/100 g), and sodium (1.9–4.3 mg/100 g). The majority of nutritive microminerals were silicon (362–3993 μg/100 g), manganese (714–5866 μg/100 g), zinc (151–301 μg/100 g), nickel (72–128 μg/100 g), copper (67–128 μg/100 g), and iron (72–126 μg/100 g). Of the non-nutritive microminerals, the ones that were most evident in the three were aluminium (301–2459 μg/100 g), strontium (50–504 μg/100 g), and boron (95–198 μg/100 g). The toxic microminerals that stood out were barium (150–676 μg/100 g) and lithium (91–257 μg/100 g) [42].

Stintzing et al. (2003) studied several properties and characteristics of three Italian PP ecotypes: red, orange–yellow and white–green. The densities for each ecotype were 1.0543 g/cm^3, 1.0479 g/cm^3 and 1.0535 g/cm^3, respectively. The pH ranged from 5.7 to 6.3, the content of TSS ranged from 12.3 to 13.7 °Brix, and the acidity content ranged from 0.05% to 0.11% [61].

Table 2. Physicochemical and nutritional composition of several PP ecotypes in different regions of the world.

Country	Ecotype	DM (%)	Water (%)	Ashes (%)	Carb (%)	Prot (%)	Lip (%)	DF (%)	AA (mg/100 g)	Carot (µg/100 g)	PC (mg/100 g)	Ref.
Mexico	Values for 18 wild and cultivated varieties of Potosino-Zacatecano	10–15	85–90	---	T: 10–17 RS: 5–14	1.4–1.6	0.50	2.40	4.6–41	TE	---	[58]
	Naranjona (yellow/orange)	---	---	---	---	---	---	---	1.5 –2.0	50–100	10–65	[59]
	Pelon rojo (red)	7.52	92.48	13.14	---	TE	0.94	SDF: 8.12 IDF: 19.39	---	1.5×10^6	1.54	[27]
		---	---	---	---	---	---	---	1–1.5	<50	50–95	[59]
	Alfajayucan (green)	5.41	94.59	17.07	---	TE	1.40	SDF: 7.98 IDF: 34.95	---	---	2760	[27]
Italy (Sicily and Apulia)	White	---	---	---	Glu: 6.4 Fru: 5.7	0.33	---	---	33	---	---	[60]
	Red/purple	---	---	---	Glu: 6.0 Fru: 5.4	0.33	---	---	31	---	---	
		---	---	---	Glu: 5.6 Fru: 4.3	---	---	---	22	---	---	[61]
		16.70	83.30	---	Glu: 1.88 Fru: 0.78	---	---	---	36.6	---	89.2	[62]
	Yellow/orange	---	---	---	Glu: 6.2 Fru: 6.0	0.34	---	---	38	---	---	[60]
		---	---	---	---	---	---	---	26.9	---	0.746	[26]
		---	---	---	---	---	---	---	29	1.5	---	[63]
		16.10	83.90	---	Glu: 2.14 Fru: 1.04	---	---	---	30.2	---	69.8	[62]
Spain	Green	17.99	82.01	0.41	---	0.87	0.48	5.65	17.1	---	45.0	[64]
	Orange	17.39	82.61	0.37	---	0.94	0.53	4.86	17.2	---	45.4	
	Red	---	---	---	---	---	---	---	18.5	2.58	218.8	[65]

Table 2. *Cont.*

Country	Ecotype	DM (%)	Water (%)	Ashes (%)	Carb (%)	Prot (%)	Lip (%)	DF (%)	AA (mg/100 g)	Carot (µg/100 g)	PC (mg/100 g)	Ref.
USA (Texas and California)	Green	---	---	---	---	---	---	---	45.8	290	---	[66]
		---	---	---	---	---	---	---	51.1	---	242	[2]
	Orange	---	---	---	---	---	---	---	70.2	---	247	
	Red	---	---	---	---	---	---	---	67.9	---	335	
	Purple	---	---	---	---	---	---	---	95.4	---	660	
Argentina	---	15.80	84.20	0.51	10.27	0.99	0.24	3.16	22.56	---	---	[6]
	Yellow	14–18	82–86	---	---	---	---	---	25–32	---	60–77	[67]
Chile	---	16.20	83.80	0.44	14.06	0.82	0.09	0.23	20.33	530	---	[6]
	Orange	---	---	---	---	---	---	---	---	984	371.95	[68]
	Purple	---	---	---	---	---	---	---	---	199.9	777.43	
North Africa (Morocco and Algeria)	Morocco–Yellow	14.49	85.51	0.29	RS: 15.23	0.28	---	---	---	---	---	[69]
	---	---	---	---	---	---	---	---	---	---	1.50	[25]
	Morocco–Red	---	---	---	---	---	---	---	---	---	1.78	
	Algeria	5.60	94.40	1.0	Glu: 29 Fru: 24 Sac: 0.19	1.45	0.70	---	---	---	---	[70]
Egypt	Average values of six local varieties	17.72–19.24	80.76–82.28	1.85–2.12	Suc: 14.9	5.09–7.62	2.52–3.17	---	1050–1200	---	---	[71]
	---	14.90	85.10	0.40	---	0.80	0.70	0.10	25	---	---	[72]
Portugal	White (only fruit)	17.4	82.6	0.43	T: 14.3 RS: 10.7	0.49	0.15	TDF: 1.5	26.5	42800	37.3	[42]
	Orange (only fruit)	15.5	84.5	0.57	T: 18.7 RS: 14.2	0.56	0.12	TDF: 1.7	21.4	25600	43.9	
	Red (only fruit)	15.2	84.8	0.49	T: 17.5 RS: 12.9	0.50	0.10	TDF: 1.2	20.6	18400	46.2	

Table 2. *Cont.*

Country	Ecotype	DM (%)	Water (%)	Ashes (%)	Carb (%)	Prot (%)	Lip (%)	DF (%)	AA (mg/100 g)	Carot (µg/100 g)	PC (mg/100 g)	Ref.
South Africa (Cape)	Five different cultivars	15.3–19.2	80.8–84.7	---	T: 8.26–11.4 Glu: 4.44–6.18 Fru: 3.25–5.24 Suc: 0.08–0.24	---	---	---	---	---	---	[19]
Saudi Arabia	Green to brownish-orange	14.40	85.60	0.44	12.8 (60:40 Glu/Fru)	0.21	0.12	0.02	22	TE	---	[73]
South Korea	---	9.30	90.70	12.12	Suc: 68.7 Fru: 18 Glu: 12.8 Man: 0.5	4.24 (extract free of N = 69.2%)	1.35	3.79	163.8	---	497.6	[74]

Legend: DM = dry matter; Carb = carbohydrates; Prot = proteins; Lip = lipids; DF = dietary fibre; AA = ascorbic acid; Carot = carotenoids; PC = phenolic compounds; T = totals; RS = reducing sugars; Glu = glucose; Fru = fructose; Sac = saccharose; Suc = sucrose; Man = mannose; TE = trace elements; TDF: total dietary fibre; SDF = soluble dietary fibre; IDF = insoluble dietary fibre.

Table 3. Physicochemical composition of several PP ecotypes in different regions of the world.

Country	Ecotype	pH	Acidity (%)	TSS (°Brix)	Pig (mg/100 g or mg/100 mL)	Ref.
Mexico	Values for 18 wild and cultivated varieties of Potosino-Zacatecano	6.5–7.1	0.021–0.049	11–16	Chlorophyll: 0.2–1.1	[58]
	Naranjona (yellow/orange)	5.7–6.6	0.05–0.09	9–12.5	Xanthophyll: 45–69	[75]
	Pelon rojo (red)	---	---	11.9–14	---	[59]
Italy (Sicily and Apulia)	White	6.4	0.02	---	---	[60]
	Red/purple	6.40	0.02	---	---	
		---	---	---	Indicaxanthin: 2.61 Betanine: 5.12	[76]
		5.70	0.11	13.6	---	[61]
		5.89	---	12.7	Betacyanin: 39.3	[62]
	Yellow/Orange	6.02	---	12.1	Betacyanin: 3.6	
		6.44	0.02	---	---	[60]
		---	---	---	Indicaxanthin: 8.42 Betanine: 1.04	[76]
		5.70	0.06	12.3	---	[61]
		---	---	---	Indicaxanthin: 9.3 Betanine: 1.21	[63]
Spain	Green	6.39	0.072	14.98	---	[64]
	Orange	6.22	---	14.05	---	
		---	---	---	Indicaxanthin: absence Betaxanthin: 25	[77]
	Red	---	---	---	Indicaxanthin: 19 Betaxanthin: 25.4–30 Betacyanin: 15.2	
USA (Texas and California)	Green	6.5	---	14.2	Betaxanthin: 4 Betacyanin: 1	[2]
	Orange	6.3	---	12.6	Betaxanthin: 763 Betacyanin: 66	
	Red	5.6	---	14.8	Betaxanthin: 679 Betacyanin: 1200	
	Purple	6.3	---	12.8	Betaxanthin: 1958 Betacyanin: 431	
Argentina	---	5.95	0.14	15.41	---	[6]
	Yellow	---	---	14–16	---	[67]
	Dark purple	---	---	11.3	Betaxanthin: 14 Betacyanin: 34.4	[78]
	Purple	---	---	15	Betaxanthin: 11.8 Betacyanin: 28	
	Green	---	---	15	Betaxanthin: 0.31 Betacyanin: 0.08	

Table 3. *Cont.*

Country	Ecotype	pH	Acidity (%)	TSS (°Brix)	Pig (mg/100 g or mg/100 mL)	Ref.
Chile	---	6.37	0.06	14.1	---	[6]
	---	---	0.078	16.5	---	[79]
	Orange	---	---	---	Indicaxanthin: 8.94 Betaxanthin: 2.93	[68]
	Purple	---	---	---	Indicaxanthin: 0.21 Betaxanthin: 11.1	
North Africa (Morocco)	Yellow	6.22	0.056	14.9	---	[69]
		---	---	---	Indicaxanthin: absence Betaxanthin: 3.78	[25]
	Red	---	---	---	Indicaxanthin: 5.65 Betaxanthin: 4.59	
Egypt	Average values of six local varieties	---	0.24–0.32	12.87–12.94	---	[71]
	---	5.8	0.05	13.2	---	[72]
Portugal	White (only fruit)	5.5	---	16.9	---	[42]
	Orange (only fruit)	5.4	---	19.1	---	
	Red (only fruit)	5.5	---	16.6	---	
South Africa (Cape)	Five different cultivars	6.13–6.38	0.02–0.03	10.2–13.9	---	[19]
Saudi Arabia	Green to brownish-orange	5.75	0.18	14.2	---	[73]

Legend: TSS = total soluble solids; Pig = pigments.

Medina et al. (2007) also carried out a similar study with two ecotypes of the PP (orange and green), from the island of Tenerife, in Spain. The different locations of PP resulted in some differences in their physicochemical composition. The refractive index of the orange ecotype was 1.35 and the refractive index of the green ecotype was 1.36, and for both the average pH value was 6.32, the average TSS content was 14.58 °Brix, and the average acidity content was 0.078% [64].

Table 4. Mineral composition of several PP ecotypes in different regions of the world.

Minerals	Mexico, Naranjona [80]	Italy, White [61]	Italy, Yellow/Orange [61]	Italy, Red/Purple [61]	Spain, Green [64]	Spain, Orange [64]	Argentina [6]	Chile [6]	Morocco, Yellow [69]	Algeria [70]	Egypt Medium Values, Six Varieties [71]	Egypt [72]	Egypt Medium Values, Five Varieties [6]	Portugal, White [42]	Portugal, Or-Ange [42]	Por-Tugal, Red [42]	Saudi Arabia [73]	South Korea [74]
									mg/100 g									
Cl	3560	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---
K	610	2321	2558	2831	159.5	156.7	78.72	217	183.9	199	400–510	90	---	140	151	152	161	2609
Mg	100	141.8	143	153	26.7	23.1	---	16.1	---	18.8	40	98.4	18.7–38.6	21	18	13	27.7	800.6
Ca	110	76.2	86.7	88.7	24.4	28.8	---	12.8	21.46	12.4	110–130	24.4	28.3–56.4	18	45	23	27.6	2087
Na	160	10.7	12.4	16.9	0.524	0.758	1.64	0.6	13.3	1.09	90–150	1.1	---	1.9	2.2	3.1	0.8	539.7
P	50	---	---	---	---	---	---	32.8	---	---	100–120	9.22	---	18	16	3.7	14.4	99.6
Mn	33	---	---	---	0.301	0.306	---	---	---	---	---	---	0.133–0.403	1.340	0.820	0.714	---	2.2
Fe	28	---	---	---	0.2	0.195	---	0.4	---	---	---	---	0.303–0.45	0.102	0.072	0.126	1.5	12.9
Zn	16	---	---	---	0.204	0.207	---	---	0.029	---	---	---	0.245–0.732	0.214	0.176	0.151	---	---
Cu	5	---	---	---	0.0384	0.0396	---	---	0.019	---	---	---	0.085–0.195	0.128	0.067	0.094	---	---
Ni	---	---	---	---	0.0289	0.0268	---	---	---	---	---	---	0.033–0.086	0.082	0.076	0.072	---	---
Mo	0.3	---	---	---	---	---	---	---	---	---	---	---	---	0.0039	0.0027	0.0035	---	---
Cr	---	---	---	---	0.0115	0.0102	---	—	---	---	---	---	0.018–0.024	0.0018	0.0034	0.0011	---	---
Co	---	---	---	---	---	---	---	---	---	---	---	---	0.017–0.029	0.0035	0.002	0.0018	---	---
Si	---	---	---	---	---	---	---	---	---	---	---	---	---	0.514	<0.362	3.993	---	---
Al	---	---	---	---	---	---	---	---	---	---	---	---	---	0.370	0.301	2.459	---	---
B	---	---	---	---	---	---	---	---	---	—	---	---	---	0.159	0.095	0.167	---	---
Sr	---	---	---	---	---	—	---	---	---	---	---	—	---	0.050	0.073	0.082	---	---
Ba	---	---	---	---	---	---	---	---	---	---	---	---	---	0.250	0.153	0.150	---	---
Li	---	---	---	---	---	—	---	---	---	---	---	---	---					---

As previously mentioned, the PP can be divided into three components: peel (30 to 40%), seeds (2 to 10%), and pulp (60 to 70%) [5]. The peel generally contains high levels of cellulose, calcium, and potassium, the seeds contain cellulose and considerable amounts of proteins and lipids, and the pulp is rich in glucose, fructose, and pectin [3,81]. El-Gharras et al. (2006) evaluated the alterations of the physicochemical characteristics of the PP in three maturity stages. The pulp's pH, sugar, protein, and calcium levels increase as it matures, while the peel, and seeds' acidity and moisture decrease [69].

In a study concerning the chemical composition of the pulp, peel, and seeds of the fruits cultivated in Algeria, Salim et al. (2009) observed that the pulp and the peel contained a higher water content (94.40 and 90.33%, respectively) than the seeds (18.05%). These had a higher protein content (4.48%) than the pulp and the peel, with 1.45%. The same observation was made for lipids, where the seeds had a more significant amount (3.66%) than the pulp (0.7%) or the peel (1.06%). Regarding the content of ashes, this was higher in the seeds (12.66%) than in the pulp (1%) or in the peel (3.05%). The content of total carbohydrates in the seeds was higher (61.15%) than in the peel (4.11%) and in the pulp (2.45%). The pulp had less sucrose and more glucose and fructose than the peel. The most representative macrominerals in the three components of the fruit were, in descending order, potassium, magnesium, calcium, and sodium [70].

The water content in the pulp is protected by the peel, which is thick and rich in mucilage. This property binds strongly to water, helping to prevent the fruit from drying out [6]. The peel is more acidic than the pulp and has a higher phenolic content. It contains considerable amounts of polyunsaturated fatty acids, particularly linoleic acid, and other fat-soluble compounds such as sterols (mainly β-sitosterol), β-carotene, and vitamin K1 [5,34,82,83].

PP seed oil is rich in unsaturated fatty acids, presenting a high linoleic acid and low linolenic acid content. It also contains tocopherols (mainly Y-tocopherol), which prevent lipid peroxidation, making this oil quite stable. Considering the physicochemical properties referred to above, as well as others, namely the refractive index, the iodine index, and the saponification number, PP seed oil has properties like sunflower or grape seed oils [5,48,83].

The PP pulp has a low acidity, with predominately, in descending order, the following organic acids: malic, quinic, shikimic, oxalic, and citric acids (may vary between 0.05 and 0.18%) [83]. In this way, the PP is characterised as a low-acid food with a pH between 5.6 and 6.5. The sugar content of the PP pulp is relatively high, ranging from 12 to 17%. The carbohydrates in this fruit consist of the reducing sugar agents glucose (53%) and fructose (47%), in a 1:1 ratio [5]. Glucose represents an instantly available energy source for the brain, while fructose enhances the flavour of the fruit [9]. Given the high sugar and low acid contents, the pulp has sugar:acid ratios in the range of 90:1 to 490:1 [1]. The levels of reducing sugars in PP pulp are superior to those of other fruits, such as apples, pears, peaches, plums, strawberries and raspberries [69].

The protein content represents only a tiny percentage, between 0.21% and 1.6%, increasing during the PP maturation phases. The amount of this nutrient in the PP is comparable to that found in other fruits such as apples, pears, apricots, pineapples, oranges, and peaches [6,58,69].

The total free amino acid content of PP pulp is higher than most other fruits and similar only to oranges and grapes, with the leading free amino acids being proline, taurine, and serine [13,59,83].

Typically, the fruit pulp contains low levels of lipids; in the case of PP, the pulp is no different. The lipid content varies between 0.1% and 1.0%, representing about 8.70 g of total lipids/kg dry weight [48]. While we can find a significant amount of neutral lipids (87% of total lipids) in the seed oil, the concentration of polar lipid compounds is superior in pulp oil (52.9% of total lipids) [57]. In this oil, the fattiest acid is linoleic acid, followed by palmitic, oleic, and linolenic acids. The main sterols reported in pulp oils are β-sitosterol and campesterol, constituting about 90% of the total sterols [13,83]. In addition, tocopherols

(α-, β- and δ-), β-carotene, and phylloquinone (vitamin K1) present in seeds and pulp oils and protect lipids from oxidative damage [57].

In the PP pulp, the TDF content can vary between 0.02% and 3.15%, a percentage significantly higher than for some of the most consumed fruits and vegetables [84]. Pectin is responsible for the PP pulp viscosity, an essential element in producing juices, marmalades, and jellies. However, more studies are needed to fully characterise the PP pulp's hydrocolloid fraction, composed of arabinose, galactose, rhamnose, and galacturonic acid [83].

The PP pulp has a high level of ascorbic acid, reaching values of 40 mg/100 g, higher than for apples, bananas, grapes, and pears but comparable to oranges, lemons, and papayas [6,7]. PP is also rich in carotenoids, mainly β-carotene (0.53 mg/100 g) [7]. Typically, the highest ascorbic acid content is present in PP with red peel and the highest content of carotenoids is present in PP with yellow peel [10]. The vitamins B, E, and K are present in the PP pulp in trace concentrations. As mentioned, carotenoids, tocopherols, and vitamin K1 play an important role in lipid protection due to their antioxidant properties [57].

Kuti et al. (2004) investigated the antioxidant compounds present in extracts of four Opuntia varieties from Texas. They observed that the predominant flavonoids in the PP with green peel were quercetin (43.2 μg/g fresh weight), isorhamnetin (24.1 μg/g fresh weight), and kaempferol (2.2 μg/g fresh weight) [13,66]. There is clear evidence that these compounds have an antioxidant power greater than several vitamins since phenolic compounds can delay the prooxidative effects in proteins, deoxyribonucleic acid (DNA), and lipids due to the generation of stable radicals [9,57].

In another study, whose objective was to investigate the amounts of total polyphenols and pigments from two PP cultivars of Moroccan origin, Khatabi et al. (2016) observed that the level of polyphenols was higher in the whole fruit than in its juice. They also concluded that the PP with red skin contained a higher quantity of polyphenols (15.34 mg/kg of juice and 17.81 mg/kg of whole fruit) than the yellow variety (15.03 mg/kg of juice and 15.03 mg/kg of whole fruit) [25].

Oniszczuk et al. (2020), executed research to determine the antioxidant properties and the content of polyphenolic compounds in PP. This study showed that PP was a rich source of phenolic compounds, particularly the benzoic acid derivatives (protocatechuic, syringic, 4-OH-benzoic, vanillic, gentisic, and salicylic), as well as the cinnamic acid derivatives (caffeic, trans-sinapic, cis-sinapic, β-coumaric, ferulic, isoferulic, m-coumaric, and 3,4-dimetoxycinnamic). In addition to this research, they produced a gluten-free pasta from rice-field bean flour that was enriched with various quantities of PP, and analysed this to determine its content of free phenolic acids, its antioxidant properties, and the sum of its polyphenols. The obtained results demonstrated that this gluten-free pasta supplemented with PP was a good source of natural antioxidants and could improve the quality of health and life of coeliac consumers [85].

Like flowers, fruits also contain betalains, and vacuolar pigments, which contain nitrogen and can be classified into betacyanins (such as betanin) and betaxanthines (such as indicaxanthin). The first gives a red colour and has absorbances at 540 nm; the latter provides a yellow colour and shows absorbances at 480 nm. In addition to these pigments justifying the range of colours available in the PP, they add important nutritional characteristics as they have higher antioxidant properties than those reported for ascorbic acid [5,6,13].

Sepúlveda et al. (2003) studied the betanin content of fourteen types of *Opuntia* from different regions of Chile. The results indicated significant variability of betanin concentrations in the fruits analysed (48.3 to 138.1 mg/100 g) [59]. In another study, Butera et al. (2002) investigated the existence of betalains and the antioxidant activity of aqueous extracts of red, yellow, and white PPs from cultivars from Sicily. Accordingly, with the results obtained, the yellow ecotype exhibited the highest amount of betalains, followed by the red and white ecotypes. Indicaxanthin represented about 99% of betalains in the white ecotype, while the betanin/indicaxanthin ratio varied from 1:8 (*m*/*m*) in the yellow

ecotype to 2:1 (*m*/*m*) in the red ecotype [76]. Two other pigments in the PP are chlorophyll (green colouration) and carotenoids (orange colouration), although in smaller quantities.

Morales et al. (2009) carried out a study to characterise the pulps of two PP ecotypes (purple and orange), from Chile. Regarding the carotenoid content, they obtained 1.999 μg/g in the edible pulp of the purple ecotype and 0.984 μg/g in the edible pulp of the orange ecotype. In both ecotypes, β-carotene, lycopene, and lutein were detected [68].

All the pigments mentioned above make the fruits and their products attractive. However, their stability is the subject of continuous study. Betalains are soluble in water, and their stability is less affected by pH than anthocyanins, another class of natural red–purple pigments, being relatively stable in pH, between 3 and 7, which allows them to be used as additives in foods with low acidity and neutral pH, like dairy products [59,86]. On the other hand, betalains are more stable than chlorophylls under heat treatment and pH variation. In this way, the products of the purple ecotype tend to be more stable than those of the green ecotype [87].

Variations in the mineral content of PP can be attributed to different origins of this fruit, namely the total mineral content in the surface soils, the fraction that is bioavailable in the soil, the pH and texture of the soil, the presence of organic matter, the oxidation–reduction conditions, and the presence of clays. As a rule, the PP is an excellent source of minerals, such as potassium (217 mg/100 g), and is low in sodium (0.6 to 1.19 mg/100 g), which is beneficial for people with kidney problems and high blood pressure. It is also rich in calcium, phosphorus, and magnesium, with levels around 15.4 to 32.8 mg/100 g, 12.8 to 27.6 mg/100 g, and 11.5 to 16.1 mg/100 g, respectively [6,9,84].

In the case of calcium, it is necessary to carry out further studies on the bioavailability of this mineral due to the calcium oxalate crystals that can be found in the OFI that present a low absorption by the organism [59,84].

Aregahegn et al. (2013) determined the concentration of several minerals (Ca, Mg, Fe, Mn, Zn, Cu, Co, Cr, Ni, Cd, and Pb) by atomic absorption spectrometry in PPs collected in five different areas of Ethiopia. The average concentration value of each mineral was 283–564 μg/g of Ca, 187–386 μg/g of Mg, 3.03–4.50 μg/g of Fe, 1.33–4.03 μg/g of Mn, 2.45–7.32 μg/g of Zn, 0.85–1.95 μg/g of Cu, 0.174–0.295 μg/g of Co, 0.181–0.242 μg/g of Cr, and 0.330–0.856 μg/g of Ni. The toxic metals Cd and Pb were not detected in the samples [72].

El-Gharras et al. (2006) evaluated the effect of the state of maturation on the physicochemical properties of Moroccan PP, in three stages of maturation. Calcium increased from 111.11 mg/kg to 263 mg/kg [69]. These results were higher than those reported for the Mexican cultivars (110–170 mg/kg) [80] and were relatively lower than those reported for Chilean cultivars (154–328 mg/kg) [73]. The potassium content ranged from 963.9 to 2338 mg/kg [69], higher than that reported for Mexican cultivars (610–720 mg/kg) [80] and Chilean cultivars (12.8–27.6 mg/kg) [73]. The sodium concentration ranged from 50.78 to 236.78 mg/kg [69], with values comparable to those reported for the Mexican cultivars (120–160 mg/kg) [80]. There was no great variation in the copper content. The amount of zinc decreased from 0.62 to 0.29 mg/kg [69], being much lower than that reported for the Mexican cultivars (12–16 mg/kg) [80].

Due to the high water content in the PP pulp, its energy value varies between 31 and 50 kcal/100 g, comparable to other fruits such as apples, oranges, peaches, and pears [6].

3.3. Medicinal Properties

According to numerous studies, a diet rich in fruits and vegetables is related to a lower incidence of cardiovascular diseases and some types of cancer [5], and this is no different for the OFI. Scientific research contains several studies that confirm that fruits and cladodes can be used as a source of nutrients and phytochemicals. In this way, OFI is valued not only for contributing to a healthy diet but also because it is rich in health-promoting substances used in the prophylaxis of various diseases [3,5,6]. Some of the medicinal properties of OFI discussed in this section are represented in Figure 3 [5,76,87–92].

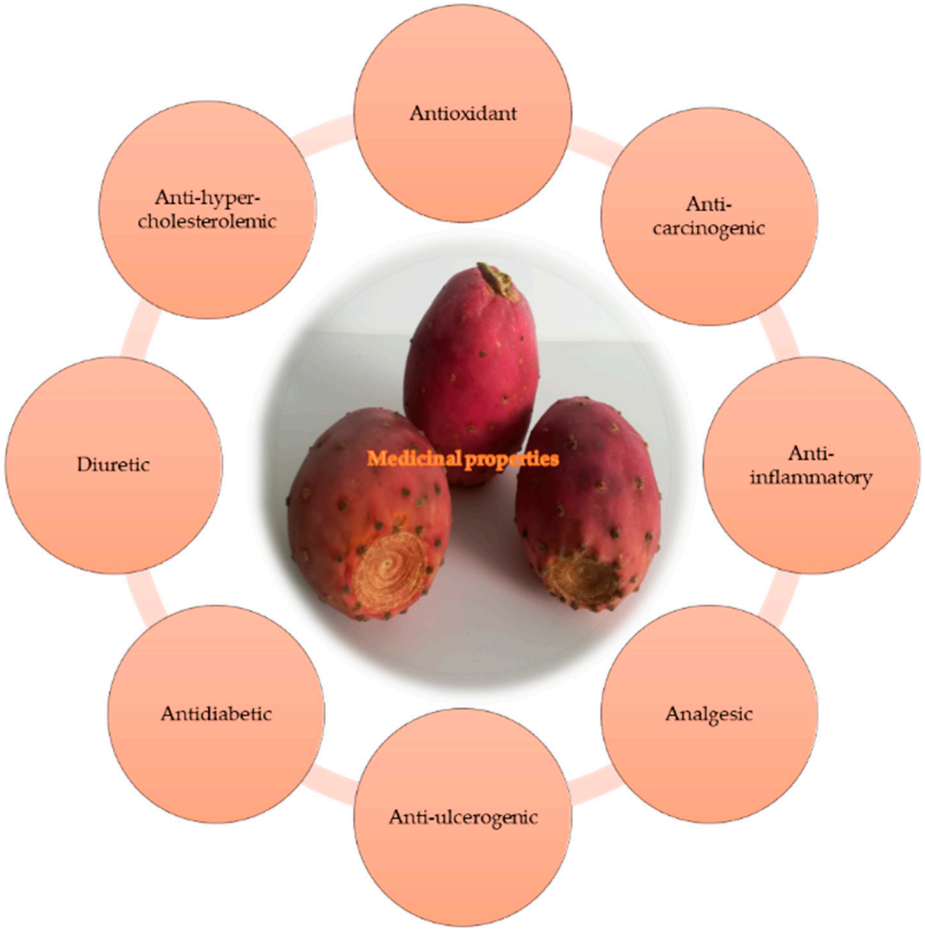

Figure 3. OFI's medicinal properties [5,76,87–92].

