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Abstract

The Stylochid flatworm, *Imogine mcgrathi* (Jennings and Newman 1996), was found to be a predator of the mussel *Mytilus galloprovincialis*. These flatworms consumed mussels at a rate of 0.032 individuals·day\(^{-1}\) or 12.6 mg·day\(^{-1}\) in laboratory trials and occurred at densities as great as 386 m\(^{-1}\) of mussel culture rope in Twofold Bay. *I. mcgrathi* has previously been found to be a predator of the oyster, *Pinctada imbricata*. In this study, *I. mcgrathi* collected from mussel ropes restricted their predation solely to mussels when offered oysters and mussels of the same size as food source. When offered only *P. imbricata*, these same *I. mcgrathi* appeared incapable of eating the oysters. *I. mcgrathi* have the potential to pose a significant threat to mussel culture and their abundance in culture warrants careful scrutiny.

Introduction

Often called the Mediterranean mussel, *Mytilus galloprovincialis* is far less parochial than the name might suggest. Found in Europe, Asia, southern Africa and Australia (McDonald et al. 1991), *M. galloprovincialis* forms the basis of several significant mussel culture industries. In Australia, mussel culture is in its infancy, particularly in New South Wales (NSW) where currently approximately 31 tonnes of mussels are cultured annually (ABARE 2000). However, proposals are in place that could see a significant increase in the production of *M. galloprovincialis* in the near future.
In Europe, one impediment to the culture of *M. galloprovincialis* is predation by *Stylochus mediterraneus*, an acotylean polyclad flatworm of the family Stylochiidae (Platyhelminthes, Polycladida) (Galleni et al. 1980). Fortunately, this flatworm has not been reported in NSW waters although the Australian east coast is host to a number of other stylochids (Jennings and Newman 1996a, b), some of which have already been implicated as predators of commercial bivalve species. Despite a general paucity of information regarding Australian polyclad flatworm fauna, one species in particular, *I. mcgrathi* has been associated with mortalities among edible oysters (Jennings and Newman 1996a), scallops (Heasman et al. 1998) and pearl oysters (O’Connor and Newman 2001). Members of this species can grow to 100 mm in length and like polyclads in general, are hermaphroditic (Prudhoe 1985).

Small numbers of the pearl oyster, *P. imbricata*, were taken to mussel farms in Twofold Bay, NSW (Fig. 1), to assess both the potential for oyster culture in southern NSW and the possibilities for polyculture with other commercial mollusks. At the time of deployment (January 2000) large numbers of small *I. mcgrathi* were observed in association with cultured mussels. This raised concerns for both the cultured mussels and the oysters, as *I. mcgrathi* had previously been found to eat *P. imbricata* cultured in Port Stephens, NSW (O’Connor and Newman 2001).

This study was undertaken to determine the possibility of *I. mcgrathi* eating cultured mussels and to estimate potential predation rates. Having confirmed predation, an assessment was made of the likelihood of *I. mcgrathi* posing an immediate threat to other mollusks being cultured concurrently at the mussel farm sites.

**Materials and Methods**

On the 22nd of March 2000, four 20 cm samples of culture rope were taken at random from longer sections collected by divers from a mussel farm in Twofold Bay, NSW (Fig. 1). The samples were transported back to the laboratory and the mussels were stripped from the culture rope. The numbers of live and dead mussels were determined and the shell height (umbo to distal margin) of the various groups was measured to the nearest mm using vernier calipers. Ten live mussels were chosen at random and the total weight and wet tissue weight were determined to the nearest 0.01 g. The mussel tissue was then dried at 100°C for 24 h to determine tissue dry weight.

The number of *I. mcgrathi* found in and on the shells of the mussels on each rope sample was recorded and a randomly selected sample (*n* = 20) of flatworms was
measured to determine average size. *I. mcgrathi* were assumed to be elliptical in shape and surface area was calculated as follows.

Surface area = $\pi \left( \frac{L}{2} \times \frac{W}{2} \right)$

where L and W are length and width, respectively.

Approximately 60 mature *I. mcgrathi* were kept alive in 400 l aerated aquaria for predation trials. Several additional specimens were collected to confirm species identification and were fixed by the frozen polyclad fixation method outlined by Newman and Cannon (1995). Species identifications were made by examination of gross morphological features of preserved specimens and by comparison of serial sections of the reproductive system with those of the holotype at the Queensland Museum (Jennings and Newman 1996a).

All mussels and flatworms used in these trials were taken from the samples collected from Twofold Bay in March 2000. The oysters used were collected from a group of hatchery reared juveniles being held in Port Stephens.

