ELSEVIER

Journal of Experimental Marine Biology and Ecology, 238 (1999) 127–137

JOURNAL OF EXPERIMENTAL MARINE BIOLOGY AND ECOLOGY

Chemical defence of the soft coral *Parerythropodium fulvum fulvum* (Forskål) in the Red Sea against generalist reef fish

D. Kelman[a,*], Y. Benayahu[a], Y. Kashman[b]

[a]*Department of Zoology, George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel*

[b]*School of Chemistry, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel*

Received 6 May 1998; received in revised form 13 November 1998; accepted 13 January 1999

Abstract

Laboratory feeding assays comparing chemical and sclerite deterrence capabilities of *Parerythropodium fulvum fulvum* revealed that the organic extract deterred feeding by the generalist reef fish *Thalassoma klunzingeri* (Fowler and Steinitz) and *T. lunare* (Linnaeus), whereas the sclerites were palatable. The mean number of pellets, containing natural extract concentration as in the living coral, eaten by the test fish was 0.25 ± 0.43, while the mean number of sclerite pellets was 7.0 ± 1.58 out of 10 pellets offered. Extracts of the two colour morphs of the studied species taken from colonies from both shallow and deep reefs deterred feeding by the wrasses even at concentrations as low as 12.5% of the natural concentration present in the coral. Feeding experiments using extracts of embryos of the *P. f. fulvum* yellow morph revealed that they are chemically protected against predation. A higher level of deterrence was found with extracts of embryos combined with the mucus in which they are embedded. The present study shows that effective defence against predation in the surface-brooded embryos of *P. f. fulvum* is accomplished by the aggregation of chemically defended embryos, embedded within mucus possessing predator repellent properties; and by the close proximity of the brood to the chemically defended parent colony.

Keywords: Chemical defence; Embryos; Feeding deterrence; Octocorallia; *Parerythropodium f fulvum*; Red Sea

1. Introduction

Soft corals (Cnidaria, Alcyonacea) are among the major benthic components

*Corresponding author. Tel.: +972-3-640-9809; fax: +972-3-640-9403.
E-mail address: kelmand@post.tau.ac.il (D. Kelman)

0022-0981/99/$ – see front matter
PII: S0022-0981(99)00016-7

occupying space in the tropical Indo-Pacific reefs (Dinesen, 1983; Huston, 1985), as well as in the coral reefs of the northern Gulf of Eilat, Red Sea (Benayahu and Loya, 1977). Their evolutionary success in areas of high levels of predation has been attributed to their production of significant amounts of secondary metabolites, especially terpenes (Tursch et al., 1978; Sammarco and Coll, 1988; 1992), many of which show predator-deterrence activity (Coll et al., 1982; Pawlik et al., 1987; Van Alstyne et al., 1994). Octocorals, however, also possess mineral-hardened sclerites which could potentially serve as an antipredator defence mechanism (Sammarco et al., 1987; Harvell et al., 1988; Harvell and Fenical, 1989). These structural skeletal components are common among a number of invertebrates, including sponges, cnidarians, platyhelminths, molluscs, echinoderms and ascidians (Kingsley, 1984). Alcyonacean sclerites are extremely variable in morphology and size, and are frequently used as a taxonomic tool (Bayer et al., 1983). For sessile marine invertebrates, sclerites and spicules appear to play an important role in colony support (Lewis and VonWallis, 1991), but their defence function is debatable. For example, Harvell et al. (1988) demonstrated that adding sclerites from the gorgonian *Pseudopterogorgia acerosa* to food strips reduced their consumption by reef-fish in field assays in Belize; while Van Alstyne et al. (1994) have demonstrated the importance of both chemical and structural defences by sclerites of three species of the soft coral *Sinularia* from Guam. However, sclerites of the sea whip *Junceela* sp. did not deter feeding of carnivorous reef-fish (reported in Van Alstyne and Paul, 1992). Studies on other benthic invertebrates, such as sponges (Pawlik et al., 1995) and ascidians (Lindquist et al., 1992) have shown no effect of spicules on fish feeding behaviour.