In Latin America, cladodes are used in traditional medicine to treat bruises, burns, wounds, and infections. In Mexico, PP juice is valued as a laxative and diuretic, while the flowers are used for chest pains [28].

In general, the OFI has applications not only in preventing hangovers and in reducing lipid oxidation but also mainly in preventing the development of some types of cancer and reducing the risk of developing type 2 diabetes and cardiovascular disease, two of the most common causes of death [28].

As previously mentioned, the PP has a high antioxidant activity, attributed to ascorbic acid, carotenoids, flavonoids, polyphenols, and betalains [5,6,9,10,13,25,57,66,86]. The antioxidant activity of the PP is twice as high as that of other fruits, such as pears, apples, tomatoes, bananas, and white grapes, and has similar levels to red grapes and grapefruit [5]. This beneficial effect is generally attributed to the ability of these compounds to fight oxidative stress and modulate the activity of various enzymes and cell receivers [17].

Butera et al. (2002) analysed aqueous and methanolic red, yellow, and white PP extracts. This study revealed high antioxidant activity, in both the chemical and in vitro biological assays. The results concluded that betalains are one of this fruit's most important antioxidant components. The white PP extract exhibited the greatest protection in all in vitro lipid oxidation models, and the acid ascorbic did not represent more than 40% of the antioxidant activity measured in the different extracts [76].

Dok-Go et al. (2003) evaluated flavonoids' protective effects against oxidative neuronal damage induced in mouse cortical cells. Quercetin, dihydroquercetin, and quercetin 3-methyl ether were isolated from ethyl acetate fractions of fruits and cladodes and identified as neuroprotective compounds. Quercetin 3-methyl ether was the most potent of the isolated flavonoids [13,93].

Sirivardhana and Jeon (2004) demonstrated the antioxidant effects of the PP extract in inhibiting lipid peroxidation in oils and emulsions. The characterisation of these properties proved that the antioxidant compounds present in PP are stable, offering a natural source for stabilising edible oils [94].

Fernández-Lopéz et al. (2010) investigated the presence of antioxidant compounds and the respective in vitro antioxidant capacity in extracts of three Spanish species of *Opuntia* with red peel. This study confirmed the potential of the three extracts as important sources of bioactive compounds, including ascorbic acid, carotenoids, taurine, and flavonoids. The OFI extract contained the highest antioxidant capacity and taurine content [3,65].

Lee et al. (2002) evaluated an ethanol extract from cladodes to determine the mechanisms of its antioxidant activity. The extract showed elimination activity dose-dependent free radicals, including 2,2-diphenyl-1-picrylhydrazyl radicals ($DPPH^{\bullet}$), the superoxide anion ($O_2^{\bullet -}$), and hydroxyl radicals ($OH^{\bullet}$), using different systems of rehearsal. This extract was also considered effective in protecting plasmids against chain rupture, induced by hydroxyl radicals in the Fenton reaction. The researchers identified a high quantity of phenolic compounds responsible for the extract's antioxidant properties [95].

El-Hawary et al. (2019) characterised the polyphenolic constituents of extracts from OFI cladodes, fruit peel and fruit pulp, and investigated their antioxidant and neuroprotective activities. They characterised 37 secondary metabolites using HPLC-MS/MS, and in DPPH assays the extracts exhibited significant antioxidant activities. In vivo, the extracts demonstrated considerable neuroprotective activity against $AlCl_3$-induced neurotoxicity (Alzheimer's condition): OFI extracts significantly decreased the elevated brain levels of proinflammatory cytokines and increased anti-inflammatory cytokine and monoamine neurotransmitters compared with the positive control group. Thus, they concluded that OFI responds to oxidative stress and may be a good candidate for the treatment of several neurological disorders [96].

Diabetes mellitus is another of the biggest problems in public health, characterised by defects in insulin secretion, insulin action, or both [87]. Natural products have long been used in traditional medicine for diabetes. In Mexico, local healers recommend drinking fresh juice from cladodes, and fresh, fried, or grilled cladodes to treat type 2 diabetes [97].

Several studies demonstrate the hypoglycaemic and antidiabetic activity of extracts of OFI. Butterweck et al. (2001) studied the effects on blood glucose concentration and plasma insulin in normal mice of an aqueous extract prepared from cladodes and a mixture of cladode peel and fruit. The results showed that the aqueous extracts reduced blood glucose levels and increased plasma insulin when given orally at low doses (6 mg/kg). Additionally, the peel mixture directly affected pancreatic β-cells [87].

Alarcon-Aguilar et al. (2003) investigated whether polysaccharides isolated from OFI had hypoglycaemic effects. Traditional OFI preparations were evaluated in temporarily hyperglycaemic rabbits, alloxan-diabetic rabbits, regular volunteers, and type 2 diabetic patients. The results suggest that the hypoglycaemic effect produced by the OFI can be explained by a mechanism that reduces the intestinal absorption of glucose due to some effect of the SDF present in the OFI. Clinical trials indicate that the OFI helps stabilize blood sugar levels, effectively treating type 2 diabetes [97]. One of the possible mechanisms of action for the hypoglycaemic activity of OFI is due to the high content of SDF present in this plant. These increase the viscosity in the intestinal tract, slowing down or reducing the absorption of sugar, and may also reduce the fat absorption in the intestine and contribute to improving lipid profiles and weight loss. The SDF content is not the only mechanism of action since fasting glucose levels in the blood are also affected [98].

A study by Ennouri et al. (2005), whose objective was to determine the fatty acids from PP seed oil and evaluate the effects of a supplementary diet with this oil (25 mg/kg) in mice, proved that this food supplement reduced the concentration of serum glucose, and was associated with the formation of glycogen in the hepatic and skeletal muscles. These observations were explained by a potential induction of insulin secretion, which stimulates the conversion of glucose to glycogen [13,99].

Current treatment strategies for type 2 diabetes generally involve pharmacological treatment aimed at stimulating insulin secretion or increasing sensitivity to insulin. Thus, the results obtained in these studies are promising since there is an opportunity to develop pharmaceutical treatments to improve B cell function and reduce insulin resistance, which is crucial to improving metabolic control and delaying the development of diabetic complications [87].

Hwang et al. (2017) investigated the antidiabetic effects of aqueous extracts of cladodes in preventing and treating diabetes. In this way, they performed glucose tolerance assays, and measured α-glucosidase activity and dietary fibre content in streptozotocin-induced diabetic mice. Based on the results obtained, the authors suggested that the investigated extracts could be considered as a food supplement in the prevention and/or treatment of diabetes [100].

Chemoprevention is an approach in which chemical agents prevent, reverse, or block the onset of cancer in certain risk groups. Although it is a promising technique in some epithelial cancers, available preventive agents currently are limited, expensive, and have side effects. Therefore, some natural products, such as grape seeds, green tea, and some herbs, which demonstrate anticancer effects, have been investigated intensively for their possible effects [9,29,48,88,101].

Zou et al. (2005) investigated the aqueous extracts of PP from Arizona in cancer cells from the ovary, cervix, and bladder. Depending on the dose and time of treatment, these extracts can inhibit the in vitro proliferation of tumours, comparable to the inhibition demonstrated by the synthetic retinoid N-(4-hydroxyphenyl) retinamide (4-HPR), which is a chemopreventive agent widely used in the chemoprevention of ovarian cancer. The inhibition of the in vitro growth of cancer cells was associated with an increase in apoptotic cells and cell cycle arrest in the G1 phase. The mechanism of action of the anticancer effect of PP extracts is not yet understood, but it appears to be dependent on the P53 pathway, which is the primary tumour suppressor. This study also demonstrated that the extracts of PP could also suppress ovarian cancer growth in the in vivo mouse model. The intraperitoneal administration of the PP extract solution did not affect the mice's body weight, indicating that the extract showed no toxic effects [88].

Several assays suggest that OFI contains anti-hyperlipidemic and anti-hypercholesterolaemic properties, as it reduces blood cholesterol levels and modifies the composition of low-density lipoproteins (LDLs), which are considered an atherogenic risk due to their affinity for cholesterol in corporation and resultant deposits (atherosclerotic plaques) on blood vessel walls [102].

In a series of studies with guinea pigs, Fernandez et al. (1990, 1992, and 1994) showed that the reduction in blood lipid concentrations, triggered by pectin isolated from OFI, was due to binding to bile acids. In this way, the reduction of bile absorption in the colon alters the enterohepatic circulation [103–105].

A study by Wolfram et al. (2003) showed that daily consumption of 250 g of PP pulp reduced the risk of thrombosis in patients suffering from hyperlipidaemia and diabetes [20]. Galati et al. (2003) studied the influence of daily administration of cladodes lyophilised in the lipid metabolism of hypercholesterolaemic mice, having evaluated the levels of cholesterol, high-density lipoprotein (HDL), LDL and triglycerides. The treatment was more effective after 30 days, with statistically significant reductions in cholesterol, LDL, and triglyceride levels in plasma. The effects are probably due to the high fibre content from cladodes, but other active ingredients may work together with this one [92].

Ennouri et al. (2005) observed a decrease in cholesterol and LDL concentrations in blood, with no changes in HDL concentrations, after adding PP seeds to the diet of mice. These results support the nutritional value of OFI as a natural source of edible oil that contains essential fatty acids [13,99].

LDL particle size has appeared as a significant predictor of cardiovascular disease and progression of coronary heart disease. Therefore, the quantity and quality of particles, mainly small and dense LDL (sdLDL), are essential in determining cardiovascular

disease risk. Giglio et al. (2020) did the first intervention study suggesting that pasta enriched with an OFI extract might have beneficial effects on some metabolic parameters and the LDL particle size and distribution, reducing atherogenic sdLDL (−45%). For one month, 49 patients with one or two criteria for the metabolic syndrome consumed 500 g of pasta supplemented with 3% OFI extract weekly. After one month of pasta supplementation, plasma glucose, triglycerides, plasma creatinine, urea, and aspartate transaminase significantly decreased [106].

The severity of hangovers from alcohol consumption may be related to the inflammation induced by impurities in alcoholic beverages and by-products of alcohol metabolism. The best hangover prevention is, of course, abstinence from alcohol. Still, it has never been shown to stop alcohol consumption effectively, and no evidence indicates that symptom relief results in increased consumption. In addition to the various symptoms (nausea, headache, dry mouth, weakness, tremors, diarrhoea, and dizziness) associated with a hangover, it also poses a risk of injury in the workplace, which can lead to cognitive changes and reduce dexterity, and visual and spatial awareness. To evaluate the effect of extracts from the PP peel in the reduction of the symptoms of a hangover from average alcohol consumption, Wiese et al. (2004) conducted a randomised crossover study with 64 healthy young adults. The results obtained showed that the analysed extract had a moderate effect in reducing hangover symptoms, inhibiting the production of inflammatory mediators, accelerating the synthesis of shock protein heat during periods of stress, and decreasing oxidative damage [90].

Park et al. (1998) prepared ethanol extracts from the fruit and cladodes to evaluate their pharmacological effects in mice. Both extracts showed an analgesic effect like acetylsalicylic acid, even at the highest doses, without toxic effects. Furthermore, the extracts suppressed the release of β-glucuronidase, one of the lysosomal enzymes released by inflammatory cells infiltrating tissue damaged by phagocytosis. This result indicates that the extracts' effects may inhibit the release of inflammatory mediators. A protective effect was also observed in the layers of the gastric mucosa, suggesting that the extracts contained a protective effect against gastric lesions [2,3,21,105].

Park et al. (2001) evaluated several methanolic extracts of OFI cladodes for their ability to treat wounds in mice. The n-hexane and ethyl acetate fractions showed significant activity when administered topically. These results demonstrated that methanolic extracts showed an anti-inflammatory action and advantages in using OFI in wound healing [3,107,108]. Park et al. (2001) continued to study the methanolic extracts of cladodes through a model of induced chronic inflammation in mice, and the anti-inflammatory principle active was isolated and identified as β-sitosterol. However, its activity appears relatively lower than hydrocortisone's [3,109].

The effects of PP powder were investigated by Lee et al. (2001), regarding gastric injuries and ulcers in mice. The results indicated an inhibition of the gastric lesions induced by hydrochloric acid/ethanol and hydrochloric acid/acetylsalicylic acid. The rate of gastric juice secretion and the pH value remained constant. These data showed that OFI has an inhibitory action on gastric lesions in mice [91].

Galati et al. (2001, 2002, and 2003) also confirmed these results. They studied the effect of administering lyophilised cladodes, which showed significant anti-ulcerogenic activity in the experimental model, on ethanol-induced experimental ulcers in mice. The curative and preventive treatments showed different results. In the curative treatment, gastric mucosal epithelial cells appeared injured. The acute treatment with lyophilised cladodes probably did not have time to restore mucosal defence factors upon ethanol induction [110]. Nonetheless, when lyophilised cladodes were administered as a preventive therapy, these maintained the gastric mucosa under normal conditions, preventing the mucus dissolution caused by ethanol and favouring mucus production [89].

In another study, Galati et al. (2003) inferred that the preventive administration of PP juice inhibited the ulcerogenic activity of ethanol in mice. The obtained results indicated increased mucus production and restoration of the standard mucosal structure. These

studies demonstrated an evident protective activity of cladodes and the juice of the PP against ethanol-induced ulcers [26].

Similar results were obtained by Khémiri et al. (2019). They carried out a study to investigate the preventive and curative effects of the PP seed oil in an ethanol-induced gastric ulcer model in mice. The oil showed high efficiency in protecting the structure and function of the gastric mucosa against damage caused by ethanol ingestion. Mucus production was stimulated, the volume of gastric juice was reduced, and its pH increased. They also concluded that the fatty acids in the oil, mainly unsaturated and triglycerides, contributed to the repair of the lipid bilayer of the cell membrane during the gastric ulcer healing process [111]. The protective activity of PP juice, namely betanin, was evaluated against acute gastric disorders caused by induced stress in mice.

Kim et al. (2012) observed that the pre-treatment made with lyophilised powder containing PP juice and maltodextrin significantly reduced lesions. Furthermore, it effectively prevented the decrease in gastric mucus content, which may be related to the production of pro-inflammatory cytokines [112].

Galati et al. (2002) investigated the acute and chronic diuretic effects of flowers, fruits, and cladode extracts in mice. The flower and cladode extracts increased diuresis but did not influence the uric acid cycle. On the other hand, the fruit extract showed diuretic and anti-uric activity. All extracts analysed showed a modest but insignificant increase in sodium and potassium urinary levels. There are some possibilities for the observed diuretic effect, which still needs to be investigated. On the one hand, these natural compounds can act synergistically or individually, promoting initial vasodilation. On the other hand, it is possible that the extracts manifest an accumulation of several substances and/or it is due to secondary active metabolites. Another possibility for this effect may be due to indirect changes in some physiological parameters before blood filtration. The anti-uric effect of the PP infusion cannot be explained only by increased diuresis or urinary excretion, which may be linked to an alteration of some enzymatic activity [3,92].

Monoamine oxidases (MAOs) are enzymes whose function is to degrade monoamines, preventing them from accumulating (in the case of endogenous monoamines) or generating undesirable effects (in the case of exogenous monoamines), being involved in the catabolism of catecholamines. These constitute a class of chemical neurotransmitters and hormones that occupy critical positions in regulating physiological processes and developing neurological, psychiatric, endocrine, and cardiovascular diseases [113].

Han et al. (2001) performed several assays, including anticoagulant activities, dopamine β-hydroxylase, and MAO, in several extracts of cladodes and fruits of Korean OFI. Methyl esters derived from organic acids have been identified as MAO inhibitors. The aqueous extracts showed the lowest inhibitory activity, followed by the n-butanol fraction and the hexane extract, while the ethyl acetate fraction exerted the most significant inhibitory action [2,114].

3.4. Applications and Agro-Industrial Uses

Applications

Considering the nutritional and pharmacological properties of OFI, it has several applications, both in human and animal nutrition, in the food, pharmaceutical and cosmetic industries. OFI is also used in civil construction and alternative fuels (Figure 4) [2,5,17,18].

The most notorious application of OFI is consuming the fruit after peeling. In some countries, such as Mexico and Chile, PP juice is consumed at home, in restaurants, or in local stores [115]. In these countries, the PP fermented juice, without the seeds, is known as a beer called "colonche" [116].

OFI flowers are used for teas, and cladodes are consumed as fresh vegetables, being used as an ingredient in several dishes, including sauces, salads, soups, snacks, drinks, and desserts [5,28].

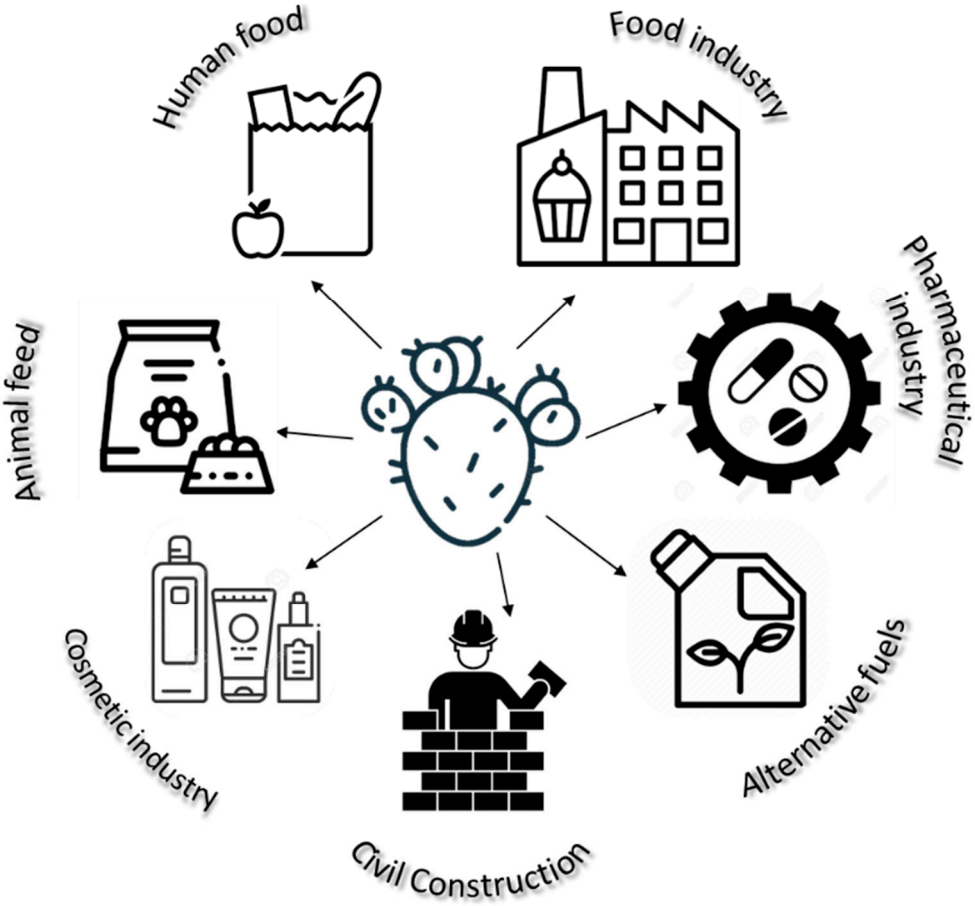

Figure 4. Applications of OFI fruits and cladodes [2,5,17,18].

The properties of OFI make it advantageous for a wide variety of products in the food industry. Beyond the fresh fruit, minimally processed fruit, dehydrated or preserved fruit, we can find in the market juices, fermented drinks, liquid sweeteners, pulps (frozen or dry), gums or gels, jams or jellies, flours, seeds, natural dyes, dietary fibres, and thickeners [6,8,10,16,117–119].

In addition to human food, the OFI also contributes to animal feed. Cladodes and fruits are profitable feed for ruminant animals, especially when pasture supply is lower and of low quality [5,116].

OFI also has advantages in the pharmaceutical industry due to its medicinal properties. Powdered extracts of PP, cladodes, and flowers are commercialised in capsules to protect the gastric mucosa, regulate weight, regulate blood sugar, or increase fibre intake. Cladode-based gels have a cooling effect, like aloe vera preparations, relieving the skin and contributing to wound healing [2,6].

Another OFI application is in the cosmetic industry, as the PP, cladodes juices and the seed oil can be found in shampoos, conditioners, creams, lotions, soaps, face masks, and sunscreens [2,6,117].

Ammar et al. (2012 and 2015) studied the presence of bioactive substances with antimicrobial action on the OFI flowers. This investigation pointed to the feasibility of using flower extracts against various microorganisms, such as *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Escherichia coli*, and these have been recommended as a preservative in multiple fields of application, including agri-food, cosmetics, and pharmaceutical [3,14,120].

Typically, OFI is used as a hedge or as an ornamental plant. Still, it also has several environmental advantages, namely in protecting fauna, combating desertification, and as a source of nectar for bees [7,121]. OFI is one of the most efficient plants in water use, protecting against soil erosion [121].

In a study carried out by Barka et al. (2013), OFI in an aqueous solution showed a bioabsorption capacity for cadmium and lead, and can be considered as an effective, low-cost, natural, and ecological absorbent for these two metals (a process known as phytoremediation), being an alternative to the current expensive methods of removal of metals from wastewater [122]. In addition to the adsorption capacity, OFI also presents compounds with antimicrobial activity and coagulant properties, which can be used to decontaminate river water and wastewater [4,18].

The coagulant, thickener/gelling, and polyelectrolytic activities of OFI are mainly to its polygalacturonic acid content. The clotting activity can be due to carbohydrates and several phytochemicals, namely phenols, carotenoids, flavonoids, betalains, vitamins, minerals, amino acids, amines, organic acids, lipids, and terpenes. These compounds have diverse chemical structures and functional groups, allowing the adsorption of various contaminants [18].

OFI plantations are also crucial for reproducing cochineals (*Dactylopius coccus*), an insect genus that thrives on this plant. These insects are important in biological control against invasive cacti and are a source of carminic acid, a red pigment used in the colouring of foods, cosmetics, pharmaceuticals, and fabrics. This pigment has been investigated for its antioxidants and antimicrobials, which point to potential applications in immunology and wastewater treatment [3,5,64].

OFI also has applications in the civil construction area, as a protective agent against corrosion, due to mucilage, and as a building material to improve stability and compressibility [2,123].

Through the fermentation of cladodes, OFI has advantages in producing alternative fuels, namely biogas, a viable and essential form of energy in agricultural and rural areas. Biogas is obtained from the transformation of organic waste through anaerobic digestion. This plant is recommended as an alternative energy source since it has a high potential to produce biomass [2,5,124].

4. Physical, Chemical, and Microbiological Changes in Prickly Pear

PP has a short shelf life of about 3 to 4 weeks, a pH of 5.6 to 6.5, and a low acid content (0.05 to 0.18% citric acid equivalent), which compromises its prolonged storage and distribution locally and worldwide [9]. Considering its physicochemical composition, the PP is susceptible to physical, chemical, and biological modifications, and there may be losses of some food constituents, especially nutrients [7]. In Mexico, it is estimated that during the commercialisation of PP there are losses of about 15% of the harvested fruits [11,125].

In general, harvested fruits' deterioration rate is proportional to their breathing rate. The PP is a non-climacteric fruit with a low respiration rate at 20 °C (20 mL CO_2/kg/h) and a reduced ethylene production (0.2 µL C_2H_4/kg/h). These values are similar to oranges [11,125].

During the conservation of the PP, the physical damages are the leading cause of the alteration of this fruit. These changes are due to its physiology and post-harvest handling, mainly due to removing glochidia [7]. PP susceptibility to physical injury increases with maturity [5]. The discolouration is one of the main post-harvest problems and is caused by the oxidation of phenolic compounds catalysed by the enzymatic actions of polyphenol oxidase (PPO) [126]. There are differences in susceptibility between the various ecotypes because the green PP appears to be more sensitive to this problem [11,125]. In addition to the visual aspect, the discolouration of the fruits leads to changes in flavour and losses in the nutritional quality of the fresh fruits, which affects consumer acceptability [11,125,126].

PPs are very sensitive to water loss, making them perishable [127]. The PP begin to show signs of rotting nine days after harvest, and twenty days after harvest PP shows water losses of between 70 and 80% [8]. These losses can also be justified by the damage caused by the fruit fly. The larvae in the damaged fruits can develop and evolve to the adult state, infecting the fruits that are not damaged and stored [7].

OFI is a tropical plant that can suffer cold injuries and damage caused by temperatures above freezing (0 °C). Flowers and fruits are more susceptible to this damage. Leaves may turn purple or reddish and, in some cases, wither. The sensitivity OFI presents to this type of lesion varies according to the cultivar, the fruit maturity, the environmental conditions, and the storage humidity. Usually, these lesions are not evident during cold storage, appearing during commercialisation when the fruits are transferred to higher temperatures [11,125]. Therefore, the fruits can be refrigerated for a maximum period of two months at temperatures of 0 ± 0.5 °C and 85 to 90% relative humidity [6].

Like most non-climacteric fruits, the PP does not contain starch as a reserve of carbohydrates. Therefore, after harvesting, the physicochemical parameters that tend to decrease are total soluble solids (TSS), sugars, and organic acids. If the PPs are stored at room temperature, the ascorbic acid content will rapidly decrease, but this content becomes relatively stable at low temperatures, despite the high pH [5,11,125].

The pH, acid content, and TSS values presented by PPs make their pulp a very attractive medium for the growth of microorganisms, which limits their useful shelf life in the fresh state [84,127,128]. Microbiological damage is favoured mainly by pathogenic microorganisms, namely *Fusarium* spp., *Alternaria* spp., *Chlamydomyces* spp., and *Penicillium* spp. [8]. In the case of purple PPs, the microbiological damage is minimised because betalains have greater stability than chlorophylls under the same heat treatments and pH variations [6].

Various means to minimise nutritional and sensory losses and microbiological growth include the storage of the PP in refrigerated conditions, guaranteeing adequate sanitary conditions, packaging in an active or passive modified atmosphere, and using edible coatings [6,7]. In addition to conservation processes, transformation processes are also essential to avoid post-harvest losses and increase the lifetime of the PP, although some technologies modify some fruit constituents [7,17].

5. Preservation Methods of Prickly Pear

The agro-industrial applications that can be applied to fruits and vegetables aim to make full use of them, reduce production losses and enable their valorisation through appropriate processing to diversify the offer of new products, with an extended useful life and with a reduction of transport, packaging, and storage costs [7].

After harvest, refrigeration is the most used technique to extend PP shelf life, and delay the physicochemical and microbiological changes mentioned previously. These procedures' main objectives are to reduce transpiration and respiration rates, increase fruit tolerance to cooling temperatures, and prevent microbial development [5].

Conservation and transformation systems include technologies based on physical, chemical, or biochemical methods (Figure 5). Physical methods include those that use heat transfer as a means of preservation, immobilisation of water, or the reduction of water activity (a_w). Examples of physical methods applied to food preservation are freezing, drying, evaporation, and lyophilisation. Chemical processes include the addition of sugars, acidification, and the use of preservatives. Biochemical methods are based on lactic or alcoholic fermentations [6].