**Flatworm predation**

Eight 160 µm mesh screens were held in a downwelling system following the method of O’Connor and Newman (2001). Four of the screens were each stocked with 10 *I. mcgrathi*. Each of the eight screens was then stocked with 10 mussels (30 to 40 mm shell length) and left for a period of one week. After one week, five mussels were removed from each screen and replaced with five oysters of a similar shell size (30 to 40 mm). Together, the flatworms, mussels and oysters were held further for two weeks before the five remaining mussels were removed and replaced with oysters. The flatworms and oysters were then held concurrently for two more weeks.

Throughout the trial, each screen was checked daily and the number of dead mussels or oysters was recorded. Predation was considered to have occurred only when the soft tissue had been removed from the shells. The shells of dead bivalves were removed and measured before being replaced with a similar sized animal of the same species. Seawater (33 g l$^{-1}$ salinity) in the downwelling systems was held at 22°C and a mixture of three algal species was added daily to feed mussels and oysters. Water in the system was changed thrice weekly.

**Results**

**Field observations**

On the average, samples from Twofold Bay held the shells of 725 mussels m$^{-1}$ of culture rope. The mean shell length of live mussels was 44.1 mm ± 0.64 mm (SE) and mean dry weight of mussel flesh was 0.391 g ± 0.03 g
At the time of collection 36% of the mussels were dead (260 ± 25.6 m⁻¹ of culture rope; mean ± SE). Their shells (mean length 38.3 mm ± 0.62 mm; mean ± SE) showed no signs of physical damage and in most cases the valves remained attached at the hinge. Size frequency distributions for live and dead mussels are shown in figure 2. The density of *I. mcgrathi* present was 386 ± 23.1 m⁻¹ of culture rope. The average length, width and size (approximate surface area) were 24.9 ± 1.3 mm, 13.5 ± 0.6 mm and 266.0 ± 20.3 mm² (mean ± SE), respectively.

**Flatworm predation**

*I. mcgrathi* was shown to be a predator of *M. galloprovincialis*, with each flatworm consuming an average of 0.032 mussels·day⁻¹ or a dry weight of 12.6 mg mussel flesh·day⁻¹. When held concurrently with oysters, the rate of mussel predation by flatworms was not significantly affected ($X^2 = 1.43, P = 0.23$) and no predation of oysters occurred. Upon removal of mussels from the upweller screens, all predation ceased (Fig. 3). Throughout this trial no mortality occurred among oysters and mussels held in control screens, however, four dead mussels with flesh intact were taken from the screens containing flatworms during the trial.

Mussel predation occurred at night with the flatworms spending the day sheltering beneath oysters or mussels on the screen (pers. obs.). Despite the preference for mussels as prey, the flatworms showed a preference for sheltering beneath oysters when held with both species concurrently. Commonly two to four flatworms were found within the shells of eaten mussels, however inspection of the flatworms during the removal of the shells found that only one worm had a distended body, suggesting that the worm had consumed the entire mussel. On two occasions during the removal and replacement of dead mussels, the flatworm assumed to be responsible for the attack regurgitated a relatively intact mussel when disturbed. It was also common to observe a single layer of *I. mcgrathi* eggs on the inner surfaces of mussel shells the morning following predation.

**Discussion**

The occurrence of *M. galloprovincialis* on the east coast of Australia has not precluded it from stylochid flatworm predation, indeed, predation by *I. mcgrathi* was found to be similar to that reported for *S. mediterraneus* on the Italian coast (Galleni et al. 1980). In laboratory trials, *S. mediterraneus* demonstrated a preference for smaller mussels (< 25 mm) and consumed them at a rate of between 0.07 individuals and 0.33 individuals·day⁻¹ (Galleni et al. 1980). Here, *I. mcgrathi* were only offered larger mussels (approx. 44 mm) and predation rates were comparatively low, 0.032 individuals·day⁻¹. However, dry weights of mussel flesh consumed by *S. mediterraneus* (26 to 35 mm in length) and *I. mcgrathi* (mean length 24.9 mm) were similar, at 11.2 and 12.6 mg·day⁻¹, respectively.
It is possible that *I. mcgrathi* may increase predation rates in the presence of smaller mussels as observed by Galleni et al. (1980) with *S. mediterraneus*. But in previous trials in which *I. mcgrathi* were offered oysters of various sizes, there was a tendency for larger flatworms to eat larger oysters rather than increase predation on small oysters (O’Connor and Newman 2001). In these earlier trials, *I. mcgrathi* was found to prey upon oysters at a similar rate to *S. mediterraneus*, approximately 0.035 to 0.057·day$^{-1}$. However, *I. mcgrathi* consumed in total a greater dry weight of mussel flesh than that of oysters (12.6 mg and 4.9 mg·day$^{-1}$, respectively). The reason for this is unclear, but may in part be due to water temperature. Landers and Rhodes (1970) found predation by *S. ellipticus* increased by as much as 72% over a 5 to 6°C temperature range. Water temperatures in this study were on the average 3°C warmer than in the earlier study of oyster predation (22°C and 18 to 20°C, respectively). In common also with *S. ellipticus, I. mcgrathi* showed a first, marked peak in predatory behavior after three days (Fig. 3). Landers and Rhodes (1970) observed these “first peaks” in all of their trials within 2 to 4 days.

