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BIOACTIVE METABOLITE FROM MALAYSIAN GREEN SEAWEED; *Ulva* sp. FOR ITS ANTIFOULING ACTIVITY

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Marine natural products mainly seaweeds gained much attention to combat the biofouling issues in marine environments. Secondary metabolites of seaweeds are attributed to wide biological activities including antibacterial, antimycotic, antifouling, and antifungal. In this study, *Ulva* sp. (UL) from Tanjung Pengelih, Johor, was determined for its antifouling, antibacterial activity and characterized the metabolites involve. The methanolic crude extract (MCE) was prepared through maceration using methanol (1:10 w/v). Metabolites of MCE was profiled using thin layer chromatography (TLC). The crude extracts were determined for their antibacterial activity using disc diffusion method against Gram-positive (*Bacillus cereus*, *Streptococcus uberis*) and Gram-negative bacteria (*Pseudomonas aeruginosa*, *Escherichia coli*, *Vibrio parahaemolyticus*, *Salmonella* sp.). Then, it was screened using crystal violet assay against biofilm-forming bacteria, *P. aeruginosa* at concentration 0.25 mg/mL to 0.0078 mg/mL and was further tested in laboratory and field tests. The concentrations of MCE prepared for coated panels were 5% and 10% (w/v). As for field tests, panels were deployed at two sites (Tok Jembal Beach and Kuala Kemaman, Malaysia) for three months to monitor the fouler's growth. The MCE was fractionated using Liquid-Liquid Extraction (LLE) and Column Chromatography. Then, the isolated compound was characterized using Fourier Transform-Infrared Spectroscopy (FTIR), Nuclear Magnetic Resonance (NMR) and

Liquid Chromatography-Mass Spectrometry (LC-MS). The results showed growth of biofilm produced by *P. aeruginosa* was inhibited by MCE at concentrations of 0.0156 mg/mL. Besides, MCE inhibited the growth of two tested strains, *Salmonella* sp. and *V. parahaemolyticus* with moderate activity of antibacterial test. The aquarium test indicated UL 5% demonstrated a higher bacterial reduction of bacterial colonies with 1.903×10^6 CFU/mL better than blank paint (2.797×10^7 CFU/mL). According to the field test, crude panels of 5% were successful in reducing the settlement of fouling organisms due to less macrofouler growth compared to blank paint. The isolated compound A4 was identified as hexadecanoic acid ($C_{16}H_{32}O_2$) through NMR with molecular mass of 256 g/mol detected using LCMS. The characterization through FTIR obtained functional groups consist of CH_3, CH_2, C=O and OH. Therefore, *Ulva* sp. yields hexadecanoic acid as one of the promising eco-friendly antifouling agents from seaweeds.

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METABOLIT BIOAKTIF DARIPADA RUMPAI LAUT HIJAU DI MALAYSIA; *Ulva* sp. UNTUK AKTIVITI ANTILEKATAN

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Produk semula jadi marin terutamanya rumpai laut telah mendapat banyak perhatian dalam usaha memerangi isu biofouling yang berlaku dalam persekitaran marin. Metabolit sekunder rumpai laut dikaitkan dengan aktiviti yang luas seperti antibakteria, antimikotik, antilekatan dan antikulat. Dalam kajian ini, *Ulva* sp. dari Tanjung Pengelih Johor, telah ditentukan untuk aktiviti antilekatan, antibakteria dan ciri metabolit yang terlibat. Ekstrak mentah methanol (MCE) disediakan melalui proses rendaman menggunakan methanol (1:10 w/v). Metabolit MCE diprofilkan menggunakan kromatografi lapisan nipis (TLC). Ekstrak mentah telah ditentukan untuk antibakterianya menggunakan kaedah resapan cakera terhadap Gram-positif (*Bacillus cereus, Streptococcus* uberis) dan bakteria Gram-negatif (*Pseudomonas aeruginosa, Escherichia coli, Vibrio parahaemolyticus, Salmonella* sp.). Kemudian, ia disaring menggunakan ujian ungu hablur terhadap bakteria pembentuk biofilem, *P.aeruginosa* pada kepekatan 0.25 mg/mL hingga 0.0078 mg/mL dan seterusnya diuji pada ujian makmal dan lapangan. Kepekatan MCE yang disediakan untuk panel bersalut ialah 5% dan 10% (w/v). Bagi ujian lapangan, panel telah ditempatkan di dua tapak iaitu (Pantai Tok Jembal dan Kuala Kemaman, Malaysia) selama tiga bulan bagi memantau pertumbuhan fouler. MCE telah difraksinasi menggunakan pengestrakan cecair-cecair (LLE) dan kromatografi lajur. Kemudian, sebatian terpencil diperoleh dicirikan menggunakan Spektroskopi inframerah fourier

transformasi (FTIR), Resonans magnetic nuklear (NMR) dan Kromatografi cecair-spektrometri jisim (LC-MS). Keputusan menunjukkan pertumbuhan biofilem yang dihasilkan oleh *P. aeruginosa* telah dihalang oleh MCE pada kepekatan 0.0156 mg/mL. Selain itu, MCE menghalang pertumbuhan dua strain, *Salmonella* sp. dan *V. parahaemolyticus* dengan aktiviti sederhana ujian antibakteria. Ujian makmal (akuarium) untuk MCE pada UL 5% menunjukkan pengurangan bakteria koloni yang lebih tinggi dengan 1.903×10^6 CFU/mL lebih baik daripada cat kosong (2.797×10^7 CFU/mL). Mengikut ujian lapangan, panel ekstrak sebanyak 5% berjaya mengurangkan pertumbuhan organisma fouling kerana didapati pertumbuhan makrolekatan kurang berbanding cat kosong. Sebatian yang diperincikan telah dikenalpasti sebagai asid heksadekanoik ($C_{16}H_{32}O_2$) berdasarkan NMR dengan jisim molekul 256 g/mol dikesan menggunakan LCMS. Perincian melalui FTIR yang diperolehi menunjukkan Kumpulan berfungsi terdiri daripada CH_3, CH_2, C=O dan OH. Oleh itu, *Ulva* sp. menghasilkan asid heksadekanoik yang boleh bertindak sebagai agen antilekatan mesra alam daripada rumpai laut.