Some of the products from the various processing methods mentioned above are fresh preserved PPs, minimally processed PPs, dehydrated PPs (dry or osmotically dehydrated), preserves, juices, fermented beverages, sweetener liquid, the pulp (frozen or dehydrated), gums or gels, jams or jellies, flours, seed oil, natural dyes, dietary fibres, and thickeners (Figure 6) [5–8,129,130]. Considering the effects of seasonality and the PP distribution, selecting the most suitable processing method depends on the intended purpose and the conservation to be applied [7]. As the handling of the raw material affects the quality of the final product, some steps must be carried out before the conservation phase, which are common to all the food industries: reception of raw materials, cleaning, selection, and washing of the sample. Usually, the cleaning step is mechanical, with the fruits going through rotating brushes to eliminate the glochidia smoothly and without damaging the epidermis of the fruits. Then, the fruits are selected on a purpose-specific basis. The fruits

that are damaged, rotten, or inadequately ripe are removed. Subsequently, the fruits are washed with potable water and, if possible, chlorinated at room temperature to reduce the microbial load. If a transformation method is applied, in addition to these steps, the peeling and cutting operations are also necessary [7,131–133].

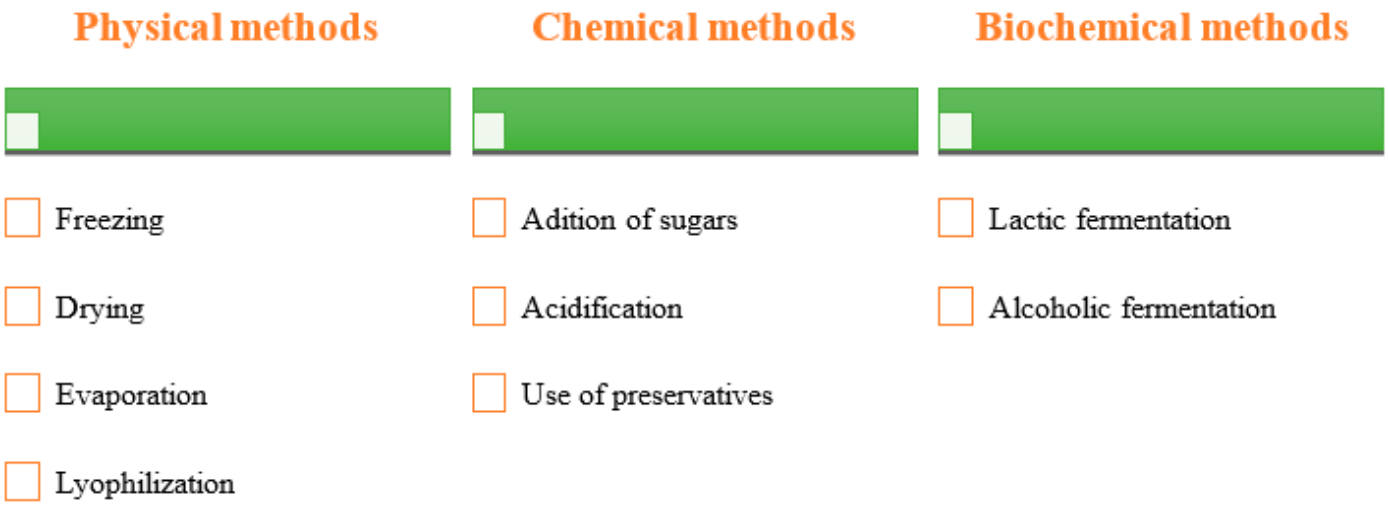

Figure 5. Conservation and transformation methods applied to the PP [6].

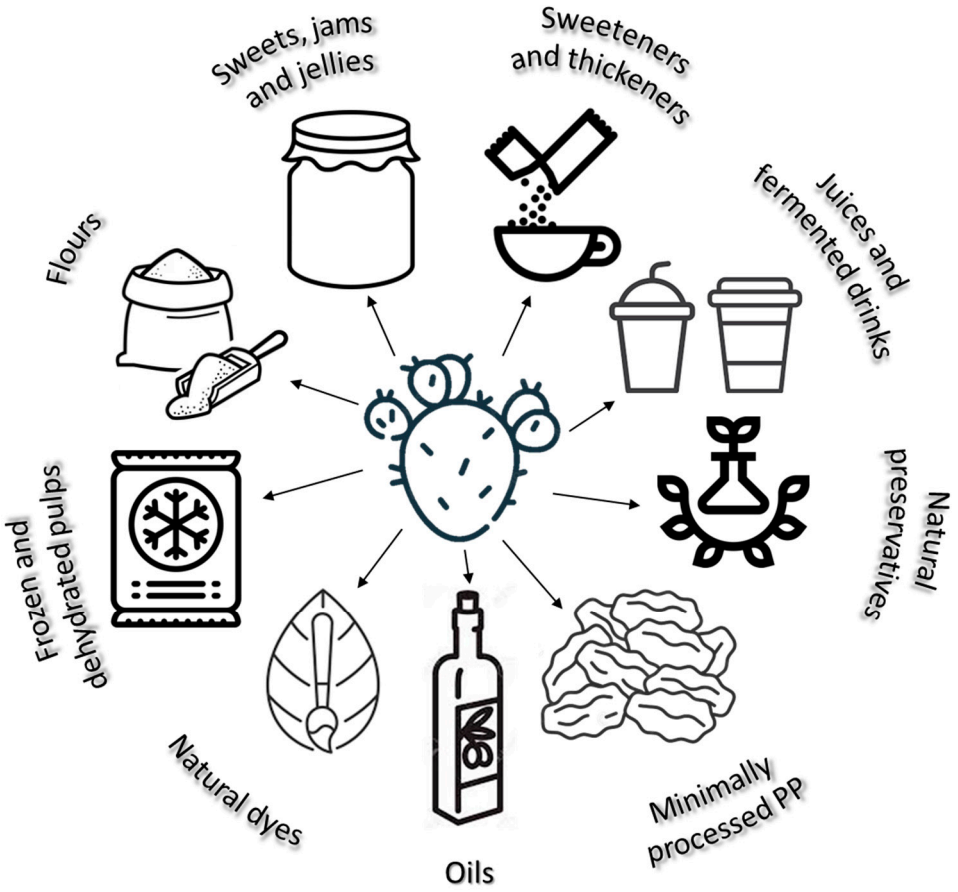

Figure 6. Products from conservation and transformation methods applied to the PP [5–8,129,130].

The harvest and post-harvest handling of the PP must be careful to avoid physical damage and contamination. The fruits must be whole and in excellent maturation, a homogeneous size, in good physical condition, and with an absence of defects [7]. The removal

of glochidia is the most critical post-harvest operation, being performed mechanically. The fruit selection is made manually by operators, who must wear gloves and visual protection. Mechanical cleaning is carried out by passing the fruit through a series of brush swivels with firm hairs, which smoothly eliminate the glochidia without damaging the fruit epidermis [7,133]. Subsequently, the fruits are selected, washed, and sanitised. If it is necessary to carry out the peeling of the PP, this is accomplished manually with the use of knives. First, the ends of the fruit are cut, followed by a longitudinal cut, so that the epidermis is extracted in one piece [5,7].

Fresh preserved PP is obtained by waxing, by immersion or spraying of wax, to control the loss of water by transpiration, reduce the intensity of the fruit's gas exchange, improve its appearance, and prolong its conservation [8]. This product's most used packaging is wood, plastic, or card. After packaging, the PP must be stored between 5 and 8 °C. The effectiveness of this type of conservation depends on several factors, such as storage time, type of packaging, and time of harvest [7].

According to the *Codex Alimentarius*, the fresh PP commercialisation must comply with several minimum quality requirements, such as being whole, healthy, clean, devoid of glochidia, free from pest damage, free from abnormal external humidity, free from damage caused by low temperatures, free from any extraneous aroma and taste, and free from pronounced stains, and have a fresh appearance, a firm consistency, and a satisfactory degree of ripeness. PPs can be classified into three categories [47]:

- "Extra" category, which includes superior quality fruits that do not contain defects, except superficial ones, that are very light, and that do not affect the general appearance of the product and its state of conservation;
- Category I, which includes good quality fruits, with defects allowed being light in shape, colour, and skin, such as spots, scabs, and other blemishes on the surface areas, provided that the total area affected does not exceed 4% and that the defects do not affect the pulp of the fruit;
- Category II includes fruits that do not fit the categories above, with the surface affected by the defects, referred to in category I, not exceeding 8%.

As an alternative and without compromising the nutritional qualities of the fruit, there are minimally processed PPs, with characteristics like those of fresh PPs, but with a longer lifetime. After washing the fruits, they can be peeled and cut, and then subjected to natural or artificial drying [7]. Despite the advantages of this processing, the physical damage caused to plant tissues makes these products more perishable than when intact due to the acceleration of metabolism from cutting [129]. To avoid this problem, these products are packaged in modified atmosphere packaging (MAP) and stored at refrigerated temperatures (5 to 6 °C) [7].

In MAP, the modification of the atmosphere is the result of breathing in the packaged product depending on the permeability of the packaging and temperature (passive modification) or due to the injection of a gaseous mixture in the free space of the package, so that the atmosphere is determined by the interaction between the product, the packaging, and the environment (active modification) [12,132]. MAP implies a decrease in the concentration of O_2 and/or an increase in CO_2 levels, reducing the rate of respiration, production, and action of ethylene, the degradation of chlorophyll, the loss of texture, and the delay in the maturation of the product [134]. Decreased O_2 and high CO_2 concentrations can also reduce or inhibit the food spoilage and the growth of pathogenic microorganisms [135]. The main constituents of the films used in MAP are low-density polyethylene (LDPE), polypropylene (PP), polyvinyl chloride (PVC), polyethylene terephthalate (PET), polystyrene (PS), and cellulose. The increase in the product's useful lifetime is due to the balance between it and the packaging. Therefore, it is important to adjust the permeability of the film to the O_2 and CO_2 concentrations, according to the respiratory rate of the PP, represented in Figure 7 [7].

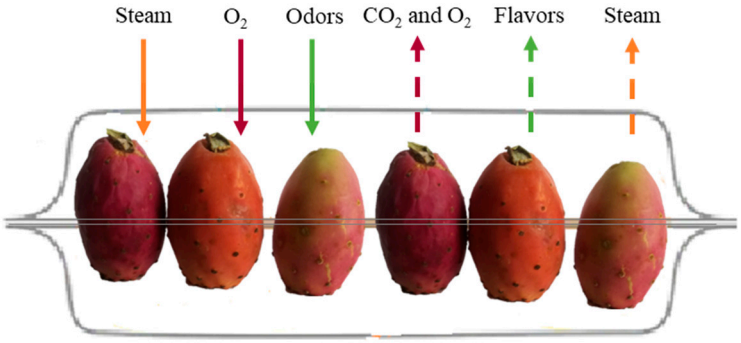

Figure 7. Transfers that occur through modified atmosphere packaging [7].

Piga et al. (2003) investigated possible changes in ascorbic acid and polyphenols levels in whole PP, manually peeled and packaged in passive MAP. The mass decrease of the PP was low, with a maximum of 0.15 g/100 g, after nine days at 4 °C. The minimal processing applied to the PP did not indicate a significant decrease in ascorbic acid and polyphenols levels nor in their antioxidant capacity. Despite being significantly altered during storage, other parameters, such as acidity and pH, did not harm sensory parameters [131].

Inglese et al. (2002) mentioned using polyethylene films in PP stored at 6 °C for six weeks and commercialised at 20 °C [136]. Corbo et al. (2004) found that PP storage at 5% O_2 and 30% CO_2 caused a selective suppression of the growth of different microbial populations [137].

Cefola et al. (2011) studied the effects of different storage temperatures and modified atmospheric conditions on commercialising ready-to-eat PP. The storage at 4 °C, both in passive and active atmospheres (10 kPa O_2 and 10 kPa CO_2), improved marketability by 30%, while storage at 8 °C caused a reduction in the partial pressure of O_2 in the MAP due to the increase in the metabolic activity of fruits, which contributed to the loss of commercialisation of the PP. The researchers concluded that it is possible to store PP in the yellow-green maturation phase, in active or passive MAP, at 4 °C for 9 days [128].

A complement to MAP is the application of edible coatings, which are thin pieces of edible material formed on a food surface or placed between food components [138]. Edible coatings applied to whole fruit and/or cut fruit can control water loss, delay ripening and colour changes, improve appearance and texture, prolong the lifetime, and create a barrier against various hazards. In addition to these advantages, these coatings can be used as carriers of antimicrobials or antioxidants. Examples of edible coatings are proteins and polysaccharides from renewable sources, such as starch, chitosan, carrageenan, alginates, soy proteins, corn zein, wheat gluten, casein, egg albumin, and fish proteins [127].

Del Nobile et al. (2009) tested different packaging designed to prolong the lifetime of minimally processed PP. Fresh fruits were coated with sodium alginate, agar-based gel, and fish-protein-based gel. Then they were wrapped in a film and biodegradable material. The results indicated that only sodium alginate coating extended the shelf life of minimally processed PP to about 13 days, responding to an increase of about 40% compared with the control sample. The pH values of all samples steadily decreased during storage, probably due to microbial fermentation, mainly attributable to yeasts. These results are also positive from an environmental point of view, reinforcing the need to replace synthetic materials with biodegradable films in various packaged foods [127].

Ochoa-Velasco et al. (2014) conducted a study to assess the effect of chitosan coatings containing different concentrations of acetic acid on the physicochemical, antioxidant, microbiological, and sensory characteristics of white- and red-peeled PP. Chitosan is produced through the deacetylation of chitin. It is a natural cellulose-like compound widely used for covering fruits and vegetables due to its ability to inhibit the growth of pathogenic microorganisms. The chitosan coatings form a semipermeable barrier capable of controlling gases and moisture. PPs treated with chitosan, containing 1% of acetic acid and stored at 4 °C and 85% relative humidity, maintained their quality for 16 days, while chitosan-coated PPs,

including 2.5% of acetic acid, did not appreciate well due to the acidity present. Chitosan coatings retarded microbial growth in both PP varieties, regardless of the amount of acetic acid used. The main factor that limited the validity period of the white PP was the moisture loss; for the red PP, it was the loss of firmness [138].

Due to the difficulty in peeling PP, there is a great demand in the market for fresh-cut PP, i.e., ready-to-eat PP. As little information has been published about the conditions necessary for the post-harvest storage of ready-to-eat PP, Kahramanoğlu et al. (2020) selected five biomaterials (*aloe vera* gel, *Portucala oleraceae* extract, *Vitis vinifera* leaf, cactus gel, and jam) to coat the fresh-cut PP. The results suggested that the duration of post-harvest storage of ready-to-eat PP could be extended using the different biomaterials evaluated. In this context, they found that the *aloe vera* gel and the cactus gel were more effective in protecting against mass loss and microbial spoilage in the quality sensory input and changes in TSS concentration [126].

Dehydration is another food preservation method that has been used for centuries. This alternative consists of eliminating water by evaporation to reduce the risks of microbiological contamination and avoid chemical reactions, allowing the preservation of the fruit's characteristics and to increase its lifetime at room temperature [7]. Dehydration is based on the reduction of a_w, which is a measure that represents the amount of free water or available water in a food. Water availability in plant tissues is variable and depends mainly on the composition of the fruits, since some components, such as hydrocolloids, have a more remarkable ability to retain water [5]. Fresh fruits and vegetables have an a_w close to 1.0, hence their susceptibility to microbial attack and rapid perishability [6]. Microbial growth can be controlled by decreasing a_w, because each microorganism has a critical a_w, below which it cannot multiply [5]. Beyond the indicated advantages, dehydrated products generally do not contain additives, and are considered safe natural foods [6].

After the post-harvest stages, inherent to any technological process and mentioned above, the PP can be cut and dried at 60 °C to reduce the final fruit moisture content (<12%). After packaging, the dehydrated PP must be kept in a cool, dry place and protected from light. The conservation period is at least six months and can reach up to a year, depending on the moisture content of the air, after drying [7].

As an alternative to the traditional dehydration process (drying), there is osmotic dehydration (OD). This process consists of the partial removal of water by pressure caused when the product, whole or in pieces, is placed in contact with a hypertonic solution of sugars, without phase change, due to the difference in osmotic potential that checks between the products and the dehydrating hypertonic solution. In addition to its low-cost energy, it is a suitable method for all production scales, being an alternative technology to reduce post-harvest losses [43]. The final product must be packed in containers impermeable to gases and water vapour and kept in the dark at room temperature. In general, drying does not change the flavour of the PP. The OD increases the strength of the fruit structure and improves the final product's flavour and colour. Despite these advantages, these alternatives have the same disadvantage, which is the hardening of excess pulp. As the PP has many seeds, which is evident with water removal, the final product becomes difficult to chew [7].

We can obtain dry mesocarp or dehydrated pulp to circumvent the inconvenience of seeds. In the case of dry mesocarp, after cutting the ends of the already peeled PP, the mesocarp is removed by longitudinally cutting it [5]. This is cut into strips and subjected to the drying process at 60 °C. The final product does not contain seeds and has a pleasant taste and a good texture. Its conservation is at least six months and it can be implemented in cereal flakes or in various snacks [7]. In the case of PP pulp, this is obtained through the pulping of the fruit, carried out through a sieve, intending to separate the pulp from the rind, fibres, and seeds [5]. The seedless pulp undergoes homogenisation, after which it is subjected to a formulation, with adjustment of the TSS content. After the pulp is processed, it has a porous appearance, with tiny rudimentary seeds, sweet flavour and light acidity, and is ready for consumption or to produce diversified, value-added products through

other technological processes [7]. One of these products is dehydrated pulp that can be processed or mixed with other fruits, such as apples or quinces, to give body to the product without influencing the final taste [5]. The mixture is subjected to a drying temperature of about 60 °C, with ventilation, until a dehydrated chewable product is obtained. After dehydration of the pulp, with about 10 to 15% of humidity, it is cut into different sizes and thickness strips, serving as a raw material to produce various snacks, muesli, and other food products [7]. This approach has had a substantial degree of acceptance at the level of gourmet products, being considered as a ready-to-eat food consumed and available throughout the year [5]. The most suitable packaging for these products is plastic, impermeable to light and water vapour, and then placed in cardboard boxes and kept at room temperature [7].

El-Samahy et al. (2007) carried out a study with the dehydrated pulp of yellow–orange PP, to evaluate different drying temperatures (60 °C and 70 °C) and sucrose concentrations (0, 1, 2, 3, 4, 5, and 10%). The pulp was spread to a thickness of 10 mm and dehydrated in an air convection oven for 44 h. The final product consisted of dry PP leaves, which were organoleptically evaluated. The products with the best acceptance level were those prepared with 2 and 3% sucrose [139].

Within the dehydration techniques, a method widely used for drying pulps and fruit juices is foam layer drying. In this method, the pulp and/or juice are transformed into a stable foam through a mixer and incorporation of air or other gas, and subjected to drying with heated air until the growth of microorganisms, and chemical and/or enzymatic reactions is prevented [140]. This technique is simple, inexpensive, and fast, resulting in a porous product that is easy to rehydrate. Usually, thickening, emulsifying and stabilising agents are used, which are intended to keep the foam stable throughout the process [141]. This method's main advantages are the need for low dehydration temperatures and a shorter drying time due to the larger surface area being exposed to air, which increases the speed of water removal [140]. Melo et al. (2013) evaluated the influence of foam thickness and drying temperature in drying the mandacaru fruit's pulp in a layer of foam, the former being a plant belonging to the *Cactaceae*. The data obtained during the drying process concluded that the drying temperature influenced the process and foam thickness, with the fastest occurring at the smallest and highest thickness [140].

Another conservation method based on a_w reduction is evaporation or concentration, which may have some effects on the product. This process consists of the loss of water, being always associated with the loss of volatile compounds responsible for the flavour and aroma of the raw material. Therefore, concentrated products generally have a less intense aroma [5]. In addition, there is also a reduction in the volume of the product, which is an advantage for storage and transport. From the consumer's point of view, concentrated products are helpful because they are easier to use, take up less space, are stored at room temperature, and can be consumed in the desired portion, while the remainder can be safely stored [6].

Freezing is a very efficient conservation technology as it allows for preserving food's colour, aroma, texture, and nutritional and functional properties [6]. This method combines the effects of low temperature with a decrease in the water reactivity due to the formation of ice crystals. Therefore, microorganisms cannot grow, chemical reactions are reduced, and cellular metabolic reactions are delayed [5]. The PP pulp can be frozen in freezer chambers at a temperature of −30 °C and stored at −18 °C [7]. The faster the product freezes, the smaller the ice crystals that form and the better the quality of the final product. The tiny ice crystals do not damage the product's structure. At the agro-industrial level, cold air tunnels are used (temperature of −40 °C) or the fruit is sprayed with liquid nitrogen (temperature −196 °C) [5]. The packaging material for this type of product must offer protection against oxidation, moisture loss, and changes in sensory characteristics [7]. The disadvantage of freezing is that it is an expensive technology that requires developing and maintaining a cold chain from production to consumption to guarantee the quality and safety of frozen products. Frozen pulps can be sold directly to the final consumer or

manufacturers as a constituent of flavoured drinks, yoghurts, desserts, ice creams, cakes, pastries, or confectionery [6,13].

Lyophilisation is an alternative drying process for foodstuffs such as onions, apples, bananas, ginger, or pineapples. This technique consists of removing water by sublimation under reduced pressure conditions. In this way, the driving force for water removal is the vapour pressure difference between the ice and the surrounding environment. The water in the foodstuff passes from the solid phase directly to the gas without passing through the liquid phase [142,143]. Thus, lyophilisation is carried out to convert the ice into vapour without entering the liquid phase. The first step is to freeze the foodstuff to be freeze-dried at a temperature below 0 °C. Subsequently, the next step is the primary drying phase, with the sublimation of ice on the surface of the product. At this stage, the required temperature is around −10 °C, and the absolute pressure is approximately two mmHg (2.6 mBar). The last step is secondary drying, which corresponds to removing adsorbed water through evaporation. For this, applying a high-pressure gradient and increasing the temperature is necessary, but without causing damage to the product [144].

Lyophilisation can be carried out under vacuum or atmospheric pressure. The most used is the first because it allows for a final product of better quality. However, lyophilisation under atmospheric pressure has a lower energy consumption and a slightly shorter drying time [143]. Despite the high costs associated with this drying method, it becomes advantageous because it avoids the degradation of the final product due to thermal decomposition, oxidation, or enzymatic reactions [145]. On the other hand, as the lyophilisation is carried out at low temperatures, there are no changes in colour, aroma, and most of the nutritional composition of the foodstuff [143].

According to various studies, Hamad et al. (2022) summarised the physical characteristics (colour, taste/odour, and thermal properties) and the chemical characteristics (moisture content, total phenolic compound, and anthocyanin content) that changed during the spray drying (SD) and freeze drying (FD) of berry fruits. They concluded that juice taste and colour of berry powders that FD produced were better than SD, and this technique better preserved the nutritional value and bioactive compounds. On other hand, the morphology of the berry powders that underwent SD was better than the result with FD, and the losses of phenolic compounds and anthocyanin content of SD berries were much lower than FD [146].

6. Transformation Methods of Prickly Pear

6.1. Juices

One of the most common technologies for preserving fruits is the production of juices. As the PP pulp presents a variety of colours, the juices obtained become even more appealing [7]. Fresh juices should be consumed as soon as possible, preferably after their production. For a longer useful life, the juices should undergo a heat treatment to avoid pathogenic microorganism appearance without affecting the final product's taste and appearance [5]. Generally, a high-temperature and a short-term duration heat treatment is chosen (HTST—high-temperature short time) so that the product undergoes a shorter deterioration. Subsequently, the juice must pass quickly through a system of cooling, at about 20 °C, for 30 min, to prevent overheating, which causes organoleptic and nutritional changes in the final product. In cases where the juice has been pasteurised, it must be stored at refrigeration temperature (8 °C to 10 °C), and if it has been sterilised it must be stored at room temperature (28 °C to 30 °C) [7].

The stability of juices depends not only on the raw material but also on the processing, packaging, and storage conditions. These factors can cause microbiological, enzymatic, chemical, and physical changes that damage the sensory and nutritional characteristics of the juice. Consumers demand fluids with minimal processing and no added sugar, resembling the original fruit [130]. Thus, the transformation of the PP into juices must be well established to satisfy the growing demands of consumers and encourage the consumption of this fruit [6].

The parameters that change the most during PP juice processing are the water content and the viscosity. Acidity, pigments, and aromatic compounds are the most important parameters in PP juice processing [6]. Considering some studies on PP juices, the purple ecotype seems to be the most promising in transforming the fruit into juice due to the stability of betalains. The green PP is the most challenging for juice production due to the presence of chlorophyll and its instability when subject to heat treatments, which cause changes in colour and flavour. The orange PP appears to be a good alternative for juice production but requires more studies [115].

El-Samahy et al. (2007) performed a study with orange–yellow PP juices. After obtaining the pulp, it was mixed with a sugar solution at 15 °Brix and pH 5 in a 1:1 ratio. The prepared juice was divided into three parts: the first underwent direct pasteurisation at 95 °C; the second was treated with 100 mg/L of sodium benzoate and then pasteurised at 95 °C; and the third underwent direct sterilisation at 121 °C. Subsequently, the chemical, microbiological, and sensorial characteristics of the obtained juices were evaluated during storage at room temperature (28 °C) and refrigerated temperature (8 °C) for 6 months. All juices produced were microbiologically stable during the storage period, and pasteurised juices (with or without sodium benzoate), mainly stored at refrigeration temperature, showed the best organoleptic results [139].

Kgatla et al. (2010) investigated the effects of PP juice processing and preservation on its organoleptic attributes (colour, flavour, aroma, astringency, visual appearance, and general acceptability). The PP pulp was treated with pectinase, and the treatments applied to obtaining the juice were the addition of sugar, acidification, heat treatment, refrigeration, freezing, and thawing. The reddish-purple colour of the PP remained stable for all processing and conservation. The untreated juice tasted bitter, with a high astringency, while the treated juice was significantly sweeter. The authors observed a significant difference in the acceptability of untreated and treated juices: the juries rejected the first, and the second had a good sensorial acceptance. With this study, the researchers concluded that the processing and conservation of PP juice had the positive attributes of the respective organoleptic properties [130].

Sometimes, when the heat treatments are long, the final product will likely have an unpleasant taste and/or undesirable aroma. In this way, one can resort to mixing it with other fruit juices to improve the quality of the final product [101]. Sáenz and Sepúlveda (1999) tested different formulations by mixing purple PP juice with pineapple juice, citric acid, water, and sugar. The dilution of PP juice proved to be an advantage in minimising its viscosity, and the addition of citric acid and pineapple juice lowered the pH, which reduced the risk of microorganism growth [115].

Since PP juices have a short shelf life, Ferreira et al. (2022) conducted a study where they applied thermal (TP) and high-pressure (HPP) pasteurisation to OFI juices from three different cultivars from Idanha a Nova (Portugal). They also evaluated the impact of these methods on microbial safety, physicochemical properties, and nutritional content over storage at 4 °C. They concluded that TP at 71.1 °C for 30 s increased the shelf life by 22 days, and HPP at 500 MPa for 10 min increased the shelf life by 52 days relative to microbial growth and to the preservation of physicochemical properties. Applying these pasteurisation methods retarded the physicochemical changes, namely in titratable acidity, °Brix, browning, polyphenolic content, and antioxidant activity of the juices [147].

Other types of products can be made from the PP juice, such as concentrates or nectars, prepared by direct concentration or by adding sugar. In the case of nectars, the production steps are identical for obtaining juice, incorporating sucrose or corn syrup, and sometimes some additives, such as carboxymethylcellulose, to give body to the final product [7]. The main advantage of these products is having a lower a_w in relation to juices, which leads to an increase of the product shelf life due to lower chemical reactivity and greater protection against the development of microorganisms [84].

6.2. Fermented Products: Beverages and Vinegar

Artisanal alcoholic beverages can be obtained by juice fermentation, such as "colonche" (4 to 6% alcohol), wine (reaching about 11% alcohol), and brandy (reaching up to 56% alcohol). Other types of products that have been emerging are vinegar and liqueurs based on PP or based on mixtures with other fruits [7].