Larval chemical defence has been demonstrated in several invertebrate groups (Lucas et al., 1979; Young and Bingham, 1979; Lindquist et al., 1992; Lindquist and Hay, 1995; 1996; McClintock and Baker, 1997). These studies have shown that unpalatable larvae are common in a wide assortment of sessile invertebrates, which are known to be chemically rich. These findings suggest that the ability of adult colonies of sessile invertebrates to produce noxious compounds is coupled with the species′ ability to chemically defend its larval stages (Lindquist and Hay, 1996). However, in some species where adult colonies were shown to be palatable to fish, their larval stages were found to be unpalatable (Lindquist and Hay, 1996). Octocorals that brood their sexually produced offspring do so either on their external surfaces or, more commonly, within their internal cavities. External brooders, such as *Briareum asbestinum* (see: Brazeau and Lasker, 1990), *Paramuricea clavata* (see: Coma et al., 1995), *Parerythropodium fulvum fulvum* (see: Benayahu and Loya, 1983) and *Clavularia hamra* (see: Benayahu, 1989) produce large numbers of developing larvae that adhere to the surface of the parent colony for the first 3–6 days after fertilisation, providing an easy target for potential predators. Larval protection is thus essential (Lindquist and Hay, 1996).

Parerythropodium fulvum fulvum is an encrusting soft coral commonly found between 3–40 m deep among the coral reefs of the Gulf of Eilat, Red Sea (Benayahu and Loya, 1977). Two colour morphs exist: yellow-brown and grey, with no taxonomic differences between them (Verseveldt, 1969). The yellow morph is found at a wide range of depths, while the grey one is common usually below 20 m (Benayahu and Loya, 1983). The species is gonochoric and reproduces annually from the end of June to the beginning of

August (summer), by a unique mode of reproduction that involves the surface brooding of embryos in a layer of mucus (Benayahu and Loya, 1983). The development of these embryos to a mature planula-stage lasts 6 days, at which time they detach from the colony surface and sink to the reef bottom. It has been suggested that surface brooding provides protection for the embryos during their development (Benayahu and Loya, 1983). Like many soft corals, *P. f. fulvum* contains a rich assemblage of secondary metabolites (Green et al., 1992), mainly comprised of sesquiterpenes such as the volatile dye, fulfulvene, which gives the living colony its yellow colour. The biological function of these metabolites is still unknown.

In the present study we examined the palatability to two species of reef fish, *Thalassoma klunzingeri* and *T. lunare*, of organic extracts and sclerites of the two colour morphs of *P. f. fulvum*, as well as of organic extracts of embryos and of the mucus in which they are embedded, to determine whether chemical defence of adult colonies of this species is coupled with defence of its larval stages.

2. Materials and methods

2.1. Collection and extraction

Samples from the yellow and grey morphs of *P. f. fulvum* were collected from shallow (3–10 m) and deep (25–30 m) reefs across from the Marine Biological Laboratory (MBL) at Eilat. Larval stages were collected only from colonies of the yellow morph during the breeding season (July to August 1996). Brooded embryos with mucus were collected from the surface of female colonies using a 50 ml syringe. In the laboratory, some of the embryos were washed twice with filtered seawater to separate them from the mucus, while others were left embedded in the mucus. Additional colonies were collected from the same locations in October 1996 for extraction of sclerites. All samples were frozen at $-70^{\circ}C$, transported on ice to Tel Aviv and kept at $-20^{\circ}C$ for further use in assays of feeding deterrence.

The volume of each coral sample (at least three samples for each colour morph and depth) was measured by water displacement using a cylinder, in order to calculate the natural volumetric concentration of the organic extract in the living coral. The samples were then cut into small pieces, sonicated for 5 min and extracted in dichloromethane for 24 h at room temperature. The organic extracts were separated from the phase using a 20 ml separation funnel, filtered, and the solvent removed by rotary evaporator under vacuum at 10–15°C bath temperature. This procedure was employed in order to avoid evaporation of the volatile compounds known for *P. f. fulvum* (see: Green et al., 1992). The embryos and mucus were similarly extracted. All extracts were weighed and kept at $-20^{\circ}C$ prior to the deterrence assays.

2.2. Extraction of sclerites

The volume of 10 coral samples was also determined by water displacement, and the sclerite content was obtained by placing the frozen colonies in 50 ml tubes filled with

sodium hypochlorite (5.25%). After the solution stopped bubbling (1–5 h), the supernatant was carefully decanted and fresh bleach was added. This process was repeated until the addition of fresh bleach resulted in no further bubbling (usually 2–3 treatments). After final treatment, a pellet of sclerites remained on the bottom of the tube, and the bleach was decanted. The pellet was then rinsed 3 times with distilled water, and the water replaced with a 1 M solution of sodium thiosulfate to neutralise any residual bleach. After 10–15 min the sclerites were again rinsed 3 times with distilled water, dried by heating (100°C), weighed, and used for the feeding deterrence assays (see below).