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**Fig. 2.** Size frequency distributions for live and dead mussels collected from culture ropes in Twofold Bay, NSW, Australia.

**Fig. 3.** Predation by *I. mcgrathi* when held concurrently with mussels (days 0-21) and oysters (days 7-35).
days, but were unsure of the cause. The possibility that the absence of food during the pre-experimental period may have accentuated the initial burst of predatory behavior was suggested (Landers and Rhodes 1970). Yet in this trial, flatworms could only have been considered to be without food during transport to the laboratory. At this time, the flatworms were still on the mussel samples but may have been unable to feed while emersed.

The behavior of *I. mcgrathi* in the presence of mussels was similar to that described previously for oysters although we had been unable to observe the method used by *I. mcgrathi* to consume the bivalve. With mussels, *I. mcgrathi* were seen on two occasions to slip between the valves of the mussel without evoking any obvious response from the mussel. When the mussel was opened some 30 to 45 minutes later, the posterior adductor muscle had been detached from the shell and the flatworm had everted its pharynx and begun to engulf the mussel flesh from the distal margin inwards.

As observed with *S. ellipticus* (Landers and Rhodes 1970) and suggested for *S. mediterraneus* (Galleni et al. 1980), *I. mcgrathi* collected from mussels appeared to demonstrate what has been called “ingestive conditioning”; a behavior in which predation is limited to the species with which the predator had been associated in nature. *I. mcgrathi* collected from Port Stephens have been demonstrated to eat *P. imbricata* but their conspecifics from Twofold Bay failed to do so in these trials. This was despite the likelihood that at the culmination of the trial many of the worms had not been fed in over a month.

*I. mcgrathi* is thought to have the potential to be a major pest to the cultured mussel industry. It eats mussels in the laboratory and the large numbers of dead intact shells on the culture ropes are consistent with flatworm predation, however, the size of the empty shells raises several questions. Assuming *I. mcgrathi* is responsible for the observed mortality, the mussel size frequency data (Fig. 2) suggests that predation is a relatively recent phenomenon. Given the absence of empty shells less than 30 mm long, a mussel growth rate of 1 to 1.5 mm a week and the smallest live mussels present being 34 mm long, it is likely that there has been some 3 to 4 weeks since mussels of 30 mm shell length were present and thus 3 to 4 weeks since predation commenced. Assuming that predation began progressively and that flatworms consumed mussels at approximately the same rate as observed in the laboratory (≈ 1 month⁻¹), 260 empty shells for 386 flatworms is not inconceivable for one month’s predation. However, the impetus for this study arose from observations of *I. mcgrathi* two months earlier and given their ability to eat 10 mm *P. imbricata*, they should be capable of consuming small *M. galloprovincialis*. This would suggest that the shells of predated mussels are dislodged from the ropes or decompose within weeks and that the overall losses from *I. mcgrathi* may be greater than the 36% calculated from dead shell numbers.

The presence of empty shells in the largest mussel size class suggests that predation is ongoing and that mussels have not grown to a size at which they are beyond attack. Thus, if flatworm numbers remain constant and they eat mussels at ≈ 1 month⁻¹, they have the ability to consume the standing crop within six weeks. Subsequent to this study, the mussels present were harvested.
and no further observations were possible but, so dire are the potential consequences, that flatworms warrant close attention. Farmers reported the harvest was extremely poor, although large numbers of mussels were lost in heavy seas (M. Bamford pers. comm.), and this was thought to be exacerbated by the increasing numbers of dead shells that were no longer byssally attached to the culture rope. Strategies for the control of *I. mcgrathi* have been discussed with mussel farmers, including freshwater or brine baths (O’Connor and Newman 2001), but the efficacy of these treatments with respect to the tolerances of *M. galloprovincialis* and the farming practices used need to be evaluated.

Whether the flatworms present on mussel culture ropes pose a threat to other mollusks remains to be seen. The possibility of “ingestive conditioning” augers well for the survival of *P. imbricata* in Twofold Bay in the short term, however, it was assumed that given the size of the population, juvenile flatworms may recruit to the oyster cages. Ultimately this did not occur and the oysters were harvested several months later without significant losses. There are many potential ecological explanations for this such as unsuitable environmental conditions for flatworm recruitment and dispersion of flatworm larvae to other areas by prevailing currents, etc. The threat that flatworms may recruit to other bivalve culture systems in coming seasons, however, remains a concern.

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