Lee et al. (2000) fermented various mixtures of PP juice (PPJ) and grape juice (GJ) to produce alcohol. Fermentation was carried out using *Saccharomyces cerevisiae* with the addition of sulphur dioxide (SO_2), sodium sulphite (Na_2SO_3), and tartaric acid ($C_4H_6O_6$) to adjust the acidity. The fermentation of the PPJ could have been more successful, but it was progressing with the addition of the GJ. The mixture of PPJ (25%)/GJ (50%) was fermented at 30 °C, for 7 days, and presented an alcohol content of 9.2% (*m*/*V*). The mixture of PPJ (70%)/GJ (30%) produced an alcoholic beverage with 6.9% (*m*/*V*) of alcohol, and alcoholic fermentation of the mixture of PPJ (50%)/GJ (50%) was carried out at 22 °C for 6 days [148].

Turker et al. (2001) used PPJ as a raw material for fermentation and *Saccharomyces cerevisiae* as a fermentation microorganism. The fermentation process allowed the conversion of 95.54% of the fermentable sugar, with an ethanol yield of 55.3 mL/L. According to the results obtained, the authors concluded that the fermentation process did not affect the thermostability of PP betalains [149].

Pérez et al. (1999) prepared vinegar from orange PP. To carry out the alcoholic fermentation at 13.5 °GL, they used *Acetobacter pasteurianus*, and for the fermentation of PPJ with added sugar at 22 °Brix they used *Acetobacter xylinum*. The vinegar produced in both cases had a clean, bright, intense amber–yellow colour, and an acidic, fresh, and intense aroma. Different kinds of vinegar can be developed depending on the wide range of colours of the PP [5].

Es-sbata et al. (2022) produced PP vinegar using two types of acetification processes (surface and submerged culture), at different temperatures (30, 37, and 40 °C), by using two different species of thermotolerant acetic acid bacteria (*Acetobacter malorum* and *Gluconobacter oxydans*). Then, 15 polyphenols and 70 volatile compounds were identified and quantified in the PP vinegar samples produced by both acetification processes, but the phenolic content from surface culture acetification was higher, and the submerged culture was a faster and more efficient acetification method because of the higher concentration of acetic acid in the PP vinegar. With this study, they verified that PP vinegar can be successfully produced at higher temperatures than usual by employing thermotolerant bacteria and that the type of acetification method significantly affects the final quality of the vinegar produced [150].

6.3. Sweets, Jams, and Jellies

Another technological alternative is sweets, jams, and jellies, which are obtained by boiling the PP pulp with ingredients such as sugar, citric acid (implements taste and microbiological stability), pectin (responsible for viscosity due to its water holding capacity), and preservatives such as sodium sorbate or potassium benzoate (they preserve the quality of the product after opening the package), followed by concentration by evaporation to ensure a certain degree of gelation [6,7].

Sawaya et al. (1983) reported the manufacture of PP jam. In this work, they studied the effects of the sugar:pulp ratio, acidifying agents (various types and amounts), the pectin dosage, aroma, the date:PP ratio, and the bleaching effect of fruit on the quality of the compote. The best sensory results were obtained with the following recipe: pulp:sugar ratio of 60:40, adding 1.25% pectin, citric acid, or a combination of citric and tartaric acids (1:1), with a 20% date ratio. Regarding the added flavours, the favourites were clove, grapefruit, orange, and almond [73].

Saenz et al. (1997) studied green PP pulp gels to which sugar and carrageenan were added. Although a significant colour change was observed because of the chlorophyll phaeophytinization due to decreased pH, the product maintained its physicochemical and sensory properties for more than 14 days when refrigerated between 4 and 6 °C. They also

concluded that refrigeration could be avoided if they increased the concentration of sugar, which is a common commercial practice [6].

Razafindratovo et al. (2022) carried out a study focused on the nutritional characterisation of PP pulp and processed products (jam and syrup). They concluded that the jam and syrup made from the PP pulp were products with interesting nutritional, organoleptic, and physicochemical characteristics, which complied with manufacturing standards [151].

Abu-shama et al. (2022) studied jelly candies prepared by using six formulas of PP juice and peels (yellow and red cultivars). The obtained results showed that the jelly candies produced were an important source of total polyphenols, flavonoids, carotenoids, and betalains. The jelly candies produced could be stored for more than four weeks during cold storage and had an acceptable quality to consumers: the sensorial evaluation showed that red peels, red juice, and yellow juice jelly candies were the most accepted by panellists [152].

6.4. Oil

The PP seed oil has several physicochemical and nutritional supplements, making it like other edible vegetable oils, such as corn or grape [84]. After extracting the seeds from the fruit, they are cleaned and dried before starting the cold extraction process [7].

On the one hand, this oil can be used as a fat substitute in confectionery, and the by-product obtained from its extraction can be used for animal feed. On the other hand, PP seeds produce a low of oil yield due to the number of seeds obtained, and are only profitable if the extraction is associated with processing, whether of pulps, juices, or jams [5,7,84].

Ammar et al. (2017) carried out a study focusing on evaluating the quality and oxidative stability of olive oil (very important in Mediterranean cuisine) with added OFI flowers. The quality, fatty acids profile, total phenol contents, and thermal properties of the olive oils enriched with OFI flowers were studied. The oxidative stability of olive oils was improved, mainly in olive oil enriched with 5% OFI flowers, after 15 and 30 days of storage at 60 °C. The olive oil was also nutritionally enriched due to the increase in its phenols content [153].

6.5. Flour

Another product from various OFI processing techniques is flour from cladodes, seeds, and PP peel. The OFI flour can be used to make cookies, puddings, cereals, tortillas, and various snacks, or to produce food supplements in the form of capsules or tablets, allowing increased daily dietary fibre intake [5].

OFI flour can be obtained by dehydrating and grinding cladodes, previously degreased, washed and cut, or by grinding the seeds and/or peel, after which it passes through two sieves of different granulometry to separate the fibres [6,7]. The result is simple and inexpensive flour that can be mixed with other flour to improve its taste, smell, colour, and texture [5].

Ayadi et al. (2009) studied the fortification of wheat flour with OFI cladode flour for obtaining cakes. As a source of fibre, cladodes increase the flour's water absorption capacity and, consequently, the cake dough's properties. The results indicated that adding cladode flour increased the tenacity and decreased the elasticity of the cake dough. In terms of colour, the cakes containing cladode flour became greener inside and darker outside. The best sensory results were with the cakes with 5% of cladodes flour [154].

In the study of Msaddak et al. (2017), the effects of OFI cladodes powder substitution of wheat flour on dough's rheological, physical, and antioxidant properties and sensory bread characteristics were analysed. It was found that, in wheat bread formulation, a substitution of up to 5% of wheat flour by cladodes powder was possible without changing the physical and sensory properties. This change improved the total phenolic content and the bread's antioxidant potential without negatively affecting its sensory acceptability [155].

6.6. Snacks

Snacks are defined as light meals consumed between regular meals and may include a wide range of products, such as cookies, cereal bars, yoghurts, and even ice cream [156]. Within this category, cookies are the most popular bakery products consumed worldwide due to their various characteristics: ready-to-eat, reasonable cost, high nutritional value, long shelf life, and availability in different colours, shapes, and flavours [157].

The production of cookies of acceptable quality is based on the selection of flour and the appropriate processing steps, such as mixing, aeration, fermentation, cooking, cooling, and packaging [158]. The main constituents of cookies are flour, wheat, sucrose, and fat, which make them a very dense food. Due to their low moisture content, cookies can serve as a source of various nutrients. There is a tendency to produce functional cookies made from wheat flour, with a good source of calories, different nutrients, and other flours containing active compounds [159,160].

Abou-Zaid et al. (2022) determined the chemical characteristics of fresh juice and peel of both OFI (yellow cultivar) and *Opuntia littoralis* (red cultivar), and used them in cookies production and evaluated the quality of the cookies. The results showed that fresh PP juices and peels could be used to produce healthy and organoleptically appealing cookies. The cookies produced with juices had a higher moisture content than those produced with peels, and those produced with peels had the highest crude fibre contents and the highest weight and hardness [30].

To take advantage of the by-products of fruits and vegetables and recover the individual dietary fibres, bioactive compounds, and antioxidants, Elhassaneen et al. (2016) produced snacks with flour from PP and potato peels. The peels were dehydrated under vacuum, at 70 °C, for 3 h to obtain flours with 7% humidity. Subsequently, they incorporated the flour into the cookie dough at levels of 5%. Biscuits enriched with the by-products studied showed higher fibre contents, total dietary intakes, carotenoids, and total phenolic compounds in comparison with control biscuits. In this way, the authors concluded that, by incorporating the flours from the PP and potato peels, it was possible to improve the nutritional and functional quality of the cookies without affecting their sensory characteristics [156].

Mahloko et al. (2019) developed and evaluated cookies obtained by mixing flour from PP and banana peels in another study with a similar objective. The flour was dehydrated at 60 °C overnight and incorporated into the wheat flour for the biscuit production. The results obtained indicated that the cookies enriched with the flours from PP and banana by-products contained higher levels of fibre, total phenolics, and flavonoids than the control. In general, they concluded that incorporating flour from PP and banana peels improved cookies' functional properties, colour, and antioxidant activity [160].

El Samahy et al. (2007) produced another type of value-added snack based on PP pulp. In this study, they concentrated the yellow–orange and red PP pulps to 40 °Brix and added them to the rice flour. Different formulations were tested, varying the relationship between rice flour and PP pulp concentrate. The products that obtained the best functional, nutritional, and sensory characteristics contained 5% and 10% PP pulp concentrate [13,161].

6.7. Dairy Products

Another way to use PP is to obtain dairy products. One of these products is PP "cheese", obtained from concentrated juice, which is boiled until reaching the desired consistency. After cooling, the product is a compact, malleable mass with a high sugar content, like caramel. Then it is struck on a smooth, moist stone platform to allow air to enter and prevent the formation of sugar crystals. Before packaging, the dough is shaped and kept for 12 to 15 h. This product must be slightly heated and can be enriched with vanilla, pine nuts, hazelnuts, walnuts, shredded coconut, raisins, or almonds [7].

Milk permeate is a by-product of milk ultrafiltration, containing 80% of the lactose starting point of treated milk, and is an excellent source of vitamins and minerals [162]. To reduce the environmental pollution inherent in the elimination of permeate, dairy industries

try to arrange new destinations for the permeate, such as the preparation of fermented milk or the production of chocolate milk [163,164]. Bearing this perspective in mind, Jambi et al. (2017) looked at producing PP-based beverages by mixing different concentrations of PP pulp and permeate. They studied the different mixtures' physicochemical, microbiological, and organoleptic properties after production and storage at 4 °C, over 7, 14, and 21 days. The obtained results indicated that the acidity and the levels of phenolic compounds and ascorbic acid increased as the PP pulp concentration increased in the beverages, unlike the pH, which decreased. Over storage time, phenolic compounds and ascorbic acid content decreased in all mixtures. The lactose content of the drinks also reduced over time of storage. The beverage that contained 30% of PP pulp and 70% permeate had the highest organoleptic ratings, both immediately after its preparation and after 21 days of storage at 4 °C [165].

6.8. Sweeteners

Based on juice technology, there is the possibility of obtaining natural sweeteners with a PP base through the treatment of enzymatic clarification, using pectinolytic enzymes with high arabinase activity. During the process, the acidity of the PP juice is corrected with citric acid to pH 4.2 to 4.5, so the enzyme works. This is followed by filtration, decolourisation with activated carbon, a new filtration, and concentration until a product with 60 to 62 °Brix is obtained. The final product has a yellowish colour and a sweetness like other commercial liquid sweeteners, and is packaged in glass or polyethylene bottles and stored at room temperature [7]. Saenz et al. (1996, 1998) were the first to develop a process for obtaining a natural liquid sweetener from PP juice. The final product, light golden yellow, had 60 °Brix (56% glucose and 44% fructose), a 1.29 g/mL density, a_w of 0.83, and a viscosity of 27.1 cps, like honey or marmalade [84].

6.9. Natural Dyes

In addition to obtaining flour, there is another type of recovery of the PP peel: extraction of its pigments, which differ depending on the ecotype, and the extraction of pectin [16,119]. The pigments that PP can offer range from chlorophyll (green colour), betacyanin (purple colour) and betaxanthin (yellow–orange colour), and they are a promising source of natural dyes. Aqueous solutions of mucilage extracted from the fruit can be used as a source of fibre, as a thickening agent for culinary purposes, and as an edible coating to protect fresh fruit [5–7].

Del-Valle et al. (2005) studied the mucilage extracted from PP as an edible coating to prolong the shelf life of strawberries stored at 5 °C. In addition, to investigate different methods of mucilage extraction to obtain the best coating, they also analysed the effects of the other coatings on the colour, texture, and sensory quality of strawberries. According to the results obtained, they concluded that, besides the mucilage coatings increasing the shelf life of strawberries, the latter also maintained their texture and flavour, with no deterioration after 9 days of storage [166].

PP peels are cheap, readily available, and their physicochemical properties can be used to improve food product parameters such as shelf life, sensory characteristics, and viscosity. They may also be applied as fat or sugar substitutes to stabilise food oxidative processes, and as oil and water retention capacity enhancers [3].

Chougui et al. (2015) conducted a study to evaluate the use of PP peel hydroethanolic extracts as a substitute for vitamin E, used as an antioxidant in margarine conservation. The tests indicated that the margarine prepared with three different concentrations of PP peel extract was more resistant to oxidation than margarine containing vitamin E. In addition, the physicochemical and microbiological properties of margarine were not modified. The lowest concentration of analysed extract (50 mg/kg) showed a higher margarine shelf life [167].

7. Case Study: Agro-Industrial Uses in Portugal and Prospects for Market

The PP is a fruit with potential agro-industrial expansion as it is a source of several nutrients and an efficient system to produce various foods, such as juices, jellies, pulps, oil, or flour [7]. The technological transformation that this fruit can undergo becomes very relevant due to the decrease in water resources, the increase in global desertification, the maximum use of resources, and how the plant adapts to arid lands and severely degraded soils, unsuitable for traditional cultures, and is important from the sustainability point of view [5]. On the other hand, the PP has characteristics of specific seasonality and rapid post-harvest deterioration, which compromises its storage and marketing, so the agro-industrial applications aim at better use of this fruit, a reduction in production losses, and a diversification in the number of new products derived from this fruit [7,42].

Portugal, namely the Alentejo and Algarve, has soil and climate conditions making it possible to obtain high-quality PP production compared with other countries [7]. In Portugal, the PP crop is expanding, with more than 800 ha cultivated between two to four years [40].

Since 2008, the PP crop has existed in an orderly fashion in Portugal under the rural development programme (ProDer). The objectives of this programme are to support investment in agriculture, boost young farmers, and help the development of small and micro-enterprises [7]. To this end, a project was started that encompasses the study of culture, the creation of a collection of national PP ecotypes, the study of physicochemical and nutritional characteristics of the fruit, and the development of innovative food technology. Subsequently, and with the increase in the planted area, associations have been created within the scope of production, marketing, and cultural dissemination around the culture of the prickly pear [7].

Portugal has adopted specific production systems to prolong the harvest season to satisfy market needs and compete with other countries' PP exporters. Using agronomic techniques, it is possible to change the biological cycle of the OFI, such as prolonging the flowering season of the plantation, which promotes late flowering [40,42].

Bearing in mind that PP is characterised as a tropical fruit with several nutritional advantages, the introduction of national production in the domestic market is simply due to the growing interest in healthy foods and lifestyles. On the other hand, consuming this fruit and its processed products can increase its commercial and economic value at the national level [7,42,57].

The PP is for sale in some local markets and distribution chains, and its prices vary between EUR 2.99 and 14.99 /kg. This price variation is due to several factors, such as country of origin, type of packaging, size, and fruit quality [7]. As for exports, the percentage is still residual, and although no data statistics show international sales volumes, potential countries' PP buyers are England, France, and Japan [7].

8. Conclusions

In summary, the physicochemical and nutritional composition of OFI is only partly known because most of the investigations were done 10–20 years ago. Hence, this needs to be validated with up-to-date methods. This is an essential issue for OFI applications and better agro-industrial use.

The PP is a source of several nutrients and an effective system to produce varied foods, which have several advantages from a nutritional, sensory, economic, and shelf-life point of view. The technological transformation that this fruit can undergo becomes very relevant due to the decrease in water resources, the increase in global desertification, and the maximum use of resources. Due to its adaptation to arid lands and severely degraded soils, which are unsuitable for traditional cultures, it is also important from a sustainability point of view. On the other hand, the PP has specific seasonality characteristics and rapid post-harvest deterioration, compromising storage and marketing. Therefore, agro-industrial applications aim to use this fruit better, reduce production losses and diversify the offer of new products derived from this fruit.

Finally, it is essential to note that about one-third of the Portuguese territory is highly susceptible to desertification. Climate change could exacerbate the effects of droughts, and accelerate soil degradation and, consequently, the desertification of the region, severely conditioning the development of extensive rural areas. OFI cultivation can contribute to revitalising these rural areas and the dynamism of local economies, combating the depopulation process that affects them. In addition to producing fruits with excellent nutritional properties, its production allows owners of uncultivated or underused land to obtain a significant and sustainable income and stimulate upstream and downstream economic activities.

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ARTICLES FOR FACULTY MEMBERS

FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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Review

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Trends in research on cacti: the food of the future

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Abstract

Cacti are a distinguished group of plants that stand out for their great nutritional values, diverse uses, and unique morphology, allowing them to grow and thrive under different conditions such as dry, xeric, and even low-temperature environments. The world is going through significant climate changes that are affecting the agriculture system. Therefore, sustainable and multifunctional crops, as many species of the Cactaceae family are, might be a good alternative in the near future. In this work, the uses of cacti in human food were analyzed through a scientific prospection from the point of view of their temporal and spatial distribution and potential uses. Brazil is the country with more publications related to the scope of this work, followed by Mexico. The presence of cacti in these countries can influence their interest in these species, which might reflect the results encountered in this study. The uses and ethnobotanical applications of cacti vary in different countries worldwide. Cactus is consumed fresh (in salads), in preparations (jams and sweets), and juices, being also present in traditional dishes in countries like Mexico. This study emphasizes cacti's importance in people's diets and ongoing world changes. Their ability to thrive even in hot environments with low water resources will lead to a greater focus on these species in the upcoming years. Furthermore, these plants have great flavor and contain several beneficial chemical compounds with desirable nutritional and health properties. Therefore, knowledge dissemination combined with technological innovations will allow greater use of these multifunctional species for human consumption.

Keywords: unconventional food plants; Cactaceae; cactus species; human nutrition; food demand

INTRODUCTION

Cacti are remarkable endemic plants growing in arid and semi-arid regions of the world. They represent a group of more than 1600 species and approximately 130 genera. The Cactaceae family is divided into four subfamilies: Cactoideae, Pereskioideae, Maihuenioideae, and Opuntioideae, the latter being the most common, with more than 300 species.[1,2] Most cactus species are endemic to America, and Mexico is the greatest center of diversity, with over 600 species recorded, followed by Brazil, Argentina, Bolivia, and Peru.[3] However, many species have become naturalized in other parts of the world, such as Australia, Hawaii, and the Mediterranean.[4]

Cactus species exhibit distinct physiological and morphological features, such as mucilaginous tissue, thick cuticles, shallow roots, and the ability to retain water.[5] With these drought-tolerant characteristics, cacti grow and thrive in environments that are stressful for most other species.[6] Owing to their adaptability and rich phytochemical profile, they represent a live forage reserve and an important biological resource for increasing food security, economic development, and livelihood in semiarid regions.[5,7,8]

Members of the Cactaceae family are used as ornamental crops, construction material, and cattle feed, and in traditional medicine and cosmetics. In addition, cactus species such as *Hylocereus* spp., *Opuntia* spp., *Pilosocereus* spp., and *Cereus* spp. are used in human food and have different applications in technological and food industries.[1] These edible species can be used as sources of ingredients for pharmaceuticals, natural dyes, and flour substitutes, as well as biofuel production.[7] Furthermore, they have a great flavor and contain several beneficial chemical compounds with desirable nutritional and medicinal properties.[1,6]

Despite these beneficial features, the consumption of cacti remains limited to local ethnic markets, and only a few countries, such as Mexico, Italy, and South Africa, produce them commercially.[9] They are used to prepare desserts, cakes, or juices, and are also consumed as fresh vegetables in salad dishes. Cladodes, fruits, and mucilage are the most frequently used parts.[5] They contain significant amounts of vitamin C, minerals, free amino

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acids, polysaccharides, and pigments such as betaxanthins and betacyanins.[4,8]

Although they constitute an unexploited and neglected crop worldwide,[4] cacti can provide fresh and processed foods with functional and medicinal properties. Thus, they can become vital foods in societies of the future.[1] In this sense, the use of scientific prospecting to map what has been published about these plants is relevant for the mining and dissemination of new information. In addition, prospective studies can identify potential strategic areas for the development of and benefits to the environment, economy, industry, and society.[10,11] Therefore, this study aimed to analyze the use of cacti in human food through scientific prospection.

MATERIALS AND METHODS

A literature search was conducted in March 2023, and no cut-off dates were established. Four databases were used for data collection: SpringerLink, Web of Science, ScienceDirect, and Scopus. The queries used were Cact* AND 'human food,' Cacti AND 'human food,' Cactus AND 'human food,' and Cactaceae AND 'human food.' The Boolean operator AND was used to direct the search according to the intersection of the words investigated. The truncation symbols (*) and ("") were also used to retrieve any endings from the searched terms and find the terms precisely as they were written, respectively. The search strategy, steps followed, and quantitative results of the search and analysis of the databases are presented in Fig. 1.

A total of 502 articles were identified in the search. Of these, 97 were duplicates and were excluded. After screening, 405 remaining studies were analyzed by title and abstract to identify which studies dealt with the theme investigated in this prospective study. Regarding eligibility, 52 articles were read entirely; of these, 20 fit the criteria described. In the other 32, although the title and abstract mentioned cacti, they were not focused on their use in human food [e.g., studies on agricultural production for human food, genetic characteristics and genetically modified organism (GMO) production, and antimicrobial studies using cacti].

EndNote Web Reference Manager software was used to analyze duplicate articles. The included studies were subjected to a bibliometric analysis of the following extracted metadata: keywords, country of origin, and year of publication. VOSviewer, Visme, Canva, and Microsoft Excel were used to build the graphical representations and tables. In addition, all the authors discussed and grouped the information contained in the articles to strengthen the argument regarding the physiological and morphological adaptations that have allowed the dispersion and

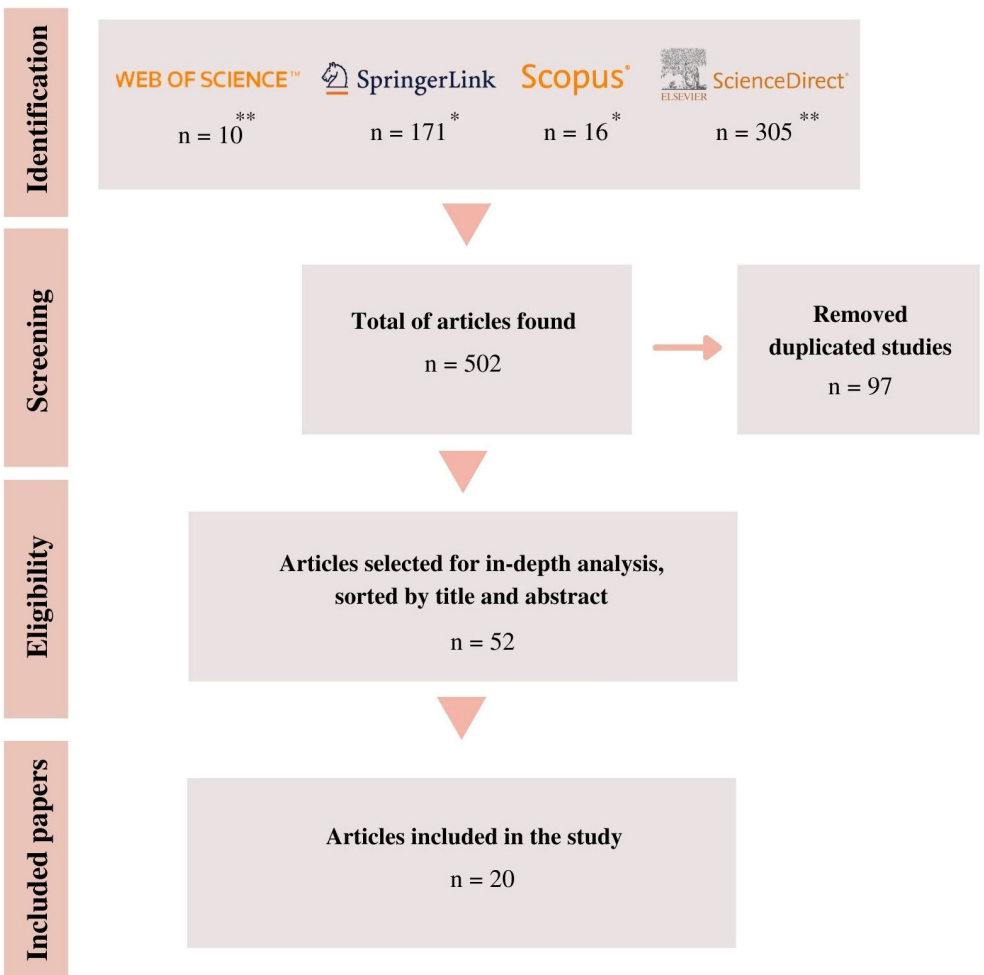

Figure 1. PRISMA flow diagram for articles selection. * = Cact* AND 'human food.' ** = Cacti AND 'human food'/Cactus AND 'human food'/Cactaceae AND 'human food.'

use of these species around the world to date, as well as the potential uses of cacti in the changing society.

RESULTS AND DISCUSSION

Overview

Bibliometric analyses are used in literature reviews to identify and map themes studied on the topic of interest and to identify new research trends.[12] Thus, a keyword co-occurrence analysis (Fig. 2) was used to provide an overview of the 20 articles included in the study and the existing connections between the conceptual items involving the use of cacti in human food. This technique assumes that each pair of co-occurring keywords, represented as linked keywords, has a thematic relationship. Thus, the network formed allows us to understand the thematic field from the patterns of connections between these keywords.[13]

Figure 2(a) shows the clusters formed based on the threshold for two occurrences of the same keyword. The keyword *Opuntia ficus-indica* exhibited the most significant co-occurrence frequency as it was tightly linked to two clusters. Also known as the prickly pear, this species is the most produced, distributed, and consumed species in the cactus family.[5,14] In cluster 1 (green), several cactus species were related to 'phytochemicals.' These compounds, which are significant in cactus species,[15] have several 'health benefits' and 'industrial potential.'

In cluster 2 (yellow), the keywords 'functional properties' and 'cladodes' were linked to some nutritional components found in cactus species. Cactus cladodes are characterized by the presence of considerable amounts of 'minerals' and 'polysaccharides,' mainly 'fibers.' which have functional properties that favor the growth of beneficial microorganisms ('probiotics') and modulate intestinal microbiota.[16,17]

Figure 2(b) shows the clusters formed with a threshold of one occurrence of the keyword, adding keywords with lower frequency of use. Four clusters were identified. The keyword 'nutritional composition' links two clusters: red and purple. The red cluster is related to the composition of 'Brazilian fruit' with antioxidant capacity to combat 'oxidative stress.' Studies have highlighted the levels of 'bioactive compounds' with anti-inflammatory and 'antioxidant capacity' in many Brazilian fruits.[18,19]

The red cluster also addresses the 'Caatinga,' the Brazilian semi-arid region, a biome marked by many cactus species and other 'wild plants' with 'food potential.'[20,21] In the purple cluster, 'Mexico' is listed with 'cultivation' and 'fruit crop varieties' since this country represents 44% of the world's production of *Opuntia* spp.[22] and is the largest trader of this type of cactus.[9] Furthermore, Mexico is the largest center of cactus diversity,[4] having achieved the 'domestication' of these species with 'morphological variations' many centuries ago.