2.3. Preparation of food pellets

Food pellets were prepared according to Pawlik et al. (1995). To each vial containing concentrated extract from 10 ml coral tissue dissolved in 0.5 ml ethanol, a mixture of 0.3 g alginic acid (sodium salt, low viscosity, Sigma) and 0.5 g of freeze-dried powdered brine shrimp was added with distilled water to yield a final volume of 10 ml. The volume of the coral sample was determined prior to extraction and the natural extract concentration of 28 mg/ml was used in these assays. Because it was difficult to volumetrically determine the concentration of extract for the embryos, and particularly for the mucus in which they are embedded since the latter contains mostly water, we added extracts of embryos or embryos with mucus at a volumetric concentration corresponding to that of the adult colonies. As these embryos are surface brooded, we considered this concentration to be a close approximation to that of the embryonic stages plus mucus. The mixture was vigorously stirred until homogenised, and then loaded into a 10-ml syringe. The syringe tip was submerged in a 0.25 M $CaCl_2$ solution, and its contents emptied, forming a spaghetti-like strand. A few minutes later the hardened strand was removed, rinsed in seawater and chopped into 4 mm long pellets. Sclerite pellets were made in the same manner, using natural concentration (wt/v) of sclerites (0.216 g/ml) instead of extract. Control pellets were similarly made, with the addition of the same concentration of ethanol, but without extract or sclerites.

2.4. Collection and maintenance of fish

The wrasses *Thalassoma klunzingeri* and *T. lunare* were caught at the MBL reef with hand nets, using live brine shrimp as bait, at depths of 2–5 m. One to two specimens of either *T. klunzingeri* or *T. lunare* were placed in each of the 75 l aquaria supplied with flowing seawater. A total of 12 test fish were used in these assays, six of each species. The fish quickly acclimated and began accepting control food pellets (see above).

2.5. Assays for deterrence to fish in the laboratory

Aquarium assays were performed as described by Pawlik and Fenical (1992). Test fish, consisting of 1–2 fish of the same species, were randomly chosen and offered either treated or control food pellets, followed by a randomly chosen second pellet. If the pellet offered was a treated one, and rejected by the fish, a control was offered to determine

whether the fish had simply ceased feeding. Test fish that would not eat control pellets were not included in the assays. A pellet was considered rejected if not eaten by one or more fish after a minimum of three feeding attempts, or if the pellet was approached and ignored after one such attempt. Each trial set consisted of 10 treated and 10 control pellets and the number of pellets consumed was recorded. The significance of differences in consumption of treated vs control pellets and between various treatments, was evaluated using Fisher exact tests and R × C tests of independence using G-statistic (Sokal and Rohlf, 1995).

3. Results

Our laboratory feeding experiments comparing the chemical and sclerite deterrence capabilities of *P. f. fulvum* revealed that the organic extract deterred feeding by the generalist reef fish *T. klunzingeri* and *T. lunare*, while the sclerites were palatable to them. The mean number of extract pellets, containing the same natural concentration found in the living coral, eaten by the test fish was 0.25 ± 0.43 ($N = 4$ trial sets), while the mean number of sclerite pellets eaten was 7.0 ± 1.58 ($N = 4$ trial sets) out of 10 pellets offered. The difference in deterrence was significant ($p < 0.001$, Fisher exact test). Comparison of chemical deterrence among different colour morphs of the coral and colonies from different depths resulted in a similar deterrence of 0.7 ± 0.9 pellets out of 10 (Fig. 1).

Comparison of chemical deterrence among different concentrations of extract yielded varying results ($p < 0.001$, R × C test of independence using G-statistic); however all

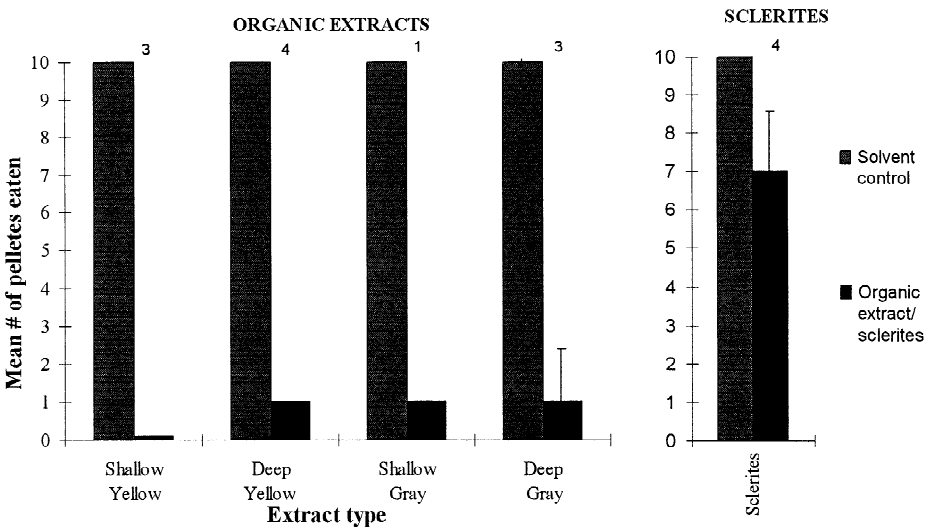

Fig. 1. Mean number of treated pellets, containing various extracts or sclerites of *Parerythropodium fulvum fulvum* and solvent controls eaten by the wrasses *Thalassoma klunzingeri* and *T. lunare*. Number of trial sets is indicated above bars.

