Liquid extracts of the brown seaweed *Ascophyllum nodosum* for the development of antimicrobial marine paints

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The transition toward more sustainable and environmentally friendly active chemical substances prompted the search for replacements of the banned tributyltin (TBT) (2008). TBT is a chemical often used as an antifouling agent in marine paints that, due to its toxicity, prevents the growth of algae, barnacles and other organisms from growing on the hull of a ship; however, when leached into the environment, TBTs exhibit an endocrine disruptive impact on marine organisms [1], [2], [3]. In the literature, antifouling, antimicrobial and antioxidant properties have been ascribed to certain seaweed compounds derived from brown seaweeds, including alginate, fucoidan, and fucoxanthins [4]. Hence, this study explored the potential of these compounds as antimicrobial agents for use in maritime paints. Several brown seaweeds (e.g., *Ascophyllum nodosum* and *Sargassum muticum*) were harnessed for component separation and extraction using a microwave-assisted extraction method [5]. A biphasic extraction system was used to separate the components of interest in the organic phase (e.g., polyphenols) and remove microorganism nurturing compounds (e.g., mannitol) via the water phase. Additionally, one-phase (organic solvent) extraction was also applied. Two organic solvents were investigated, which (i) have relatively low boiling points, (ii) are susceptible to microwave radiation, (iii) are immiscible with water and (iv) have good miscibility with common marine paints (e.g., Sigmacover 456). Both ethyl acetate (EthAc) and methyl isobutyl ketone (MIBK) were found to meet these criteria. First, seaweed extracts were produced according to the extraction method described in previous work, with a few modifications [6]. Briefly, a total volume of 400 mL (1:1 organic:water) was used, and 20 wt% dried seaweed powder was added to the mixture, which was extracted at 120°C for 15 min. Next, the extracts were concentrated by evaporation until 15 mL of organic solvent remained, after which the mixture was subsequently processed (16.7 v%) into a commercially available two-component resin marine paint (Sigmacover 456). The antimicrobial and antibiofilm characteristics of the samples were evaluated using a plating assay and an MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay on one gram-positive (*S. aureus*) and one gram-negative (*P. aeruginosa*) bacterium, respectively [7]. The results showed inhibitive effects on *S. aureus* (5-10 times fewer cells) treated with one-phase *Ascopyllum nodosum* and *Sargassum muticum* EthAc extracts; however, an antibiofilm effect was not demonstrated. Further research is needed to optimize the dose of the extract used to treat paints and elucidate the underlying mechanisms involved.

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