Finally, two isolated clusters were observed in blue (dark and light blue). The dark blue cluster presents keywords related to 'sensorial analysis,' such as 'word association' to observe 'consumers' perceptions' of cacti, since these 'unfamiliar foods' depend on cultural acceptance.[23] The light blue cluster relates the 'non-food uses' of cacti (e.g., ornamentation, folk medicine, animal feed) with 'rural development' by opening local markets and increasing economic resilience[24] beyond their global importance as carbon sequestrations. Thus, cactus species have been receiving some attention in human food due to their nutritional, industrial, and health potential combined with environmental benefits.

Adaptability mechanisms

Cactus species are unusual and distinctive plants with several anatomical and physiological adaptations that enable survival in extremely hot and dry environments.[5,25] Although they survive in diverse habitats, such as coastal areas, deserts, and mountains, they are more abundant in arid and semiarid regions. In this sense, the origin of the 20 articles (Fig. 3) might be related to the development of studies about cacti in human food, as all countries (Brazil, Mexico, India, Morocco, the United States, Spain, Tunisia, and South Africa) have or are situated in dryland regions.

Drylands can be classified as hyper-arid, arid, semiarid, and dry subhumid areas[26] and cover approximately 50% of the terrestrial surface,[27] where more than two billion people live.[28] Generally, these regions are characterized by high temperatures, low rainfall, and poor soils, which underlie low agronomic productivity and represent a challenge for conventional cropping systems.[27,28] In addition, the temperatures, frequency, and severity of droughts globally are increasing with climate change.[29]

However, with the growth of the world's population and the increase in food demand, the diversification of crop production and use of drylands for agriculture will become vital for both human food and animal feed in the coming years.[27,30] Given the characteristics of these areas, along with irrigation restrictions due to water scarcity, the production potential of crops such as cereals and beans has reduced, increasing their market prices.[7,28]

Therefore, the use of species that can be cultivated in drylands and are resistant to drought can be an alternative to conventional agricultural systems. Furthermore, the exploration and popularization of crops such as those of the Cactaceae family will offer sustainable alternatives because both cladodes and fruits are nutritious and can be used as food.[6,7,28]

As it has adapted to dry or xeric environments, the Cactaceae family has the physiology and morphology to survive in regions where the annual precipitation is only 300 mm.[31] For example, cacti have a shallow, extensive, and highly fibrous root system, allowing more effective water uptake and absorption. Moreover, to absorb water after rains, it can produce tiny 'rain roots' that die in the absence of water.[4,28]

Another adaptation in the species of this family is the fleshy stem and cladodes. Both structures are succulent because they contain mucilaginous and aqueous tissues that can store large amounts of water in the aquifer parenchyma during unfavorable weather conditions. These structures are further protected by a thick waxy cuticle that reduces water loss, allowing them to survive drought periods. Additionally, the vertical position of the cladodes decreases heating by intercepting less midday light.[4,28,32]

However, the most important adaptations for survival in hot, dry environments are the presence of spines and a special carbon dioxide (CO_2) fixation pathway known as Crassulacean acid metabolism (CAM).[1] The evolution from leaves to spines reduces water loss through evaporation by decreasing the surface area.[4,32]

CAM is a photosynthetic pathway that captures CO_2 at night. Thus, stomata only open at night and remain closed during the day, reducing the loss of water for plant cooling.[1,28] This pathway, combined with fewer stomata, reduces the surface area through which water vapor can be lost, increasing the water conservation capacity. Under extreme water conditions, stomata can remain closed at night, preventing transpiration and CO_2 capture.[4,28]

Furthermore, this metabolic pattern allows for greater efficiency (four to five times greater) in converting water into dry matter

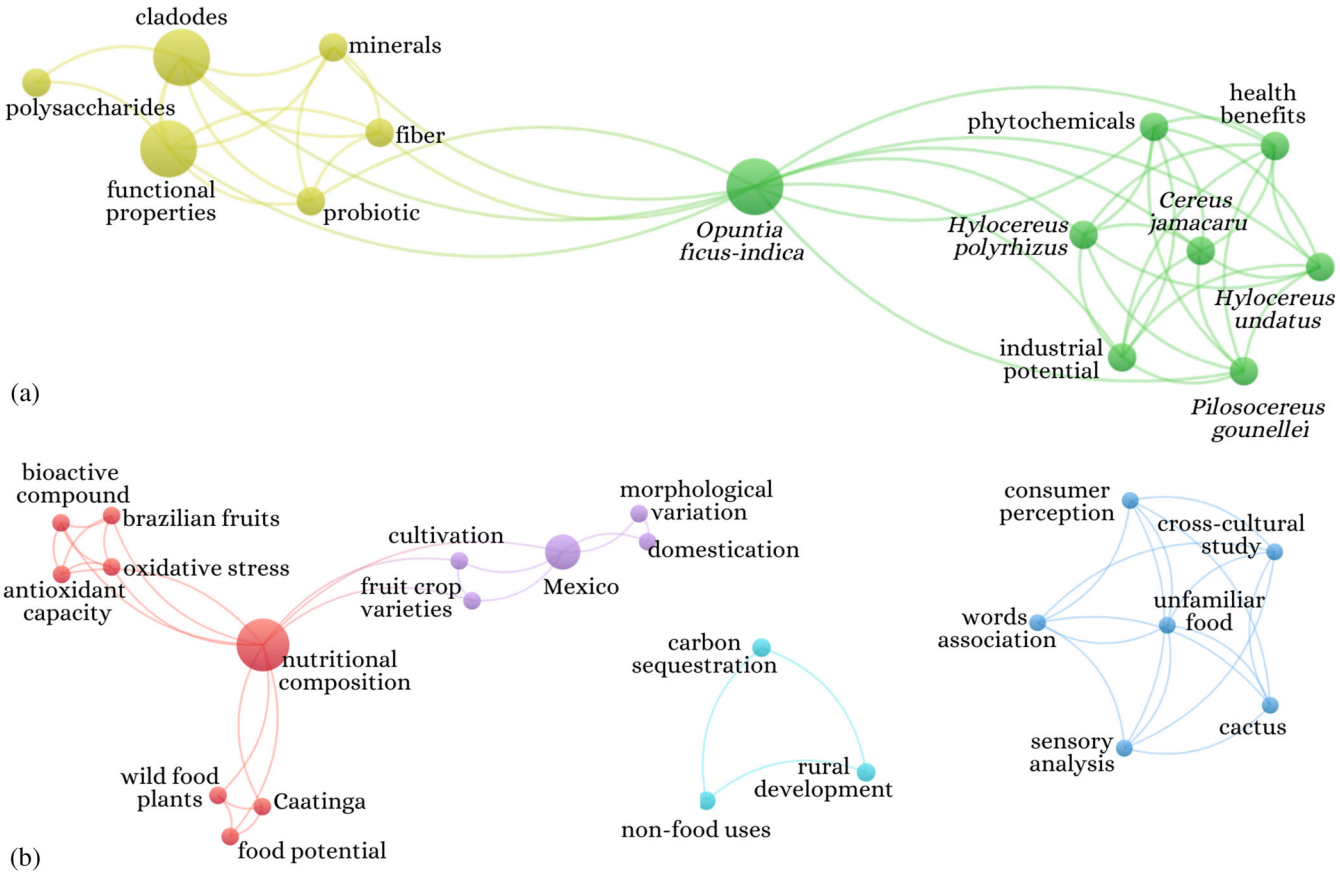

Figure 2. Keywords co-occurrence analysis with a threshold of two (a) and one (b) occurrences in articles on cacti in human food.

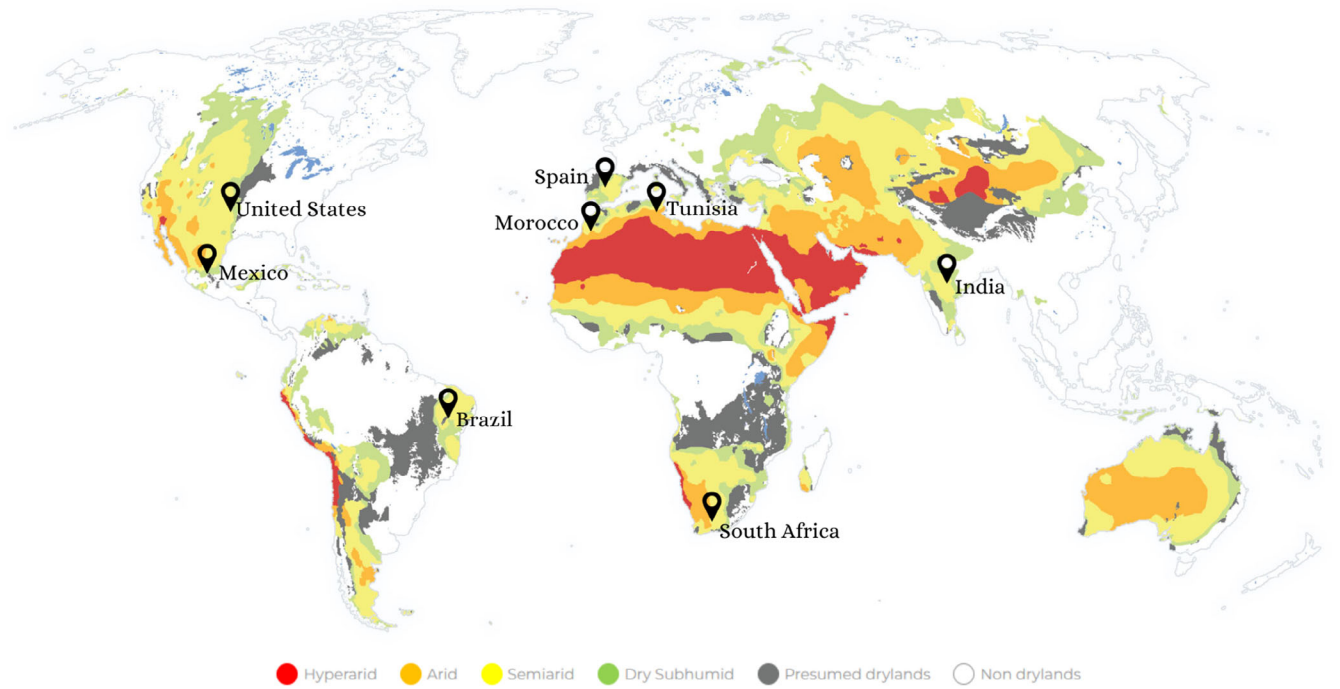

Figure 3. Dryland areas and origin location of publications on cacti in human food.

because such plants absorb CO_2 at night when solar radiation is zero and the air temperature and atmospheric water vapor pressure are low.[28,31,33] López[31] highlighted that *O. ficus-indica* can produce 10–20 tons of dry matter ha^{-1} yr^{-1} even without irrigation. These values are higher than those obtained with cereal and legume cultivation (e.g., corn, rice, wheat, sorghum, and soybeans) under normal conditions.

In addition to tolerating high temperatures (45 °C or more), cacti can survive in regions with low temperatures (5–10 °C), with *Opuntia* spp. species recorded in Alberta, Canada; Patagonia, Argentina, and mountains in Switzerland, demonstrating its tolerance to cold.[28,33] Thus, cacti species are important crops for producing carbohydrates and vitamins while absorbing water and mineral nutrients under adverse conditions, potentially contributing to the survival and health of humans and animals in dryland regions and beyond.[28,34]

Besides their value as food and fodder, these plants also contribute to the anticipated adverse impacts of global climate change. As carbon levels in the atmosphere increase, the global average temperature will increase, leading to changes in annual precipitation.[4] This way, wet regions will become wetter and dry areas will become even drier. One way to avoid this is to remove CO_2 from the atmosphere, and cactus species can contribute to the accumulation of organic carbon in the soil.[4,9]

Furthermore, cacti can act as a valuable genetic resource; their genes can be transferred to other crops to make them resistant or tolerant to high temperatures and drought.[2,4] Therefore, the importance of cacti will steadily grow over the years, with them already being considered as food of the future owing to their importance in food security.[35,36]

Dissemination and utilization through the years

Archeological studies indicate that the use of cacti as human food dates back thousands of years, probably before the history of agriculture itself.[4] The origin and history of *O. ficus-indica* are related to the ancient Mesoamerican civilizations, mainly the Aztec culture.[34,37] This species has a long tradition of use, being cultivated for the first time by indigenous populations that settled in the semiarid regions of Mesoamerica.[37] For these reasons, cacti have a tradition of use so profoundly rooted in Mexico, being one of the emblems of the country and presenting more than 200 culinary uses.[23,33,34] According to Russell and Felker,[33] the consumption of cladodes and cactus fruit was reported to be common when Bernal Diaz del Castillo and Hernando Cortes entered Tlaxcala in 1519, as described in *The Discovery and Conquest of Mexico*.

The introduction of cactus species to the Old World occurred after the discovery of America, when Christopher Columbus took specimens of exotic flora of the New World to Spain.[31,37] At the beginning of the 16th century, Spanish conquerors introduced cacti to the unproductive soils of the Iberian Peninsula to cultivate cacti to serve as food for the cochineal insect (*Dactylopius coccus* Costa) used to produce dye. This idea was unsuccessful, and the cacti spread like wild plants, ultimately being used as natural fences, cattle feed, and human food.[9,28]

When the last Moors were expelled from the Iberian Peninsula in 1610, cacti reached North Africa, where they became naturalized.[33] In the 17th century, cacti were also introduced by the British in India to produce dyes, as attempted by the Spaniards. Once again, the plan failed due to pests and floods occurring in the country.[25] Thus, by the end of the 18th century, cacti had spread across the Mediterranean basin, Africa, and Asia,[28,33] being found in several countries (e.g., Greece, Italy, South Africa, Madagascar, Saudi Arabia, India, and Australia).[31]

In Mexico, modern commercial plantations only started in the middle of the 20th century.[38] In fact, for some years, the prices of *O. ficus-indica* fruit in the Mexican market were even lower than those of fruits such as apples, oranges, and peaches.[38] These fruits were being sold as dessert fruit in markets located in American states with Latin American and Mediterranean populations, such as California and Washington DC.[33]

At the end of the 20th century, commercial plantations of *O. ficus-indica* already existed in Algeria, Argentina, Brazil, Chile, Mexico, South Africa, and other semiarid regions of Latin America and Africa, with Brazil being the country with the largest plantations (about 300 000 ha).[33] Even so, their consumption remained limited to local ethnic markets with little export.

Because these fruits are highly attractive to consumers owing to their quality and variety of colors,[28] their cultivation areas have expanded dramatically over the years. According to de Araújo *et al.*,[7] some cactus species are already commercially exploited, mainly *Opuntia* spp. Mexico is the largest producer in the world (45% of production),[9] producing large amounts of fruit and overcoming market demand.[25]

The fruit, called *nopal* or *tunas*, is commercialized in markets worldwide and is consumed as an exotic food,[4,23] especially those from South Africa, which are very popular in European markets.[39] They are used fresh, turned into jams and other products, or dried as raisins for consumption or to prepare *mole*, a traditional Mexican sauce.[40]

Young cladodes (3–4 weeks), known as *nopalitos*, are usually consumed as fresh green vegetables in Mexico and some United States regions with Mexican influence (e.g., Texas).[4,31,33,34] This part of the plant is considered a key ingredient in several dishes from Mexico and is consumed cooked, baked, pickled, or raw in drinks (*batidos*).[8,31]

Other areas producing *O. ficus-indica* fruit are Morocco (150 000 ha), Tunisia (25 000 ha), Italy, and South Africa (3000 ha each); beyond southern Spain, Angola, and India, Mexico produces almost half of the world's fruit.[22,28,39] Its production is intended for fruit and cladode consumption, dietary supplements, and nutraceuticals.[27]

Despite their importance, these species have been ignored as human food by the scientific community for many years and have only been mentioned as fodder. This scenario only changed in the early 1980s, when studies on the benefits of *O. ficus-indica* were published.[37] This explains why the first article related to the scope of this scientific prospect dates back to 1987 (Fig. 4) in the United States, with a review proposed by Russell and Felker[33] on the importance of using *Opuntia* spp. in animal and human food.

Seven years later, another article was published in Mexico, another the following year, and again in 2005, after 10 years without any study on the subject. From 2011 onwards, there was a more significant number of publications. Between that year and 2018, twice the number of articles were found (eight), of which five were from Brazil. The growing number of studies published on this topic, particularly in recent years, reflects the expansion of academic interest in cacti as human food.

Brazil is considered the third-largest center of diversity and importance for the Cactaceae family in the world, after Mexico and the United States,[41] and is the country with the largest cultivated area of *O. ficus-indica* at approximately 500 000 ha.[28] However, according to Cardoso *et al.*,[5] cacti are mainly used for animal feed and are still not present in the Brazilian diet, especially in urban areas.

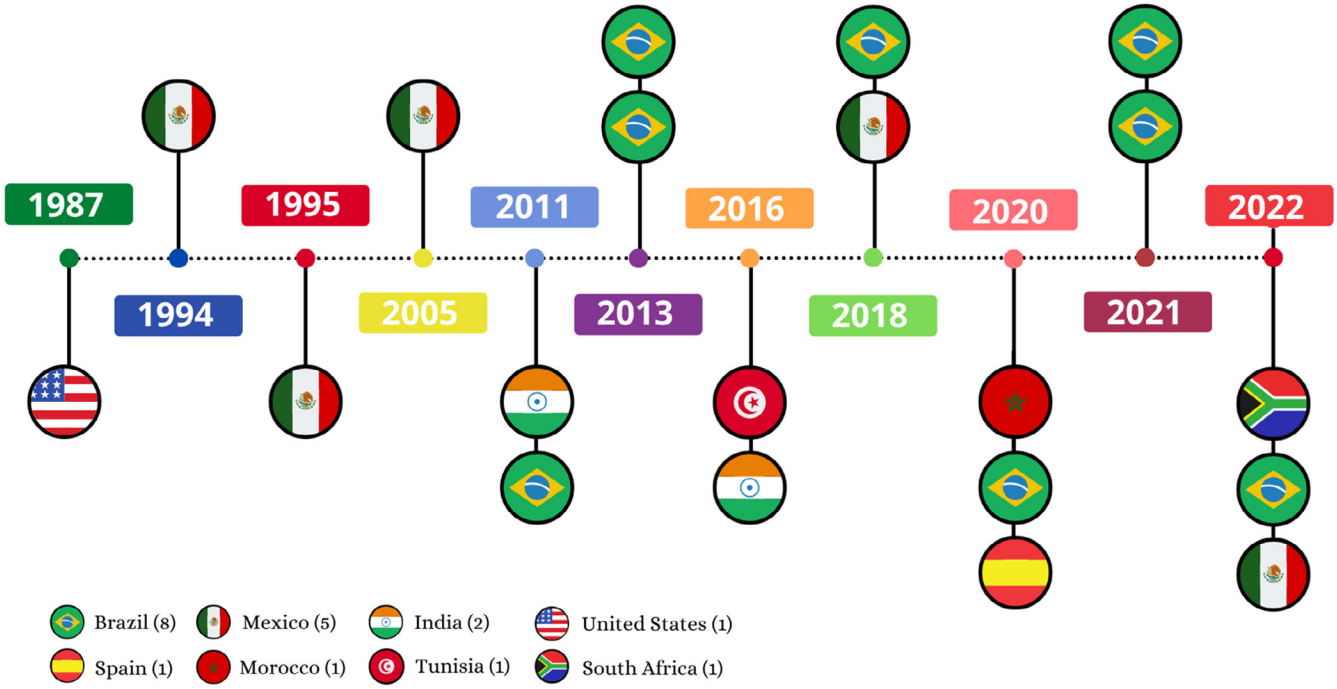

Figure 4. Annual distribution by country of published studies on cacti in human food.

In a study on a local population in north-eastern Brazil, Cruz *et al.*[42] observed that the lack of interest in testing these foods was related to the lack of knowledge that these plants could be used for human consumption. Therefore, to encourage the use and consumption of cacti in Brazil and around the world, Cruz *et al.*[42] highlighted the importance of teaching people that these plants can be used for human food, dispelling the myths surrounding their consumption, and promoting the diversification of uses based on new products and forms of preparation.

Cactus health benefits

Studies have highlighted cacti for their consumption and beneficial nutritional, bioactive, and health properties in humans (Table 1).[44,46] Furthermore, there is a growing global demand for healthy food products and nutraceuticals to improve health and prevent diseases.[34,37,46] Owing to their composition, cacti can be considered excellent candidates for dietary and food supplements and functional ingredients in nutraceuticals.[44]

Although variations in composition are related to edaphoclimatic conditions and the degree of maturation, cactus cladodes contain high moisture (910–950.5 g kg^{-1} fresh weight) and high water activity (0.97). Despite the low lipid content (10.2–30.5 g kg^{-1} dry weight), they are a source of omega and essential fatty acids, mainly eicosadienoic, oleic, palmitic, and γ-linolenic acids.[34] They are rich in carbohydrates (e.g., galacturonic acid, arabinose, and glucose) and proteins.[7,31] These proteins have a high biological value (72.6%) compared to chicken eggs, with 17 different amino acids (e.g., arginine, proline, taurine, serine, aspartic acid, and glutamic acid) reported to be present.[4,7,28,31,34]

Fiber and ash content have also been reported (mainly calcium, with higher levels than other vegetables included in the diet, in addition to magnesium and potassium).[4,7,31] The mineral content is beneficial in preventing various chronic diseases and in combating nutritional deficiency-related diseases.[34]

The ingestion of cladodes has hypoglycemic effects, decreasing glucose levels (30–40 mg dL^{-1} in 3 h) and stimulating pancreatic activity, which can reduce blood glucose in obese patients by up to 22%.[2,31,34]

The cactus fruit tends to have low acidity, with a pH close to neutral (6.5–7.5), requiring a more severe thermal treatment or pH reduction (< 4.5) for better preservation using classical methods.[31,38] Furthermore, they have a low percentage of titratable acidity (0.015–0.049%), being rich in organic acids (e.g., ascorbic, citric, lactic, and malic acids).[7,38]

The chemical composition of cactus fruit is similar to that of other conventional fruits (e.g., apple, mango, melon, and peach), with low protein values.[28,31] Prominent amino acids are proline, glutamine, serine, and taurine.[4,7] The fruits contain reasonable amounts of carbohydrates (arabinose, galactose, glucose, mannose, and xylose); minerals (mainly calcium, magnesium, phosphorus, and potassium); and C, E, and B-complex vitamins.[4,7,28,31,38]

It is important to note that the skin of cactus fruit is similar in composition to pulp, but with higher ash, fibrous and non-fibrous carbohydrates (mainly pectin), and low sugar content.[7] Moreover, the seeds contain lipids.[7] Therefore, as López[31] points out, the cactus fruit is quite valuable.

The fruit also contains several bioactive compounds produced in response to biotic and abiotic factors to which the plant is exposed. Among them are polyphenols (e.g., flavonoids, isorhamnetin, kaempferol, phenolic acids, quercetin, tannins, and derivatives) and pigments (e.g., chlorophyll, carotenoids, and betalains).[1,4,5,7,8,38,44] These fruits also have a high ascorbic acid content, which, along with the other bioactive compounds mentioned earlier, has antioxidant capacity.[4,8,44,45]

According to da Costa Nunes *et al.*,[35] identifying new natural sources with antioxidant compounds is one of the main focuses of current research. Foods with antioxidant properties, such as cactus fruit, help reduce oxidative stress in the body and have

Table 1. Beneficial nutritional, biological, and health properties of cacti according to the studies analyzed

Species	Fraction	Nutritional properties	Bioactive properties	Health properties	Reference
Opuntia spp.	Cladodes	—	Total phenolic (24.65–86.36 g GAE kg^{-1})[a], phenol (91.20–853.8 g GAE kg^{-1})[a], total flavonoid (16.4–45.1 g QE kg^{-1})[a], gallic acid (91.23–853.78 g kg^{-1})[a], and quercetin (17.10–39.42 g kg^{-1})[a] content.	Antiallergic, antiatherogenic, anti-inflammatory, antimicrobial, and antioxidant activities.	Dávila-Aviña *et al.*[8]
	Dehydrated cladode	Protein (70.25 g kg^{-1})[a], calcium (28.35 g kg^{-1})[a], and fiber (180.73 g kg^{-1})[a] content.	—	-	Ranjan *et al.*[4]
	Fruit pulp	Total sugars (120.00–170.00 g kg^{-1})[b] and calcium (3.16 g kg^{-1})[a] content.	Ascorbic acid content (0.05–0.35 g kg^{-1})[b].	Anti-hyperglycemic and hypocholesterolemia effects.	Ranjan *et al.* and Pimienta-Barrios *et al.*[4,38]
	Fruit seeds	Lipid content (60.4–200 g kg^{-1})[a], mostly linoleic (60–77%)[a], oleic (11–23%)[a], and palmitic (9–16%)[a].	—	—	Pimienta-Barrios *et al.*[38]
Opuntia ficus-indica	Cladodes	Protein (3.20–140.80 g kg^{-1})[a] and total fiber (110.00–230.33 g kg^{-1})[a] content, calcium (17.52–34.40 g kg^{-1})[a] and magnesium (8.80–11.20 g kg^{-1})[a] bioavailable amounts, 17 different amino acids, being nine essential, and important amounts of omega and essential fatty acids (mainly omega 6).	Total phenolic content (3.62–18.50 g GAE kg^{-1})[a].	Prevent diabetes and obesity, hypoglycemic, hypercholesterolemia, antioxidant, anticarcinogenic, anti-inflammatory, and antidyslipidemic effects.	Dávila-Aviña *et al.*, López *et al.*, Hernández-Becerra *et al.*[8,31,34]
	Mucilage	Arabinose (42%)[a], galactose (22%)[a], and xylose (22%)[a] content.	—	—	López *et al.*[31]
	Fruit	Total carbohydrate (0.58–0.93 g kg^{-1})[a], vitamin C (0.72 g kg^{-1})[b], calcium (14.16 g L^{-1})[a], magnesium (2.76 g L^{-1})[a], potassium (5.59 g kg^{-1})[a], and folic acid (0.71 g kg^{-1}) content.	Phenolic compounds (0.90 g GAE L^{-1}), phenolic acids (8.05–23.13 g kg^{-1}), total polyphenols (3.42 g kg^{-1}), β-carotene (0.48 g kg^{-1}), and betalain (2.40 g kg^{-1})[b] content.	Antiulcerogenic, anti-inflammatory, antidiabetic, anticancer, antioxidant, neuroprotective, hepatoprotective, antiproliferative, anti-hyperglycemic, and hypocholesterolemic activities.	Ranjan *et al.*, Cardoso *et al.*, de Araújo *et al.*, Andreu-Coll *et al.*[4,5,7,9]
	Fruit pulp	Carbohydrate (120.89 g kg^{-1})[b], potassium (1.69 g kg^{-1})[b], and phosphorus (0.21 g kg^{-1})[b] content.	Ascorbic acid content (0.19 g kg^{-1})[b].	—	López *et al.*[31]
Opuntia dillenii	Fruit	—	—	Inhibit stomach ulcer and anti-inflammatory, anti-hyperglycemic, hypocholesterolemic, neuroprotective, and analgesic effects.	Ranjan *et al.*[4]
Opuntia elata	Fruit	—	Phenolic compounds (mainly hydroxybenzoic acids and flavonols).	Antioxidant activity.	Rockett *et al.*[43]
Opuntia aequatorialis	Fruit	—	Ascorbic acid (0.24 g L^{-1})[b].	Antioxidant activity.	El Kharrassi *et al.*[44]
	Seed oil	—	Phytosterol (1.21 g kg^{-1})[b] and total tocopherols (0.67 g kg^{-1})[b] levels.	—	El Kharrassi *et al.*[44]