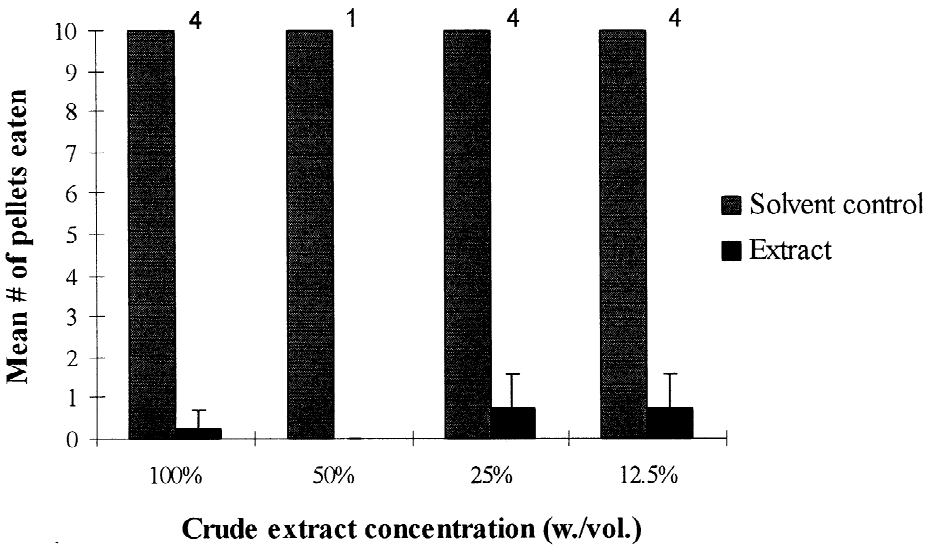

Fig. 2. Mean number of treated pellets, containing different concentrations (% of natural extract concentration) of extract of *Parerythropodium fulvum fulvum* and solvent controls eaten by the wrasses *Thalassoma klunzingeri* and *T. lunare*. Number of trial sets is indicated above bars.

concentrations of extract were highly unpalatable (Fig. 2). Feeding experiments performed on different extract concentrations of the embryo crude-extract revealed that the natural extract concentration was unpalatable to the test fish, but the level of deterrence decreased with decreasing concentration of extract (Fig. 3). A similar pattern was found for the embryos combined with mucus extracts (Fig. 4), which however, exhibited higher deterrence. Comparison of deterrence of natural extract concentrations of the coral, of embryos and of embryos combined with mucus, showed no significant differences. Comparison performed with concentration of 12.5% of the natural extract concentrations of these extracts revealed highly significant differences ($p < 0.001$).

4. Discussion

Our data from the feeding experiments, using the same natural extract and sclerite concentrations found in the living colonies, revealed that *P. f. fulvum* is protected from generalist predatory reef-fish by an effective chemical defence, rather than by its sclerites. This chemical defence is present in both colour morphs and in colonies from both shallow and deep reefs. The deterrence was consistent even when the extract concentration in the test pellets was decreased to as low as 12.5% of the natural concentration present in the coral. The merits of using *Thalassoma* fish for aquarium bioassays have been detailed by Pawlik et al. (1987). These fish are highly abundant on Red Sea reefs and are generalist predators, known to feed on a wide assortment of benthic invertebrates (Randall, 1983). They were chosen for the present study because

