tumorigenic potential by stimulating DNA synthesis. Both autoradiograms and DNA labeling revealed a significant increase in DNA synthesis (approximately 4–6 fold) in the nuclei of neoplastic cells following endoperoxide plus MCA administration compared to that of MCA alone, or control epidermal nuclei. It is also possible that endoperoxide analogs and thromboxanes accelerate the tumor cell growth by modulating the basal tumor cell cAMP levels[11]. Ultrastructurally, these are poorly differentiated and more invasive neoplastic cells.

The experiments described here conclusively implicated the endoperoxide analogs U-46619 and U-44069 in the control of tumor cell proliferation and function. Thus, research regarding the role of prostaglandins and their intermediary products, endoperoxide analogs, became an exciting field in oncology.

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The sperm attractant of *Hormosira banksii* (Phaeophyceae, Fucales), a seaweed common to Australia and New Zealand[1]

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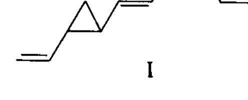
Summary. Hormosira banksii is a taxonomically isolated brown seaweed endemic to Australia and New Zealand. The sperm attractant of this species has been isolated and identified as trans-1-vinyl-2-(1*E*, 3*Z*-hexadienyl)-cyclopropane (**I**) (hormosirene). *Hormosira* is the first organism in which a cyclopropane derivative has been found to act as a hormone in sexual reproduction. The implication of this finding in relation to phylogeny and phytogeography is discussed.

Chemical communication during sexual reproduction is a well-known and widespread phenomenon in lower plants[3]. Within the last 10 years marine brown algae have proved to be exceedingly well suited to the study of this problem, and the chemical structures of seven sex hormone factors have been identified recently in joint efforts by biologists and chemists[4,5]. Monocyclic molecular structures have been found in the genera *Ectocarpus*[6], *Cutleria*[7], *Dictyota*[8], *Desmarestia*[9], and *Syringoderma*[10]. Linar conjugated olefines with chain lengths of 8 and 11 carbon atoms have been identified in the respective genera *Fucus*[11] and *Ascophyllum*[4] which belong to the family Fucaceae within the order Fucales. Their straight-chain character and identical stereochemistry reflect a close taxonomic or phylogenetic relationship[8,12]. All organisms used for this previous work were marine brown algae, mainly from the North Atlantic.

The temperate coasts of Southern Australia have an extremely rich marine algal flora which differs significantly from that of the northern hemisphere. This region is the center of diversity of the most advanced group of brown algae, the order Fucales, and the region from which they are believed to have originated and spread. For this reason it seemed desirable to include species from this area in our studies. This paper reports the identification of the sex attractant in a second family of the order Fucales, the Hormosiraceae.

Hormosira banksii (Turner) Decaisne is endemic to Southern Australia and New Zealand. It is the dominant fucalean species in the lower eulittoral zone of many shores. It is dioecious, and in contrast to many other species in the order Fucales, *Hormosira* plants are fertile throughout the year in most localities. Reproduction is oogamous. Antheridia and oogonia exude on to the surface when the thalli are emersed at low tide. Upon contact with seawater the oogonial membranes dissolve and liberate 4 eggs per oogonium. Each antheridium releases 64 spermatozoids, which swarm around the eggs in large numbers. Gamete release and fertilization have been described in detail by Osborn[13], Levring[14], and Forbes and Hallam[15].

Materials and methods. Hormosira plants were collected from Sorrento and Point Lonsdale, Victoria (Australia), and stored at +4 °C until required. Individual plants were rinsed with cold seawater and allowed to dry at room temperature for a few minutes until the gametangia began to exude. Female thalli were then immersed in cold seawater and agitated to obtain a suspension of eggs. Batches of eggs amounting to about 5 ml volume were added to 2 l of seawater in an extraction flask. Volatile compounds were extracted by the closed-loop stripping technique[16], and adsorbed on a filter consisting of 2 mg activated carbon. After extraction periods of 24 h the filters were eluted with 30 µl of dichloromethane. These extracts were then subjected to further analysis by glass-capillary gas chromatography and mass spectrometry.



I

Standard gas chromatography with heated injection-ports (200 °C) revealed 2 substances in the extract with evidence of thermic instability and decomposition during injection. This difficulty was overcome by using the on-column injection technique[17] which avoids heating of the sample during injection. Application of this improved technique revealed a single compound in the eluates from *Hormosira* eggs. Bio-assays with this substance recondensed after gas-chromatographic separation confirmed that it represents the spermatozoid-attractant of *Hormosira.* In 9 trials the average yield of this compound in 1 h was 7.3 ± 2.0 ng per 10^6 eggs.