Table 1. Continued

Species	Fraction	Nutritional properties	Bioactive properties	Health properties	Reference
Opuntia leucotricha	Fruit	—	Ascorbic acid (0.55 g L^{-1})[b].	Antioxidant activity.	El Kharrassi *et al.*[44]
	Seed oil	—	Phytosterol (0.89 g kg^{-1})[b] and tocopherols (0.71 g kg^{-1})[b] levels.	—	El Kharrassi *et al.*[44]
Hylocereus undatus	Pulp	Potassium (3.99 g L^{-1})[a] and magnesium (0.26 g L^{-1})[a] content.	Total phenolics (3.58 g kg^{-1}) and flavonoids (4.77 g QE kg^{-1}) content.	—	de Araújo *et al.*[7]
Hylocereus polyrhizus	Pulp	—	Total polyphenols content (1.18 g GAE kg^{-1}).		de Araújo *et al.*[7]
Cereus jamacaru	Fruit	Fat content (7.5–130.60 g kg^{-1})[a].	Total phenolic content (6.95 g GAE kg^{-1}).	Antioxidant activity and relief of allergies and indigestion.	Cardoso *et al.*[5]
	Pulp with seed	—	—	Anti-cancer and anti-tumor potential.	de Araújo *et al.*[7]
	Pulp	—	Total carotenoids content (0.07 g kg^{-1}).	—	de Araújo *et al.*[7]
Cereus peruvianus	Cladodes	—	Phenolic compounds content (14.91 g GAE kg^{-1})[b].	Treatment of kidney disorders and wounds, gastroprotective, antioxidant, antifungal, hemagglutinating, and anticancer activities.	da Costa Nunes *et al.*[35]
Pilosocereus pachycladus	Fruit	—	Betalains (4.18 g kg^{-1}) content, mainly betacyanins (3.35 g kg^{-1}).	Antioxidant activity.	Cardoso *et al.*[5]
Pilosocereus gounellei	Fruit	Protein content (50.50 g kg^{-1})[a].	Phenolic compounds (0.12 g GAE kg^{-1}), total flavone/flavonol (2.41 g QE kg^{-1}), and betalains (approximately 0.98–1.10 g kg^{-1}). content.	Antioxidant activity.	Cardoso *et al.*, do Nascimento *et al.*[5,45]
	Pulp with seed	Total caloric values (1.02 cal kg^{-1}).	Flavonols content (0.22 g of quercetins kg^{-1})[b].	—	do Nascimento *et al.*[45]
Trichocereus pachanoi	Cladodes	—	—	Urinary tract infection treatment, diuretic.	Rai *et al.*[2]
Lophophora williamsii	Fruits and stems	—	—	Neurasthenia treatment.	Rai *et al.*[2]
Carnegiea gigantea	Stem	—	—	Enhance milk flow in the mother.	Rai *et al.*[2]
Pachycereus schottii	Whole plant and pulverized stems	—	—	Painkiller, and in toothache, rheumatism, asthma, and colds treatments (antibacterial effect).	Rai *et al.*[2]
Melocactus zehntneri	Fruit	—	—	Flu, cough, and throat inflammation treatments.	Cardoso *et al.*[5]

Abbreviations: GAE, gallic acid equivalent; QE, quercetins equivalent.
[a] Dry weight.
[b] Fresh weight.

anti-inflammatory; anti-atherogenic; anti-hyperglycemic; hypocholesterolemic; antidiabetic; anti-ulcerogenic; anticancer; and cardioprotective, neuroprotective, and hepatoprotective effects.[4,8,9,25,35]

The seed lipids include some saturated fatty acids (e.g., palmitic and stearic acids), but the emphasis is on unsaturated fatty acids.[7,44] The presence of high linoleic and oleic acid,[2,7] together with the absence of linolenic acid, indicates that the seeds of cactus fruit can be used to extract oil for human consumption.[38]

The presence of tocopherol, mainly γ-tocopherol, has been found in cactus seed oil.[44] In addition to health benefits due to its antioxidant action, this substance helps prevent polyunsaturated fatty acid (PUFA) activity and increases shelf life by reducing oil oxidation and rancidity. Therefore, the PUFA and tocopherol content in cactus seed oil reiterate its importance as a new ingredient in food products, supplements, and nutraceuticals.[44]

Cactus consumption and food production

The uses and ethnobotanical applications of cacti vary in different countries worldwide.[1] Cactus fruit is generally consumed fresh,[28,32] boiled, or grilled,[28] or used in the production of fruit pulp, juices, concentrated juices, nectars, jellies, jams, marmalades, cakes, ice cream, natural liquid sweeteners, mock-gherkins, vinegar, and alcoholic beverages like liqueurs and wine.[1,5,28,31,32,43] They can also be preserved in sugar syrup, canned, frozen, or sun-dried to make raisins.[1,28,46]

The fruit is the most consumed part of cactus, but the roots, stems, flowers, and especially cladodes are edible, serving as a food source in different countries.[1,25] Young cladodes can be consumed fresh as vegetables or in salad dishes.[1,25,28] They can be baked; boiled; roasted; cooked with risotto, rice, beans, and meat; or used in stews and soups.[1,32] They are used to prepare sweets, puddings, cakes, coconut candy, and cookies.[32] In addition, they can be canned or dehydrated; the flour is used in cakes and cookies or used in the preparation of couscous.[28,32,45]

Cactus fruit, mainly *O. ficus-indica*, has some established products in Mexican food culture. Products derived from the juice squeezed from these fruits include *miel de tuna*, a product similar to molasses; *queso de tuna*, a concentrated paste similar to pulled taffy; *colonche*, a low-alcohol fermented drink; *melcocha*, a kind of marmalade; and *tunas pasas*, which is the dried fruit.[31,33]

These plants are also important for the food industry to obtain food additives.[28] Cladodes are widely used to obtain industrial hydrocolloids. Because of the significant presence of mucilage, a complex carbohydrate with gelling properties due to its high capacity to absorb water, cacti can be used as natural thickeners.[25,44]

Cactus mucilage can also be used as an edible coating for fruit to increase shelf life, and in stabilizing emulsions and foams, syneresis inhibitors, fat mimics in ice cream and sorbets, gelatin substitutes, spherification, and crystallization controllers.[25,34,39] Enzymes from unripe *O. ficus-indica* fruit can be used as a source of milk clotting enzymes for the dairy industry as they provide faster coagulation than other rennets and have structural properties and a pleasant smell.[2,4]

Another industrial use of great importance is dye production.[9] In addition to using cladodes to cultivate the cochineal insect,[4] the fruit of *Opuntia* spp. is also a source of yellow and red food dyes.[9] These fruits have better technical and sensory properties than those of red beets due to the absence of geosmin, an earth-like flavor compound.[4,5]

De Araújo *et al.*[7] reported the use of *Pilosocereus gounellei* in the production of refined and wholemeal flour with high fiber and mineral content and good sensory characteristics, offering nutritional and bioactive benefits. There are also reports of other cacti being used in flour production (e.g., *P. pachycladus* and *O. ficus-indica*), which, owing to their fiber content, help in weight and cholesterol reduction.[28,31,45]

Cactus fruit characteristics enable processing (e.g., jam, jelly, juice, and marmalade). However, the pulp viscosity is very high for juice production, making it necessary to dilute and adjust the sugar content, whereas it is very low for jelly production, requiring concentration.[31] Moreover, the balance between sugars and acidity (soluble solids/titrable acidity ratio) indicates high palatability, allowing fresh consumption.[45]

Unfortunately, this balance makes the fruit susceptible to the growth of microorganisms, requiring treatments such as cold storage and hot water treatment to improve the shelf life and reduce waste. Another option widely used by the food industry to improve the shelf life of products with these characteristics is lactic acid fermentation, which offers an option for the development of new food products using cacti.

Although the Cactaceae family is not a major part of the daily lives of people in many countries, it is anticipated to have an essential role in the future. Scientific dissemination combined with technological innovations will allow for greater use of these multifunctional species, promoting their use in food systems, biotechnology, pharmaceuticals, and medicine.

CONCLUSION AND FUTURE PERSPECTIVES

Cacti are used by rural populations in some countries (e.g., Brazil) and are commercially exploited in other countries, such as Mexico. They are used in many food preparations, such as juices, snacks, and some traditional dishes. These plants are impressive because of their wide range of uses and forms of consumption. Their excellent flavor and color can attract consumers, and their nutritional, bioactive, and technical properties may be of interest to the food industry.

Thus, these species have the potential to become part of the regular human diet, but they are still consumed very little around the globe. Only a few studies have been conducted on cacti as human food. Nevertheless, the increase in the number and frequency of publications in the last decade indicates a growing interest in this topic. Because the world is projected to face unprecedented climate and health challenges, modifications in people's behaviors and habits are expected. Thus, the need for sustainable and multifunctional crops is increasing.

Known for their minimal water requirement, species from the Cactaceae family will become more popular in markets, cosmetics, pharmaceuticals, and food industries. This is explained by their high adaptability and excellent resistance to high and low temperatures, as well as their ability to retain water in dry environments and still produce fruits, flowers, and cladodes with reasonable amounts of vitamins, minerals, and bioactive compounds.

Compounds of interest can be applied in nutraceutical development and as food additives. Cactus mucilage can be used in edible film production, and the high content of healthy unsaturated and saturated fatty acids extracted from seeds can be micro- and nano-encapsulated. The high fiber content can be used in nutraceuticals aimed at cholesterol reduction, and the

flour produced by these species can improve the physicochemical properties of bakery products.

Moreover, owing to the physicochemical characteristics of its fruits, technological processes can be applied to minimize losses during harvesting and diversify its consumption. They can be used as the main ingredients in jams, jellies, candies, syrups, and beverages. Other processes that can be applied to extend the shelf life include drying and freezing, in addition to the production of canned and fermented food products.

Cacti have promising properties to position it as a food source for the future. More studies on the potential applications of these species and the dissemination of what is already known about them to society are warranted. There is a gap in information regarding anti-nutritional factors, inhibitors, and the bioavailability of compounds present in these foods, as well as information on species outside the genera *Opuntia* and *Hylocereus*.

Studies are also needed on the acceptance of the consumption of these species and their products outside their traditional locations, as social and cultural issues related to food seem to be some of the main obstacles to the inclusion of unconventional food plants in the regular diet globally. Therefore, this article may support future research on the subject, as it is the first study to map published studies on this topic.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

Data sharing not applicable to this article as no datasets were generated or analysed during the current study.

ETHICAL STATEMENT

Ethics approval was not required for this research.

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ARTICLES FOR FACULTY MEMBERS

FUNCTIONAL HYDROCOLLOIDS FROM THE CACTACEA FAMILY FOR FOOD AND PHARMACEUTICAL APPLICATIONS

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Review

Underutilized plants of the Cactaceae family: Nutritional aspects and technological applications

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ABSTRACT

This review examines the nutritional and functional aspects of some representatives of the Cactaceae family, as well as its technological potential in the most diverse industrial fields. The studied species are good sources of nutrients and phytochemicals of biological interest, such as phenolic compounds, carotenoids, betalains, phytosterols, tocopherols, etc. They also have shown great potential in preventing some diseases, including diabetes, obesity, cancer, and others. As to technological applications, the Cactaceae family can be explored in the production of food (e.g., cakes, yogurts, bread, ice cream, and juices), as natural dyes, sources of pectins, water treatment and in animal feed. In addition, they have great potential for many technological domains, including food chemistry, pharmacy, biotechnology, and many others.

1. Introduction

Currently, there is a worldwide concern not only with food security, but also with nutritional security (Farooq et al., 2019). Within this context, the exploitation of underutilized plant species becomes a way to guarantee food supply in adequate quantity and quality, once plant-based foods are widely recognized as sources of nutrients and bioactive compounds, essential to health maintenance (Araújo et al., 2020, 2019; Farias et al., 2020).

Cactus are part of a group of xerophilic plants widely distributed in arid and semi-arid regions (Abidi et al., 2009). The Cactaceae family has a wide variety of shapes and sizes, succulent stem and edible fruits. Besides having great socioeconomic importance for rural populations from the Caatinga biome in Brazil, these plants are used as ornamental crops, in human food and as fodder for animals, thus guaranteeing food/nutritional security, economic development, and the livelihood of the local population (Ramírez-Rodríguez et al., 2020; Magalhães et al., 2019; Abidi et al., 2009; Fidelis et al., 2015).

Some species of the Cactaceae family are already commercially exploited (e.g. *Opuntia ficus-indica*, *Hylocereus undatus* and *Hylocereus polyrhizus*), while others are still underutilized (*Pilosocereus gounellei* and *Cereus jamacaru*). Studies have shown that these species are good sources of nutrients and bioactive compounds (Salehi et al., 2019; Muhammad et al., 2014; do Nascimento et al., 2012; Magalhães et al., 2019). In addition, recent research indicates that the fruits and other botanical parts of these representatives have great potential as natural dyes, synthesis of nano particles, as agents to control diseases such as diabetes and obesity, and to develop edible films. (Guesmi et al., 2013; Alvarez-Bayona et al., 2019; Del-Valle et al., 2005; Song et al., 2016).

Therefore, considering the socioeconomic, nutritional and technological relevance of the Cactaceae family, this review addressed its nutritional and functional aspects, as well as its potential for applications in the most diverse technological domains, in order to encourage the cultivation and commercial exploitation of some species, namely *Opuntia ficus-indica*, *Hylocereus undatus*, *Hylocereus polyrhizus*, *Pilosocereus gounellei* and *Cereus jamacaru*.

2. Botanical characteristics

O. ficus-indica, also known as *prickly pear, Indian fig, smooth prickly pear* or *nopal cactus,* is found in arid and semi-arid climates, mainly due to its good ability to adapt to different environmental and climatic conditions (Bakar et al., 2020; Mena et al., 2018; Ramírez-Rodríguez et al., 2020; Salehi et al., 2019). This species has a wide genetic variability and flattened fleshy cladodes, which are systems of oval and flat leafless shoots, with thick bark and gloquids. Fruits are edible, have

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Table 1
Nutritional composition of cactus species.

Fruit	Fraction	Composition	Content	Units	Reference
Opuntia ficus-indica	Whole fruit	Moisture	7.65	% [a]	
		Ash	9.00		
		Protein	0.86		
		Total carbohydrate	88.85	g 100 g^{-1} [a]	Salehi et al. (2019)
		Arabinose	10.83	g 100 g^{-1} [a]	
		Galactose	2.13[a]		
		Xylose	4.26[a]		
		Mannose	2.13[a]		
		Glucose	69.30[a]		
		Calcium	14159.90	mg L^{-1} [a]	
		Magnesium	2759.63		
		Manganese	8.51		
		Potassium	2161.02		
		Phosphorus	321.98		
		Copper	177.53		
		Aluminum	4004.94		
		Nickel	6.86		
		Sodium	2807.92		
		Iron	94.32	µg g^{-1a}	
		Selenium	0.38		
	Pulp	Total acidity	0.06[b]	g citric acid 100^{-1}	García-Cayuela et al. (2019)
		pH	5.90	–	
		Total soluble solids	13.30[b]	°Brix	
	Seed oils	Lauric acid (C12:0)	0.60	g 100 g^{-1}	Brahmi et al. (2020)
		Tridecylic acid (C13:0)	0.01		
		Myristic acid (C14:0)	0.13		
		Pentadecylic acid (C15:0)	0.02		
		Palmitoleic acid (C16:1n-7)	1.77		
		Palmitelaidic acid (C16:1n-9)	0.04		
		Palmitic acid (C16:0)	20.34		
		Margaric acid (C17:0)	0.04		
		Oleic acid (C18:1n-9)	41.22		
		Vaccenic acid (C18:1n-7)	1.77		
		Stearic acid (C18:0)	1.66		
		Linoleic acid (C18:2n-6)	19.94		
		α-Linolenic acid (C18:3n-3)	0.43		
		Arachidic acid (C20:0)	0.15		
		Gondoic acid (C20:1n-9)	0.02		
		Paullinic acid (C20:1n-7)	0.19		
		Eicosadienoic acid (C20:2n-6)	0.03		
		Heneicosylic acid (C21:0)	0.01		
		Behenic acid (C22:0)	0.03		
		Erucic acid (C22:1n-9)	0.01		
		Tricosylic acid (C23:0)	0.01		
		Lignoceric acid (C24:0)	0.03		
		Pentacosylic acid (C25:0)	0.01		
		Cerotic acid (C26:0)	0.02		
		Chlorophylls	16.66	mg kg^{-1}	
		Total Carotenoids	1480.00		
		Total phenolics	25.15	mg GAE 100 g^{-1}	
		Total flavonoids	16.08	mg QE 100 g^{-1}	
	Whole fruit	Total betacyanins	1830.00	µg IE g^{-1b}	
		Total betaxanthins	760.00	µg BE g^{-1b}	
		Total Betalains	2580.00	µg g^{-1b}	
		Vitamin C	71.94	mg AE 100 g^{-1b}	Cano et al. (2017)
		Vitamin E	63.20	µg g^{-1a}	Bakar et al. (2020)
		Vitamin B1	8.60		
		Vitamin B2	6.72		
		Vitamin B3	307.20		
		Vitamin B6	21.20		
		Vitamin B9	708.80		
		Vitamin B12	4.00		
		DPPH	126.49[b]	µmol TE 100 g^{-1}	
		ORAC	38.66[b]		
Hylocereus undatus	Pulp	Moisture	85.39	%	Esgote Junior (2017)
		Ashes	0.47	g 100 g^{-1}	
		Proteins	1.31		
	Juice	Proline	415.00	mg L^{-1}	Stintzing et al. (2003)
	Pulp	Lipids	0.47	%	Abreu et al. (2012)
	Juice	pH	5.32	–	
		Dietary fibre	2.14	%	
		Total acidity	3.30	g malic acid L^{-1}	
		Total soluble solids	9.40	°Brix	
	Pulp	Calorific value	55.99	Kcal	

(*continued on next page*)

Table 1 (*continued*)

Fruit	Fraction	Composition	Content	Units	Reference
		Total carbohydrates	12.95	g 100 g^{-1}	
	Juice	Glucose	46.60	g L^{-1}	
		Fructose	18.4		
		Potassium	3995.00	mg L^{-1}	
		Sodium	33.00		
		Calcium	30.60		
		Magnesium	265.50		
	Seed oil	Saturated fatty acid	17.99	g 100 g^{-1}	Lim et al. (2010)
		Monounsaturated fatty acid	25.20		
		Polyunsaturated fatty acid	56.81		
		Myristic acid (C14:0)	0.19		
		Palmitic acid (C16:0)	12.78		
		Stearic acid (C18:0)	4.67		
		Arachidic acid (C20:0)	0.35		
		Palmitoleic acid (C16:1)	0.64		
		Oleic acid (C18:1)	24.43		
		Erucic acid (C22:1)	0.13		
		Linoleic acid (C18:2)	55.63		
		Linolenic acid (C18:3)	1.18		
	Pulp	Total phenolics	358.42	mg GAE 100 g^{-1}	
		Total flavonoids	4.77	mg QE g^{-1}	
		Ascorbic acid	28.40	mg 100 g^{-1}	
	Juice	Citric acid	132.50	mg L^{-1}	
		D-Isocitric acid	19.20		
		L-Lactic acid	153.50		
		L-Malic acid	7.20		
	Pulp	DPPH IC_{50}	0.84		
		FRAP	59.27	μmol Fe_{II} g^{-1}	
Hylocereus polyrhizus	Pulp	Moisture	89.46	%	Muhammad et al. (2014)
		Protein	1.06	%	Abreu et al. (2012)
		Ash	0.36	%	
		Lipids	0.36	%	
		Dietary fibre	3.26	%	
		Calorific value	44.87	Kcal	
		Total acidity	0.24	mg CAE 100 g^{-1}	
		pH	4.88	–	
		Soluble solids	11.00	°Brix	
		Total carbohydrates	7.93	%	
	Peel	Glucose	55.4	g L^{-1}	Stintzing et al. (2003)
		Fructose	19.2	g L^{-1}	
		Phosphorus	1.02	g kg^{-1}	
		Potassium	14.31		
		Calcium	0.41		
		Magnesium	1.47		
		Sulfur	0.14		
		Sodium	0.22		
		Copper	2.75		
		Iron	9.75		
		Zinc	8.00		
		Manganese	6.00		
	Seed oil	Saturated fatty acid	22.78	g 100 g^{-1}	Lim et al. (2010)
		Monounsaturated fatty acid	27.92		
		Polyunsaturated fatty acid	49.30		
		Myristic acid (C14:0)	0.19		
		Palmitic acid (C16:0)	16.53		
		Stearic acid (C18:0)	5.57		
		Arachidic acid (C20:0)	0.49		
		Palmitoleic acid (C16:1)	0.94		
		Oleic acid (C18:1)	26.80		
		Erucic acid (C22:1)	0.18		
		Linoleic acid (C18:2)	48.00		
		Linolenic acid (C18:3)	1.30		
	Pulp	Total polyphenols	118.57	GAE 100 g^{-1}	
		Total Betacyanin	42.84	mg 100 g^{-1a}	Yong et al. (2017)
	Juice	Citric acid	579.00	mg L^{-1}	
		D-isocitric acid	10.00		
		L-lactic acid	18.50		
		L-malic acid	4.80		
	Pulp	Ascorbic acid	20.69	mg 100 g^{-1}	
	Peel	DPPH IC_{50}	0.83	mg mL^{-1}	Luo et al. (2014)
Pilosocereus gounellei	Edible fraction	Moisture	92.5	g 100 g^{-1}	Nascimento et al. (2012)
		Ashs	1.1		
		Protein	0.4		
		Calorific value	27.5	kcal	
		pH	5.07	–	
		Total acidity	0.10	% citric acid	

(*continued on next page*)

Table 1 (*continued*)

Fruit	Fraction	Composition	Content	Units	Reference
		Carbohydrates	5.7	g 100 g^{-1}	
	Cladodes	Fuctose	5.26	mg 100 g^{-1a}	Bezerril (2017)
		Glucose	2.37		
		Xylose	1.14		
		Arabinose	0.89		
		Galactose	0.25		
		Sucrose	0.21		
		Total fiber	46.19		
		Insoluble fiber	31.36		
		Soluble fiber	14.83		
		Magnesium	3123.00		
		Calcium	2493.00		
		Sodium	412.00		
		Manganese	131.90		
		Phosphor	50.40		
		Zinc	7.78		
		Iron	2.10		
		Selenium	0.05		
		Aspartic acid	0.30	mg g^{-1a}	
		Glutamic acid	0.24		
		Serina	0.27		
	Cladodes	Glycine	0.27		
		Histidine	0.09		
		Arginine	0.54		
		Threonine	0.37		
		Alanine	0.25		
		Proline	0.28		
		Tyrosine	0.19		
		Valine	0.34		
		Methionine	0.05		
		Cystine	0.01		
		Isoleucine	0.23		
		Leucine	0.70		
		Phenylalanine	0.30		
		Lysine	0.23		
	Edible fraction	Total flavonoids	3.13	mg QE 100 g^{-1}	
	Cladodes	Total phenolics	258.21	mg GAE 100 g^{-1a}	
		Ascorbic acid	15.24	mg 100 g^{-1}	Sousa (2017)
		Carotenoids totals	422.55	µg 100 g^{-1}	
		Anthocyanins totals	2.60	mg 100 g^{-1}	
		DPPH	572.96	µmol TE 100 g^{-1a}	
		FRAP	1912.95		
Cereus jamacaru	Pulp with seed	Moisture	85.82	g 100 g^{-1}	Nascimento et al. (2011)
		Ashs	0.64		
		Protein	1.80		
		Lipids	1.98		
		Carbohydrates	9.76		
		Total Caloric Values	64.06	cal 100 g^{-1}	
		pH	4.40	–	
		Total soluble solids	10.30	°Brix	
		Total acidity	0.32	% citric acid	
		Neutral detergent fiber	457.10	g kg^{-1a}	Magalhães et al. (2019)
		Acid detergent fiber	258.50		
		Hemicellulose	198.70		
		Cellulose	252.80		
		Phosphorus	1.40		
		Potassium	9.40		
		Calcium	38.20		
		Magnesium	8.70		
		Sulfur	0.30		
		Boron	79.80	mg kg^{-1a}	
		Copper	2.30		
		Iron	28.90		
		Manganese	261.90		
		Zinc	39.40		
		Total flavonoids	0.67	mg QE g^{-1}	Santana (2016)
	Pulp	Total phenolic	29.15	mg GAE 100 g^{-1}	Santos et al. (2020)
		Ascorbic acid	12.31	mg 100 g^{-1}	Sousa (2017)
		Carotenoids totals	76.64	µg 100 g^{-1}	
		Anthocyanins totals	0.12	mg 100 g^{-1}	
		$ABTS^{+}$	9.71	µmol TE g^{-1}	
		DPPH	6.93	g DPPH g^{-1}	

[a] Values expressed on the basis of dry weight; [b] Values expressed on the basis of fresh weight. GAE: Gallic acid equivalents; IE: Indicaxanthin equivalents;CAE: Citric acid equivalent; BE: Betanin equivalents; AE: Ascorbic acid equivalents; TE: Trolox equivalentes; QE: Quercetin equivalets.

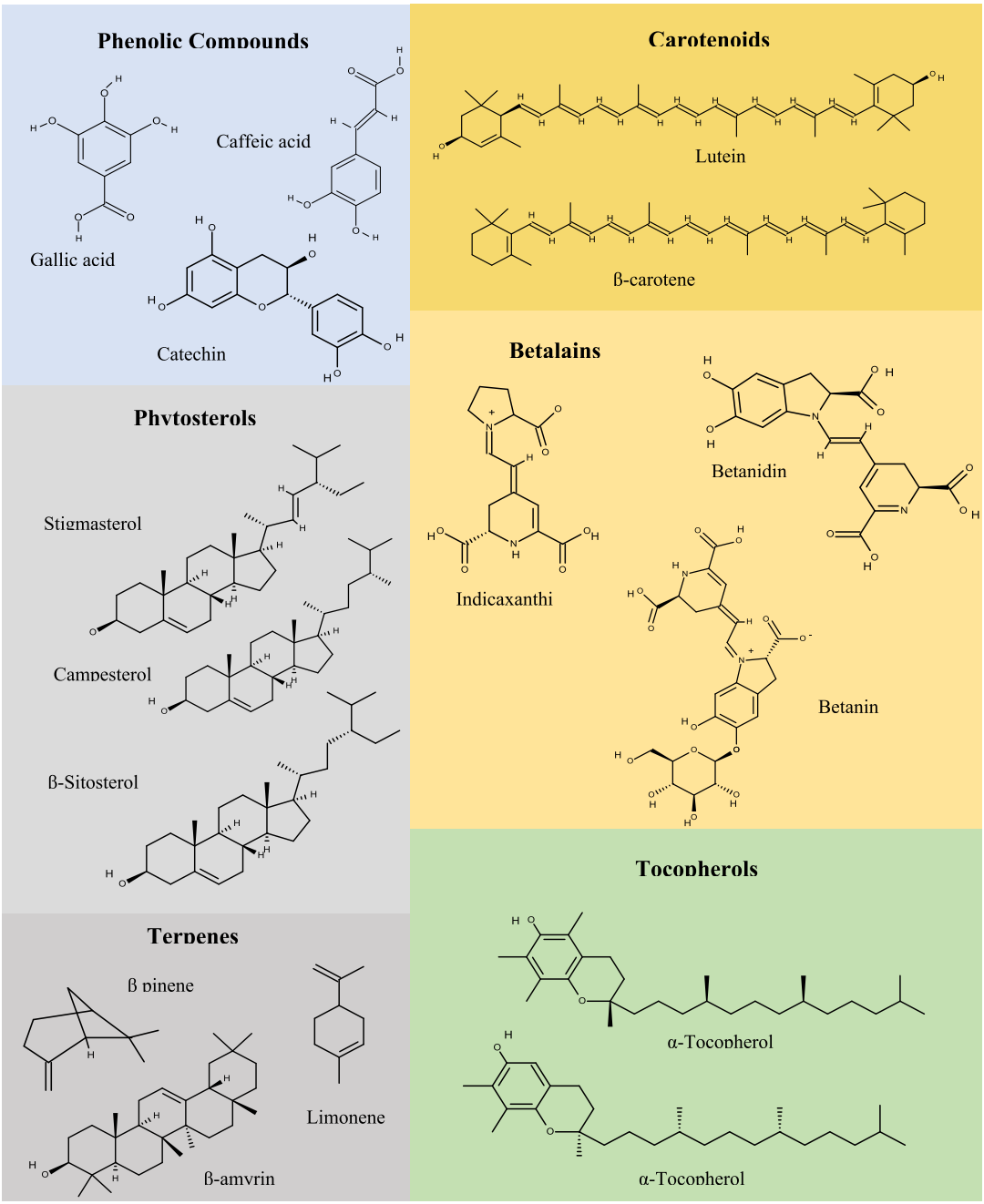

Fig. 1. Chemical structures of some phytochemicals found in species from Cactaceae family.

sweet pulp, with low acidity, weighing approximately 43 to 220 g and can vary from green, yellow, orange and red–purple, depending on the species and stage of maturation (Bakar et al., 2020; García-Cayuela et al., 2019; Guesmi et al., 2012; Mena et al., 2018; Ramírez-Rodríguez et al., 2020).