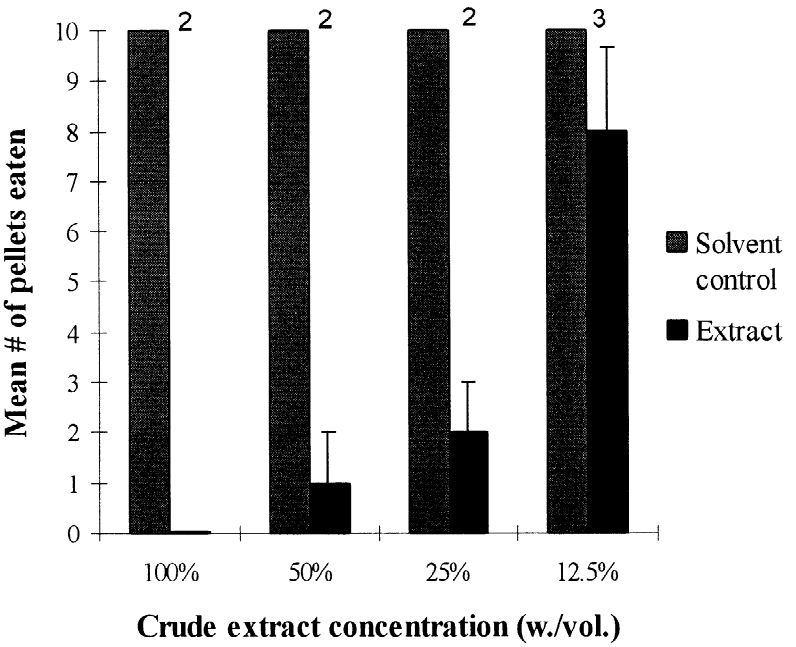

Fig. 3. Mean number of treated pellets, containing different concentrations (% of natural extract concentration) of extract of *Parerythropodium fulvum fulvum* embryos and solvent controls eaten by the wrasses *Thalassoma klunzingeri* and *T. lunare*. Number of trial sets is indicated above bars.

they represent the majority of predatory reef fish and thus antipredatory defences are expected to be directed against them in particular. Such fish are less likely to have evolved mechanisms to circumvent these defences than specialist predators (Pawlik et al., 1995).

Predation on reef octocorals is limited to a few species of specialised invertebrates and fishes (Sammarco and Coll, 1988). The gastropod *Ovula ovum* has been reported to prey upon *Sarcophyton* (see: Coll et al., 1983); the butterflyfish *Chaetodon melannotus* and *C. unimaculatus* upon *Sinularia* (see: Wylie and Paul, 1989; Alino et al., 1992) and *C. capistratus* upon Caribbean gorgonian octocorals (Lasker, 1985). These highly specialised predators are capable of consuming their toxic prey without any ill effects through adaptation such as detoxification of the noxious compounds (e.g., Coll et al., 1983). However, many octocorals and sponges have been reported to chemically deter generalist predatory reef-fish (e.g., Pawlik et al., 1987; Harvell et al., 1988; Fenical and Pawlik, 1991; Pawlik and Fenical, 1992; Van Alstyne et al., 1994; Pawlik et al., 1995). While it appears that *P. f. fulvum* is protected against generalist predatory reef fish, such as *Thalasoma* species, it is possible that this soft coral, like some other Red Sea species, is still preyed upon by specialists, such as the fishes *Pomacentrus trichourus*, *Amblyglyphidodon leucogaster* and *A. flavilatus* (A. Meroz, personal communication). Further feeding deterrence experiments are required in order to determine the extent to which *P. f. fulvum* is protected from a range of other reef fish.

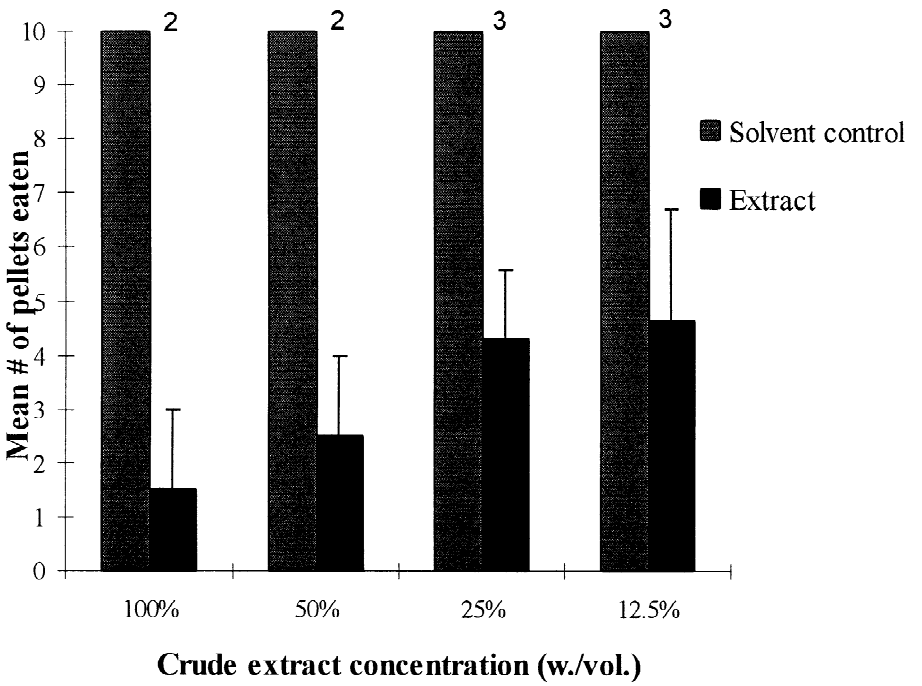

Fig. 4. Mean number of treated pellets, containing different concentrations (% of natural extract concentration) of extract of *Parerythropodium fulvum fulvum* embryos and mucus and solvent controls eaten by the wrasses *Thalassoma klunzingeri* and *T. lunare*. Number of trial sets is indicated above bars.