Bio-assays were carried out using the fluorocarbon solvent FC-72[18]. This liquid is biologically inert and forms spherical droplets when placed on the base of a well slide filled with a suspension of *Hormosira* spermatozoids in seawater. Microscopic observation under dark-field illumination followed after 3–4 min. Droplets of the pure solvent were ineffective, whereas droplets containing active concentrations of attractants showed massive accumulations of spermatozoids comparable to the effect of genuine eggs.

Results. Analytical data are presented in the table. Molecular weight and absence of hetero-atoms indicated an olefinic hydrocarbon of the composition $C_{11}H_{16}$. The thermal instability and re-arrangement of the molecule lead to the formation of ectocarpene, a 7-membered ring system, as revealed by capillary gas-chromatographic analysis. Based on this information several cyclopropane derivatives were synthesized. Comparison of their retention indices and mass spectra with those of the *Hormosira* egg product demonstrated its identity with trans-1-vinyl-2-(1*E*,3*Z*-hexadienyl)-cyclopropane (**I**). Furthermore, synthetic **I** showed attractivity to *Hormosira* spermatozoids down to a concentration of 10^{-6} M in FC-72, which corresponds to less than 2×10^{-9} M in the aqueous phase. This is comparable to the threshold concentrations of other brown algal pheromones. Thus **I** is the spermatozoid attractant of *Hormosira banksii.* This substance was first described by Pettus and Moore[19] as a component of the essential oil obtained from vegetative thalli of the brown alga *Dictyopteris plagiogramma* and named 'dictyopterene B'. Kajiwara et al.[20] found it in thallus extracts of the Japanese species *D. prolifera* and *D. undulata* but did not deal with any biological activity. Moore[21] used the term dictyopterene for various cyclopropane and cycloheptadiene derivatives of the *Dictyopteris* extract sub-classifying them by capital letters and primes. Since dictyopterene B now has been shown to act as a highly specific sexual hormone in *Hormosira* we may suggest 'hormosirene' as a more fitting trivial name in line with those for other seaweed sex-attractants.

Discussion. Eight sexual pheromones, including hormosirene, the spermatozoid attractant of *Hormosira banksii* are now identified. The molecular structure of hormosirene corresponds to the general features which have emerged: brown algal sexual pheromones are olefinic hydrocarbons with a C_{11} skeleton. Only 2 exceptions from this rule have been encountered so far: the attractant in the genus *Fucus* with a straight-chain C_8 molecules, and the *Laminaria* pheromone with a C_{11} ring structure and oxygen as a hetero-atom bound in a still unknown manner[22]. Considering structural details of the previously known pheromones, it appeared that straight-chain hydrocarbons as found in *Fucus* and *Ascophyllum* were restricted to the order Fucales and that monocyclic 5- and 7-membered ring systems characterize the rest of the Phaeophyceae. This pattern agreed with taxonomic considerations suggesting that the order Fucales forms a distinct sub-class, the Cyclosporidae which separated very early in evolution from the Phaeophyceae (sub-class Phaeophycidae[23]). Hormosirene with its 3-membered ring does not fit the hypothesis that straight-chain molecules are characteristic of the order Fucales. The cyclopropane ring represents a unique structural feature in a brown algal sexual pheromone. This differs significantly from findings obtained so far in northern hemisphere organisms. It should, however, be kept in mind that our knowledge in this field is still very incomplete. Only 2 families of the order Fucales have been studied at present. There remain 5 more families in this order, 2 of which are endemic to the southern hemisphere. Further studies on pheromone systems in these families will examine the relationship of chemical structures to phytogeographic, phylogenetic, and taxonomic distinctions in the brown algae.

According to Moore[21] vegetative thalli of *Dictyopteris plagiogramma* and *D. australis* (Dictyotales) secrete a large number of low-molecular volatile compounds termed 'dictyopterenes' in addition to (**I**), among them the hydrocarbons used as pheromones in genera of the orders *Ectocarpus* (Ectocarpales), *Dictyota* (Dictyotales), and *Ascophyllum* (Fucales). This may indicate that various compounds from the general metabolic pool have evolved independently as sexual pheromones in various groups of brown algae.

Retention indices (70 ± 0.1 °C), and mass fragments of the natural and synthetic hormosirene

Liquid phase	Hormosirene Natural	Synthetic
Silicone SE 54_i*	1177.96 ± 0.15	1177.99 ± 0.08
Silicone OV 1701_i*	1232.31 ± 0.14	1232.44 ± 0.09
Mass/charge	148 (M^+, 4%), 119 (12%), 105 (17%), 91 (74%), 79 (100%), 66 (46%)	

*'i' stands for immobilized liquid phase.

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