Pitayas, also known as dragon fruits, belong to the genus *Hylocereus* and originate in the tropical regions of Central and South America; however, due to their good ability to adapt to different types of climates and soil, they can be found in several countries (Abreu et al., 2012; Muhammad et al., 2014; Ramírez-Rodríguez et al., 2020; Ramli et al., 2014). There are several types of pitayas, but *H. undatus* and *H. polyrhizus* are the two most produced and commercialized species due to their sensory characteristics attractive to consumers (Abreu et al., 2012). These species produce fruits that are 7 to 14 cm long and 5 to 9 cm wide, with bright red skin with overlapping green bracts, juicy pulp of white color (*H. undatus*), or red–purple (*H. polyrhizus*) and small black seeds (Arul et al., 2017; Muhammad et al., 2014; Ramírez-Rodríguez et al., 2020; Zhuang et al., 2012).

P. gounellei belongs to the genus *Pilosocereus* of the subfamily Cactoideae, is popularly known as *xique-xique* and widely distributed in the semiarid region of Northeast Brazil, constituting an important natural resource for the livelihood of the local population (Bezerril, 2017; De Assis et al., 2019). It is an endemic species of the Caatinga biome that has an erect trunk, with green colored side branches and a large number of thorns (Bezerril, 2017). Fruits are berries that can weigh from 41 to 48 g, with a diameter ranging from 46 to 49 mm, length from 35 to 39 mm, thick purple-colored mesocarp that tends to crack when ripe, a pink-purple endocarp and funicular, mucilaginous pulp that contains a large number of small black seeds (A. A. da Silva et al., 2018; A. C. P. Sousa, 2017).

Table 2
Bioactive compounds quantified in cactus species.

Species	Fraction	Class	Compound	Content	Units	References
Opuntia ficus-indica	Fruit oil	Phytosterol	Campesterol	21.65	mg 100 g^{-1}	Brahmi et al. (2020)
			Stigmasterol	11.26		
			β-Sitosterol	387.44		
			Stigmastanol	47.04		
	Whole fruit	Betaxanthins	Portulacaxanthin I	12.9	μg g^{-1} [a]	García-Cayuela et al. (2019)
			Portulacaxanthin III	84.1		
			Vulgaxanthin III	25.5		
			Vulgaxanthin I	49.3		
			Vulgaxanthin II	16.3		
			Betaxanthins -amino butyric acid	13.4		
			Indicaxanthin	207.4		
			Betaxanthins -tryptophan	17.9		
	Whole fruit	Betacyanins	Betanin	292.1		
			Isobetanin	21.4		
			Betanidin	22.4		
			Gomphrenin I	31.7		
			Neobetanin	16.8		
	Whole fruit	Carotenoids	(all-E)-violaxanthin	23.78	μg 100 g^{-1}[b]	Cano et al. (2017)
			(all-E)-neoxanthin	12.24		
			(9Z)-violaxanthin	8.41		
			(all-E)-anteraxanthin	11.83		
			(all-E)-lutein	332.16		
			(all-E)-zeaxanthin	13.50		
			Lutein- 5,6- epoxide	13.50		
			(all-E)-α-carotene	3.61		
			(all-E)-β-carotene	65.81		
			(9Z)-β-carotene	6.75		
			Lycopene	26.40	μg g^{-1} [a]	Bakar et al. (2020)
	Pulp	Phenolic acids	Protocatechuic acid-hexoside	0.02	mg g^{-1} [a]	Mena et al. (2018)
			Ferulic acid derivative	0.08		
			Ferulic acid-hexoside	0.14		
			Sinapic acid-hexoside	0.10		
			Piscidic acid derivative	982.3	μg g^{-1} [a]	
			Piscidic acid	21331.80		
			4-hydroxybenzoic acid derivative	816.80	mg g^{-1} [a]	
	Pulp	Flavonols	Quercetin-hexoside-pentoside	0.01		
			Quercetin-hexoside	0.01		
			quercetin glycoside	71.50	μg g^{-1} [a]	
			Isorhamnetin glucoxyl-rhamnosyl-rhamnoside	15.10		
			Isorhamnetin glucoxyl-rhamnosyl-pentoside	8.30		
			Isorhamnetin hexosyl-hexosyl-pentoside	6.30		
			Isorhamnetin glucoxyl-pentoside	4.40		
			Rutin	15.5		
			kaempferol-glucosyl-rhamnoside	9.40		
			Isorhamnetin derivative	0.02	mg g^{-1} [a]	
			Isorhamnetin-rutinoside	0.02		
	Pulp	Flavanone	Naringin	0.04		
	Whole fruit	Terpenoids	β pinene	0.20	%	Oumato et al. (2016)
			Camphene	5.30		
			α phllandrene	0.10		
			ρ –cymene	1.40		
			Limonene	3.90		
			1,8-cineole	0.60		
			β ocimene	0.60		
			3-carene	0.80		
			γ terpinene	0.50		
			Linalool	3.80		
			α farnesene	1.00		
Hylocereus undatus	Pulp oil	Tocopherol	α-Tocopherol	24.00	mg 100 g^{-1}	Lim et al. (2010)
			γ-Tocopherol	12.70		
	Pulp oil	Phytosterol	Campesterol	198.00		
			Stigmasterol	72.00		
			β-Sitosterol	548.00		
	Peel		γ-Sitosterol	19.32		
	Peel	Phenolic acid	Gallic acid	0.20		
			Protocatechuic	0.93		
			p-Hydroxybenzoic	0.72		
			Vanillic acid	0.70		
			Caffeic	0.71		
			Syringic acid	0.21		
			p-Coumaric	0.79		
			Salicylic acid	0.98	mg g^{-1} [a]	Zhuang et al. (2012)
			Caffeotannic acid	0.27		
			Benzoic acid	0.19		

(*continued on next page*)

Table 2 (*continued*)

Species	Fraction	Class	Compound	Content	Units	References
	Peel	Monomeric flavan-3-ols	Catechin	0.29		
	Peel	Flavonols	Rutin	0.30		
	Peel	Alkane	Eicosane	1.92	%	Luo et al. (2014)
			Heptacosane	5.52		
			Nonacosane	5.02		
			Octadecane	9.25		
	Peel	Triterpenoid	β-amyrin	23.39		
	Peel	Ergostanoid	Ergosta-4,6,8(14),22-tetraen-3-one	1.46		
Hylocereus polyrhizus	Pulp oil	Tocopherol	α-Tocopherol	31.90	mg 100 g^{-1}	Lim et al. (2010)
			γ-Tocopherol	11.60		
	Pulp oil	Phytosterol	Campesterol	252.00		
			Stigmasterol	106.00		
			β-Sitosterol	676.00		
	Peel		γ-Sitosterol	9.35		
	Pulp oil	Phenolic acid	Gallic acid	0.25		
			Protocatechuic	0.93		
			p-Hydroxybenzoic	0.66		
			Vanillic acid	0.64		
			Caffeic	0.08		
			Syringic acid	0.08		
			p-Coumaric	0.78		
	Peel	Alkane	Eicosane	3.64	%	Luo et al. (2014)
			Tetratriacontane	1.04		
			Heptacosane	0.44		
			Nonacosane	1.43		
			Octadecane	6.27		
			Docosane	3.19		
	Peel	Triterpenoid	α-Amyrin	13.90		
			β-Amyrin	15.87		
	Fruit	Betacyanin	Betanin	23.81	mg 100 g^{-1}[b]	Yong et al. (2017)
			Isobetanin	3.11		
			Phyllocactin	27.17		
			Hylocerenin	6.37		
Pilosocereus gounellei	Cladodes	Fenolic acids	Gallic acid	1.74	mg 100 g^{-1} [a]	De Assis et al. (2019)
			Caffeic acid	0.26		
			Syringic acid	0.81		
			Chlorogenic acid	0.26		
	Cladodes	Monomeric flavan-3-ols	Catechin	4.71		
			Epicatechin	0.32		
			Epicatechin gallate	0.28		
			Epigallocatechin gallate	14.56		
	Cladodes	Flavonols	Quercetina 3-glucoside	0.24		
			Rutin	0.72		
			Kaempferol 3-glucoside	0.18		
	Cladodes	Flavanones	Naringenin	0.09		
			Hesperidin	1.92		
Cereus jamacaru	Cladodes	Alkaloids	*N*-Methyltyramine	33.79	mg 100 g^{-1} [a]	Davet et al. (2009)
			Hordenine	0.69		

[a]Values expressed on the basis of dry weight. [b] Values expressed on the basis of fresh weight.

C. jamacaru, also known as *mandacaru, mandacaru-de-boi, mandacaru facheiro, cardeiro,* and *jamacaru,* is a succulent, perennial and xerophytic cactus, adapted to dry conditions and rocky soils, native of the Caatinga biome. This species has a rigid trunk, with a 60 cm diameter and, approximately, 10 m high, with radial spines, which can grow up to 30 cm long, and white lateral and subapical flowers, from 20 to 30 cm long. Fruits are orange or red berries of ellipsoid shapes that can reach 5 to 12 cm in length and 7 to 12 cm in diameter, with a white and sweet edible pulp, funicular and mucilaginous appearance, and small black seeds (Medeiros et al., 2019; Santana, 2016; Santos et al., 2020; Sousa, 2017; Vencioneck Dutra et al., 2018).

3. Chemical composition

3.1. Nutritional composition

Table 1 shows the nutritional composition of the cactus studied in this review. *O. Ficus-indica* fruits have good amounts of protein, carbohydrates (arabinose, galactose, xylose, mannose, and glucose), lipids (oleic, palmitic, linoleic and linolenic acids, etc.), fiber, minerals (Ca, Mg, K, P, Fe, Cu, Se, Mn), vitamins (C, E, B1, B2, B3, B6, B9 and B12), pigments with bioactive properties (chlorophyll, carotenoids, phenolic compounds, and betalains), and a high antioxidant capacity (Bakar et al., 2020; Brahmi et al., 2020; Salehi et al., 2019). This species has nutritious cladodes rich in carbohydrate, such as rhammose (7.23 μg mg^{-1}), fucose (2.67 μg mg^{-1}), arabinose (101.86 μg mg^{-1}), xylose (23.42 μg mg^{-1}), mannose (12.59 μg mg^{-1}), galactose (26.49 μg mg^{-1}), glucose (53.81 μg mg^{-1}), and galacturonic acid (187.62 μg mg^{-1}) (Blando et al., 2019; Ginestra et al., 2009).

H. undatus has very nutritious fruits, rich in organic acids (such as ascorbic, citric, isocitric, and malic acids), carbohydrates, amino acids (proline), minerals (mainly potassium and magnesium), and lipids (Esgote Junior, 2017; Stintzing et al., 2003). Its peel can be considered a good source of dietary fiber and vitamin C, with a content of approximately 2.75 and 1.36 fold, respectively (Abreu et al., 2012). The seeds can be used in the extraction of functional lipids, as a new source of essential oil, with high content of saturated (myristic, palmitic, and stearic acids) and unsaturated (oleic, linoleic and linolenic acids) fatty acids (Lim et al., 2010).

Likewise, *H. polyrhizus* contains organic acids (citric, D-isocitric, L-lactic and L-malic acids), some natural pigments (including betacyanin and phenolic compounds), and a pulp rich in minerals, mainly K, Ca and

Mg (Stintzing et al., 2003; Yong et al., 2017). According to Abreu et al. (2012), when compared to the fruit pulp, the skin has higher amounts of dietary fiber (6.81%), ash (1.04%), and vitamin C (22.51 mg 100 g^{-1}). A study by Lim and collaborators showed that *H. polyrhizus* seeds contained a great amount of oil (18.33–28.37%), with the linoleic, oleic and palmitic ones being the main fatty acids found (Lim et al., 2010). Another study found that the fruit peel has good amounts of pectin (26.38% dw), which consisted mainly of galacturonic acid (39.11%), mannose (17.78%), rhamnose (14.47%), galactose (11.91%), glucose (10.82%), xylose (2.41%), and arabinose (3.49%) (Muhammad et al., 2014).

According to Bezerril (2017), *P. gounellei* cladodes showed high water activity (0.97) and good amounts of protein (7.49%), lipid (5.31%), ash (24.33%), total fiber (47.13 mg 100 g^{-1}), minerals (including Mg, Ca, Mn, Fe, P, Zn) and several amino acids (mainly arginine, aspartic and glutamic acids). The fruit pulp of this species contains lipid, protein, carbohydrate and energy value of 27.33; 1.08; 1.42 and 1.85 fold greater than the peel, respectively. In addition, minerals such as Ca, Cu, Fe, P, K, Mg, Zn, Mn and Se were found in both fractions evaluated (pulp and peel). However, the pulp had a content of vitamin C and anthocyanins about 1.56 and 2.32 fold higher than the peel, respectively (Sousa, 2017).

Similarly, *C. jamacaru* fruits have good amounts of carbohydrate, protein, lipid, fiber (including cellulose and hemicellulose), macro and micro minerals (P, K, Ca, Cu, Fe, Zn, Mn, etc.), and antioxidant pigments (Do Nascimento et al., 2011; Magalhães et al., 2019; Santana, 2016; Santos et al., 2020). The peel has ash values and proteins about 2.81 and 1.16 fold higher than the pulp, respectively; however, it has lower concentrations of lipid and carbohydrate and a lower energy value (Sousa, 2017). As to cladodes, no study has reported their nutritional composition yet, similar to *H. undatus* and *H. polyrhizus*.

3.2. Phytochemicals

Fig. 1 shows the molecular structures of some phytochemicals found in the cactus species studied in this review.

3.2.1. Phenolic compounds

Phenolic compounds are secondary metabolites and can be divided into phenolic acids, flavonoids, coumarins, stilbenes, condensed and hydrolyzable tannins, lignans and lignins. These compounds have increasingly aroused the interest of the scientific community, which has highlighted not only their biological effects, but also their potential for applications in the most diverse technological domains (Araújo et al., 2020; Santana, 2016). Table 2 shows some phenolic compounds identified in the species studied in this review.

Recently, a study showed that the content of (poly)phenolic compounds in young cladodes, old cladodes, fruit peel, and fruit pulp of *O. ficus-indica* was dependent on the cultivar evaluated (Mena et al., 2018). In general, young cladodes contained mainly myricetin-hexoside (4.27 mg g^{-1} dw), quercetin-3-O-rutinoside (1.80 mg g^{-1}) and isorhamnetin-rutinoside (1.22 mg g^{-1}); the old cladodes, on the other hand, contained myricetin-hexoside (2.43 mg g^{-1}), ferulic acid-hexoside (1.82 mg g^{-1}) and isorhamnetin-rutinoside (1.27 mg g^{-1}). In turn, fruit peel showed ferulic acid-hexoside (1.81 mg g^{-1}), sinapic acid-hexoside (1.72 mg g^{-1}) and dihydrosinapic acid hexoside (1.16 mg g^{-1}), while the pulp showed dihydrosinapic acid hexoside (2.39 mg g^{-1}), sinapic acid-hexoside (1.71 mg g^{-1}), and feruloyl derivative (0.08 mg g^{-1}) (Mena et al., 2018).

Regarding the identification and quantification of phenolic compounds in *H. undatus* and *H. polyrhizus*, those such as protocatechuic (0.93 mg 100 g-1), p-coumaric (0.78–1.79 mg 100 g^{-1}), p-hydroxybenzoic (0.66–0.72 mg 100 g^{-1}), vanillic (0.64–0.70 mg 100 g^{-1}), caffeic (0.08–0.71 mg 100 g^{-1}), gallic (0.20–0.25 mg 100 g^{-1}), and syringic acids (0.08–0.21 mg 100 g^{-1}) were reported in the seed oil of these two species (Lim et al., 2010). In another study, rutin, quercetin, kaempferol and isorhamnetin were also identified in the pulp of *H. undatus* (Esgote Junior, 2017).

Likewise, *P. gounellei* cladodes are good sources of flavanols and phenolic acids. Among these, epigallocatechin-gallate was the compound found in greater quantity (328.23), followed by catechin (77.13) and quercetin-3-glucoside (14.97 mg 100 g^{-1} dw) (Bezerril, 2017). Compounds such as pinostrobin, Kaempferol and quercetin were also identified in the ethanolic extract of the *P. gounellei* cladodes (Sousa et al., 2018). In turn, the edible portion of the fruits had a flavonol content of 22.76 mg quercetin equivalents 100 g^{-1} (Do Nascimento et al., 2011).

As for *C. jamacaru*, only cinnamic and valeric acids were reported in their cladodes (Medeiros et al., 2019). Vencioneck Dutra et al. (2018) showed that the extract of cladodes of this species has a total flavonoid content of 0.51 μg mL^{-1}. In another study, researchers observed that the chloroformic extract of the roots of *C. jamacaru* had a content of 104.05 mg g^{-1} and 8.87 mg g^{-1}, for total phenolics and total flavonoids, respectively (Santana, 2016).

3.2.2. Carotenoids

Carotenoids are fat-soluble pigments responsible for the yellow, orange and red coloration of many fruits and vegetables. These pigments are also responsible for photoprotection during the photosynthetic process and membrane stabilization. They are of great interest in nutrition for they can act as dietary antioxidants, and, in some cases, have provitamin A activity (Cano et al., 2017; Santana, 2016).

Despite the great importance of carotenoids, few studies have been carried out in order to identify and quantify such compounds in the species studied in this review (Table 2). In fact, no study in the literature has carried out the characterization of carotenoids in *H. polyrhizus*, *H. undatus*, *P. gounellei*, and *C. jamacaru*. However, the content of total carotenoids in *P. gounellei* and *C. jamacaru* was determined and can be seen in Table 1.

Bakar and colleagues, when evaluating the content of carotenoids in *O. ficus-indica* fruits, submitted them to different pretreatments and found that the sun-dried treatment reduced the content of β-carotene and lycopene (4.54 and 17.24 μg g^{-1} dw, respectively) in relation to fresh pulp (12.69 and 26.40 μg g^{-1} dw for β-carotene and lycopene, respectively) (Bakar et al., 2020). They identified in two varieties of *O. ficus-indica* ('Verdal' and 'Sanguinos') (all-E)-lutein (767.98–1132.51 μg 100 g^{-1}) and (all-E)-β-carotene (173.50–200.4 μg 100 g^{-1}) (Cano et al., 2017).

3.2.3. Betalains

Betalains are natural nitrogenous pigments soluble in water, synthesized via the shikimic acid from tyrosine, and are responsible for the red-violet coloration in plants, fruits and vegetables. These compounds have betalamic acid as a common structural unit, which condenses with various amino acids or structures containing indoline to form betaxanthines and betacyanins, respectively (García-Cayuela et al., 2019; Song et al., 2016).

In general, the species studied in this review are good sources of betalains, especially *O. ficus-indica* and *H. polyrhizus* (Table 2). According to the study by Cano and colleagues, the whole fruit of *O. ficus-indica* has a higher content of betacyanins (0.87–3.57 mg of betanin 100 g^{-1} fw) and betaxanthins (1.70–3.04 mg of indicaxanthin 100 g^{-1} fw) than the peel (1.17–2.52 mg 100 g^{-1} and 1.73–2.00 mg 100 g^{-1} for betacyanins and betaxanthins, respectively) and pulp (0.37–1.98 mg 100 g^{-1} and 1.70–2.61 mg 100 g^{-1} for betacyanins and betaxanthins, respectively) (Cano et al., 2017). Another study identified and purified indicaxanthin (15 mg 100 g^{-1}) and betanine (280 mg 100 g^{-1}) from *O. ficus-indica* fruits (Guesmi et al., 2012).

Recently, 13 betacyanins were identified in the *H. undatus* skin, including betanidin-5-*O*-β-glucoside, 17-decarboxy-betanin, isobetanidin-5-*O*-β-glucoside, betanidin-5-*O*-(6′-*O*-3-hydroxy-butyryl)-β-glucoside, 17-decarboxy-phyllocactin, 2′-*O*-apiosyl-phyllocactin, 2-

decarboxy-isophyllocactin (Song et al., 2016). Likewise, betanidin 5-*O*-β-glucoside, isobetanidin 5-*O*-β-glucoside, betanidin 5-*O*-(6́-*O*-malonyl)-β-glucoside, isobetanidin 5-*O*-(6́-*O*-malonyl)-β-glucoside, betanidin 5-*O*-(6́-*O*-3-hydroxy-butyryl)-β-glucoside, and decarboxy betanin 5-*O*-(6́-*O*-malonyl)-β-glucoside were identified as the main betacyanin of *H. polyrhizus* pulp (Utpott et al., 2020). As far as we know, no study in the literature has carried out the chemical characterization of betalains present in *P. gounellei* and *C. jamacaru*.

3.2.4. Other phytochemicals

In addition to the bioactive compounds already mentioned, some studies have reported the presence of other phytochemicals in the cactus species studied here (Table 2). For example, *O. ficus-indica* showed the presence of phytosterols (campesterol, stigmasterol, β-Sitosterol and stigmastanol) and some terpenoids, such as camphene, limonene, linalool, etc. (Brahmi et al., 2020; Oumato et al., 2016). Similarly, tocopherols (α-tocopherol and γ-tocopherol) and phytosterols (campesterol, stigmasterol, β-sitosterol and γ-sitosterol) were also found in the seed oil of *H. undatus* and *H. polyrhizus* (Lim et al., 2010), besides volatile compounds, including alkanes (eicosane, octadecane, nonacosane, among others) and triterpenoids such as β-amyrin in the supercritical CO_2 extract of the bark of these species (Luo et al., 2014). The presence of these compounds in plants is very interesting, since they have medicinal properties (including antioxidant, anti-cancer and anti-tumor activities) and can contribute to health maintainance (Lim et al., 2010; Luo et al., 2014). To date, none of these compounds have been identified in *P. gounellei* and *C. jamacaru*.

4. Biological properties

4.1. Antioxidant and anti-inflammatory

Recently, the antioxidant potential of these species was discovered, as well as their ability to prevent or even reduce inflammation and its complications. Smida and colleagues, by evaluating the immunoprotective activity and antioxidant properties of *O. ficus-indica* (100 mg kg^{-1}) in male Wistar rats, found that its extract was able to decrease the loss of thymocytes, thymus atrophy and hypertrophy of the spleen, reduce the levels of substances reactive to thiobarbituric acid (TBARS), normalize the activity of endogenous antioxidant enzymes (catalase, superoxide dismutase and glutathione peroxidase), increase thymic weight gain, and reduce DNA damage (Smida et al., 2017). In another study, El-Hawary and collaborators observed that the extract of this species showed antioxidant, anti-inflammatory and neuroprotective activity in male Sprague Dawley rats. According to the authors, the intake of the extract increased the antioxidant capacity, the levels of catecholamines, Interleukin 10 (IL-10), the activity of the enzymes catalase (CAT), glutathione and superoxide dismutase (SOD), improved the animals' memory and latency time, and reduced the levels of malondialdehyde, acetylcholinesterase, tumor necrosis factor α (TNF-α) and nuclear factor kappa B (NF-κβ) (El-Hawary et al., 2020).

Ingestion of *H. undatus* oligosaccharides (0; 9 and 27 mg L^{-1}) showed potential to reduce oxidative stress in Daphnia magna, since it was able to reduce lipid peroxidation and increase SOD activity and expression of genes Toll2, Toll3, Toll5, Toll7, and Pelle (Sangkuanun et al., 2020). A study by Macias-Ceja and colleagues found that *H. polyrhizus* extract (1 g kg^{-1}) had anti-inflammatory potential in Balb/c mice. According to the authors, the intake of the extract decreases the loss of body weight, the formation of hemorrhagic ulcers in the cecum, colon and rectum, the damage to the mucosa and the expression of NF-kB, avoided the degradation of IκB-α and the increased myeloperoxidase activity. It also decreased the expression of nitric oxide synthase (iNOS), cyclooxygenase-2 (COX-2), TNF-α and Interleukin 6 (IL-6) in colonic tissue (Macias-Ceja et al., 2016). Similarly, de Oliveira et al. (2021) reported that *P. gounellei* extract (125, 250, and 500 mg kg^{-1}) reduced the expression of TNF-α, IL-6, monocyte chemoattractant protein-1 (MCP1) and lipid peroxidation, besides improving SOD levels in liver, muscle, fat tissues, and reducing liver inflammation in male Swiss mice.

4.2. Anti-obesity and antidiabetic

In recent years, cactus have attracted the attention of the scientific community due to their beneficial properties in preventing some metabolic disorders such as obesity and diabetes. In this sense, a study by Berraauan and collaborators showed that the use of *O. ficus-indica* can prevent alloxan-induced diabetes. According to the authors, the administration of this species seed oil (2 mL kg^{-1}) in Swiss albino mice increased the survival rate of the animals, reduced hyperglycemia, free radicals formation, loss of body weight, and inhibited damage in pancreatic tissue (Berraaouan et al., 2015).

In another study, while evaluating obesity and insulin resistance in male C57BL/6J mice, researchers observed that the intake of betacyanins from *H. undatus* peel (50; 100 and 200 mg kg^{-1}) reduced body weight gain, improved adipose tissue hypertrophy, hepatosteatosis, glucose intolerance and insulin resistance. It also improved expression of genes related to fibroblast growth factor 21 (β-Klotho and FGFR1/2), to lipid metabolism (adiponectin receptor 2 (AdipoR2), carnitine palmitoyltransferase 1b (Cpt1b), peroxisomal acyl- coenzyme A oxidase 1 (Acox1), peroxisome proliferator-activated gamma receptor (PPARγ), insulin-induced gene 1 (Insig1) and insulin-induced gene 2 (Insig2)), and it reduced the expression of fatty acid desaturase 2 (Fads2) (Song et al., 2016). Moreover, *H. undatus* also showed inhibitory activity in the multiple stages of glycation and great potential to decrease the formation of reactive species, oxidation and glycation of proteins, thus reducing the complications caused by diabetes (Pérez-Gutiérrez & Enriquez-Alvirde, 2018). Likewise, the intake of *P. gounellei* cladodes extract (125, 250 and 500 mg kg^{-1}) reduced body weight, total cholesterol levels, LDL cholesterol and triglycerides, improved HDL cholesterol levels, tolerance to glucose, and decreased epididymal fat and steatosis in male Swiss mice (de Oliveira et al., 2021). Ingestion of the *C. jamacaru* extract (210 and 420 mg kg^{-1}) decreases the consumption of food and the body weight gain in male Wistar rats (Medeiros et al., 2019).

4.3. Anticancer and antiproliferative

Scientific evidence has shown the therapeutic potential of the species studied in this review as possible preventive agents for degenerative diseases such as cancer (Dasaesamoh et al., 2016; El-Beltagi et al., 2019; Kim et al., 2011). A study by Helal and collaborators observed that the *O. ficus-indica* extract showed anticancer potential in squamous cells of the lung (A549) and in colon cell carcinoma (Caco2). According to the authors, there was an inhibition in cell proliferation, depending on the concentration used. Furthermore, the extract also showed positive regulation for the casp-3 and Bax genes, and a negative regulation of Bcl-2 after 24 h (Helal et al., 2019). In another study, the extract of this species also had antiproliferative potential against liver cancer cells (HepG2), colorectal adenocarcinoma (Caco-2) and breast (MCF-7), with cell viability decreasing as the extract concentration increased (El-Beltagi et al., 2019).

The fermentation of *H. undatus* oligosaccharides by the gut microbiota increased the production of short-chain fatty acids, which, in turn, inhibited the proliferation of Caco-2 cells, reducing thus the risk of colon cancer (Dasaesamoh et al., 2016). Likewise, Kim and colleagues found that the *H. undatus* and *H. polyrhizus* extracts showed good antiproliferative activity against cancer cells (AGS and MCF-7) (Kim et al., 2011). Similarly, *C. jamacaru* extract also showed anti-cancer and antitumor potential, as it reduced the viability of sarcoma 180, promoted a tumor reduction of 86.07%, and inhibited the cytotoxicity of cisplatin without inducing mutagenic or cytotoxic damage in blood cells of mice and in human lymphocytes (Vencioneck Dutra et al., 2018).