Sclerites of *P. f. fulvum* did not deter feeding by the fish when tested with food pellets containing volumetrically equivalent concentrations to those in living coral tissue. Octocoral sclerites and sponge spicules may share some similar characteristics, such as morphology, concentration found in the tissues, or size range, but may differ in their ability to deter predation (Chanas and Pawlik, 1996). Sclerites of the octocorals *Leptogorgia virgulata*, *Pseudopterogorgia acerosa*, *Gorgonia ventalina* and *Sinularia* spp. deterred feeding at their natural concentration, which ranged from 31–82% of total tissue dry weight (Gerhart et al., 1988; Harvell et al., 1988; Van Alstyne and Paul, 1992; Van Alstyne et al., 1992). A further study on *Sinularia* spp. by Van Alstyne et al., 1994 attempted to determine the relative importance of chemical and physical defences at different colony parts. It was found that sclerites, assayed at concentrations found in the colony tips, which were as low as 31–47% of their dry weight, did not deter feeding. Similarly, sclerites of the sea whip *Junceela* sp. did not deter feeding at a natural concentration which was ~45% of dry weight (reported in Van Alstyne et al., 1992). A recent comprehensive study was undertaken by Chanas and Pawlik (1995) to determine the palatability of spicules of 71 species of Caribbean sponges. They found that sponge spicules did not deter feeding by generalist fish at their natural concentration, which ranged between 3–49% of dry weight, and concluded that the difference in palatability between sponge spicules and octocoral sclerites was due to differences in their chemical

composition, rather than in concentration (Chanas and Pawlik, 1995; 1996). In the present study, sclerites of *P. f. fulvum* did not deter feeding at their natural concentration, previously found to be 79.9±5.3% ($N = 30$) of dry weight (Kelman, 1998). It can therefore be concluded that the concentration of sclerites in *P. f. fulvum* does not play a significant role in deterrence. Sclerites, however, are necessary in octocorals for structural support (Lewis and VonWallis, 1991). It appears that in *P. f. fulvum* the primary function of the sclerites is thus structural rather than defensive, and that defence is accomplished by the rich content of secondary metabolites.

The current feeding experiments using extracts of yellow morph *P. f. fulvum* embryos demonstrated that the early developmental stages are chemically protected against predation. As described earlier (see Methods section), we prepared the embryo extract-pellets at a natural concentration of 28 mg/ml as determined for the adult colonies. Considering that each embryo yields ca. 16.74 μg of organic extract (Kelman, 1998), the concentration of 28 mg/ml of extract corresponds to 1672 embryos. In our study, each ml of prepared artificial food comprised 15 pellets, each containing extract corresponding to 110 embryos. At this concentration the embryos were found to be unpalatable. Feeding deterrence of embryos declined with decreasing concentration of extract in the pellets (Fig. 3), which were found to be palatable at a concentration of 12.5% of its natural level, corresponding to around 14 embryos. We therefore conclude that a few embryos alone might not be sufficiently chemically defended and that it is their aggregation which provides protection from generalist predatory fish. This assumption was also confirmed by field observations when a few embryos were released into the water column and were readily consumed by reef fish (Kelman, personal observations).

A similar level of deterrence was obtained with feeding experiments using extracts of embryos combined with the mucus in which they were embedded, at the same concentration as the mucus-free embryos. However, when decreasing the concentration of extract to 12.5% of its natural level (Fig. 4), a significantly higher deterrence was achieved with the extract of embryos plus mucus than with the mucus-free extract at the same concentration. This suggests that compounds present in the mucus may enhance embryo protection from predation. Benayahu and Loya (1983) suggested that surface brooding provides protection for *P. f. fulvum* embryos during their development. The present study demonstrates that effective defence against predation in the surface brooded embryos of *P. f. fulvum* is accomplished by the aggregation of chemically defended embryos, embedded within mucus possessing anti-predatory properties, and by the close proximity of the brood to the chemically defended parent colony.

Acknowledgements

We acknowledge the insightful critiques on the manuscript by M. Ilan and by the anonymous reviewers. We would like to thank A. Meroz and D. Goulet for their assistance with the fish collection and advice. We thank the staff of the Interuniversity Institute of Marine Biology at Eilat for their hospitality and facilities. We thank D. Wool for statistical advice, and N. Paz and C. Shapiro for editorial assistance. This paper is

part of a thesis presented as partial fulfilment of the M.Sc. dissertation submitted by D.K. to Tel-Aviv University. This research was supported in part by a grant to Y.B. from the Israel Science Foundation.

References

Alino, P.M., Coll, J.C., Sammarco, P.W., 1992. Toxic prey discrimination in a highly specialised predator *Chaetodon melannotus* (Block et Schneider): visual vs. chemical cues. J Exp Mar Biol Ecol 164, 209–220.

Bayer, F.M., Grasshoff, M., Verseveldt, J., Brill, E.J., 1983. Illustrated trilingual glossary of morphological and anatomical terms applied to Octocorallia. Leiden.

Benayahu, Y., Loya, Y., 1977. Space partitioning by stony corals soft corals and bethnic algae on the northern Gulf of Eilat (Red Sea). Helgol Wiss Meeresunters 30, 362–382.

Benayahu, Y., Loya, Y., 1983. Surface brooding in the Red Sea soft coral *Paerythropodium fulvum fulvum* (Forskål). Biol Bull 165, 353–369.

Benayahu, Y., 1989. Reproductive cycle and developmental processes during embryogenesis of *Clavularia hamra* (Cnidaria, Octocorallia). Acta Zoologica 70 (1), 29–36.

Brazeau, D.A., Lasker, H.R., 1990. Sexual reproduction and external brooding by the Caribbean gorgonian *Briareum asbestinum*. Mar Biol 104, 465–474.

Chanas, B., Pawlik, J.R., 1995. Defenses of Caribbean sponges against predatory reef fish. II. Spicules tissue toughness and nutritional quality. Mar Ecol Prog Ser 127, 195–211.

Chanas, B., Pawlik, H.R., 1996. Does the skeleton of a sponge provide a defense against predatory reef fish ? Oceologia 107, 225–231.

Coll, J.C., La Barre, S., Sammarco, P.W., Williams, W.T., Bakus, G.J., 1982. Chemical defences in soft corals (Coelenterata: Octocorallia) of the Great Barrier Reef: A study of comparative toxicities. Mar Ecol Prog Ser 8, 271–278.

Coll, J.C., Tapiolas, D.M., Bowden, B.F., Webb, L., Marsh, H., 1983. Transformation of soft coral (Coelenterata: Octocorallia) terpenes by *Ovula ovum* (Mollusca: Prosobranchia). Mar Biol 74, 35–40.

Coma, R., Ribes, M., Zabala, M., Gili, J.M., 1995. Reproduction and cycle of gonadal development in the Mediterranean gorgonian *Paramuricea clavata*. Mar Ecol Prog Ser 117, 173–183.

Dinesen, Z.W., 1983. Patterns in the distribution of soft corals across the central Great Barrier Reef. Coral Reefs 1, 229–236.

Fenical, W., Pawlik, J.R., 1991. Defensive properties of secondary metabolites from the Caribbean gorgonian coral, *Erythropodium caribaeorum*. Mar Ecol Prog Ser 75, 1–8.

Gerhart, D.J., Rittschof, D., Mayo, S.W., 1988. Chemical ecology and the search for marine antifoulants studies of a predator prey-symbosis. J Chem Ecol 14, 1905–1917.

Green, D., Kashman, Y., Benayahu, Y., 1992. Secondary metabolites of the yellow and grey morphs of the soft coral *Parerythropodium fulvum fulvum*: comparative aspects. J Nat Prod 55 (9), 1186–1196.

Harvell, C.D., Fenical, W., Greene, C.H., 1988. Chemical and structural defenses of Caribbean gorgonians (*Pseudopterogorgia* spp.) I. Development of an in situ feeding assay. Mar Ecol Prog Ser 49, 287–294.

Harvell, C.D., Fenical, W., 1989. Chemical and structural defenses of Caribbean gorgonians (*Pseudopterogorgia* spp.) Intracolony localisation of defense. Limnol Oceanogr 34 (2), 382–389.

Huston, M.A., 1985. Patterns of species diversity on coral reefs. A Rev Ecol Syst 16, 149–177.

Kelman, D., 1998. The chemical ecology of the soft coral *Paerythropodium fulvum fulvum*. M.Sc. Thesis, Tel-Aviv University, Tel Aviv, Israel.

Kingsley, R.J., 1984. Spicule formation in the invertebrates with special reference to the gorgonian *Leptogorgia virgulata*. Am Zool 24, 883–891.