4.4. Other biological effects

In addition to the potential to prevent inflammation, diabetes, obesity, and cancer, some studies have also reported other health benefits from the intake of these species. According to Khémiri et al. (2019), *O. ficus-indica* oil showed antimicrobial effect against *Enterobacter cloacae* and *Candida parapsilosis*, antifungal activity against three opportunistic cutaneous fungi (*Penicillium*, *Aspergillus* and *Fusarium*), a good wound healing effect and potential to prevent infections. In another study, Khuituan and colleagues reported that oligosaccharides from *H. undatus* (500 and 1000 mg kg^{-1}) as a dietary supplement can promote intestinal health and correct gastrointestinal motility disorders in adult male ICR/Mlac mice (Khuituan et al., 2019).

The supplementation with 5% of *H. polyrhizus* juice increased the concentration of aspartate transaminase, reduced the concentrations of alkaline phosphatase, alanine transaminase in the plasma, and the diastolic stiffness of the heart, thereby preventing liver and cardiovascular changes in male Wistar rats (Ramli et al., 2014). Another study, while evaluating the gastroprotective effect of *P. gounellei*, found that the ingestion of this species extracts of cladodes and roots (200 and 400 mg kg^{-1}) reduced the damage and gastric lesions induced by ethanol, decreased the development rates of gastric ulcers, and increased catalase activity in Swiss mice and Wistar rats (G. A. Sousa et al., 2018). Likewise, the intake of *C. jamacaru* extract (75; 150 and 300 mg kg^{-1}) in adult male mice decreased the abdominal contortions and the edema of the animals' paws, besides presenting low toxicity, being considered a potential antinociceptive and anti-inflammatory agent (Santana, 2016).

5. Application in the development of food products

5.1. Bread and cake

C. E. Silva (2019), when evaluating the cake production using *P. gounellei* flour in partial replacement of wheat flour, observed that formulations containing 20, 40 and 60% of the flour of this species had greater acceptability and purchase intention, while the formulation with 80% resulted in a wetter cake, slightly more acidic and less accepted by the tasters. In addition, he found that this partial substitution increased the content of ash (20%), protein (20%) and ascorbic acid (60 and 80%) in the formulated cake. In another study, the formulation of loaf bread using 10% flour from the skin of the *C. jamacaru* fruit – dried at 50 °C – resulted in a product with better color, flavor and appearance (do Nascimento, 2014). Msaddak et al. (2017) observed that breads formulated with *O. ficus-indica* cladodes powder instead of wheat flour (2.5; 5.0; 7.5; and 10%) showed less extensibility, deformation energy and changes in color parameters, mainly lightness (L *), redness (a *) and yellowness (b *), in addition to a 25% increase in the specific bread volume (supplementation with 5% powder), phenolic content and antioxidant capacity in 10 and 139 fold in supplementation with 10%, respectively. However, the greatest acceptability was for bread formulated with 5% of this powder (Msaddak et al., 2017). The use of flour from *H. polyrhizus* skin to replace fat (50, 75 and 100%) for the preparation of loaf bread resulted in a product with lower values of specific volume and L *, and higher values of a* and b*. The textural analysis showed that breads formulated with the flour from *H. polyrhizus* skin presented greater firmness (crumb and crust), cohesiveness, flexibility and guminess of the crumb. Overall, the breads formulated had good sensory acceptance, although breads formulated with 100% fat substitution received the lowest grades (Utpott et al., 2018).

5.2. Ice creams

Food processing can be considered an interesting strategy for industrialization and commercial fruit exploitation. In this sense, ice creams made from the pulp of *C. jamacaru* (400–500 g), *P. gounellei* (400–500 g) and *C. jamacaru* + *P. gounellei* (200 g each) had physicochemical attributes such as moisture (71, 89–79.13%), pH (5.71 to 6.19), total soluble solids (3.9–4.9%), total acidity (0.21–0.32%) and ashes 0, 70–0.90%) suitable for industrialization. Moreover, all formulations developed showed good sensory acceptance, with emphasis on the formulation containing 500 g of *C. jamacaru* pulp, which showed greater sensory acceptance and consequently greater purchase intention (Pinto, 2017). Similarly, the production of ice cream from *O. ficus-indica* and *C. jamacaru* fruits proved to be an interesting strategy to expand their commercial exploitation (Fidelis et al., 2015). In this study, all formulations showed acidic pH, and ice cream made with *C. jamacaru* (100 g of ice cream and 100 g of pulp or peel) had a higher content of total soluble solids (21.66° Brix) and total acidity (0.41%), while the formulation made with *O. ficus-indica* (1:1, w/w) had the highest vitamin C content (6.24 mg 100 g^{-1}).

Recently, Gengatharan et al. (2020) evaluated the application of betacyanins extracted from *H. polyrhizus* as a natural dye in ice cream. The results showed that the application of betacyanins caused total color changes similar to the commercial colorant (E-162). They also observed that the addition of natural dye increased the antioxidant capacity and there was greater color acceptability of ice cream with betacyanins than E-162. In another study, the application of *H. polyrhizus* skin flour proved to be an alternative to reduce fat in ice cream (73.5%), with good general acceptability, improving the overrun and the rheological behavior of the sample (Utpott et al., 2020).

5.3. Yogurt

Fidelis and colleagues, when evaluating the development of yogurt from the addition of pulp/peel of *C. jamacaru* and *O. ficus-indica* (natural yogurt + pulp/peel of each fruit-1: 1, w/v), found that yogurts formulated with pulp or skin of *C. jamacaru* had a higher content of soluble solids (9.40° Brix), pH (3.53) and total acidity (0.84%), while the formulation containing *O. ficus-indica* had the highest vitamin C content (4.87 mg 100 g^{-1}) (Fidelis et al., 2015). Similarly, Nóbrega et al. (2020), while preparing goat yogurt with 15% jelly from *C. jamacaru* pulp and passion fruit with oligofructose (3.4 g L^{-1}) and while assessing its stability during 28 days of refrigerated storage, observed an increase in water activity at the end of storage and changes in the physicochemical properties evaluated, with emphasis on acidity, pH and lactose. This way, the authors found that the addition of these ingredients had good incorporation and can be an interesting approach for the formulation of a new dairy product.

5.4. Juice

A study on the influence of clarification on the physical–chemical composition of *O. ficus-indica* fruit juice was recently evaluated (Cassano et al., 2010). In this study, showed that the use of microfiltration and ultrafiltration to clarify the juice did not change the content of total soluble solids, pH and acidity, but reduced the content of phenolic compounds (6.1%) in the permeate fraction for ultrafiltration and protein content in permeate fractions for microfiltration and ultrafiltration (76.9% and 78.3%, respectively). On the other hand, the retained fraction can be considered a source of bioactive compounds (e.g., phenolics and vitamin C.), betacyanins, and betaxanthins, with the potential to be used in the preparation of mousses, ice creams, jellies and functional foods (Cassano et al., 2010).

5.5. Gummy candies

Recently, Otálora et al. (2019) evaluated the application of encapsulated betalains extracted from *O. ficus-indica* in the making of gummy candies. The authors observed that the viscoelastic properties, gel strength and stress relaxation depended on the type of gelatin and betalain-rich capsules/gelatin ratio used. Also, the red–purple coloring of the chewing gum (characterized by the addition of betalain) had good

Table 3
Use of cactus in the most diverse technological purposes.

Species	Technique (Description)	Purpose/application	Reference
Opuntia ficus-indica	Synthesis of Au/Li nanoparticles using extract of the species	Nanoparticle synthesis using an economic and ecological method.	Alvarez-Bayona, Cortez-Valadez, Martínez-Suárez, Cruz-Rivera, & Flores-Acosta (2019)
	Prickly pear peel flour for biscuits formulation	Prickly pear flour showed better kneading ability, flavour retention and antioxidant capacity. Cookies formulated with fig flour (20–30 g/100 g) showed greater appreciation in terms of smell, taste, colour and acceptability.	Bouazizi, Montevecchi, Antonelli, & Hamdi (2020)
	Extraction of gum	The extracted gum contained mainly carbohydrates such as glucose (78.0%), arabinose (12.9%), xylose (4.8%), galactose (2.4%), and mannose (2.4%), in addition to having excellent stabilizing properties, good gelling capacity, and high viscosity capacity	Salehi, Emam-Djomeh, Askari, & Fathi (2019)
	Development of films plasticized with different polyols	Films plasticized with glycerol showed greater flexibility, water vapor permeability and lower glass transition temperature	Gheribi et al. (2018)
	Development of anticoagulant agent from the sulfation of a pectin-like polysaccharide extracted from *Opuntia ficus indica*	Sulfated polysaccharides showed greater anticoagulant activity as they prolong activated partial thromboplastin time and thrombin time	Chaouch et al. (2018)
	Elaboration of cosmetic nanoemulsions containing hydroglycolic extract of *Opuntia ficus indica* as moisturizing agent	The formulation containing the hydroglycolic extract (1%) can be considered a potential moisturizing agent, as it increased the water content in the stratum corneum for up to 5 h after application on the forearm.	Ribeiro et al. (2015)
	Addition of aqueous extract and peel powder peel for elaboration of carboxymethyl celluloseedible films	Increase in the content of bioactive compounds and antioxidant capacity of the elaborated film	Aparicio-Fernández et al. (2018)
	Elaboration of edible coating from mucilage and application for strawberry conservation	Application of edible coating increased the firmness and shelf-life of strawberries. In addition, there was a greater preference for coated strawberries at the end of the storage period (nine days).	Del-Valle, Hernández-Muñoz, Guarda, & Galotto (2005)
Hylocereus undatus	Synthesis of green synthesized fluorescent nitrogen doped carbon dots (*N*-CDs)	*N*-CDs showed low cytotoxicity, good compatibility with L-929 and MCF-7 cells and high catalytic activity.	Arul, Edison, Lee, & Sethuraman (2017)
	Anthocyanin extraction for used to develop intelligent gelatin films	The film showed the highest antioxidant activity and highest pH sensitivity.	Rawdkuen, Faseha, Benjakul, & Kaewprachu (2020)
	Rheological behavior and physical properties of spray dried mucilage	The reconstituted mucilage exhibited non-Newtonian shear thinning behavior; good lubricating effect between the particles; low trends in gel formation; and low degree of polydispersity.	García-Cruz, Rodríguez-Ramírez, Méndez Lagunas, & Medina-Torres (2013)
	Biosynthesis of zinc oxide nanoparticles (ZnO NPs)	The nanoparticles showed good biomedical capacity and good antimicrobial activity against *Bacillus subtilis*.	Vishnupriya, Nandhini, & Anbarasi (2020)
Hylocereus polyrhizus	Production of pectins	High yield of pectin extraction, high degree of esterification, high methoxyl pectin, and low viscosity.	Muhammad, Nur, Gannasin, Mohd. Adzahan, & Bakar (2014)
	Development of active and intelligent packaging	Increased the barrier to water vapor and the capacity of the barrier to ultraviolet–visible light, in addition to the mechanical, antioxidant and antimicrobial potential of films, and improvement sensitivity to ammonia.	Qin, Liu, Zhang, & Liu (2020)
	Extraction of red – purple pitaya colorant and its application in yogurt	The formulated yogurt showed better color and there was no change in aroma	Lima et al. (2020)
	Preparation of cookies from wheat flour substituted with pitaya peel flour	The addition of pitaya husk flour improved the nutritional quality but did not affect the sensory acceptability of the formulated cookies.	Ho et al. (2016)
Xiquexique (*Pilosocereus gounellei*)	Elaboration of cake using xique-xique pulp (50–100%) to replace wheat and rice flours.	The formulation with 100% of the pulp showed higher humidity, acidity and a greener color, while formulations containing 50% of the pulp showed similarities in physico-chemical composition and appearance.	dos Reis et al. (2018)
Mandacaru (*Cereus jamacaru*)	Extraction of cellulose nanowhiskers from the spines of mandacaru using alkaline treatment and bleaching, followed by acid hydrolysis.	The alkaline and bleaching pretreatments reduced the thermal stability, while the acid hydrolysis time influenced the thermal stability and degree of crystallinity of the nanowhiskers.	Nepomuceno, Santos, Oliveira, Glenn, & Medeiros (2017)
	Elaboration of biosorbent for diesel oil from mandacaru *in natura* and modified	The modified mandacaru showed higher sorption capacity (29%) compared to the commercial sorbent.	Anjos (2017)
	Elaboration of powdered biosorbent	The biosorbent proved to be efficient for discoloration of liquid flows in discontinuous and continuous systems.	Georgin et al. (2020)
	Elaboration of fermented berevage from mandacaru	The fermented beverages had an ethanol content of 82.11 g L^{-1} and quality comparable to other fruit fermented beverages.	de Almeida, da Silva, Conrado, Mota, & Freire (2011)

stability during storage at 4 °C for 30 days, without significant changes in the total color parameters.

6. Use in animal feed

The demand for food that can be used in both human and animal diets has been increasing recently. Factors such as drought and climate change can affect the production of cereals, increase their prices on the international market, and thus hamper access to this raw material. Given this, the use of nutritious species such as cactus can be an alternative for agribusiness, since these crops are multifunctional, resistant to drought conditions and grow in semi-arid and arid regions (Abidi et al., 2009).

Within this context, Souza and colleagues observed that the addition of *O. ficus-indica* silage as a substitute for 'Tifton' hay affected the morphometric measurements, the carcass compactness index, the loin eye area, and the weight of casting of commercial cuts. They also observed a reduction in meat color parameters (L*, a*, b* and Chroma) and pH, and changes in fatty acid composition in the Longissimus lumborum muscle of lambs (Souza et al., 2020). Likewise, the study showed that the replacement of barley for supplementation with *O. ficus-indica* cladodes for 84 days in the diet of lambs and goats did not have harmful effects on the digestion, growth and quality of the meat of the

animals (Abidi et al., 2009). Another study found the ingestion of this species reduced the concentration of fat in the goat carcass; however, it was responsible for a higher concentration of linoleic acid, as well as a higher amount of polyunsaturated fatty acids in the meat (Mahouachi et al., 2012).

Similarly, *P. gounellei* can be used as an alternative to feed dairy cows of medium and low production without causing changes in feed efficiency and physiological responses (Furtado et al., 2016). Supplementation with 473 to 501 g of a mixture containing *P. gounellei*, *C. jamacaru*, *C. squamosus*, *Nopalea cochenillifera* or *O. stricta* did not cause any changes in sensory quality, lipid profile and physical–chemical composition (levels of fat and total solids) of Saanen goat milk, but there were changes in the levels of protein, lactose, non-fat solids and cryoscopy point (Catunda et al., 2016). In addition, the supplementation of *C. jamacaru* in goat feeding during the dry season in the semi-arid region of Pernambuco, Brazil, resulted in less weight loss compared to animals that remained on continuous grazing in the Caatinga, whose weight loss was on average 5.25% of the live weight in relation to the initial weight (Cavalcanti & Resende, 2006).

Little information can be found in the literature on the use of pitayas in animal feed. However, a study by Matra and colleagues showed that the supplementation of *H. undatus* peel pellet (400 g) in the feeding of crossbred Holstein bulls resulted in a better formation of final products of rumen fermentation, since it increased the concentrations of blood urea nitrogen, NH3-N, volatile fatty acid, in the synthesis of microbial protein and urine purine derivatives, besides reducing rumen methane production (Matra et al., 2020).

7. Other applications

7.1. Water treatment

Chemical coagulants, widely used in water treatment to remove colloids and fine particles, despite being very efficient, can have negative impacts on the environment. Thus, some studies have shown the potential of natural coagulating agents derived from plants to replace chemical flocculating agents (Wan et al., 2019). In this sense, Bouaouine and collaborators, when evaluating the flocculating properties of *O. ficus-indica* for water treatment, observed that the solid cactus material was responsible for reducing water turbidity by more than 90%. In this study, we also found that the molecules responsible for the flocculating properties were quercetin and starch and developed a flocculation model based on the identified constituents. When used alone, quercetin removed up to 70% of the turbidity; when used in combination with starch, the removal of turbidity was 93%, with a flocculant activity of up to 84%. Finally, we observed that the mechanisms related to the flocculating properties in the proposed model involved ionic interaction (kaolinite/quercetin) and hydrogen bonding (kaolinite/starch and quercetin/starch) (Bouaouine et al., 2019).

7.2. Natural dyes

Guesmi et al. (2013) evaluated the application of indicaxanthin extracted from *O. ficus-indica* in the dyeing of modified acrylic fabrics by comparing two dyeing methods (conventional heating and ultrasound). The authors found that the optimal conditions for dyeing the fabrics were 80 °C and pH 3 for 30 min, and that the ultrasound treatment improved the dye absorption (49.62%), light fastness, and washing when compared with the conventional heating method. In another study, Utipott and colleagues observed that the pigments extracted from *H. polyrhizus* pulp had great potential as natural dyes with characteristics suitable for application in yogurts and other matrices, such as candies, soft drinks, ice cream, and bakery products (Utpott et al., 2020).

7.3. Extraction of pectins and mucilages

Polysaccharides are of great importance in the food industry, as they can be used in food systems to change their functional properties (Medina-Torres et al., 2000). Gárcia-Cruz and collaborators, evaluating the rheological and physical properties of reconstituted spray-dried mucilage obtained from *H. undatus* cladodes, verified that the reconstituted mucilage (3–6% w/v) presented Newtonian behavior in low shear rates, pseudoplastic behavior in simple shear flow and viscoelastic behavior of non-gel formation (García-Cruz et al., 2013).

Another study showed that the powder obtained from spray drying of *H. polyrhizus* and *H. undatus* juice presented physico-chemical characteristics of great interest to the food industry, such as low humidity (3.7–5.3%) and bulk density (0.38–0.40 g cm^{-3}), high solubility (131–136 s), good color-related sensory characteristics, such as chroma (2,3–47) and hue angle (86-329°), good content of ascorbic acid (10–11.7 mg 100 g^{-1}), phenolic compounds (2.2–6.1 mg gallic acid equivalent 100 g^{-1}), betacyanin (58.8 mg L^{-1}) and considerable antioxidant capacity (10.4–14.1 mg ascorbic acid equivalent 100 g^{-1}) (Lee et al., 2013).

Likewise, the *H. polyrhizus* bark proved to be rich in high-methoxyl pectin (HMP), which, due to its high degree of esterification (63.8%) and contact angle (95.5°), is hydrophobic in nature and may show hypolipidemic activity (Zaid et al., 2019). According to Zaid et al. (2019), *H. polyrhizus* is a good source of pectins of great importance for the food industry, as they can be used as dispersing, emulsifying and gelling agents in products such as jams, drinks and dairy products. More applications of the species studied in this review can be seen in Table 3.

8. Innovation potential in the industrial field

When using the terms Cactaceae, cactus, palm fruit, fig, *Opuntia ficus-indica*, dragon fruit, *Hylocereus undatus*, *Hylocereus polyrhizus*, *xique-xique*, *Pilosocereus gounellei*, *mandacaru* and *Cereus jamacaru* to check the number of patent applications granted, we observed a total of 15,184 inventions between 2001 and 2020 patented in Questel Intellectual Property Portal on the Internet: <URL: http//www.orbit.com > . The technological domain is represented mainly by basic materials in chemistry, biotechnology, medical technology, environmental technology, food chemistry, organic fine chemistry, other special machines, and pharmaceuticals.

Some of the patent applications related to these species involve the development of new food products, production of polymeric material for application in the manufacture of plastic parts, natural dyes, products for slimming, production of second generation bioethanol, cosmetics and skin moisturizers, production mucilage as a controlled release vehicle and adjuvant for regenerating the mucosa of the gastrointestinal tract, among others (Table 4).

A Chinese invention showed the potential of *O. ficus-indica* in the feeding of free-range chickens in order to reduce the presence of drug residues in the carcasses and increase the resistance to diseases and the survival rate of animals (Baocheng and Fenglin, 2013). Likewise, its potential in the development of conventional wetting agents to be used as active ingredients in cosmetic and dermatological compositions has also been shown (Quintanar et al., 2013).

H. undatus bark was used to obtain a low-cost nutritious sauce, simple to process, and rich in bioactive substances, such as anthocyanidins, pectin, dietary fibers, gamma aminobutyric acid and flavones (Li et al., 2015). *H. polyrhizus* was also used in the production of mixed fermentation wines with improved sensory, nutritional and functional characteristics (Yuhua et al., 2016). Pitaya peels were also used in the preparation of membranes for making edible packaging, with improved mechanical and barrier properties (Wu et al., 2015).

Recently, a Brazilian invention used *P. gounellei* to obtain refined and wholemeal flour, with high nutritional and functional value due to the high content of minerals and fibers, combined with unique sensory

Table 4
Overview of patents for cactus for various purposes.

Title	Publication number	Publication date	Claim	Technology domain	Applicant/Assignee
Opuntia ficus-indica					
Procedure for the transformation of dry *Opuntia ficus-indica* cactus cladodes to produce second generation bioethanol	ES2552603	2014–05-30	Development of second generation biofuels as renewable and clean energy, to face the remarkable depletion of resources in fossil energies.	Biotechnology	Universidad de Cadiz andUniversidad de Jaen
Composition comprising an extract from *Opuntia ficus-indica* for the enhancement of learning and memory	KR20120010426	2010–07-26	*Opuntia ficus-indica* extract as an active ingredient characterized in that it contains the prevention and treatment of amnesia or memory impairment associated with a pharmaceutical composition.	Pharmaceuticals	Korea Institute of Science & Technology
Use of nopal *Opuntia ficus-indica* mucilage combined with conventional moisturizing agents as active ingredients in cosmetic and/or dermatological compositions.	MX2013010936	2013–09-24	Production of cosmetic and/or dermatological compositions administered topically to the skin with enhanced moisturizing effect.	Organic fine chemistry	University Mexico NAC Autonoma
Nopal mucilage (*Opuntia ficus-indica*) as a controlled-release vehicle and adjuvant for regeneration of the mucosa of the gastrointestinal tract	WO2015/126232	2014–02-18	The invention relates to a composition comprising ranitidine, nopal mucilage and a wetting agent, for use in the treatment of gastric disorders, such as gastric ulcers, active duodenal ulcers, Zollinger–Ellison syndrome, gastro-oesophageal reflux and erosive oesophagitis.	Pharmaceuticals	University NAC Autonoma de Mexico
Hylocereus undatus					
Use of *Hylocereus undatus* fruit extract as fluorescent colorant of skin	EP3270942	2016–03-17	The present invention relates to an extract of *Hylocereus Undatus* white fruit that fluoresce in blue when exposed to light source comprising UV light, for example sunlight, and to its use as fluorescent colorant in cosmetic applications.	Organic fine chemistry andpharmaceuticals	IBR Israeli Biotechnology Research
Glass sealant based on *Hylocereus undatus* emulsion and preparation method thereof	CN104327773	2014–10-30	Obtaining a glass sealant with high resistance to water and adhesion, short curing time and reduced production cost.	Basic materials chemistry	Guangzhou Constant Silicone
Cosmetic compositions and uses thereof	EP3432859	2017–03-23	Topical composition comprising *Hylocereus undatus* fruit extract, *Aloe barbadensis* leaf extract, and a dermatologically acceptable vehicle, and wherein the composition has antioxidant capacity and/or the composition is capable of inhibiting TNF-a expression.	Organic fine chemistry	MARY KAY
Hylocereus polyrhizus					
Hylocereus polyrhizus flesh and pericarp red pigment and extraction method thereof	CN105524482	2015–12-25	Extraction of dark purple pigment (betalaines) easily dissolved in water stable to pH 3–6.	Basic materials chemistry, food chemistry, andorganic fine chemistry	Nanning Sailong Linhua Biological
Anti-aging facial cream	CN109512745	2018–12-29	The cream obtained can increase circulation and decrease blood viscosity in the skin, stimulate metabolism, reduce reactive species and achieve anti-aging effect.	Organic fine chemistry	Laf Household
Lipstick containing pitaya pigment and preparation method thereof	CN107412065	2017–08-15	The lipstick disclosed by the invention adopts the pigment extracted from *hylocereus polyrhizus* and raw materials are natural, so that the lipstick has no harms to human bodies and has an anti-oxidization effect.	Organic fine chemistry	Li Shuying and Liu Houwei
Lemon-flavor artistic pigment	CN109401400	2018–11-13	The dye obtained has natural characteristics, greater potential for the health and is less aggressive to the environment.	Basic materials chemistry	Tianmen Weiwei Bride Wedding Culture Communication
Pilosocereus gounellei					
Production of goat yoghurt added from xique-xique geleia	BR102018073372	2018–11-13	The yoghurt produced is a food with high nutritional value, uniform texture, enjoyment and excellent flavour, as well as contributing to the health of the digestive system.	Food chemistry	Federal University of Paraiba,owned by Federal University of Santa
Xique-xique goat cream cream	BR102018070346	2018–10-03		Food chemistry	

(*continued on next page*)

Table 4 (*continued*)

Title	Publication number	Publication date	Claim	Technology domain	Applicant/Assignee
			Obtaining goat cheese with good nutritional and bioactive value added from xique-xique flour		Federal University of Paraiba,owned by Federal University of Santa
Manufacture of biscuits	BR102018070273	2018–10-02	Cookie production with high nutritional value, high functional potential, in addition to unique flavors obtained from the xique-xique flour	Food chemistry	Federal University of Paraiba,owned by Federal University of Santa
Xique-xique jelly production process	BR102018073370	2018–11-13	The geleel produced is a food with well appreciated characteristics, pleasant coloration and adocicated flavour, as well as nutritional and bioactive characteristics.	Food chemistry	Federal University of Paraiba,owned by Federal University of Santa
Cereus jamacaru					
Polymeric composite with mandacaru wood (*Cereus jamacaru* dc.) and process of obtaining	BR102017004453	2017–03-06	Production of a material with a polymeric matrix of polyethylene and mandacaru for application in the manufacture of injected plastic parts.	Macromolecular chemistry, polymers, andother special machines	National Service for Industrial Training (SENAI)
Elaboration of functional goat yogurt with passion fruit jelly (*Passiflora edulis* sims.) and mandacaru fruit (*Cereus jamacaru*)	BR102017020281	2017–09-22	Elaboration of a functional beverage of easy preparation and low cost, with good nutritional and sensory quality.	Food chemistry	Federal University of Campina Grande,owned by Federal University of Rio Grande do Sul
Elaboration and processing of passion fruit (*Passiflora edulis*) and mandacaru (*Cereus jacamaru*) flavored jelly	BR102017020087	2017–09-20	Preparation and processing of jelly that is easy to prepare and low cost, with good nutritional and sensory quality.	Food chemistry	Federal University of Campina Grande,owned by Federal University of Rio Grande do Sul
Cosmetic composition containing extract from plant of the genus cereus, use of said extract and cosmetic method	WO2019/178657	2018–03-21	Preparation of a cosmetic composition to be topically applied on the skin.	Organic fine chemistry	Loccitane

characteristics (Machado et al., 2018). On the other hand, *C. jamacaru* was used in the preparation of a gluten-free cake with good sensory, nutritious and functional characteristics, being an excellent alternative to generate income for small traders, due to the appreciation of underutilized ingredients from the region (Costa et al., 2018).

9. Conclusion

The studied species have high nutritional value and can be considered excellent sources of phytochemicals, such as phenolic compounds, carotenoids and betalains, responsible for several biological properties already reported in the literature, including action against diabetes, obesity, cancer and others. In addition, they also have great potential to be explored by the food, pharmaceutical and cosmetic industries, and can make for great alternatives for animal feed, contributing to the development of agriculture in arid and semiarid regions.

However, some species of the Cactaceae family still need further research, since there is little information about its phytochemical composition and biological and technological potential, especially regarding *Pilosocereus gounellei* and *Cereus jamacaru*. Likewise, future approaches are also needed to encourage cultivation and commercialization. Research of this nature is important and can contribute to food/nutritional security and the socioeconomic development of the population in regions where these species naturally occur.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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