Lasker, H.R., 1985. Prey preferences and browsing pressure of the butterflyfish *Chaetodon capistratus* on Caribbean gorgonians. Mar Ecol Prog Ser 21, 213–220.

Lewis, J.C., VonWallis, E., 1991. The function of surface sclerites in gorgonians (Coelentrata, Octocorallia). Biol Bull 181, 275–288.

Lindquist, N., Hay, M.E., Fenical, W., 1992. Defense of ascidians and their conspicuous larvae: adult vs larval chemical defenses. Ecol Monogr 62, 547–569.

Lindquist, N., Hay, M.E., 1995. Can small rare prey be chemically defended? The case for marine larvae. Ecology 76, 1347–1358.

Lindquist, N., Hay, M.E., 1996. Palatability and chemical defense of marine invertebrate larvae. Ecol Monogr 66 (4), 431–450.

Lucas, J.S., Hart, R.J., Howden, M.E., Salathe, R., 1979. Saponins in eggs and larvae of *Acathaster planci* (L.) Asteroidea as chemical defences against planktivorous fish. J Exp Mar Biol Ecol 40, 155–165.

McClintock, J.B., Baker, B.J., 1997. Palatability and chemical defense of eggs, embryos and larvae of shallow water Antarctic marine invertebrates. Mar Ecol Prog Ser 154, 121–131.

Pawlik, J.R., Burch, M.T., Fenical, W., 1987. Patterns of chemical defense among Caribbean gorgonian corals: a preliminary survey. J Exp Mar Biol Ecol 108, 55–66.

Pawlik, J.R., Fenical, W., 1992. Chemical defense of *Pterogorgia anceps* a Caribbean gorgonian coral. Mar Ecol Prog Ser 87, 183–188.

Pawlik, J.R., Chanas, B., Toonen, R.J., Fenical, W., 1995. Defenses of Caribbean sponges against predatory reef fish. I. Chemical deterrency. Mar Ecol Prog Ser 127, 183–194.

Randall, J.E., 1983. Red Sea reef fishes, Immel Publishing, London.

Sammarco, P.W., La Barre, S., Coll, J.C., 1987. Defensive strategies of soft corals (Coelenterata: Octocorallia) of the Great Barrier Reef. III. The relationship between ichthyotoxicity and morphology. Oceologia 74, 93–101.

Sammarco, P.W., Coll, J.C., 1988. The chemical ecology of alcyonarian corals (Coelenterata: Octocorallia). In: Scheuer, P.J. (Ed.), Bioorganic marine chemistry, Springer-Verlag, Berlin, pp. 87–116.

Sammarco, P.W., Coll, J.C., 1992. Chemical adaptations in the Octocorallia: evolutionary considerations. Mar Ecol Prog Ser 88, 93–104.

Sokal, R.R., Rohlf, F.J., 1995. Biometry. the principle and practice of statistics in biological research, 3rd ed, W.H. Freeman and Co, New York.

Tursch, B., Braekman, J.C., Daloze, D., Kaisin, M., 1978. Terpenoids from coelenterates. In: Scheuer, P.J. (Ed.), Marine Natural Products Chemical and Biological Perspectives, Academic Press, New York, pp. 247–296.

Van Alstyne, K.L., Paul, V.J., 1992. Chemical and structural antipredator deterrents in the sea fan *Gorgonia ventalina* effects against generalist and specialist predators. Coral Reefs 11, 155–160.

Van Alstyne, K.L., Wylie, C.R., Paul, V.J., Meyer, K., 1992. Antipredator defenses in tropical Pacific soft corals (Coelenterata: Alcyonacea). I. Sclerites as defenses against generalist carnivorous fishes. Biol Bull 182, 231–240.

Van Alstyne, K.L., Wylie, C.R., Paul, V.J., 1994. Antipredator defenses in tropical Pacific soft corals (Coelenterata: Alcyonanea). II. The relative importance of chemical and structural defenses in three species of *Sinularia*. J Exp Mar Biol Ecol 178, 17–34.

Verseveldt, J., 1969. Octocorallia from North western Madagascar. Part I. Zool Verh. Leiden 106, 1–38.

Wylie, C.R., Paul, V.J., 1989. Chemical defenses in three species of *Sinularia* (Coelenterata, Alcyonacea): effects against generalist predators and the butterflyfish *Chaetodon unimaculatus* Bloch. J Exp Mar Biol Ecol 129, 141–160.

Young, C.M., Bingham, B.L., 1979. Chemical defense and aposematic coloration in larvae of the ascidian *Ecteinascidia turbinata*. Mar Biol 96, 539–544.