Frequency-dependent success of aggressive mimics in a cleaning symbiosis

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Batesian mimics—palatable organisms that resemble unpalatable ones—are usually maintained in populations by frequency-dependent selection. We tested whether this mechanism was also responsible for the maintenance of aggressive mimicry in natural populations of coral reef fishes. The attack success of bluestriped fangblennies (*Plagiotremus rhinorhynchus*), which mimic juvenile bluestreaked cleaner wrasses (*Labroides dimidiatus*) in colour but tear flesh and scales from fishes instead of removing ectoparasites, was frequency-dependent, increasing as mimics became rarer relative to their model. However, cleaner mimics were also more successful on reefs with higher densities of potential victims, perhaps because a dilution-like effect creates few opportunities for potential victims to learn to avoid mimics. Further studies should reveal whether this second mechanism is specific to aggressive mimicry.

Keywords: Batesian mimicry; frequency-dependence; cleaning symbioses; ectoparasites

1. INTRODUCTION

Classical Batesian mimicry is one of the best examples of Darwin's theory of adaptation by natural selection: an edible mimetic species copies the warning signal of a noxious, aposematic model species, thereby gaining protection from predators (Bates 1862). Theory developed to explain the evolution of protective mimicry invokes frequency-dependence as the main mechanism of maintaining mimics in populations (e.g. Brower *et al.* 1970; Huheey 1988). After an encounter with a noxious prey, predators learn to avoid similarly coloured individuals, but their limited memory span or the need to reassess periodically changing model and mimic frequencies results in repeated sampling of prey over time (Huheey 1964; Dill 1975). If the number of palatable mimics of noxious models in a population increases, the predator's previously learnt avoidance of the model is reinforced less frequently (Lindström *et al.* 1997). Mimics should therefore have the highest fitness when greatly outnumbered by models, with fitness declining as mimic frequency increases (Sheppard 1959; Bates 1862). Although frequency-dependence has been shown for Batesian systems (Lindström *et al.* 1997; Pfenning *et al.* 2001), there have been no tests for other types of mimicry. Here we test the applicability of the frequency-dependence model to aggressive mimicry.

In aggressive mimicry, a 'predatory' species resembles a model that is harmless or beneficial to a third victim species, the 'dupe' (Wickler 1966). Aggressive mimics vary greatly in their effect on dupes. An extreme example is that of female fireflies from the genus *Photuris*, which mimic the flashing signal of *Photinus* females to attract and

devour *Photinus* males (Haynes & Yeargan 1999). A less lethal form is found on Indo-Pacific coral reefs where bluestreaked cleaner wrasse (*Labroides dimidiatus*; family Labridae) are mimicked by fangblennies (family Blenniidae) which, instead of removing ectoparasites, strike unsuspecting clients (the dupes) and tear off fins, skin and scales (Wickler 1966). Aggressive and Batesian mimicry are similar in that dupes incur costs due to mimics; however, when attacks by aggressive mimics are not fatal, dupes might learn to avoid both model and mimic when mimics are abundant. Therefore, the success of aggressive mimics could also be frequency-dependent.

Aggressive mimicry does, however, differ fundamentally from Batesian mimicry in the relationship between model and dupe. Batesian mimics insert themselves into an antagonistic predator–prey interaction (where the models are the unpalatable prey). In contrast, aggressive mimics insert themselves into cooperative interactions. In cleaning symbioses, clients gain when visiting cleaners through a reduction in their ectoparasite loads (e.g. Grutter 1999) and cleaners gain nutrition from client-gleaned ectoparasites (Grutter 1996). The success of aggressive mimics of cleaners should therefore depend on the maintenance of this mutualism, which can attract high densities of fish (i.e. potential dupes) to cleaning stations (Whiteman & Côté 2002), but which can deteriorate if cheating occurs (Johnstone & Bshary 2002; Freckleton & Côté 2003). As a result, in parasite-poor areas where clients benefit little from cleaning interactions (Cheney & Côté 2005), cleaner–client relationships might break down easily in response to low levels of cheating. By contrast, clients might tolerate aggressive mimics more in areas where ectoparasite loads are high and successful cleaning confers higher benefits (Cheney & Côté 2005). In addition, increasing the numbers of dupes should have different impacts on models in Batesian and aggressive mimicry. In the former, a low encounter rate between predators and mimics should slow predator learning

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and be deleterious for models (Brower *et al.* 1970), whereas in aggressive cleaning mimicry, slow learning by victims should maintain cleaner–clients relationships for longer. Cleaner models and mimics may therefore be maintained at different relative frequencies in populations that vary in ectoparasite intensities and in availability of potential victims.

In this paper, we investigated the mechanisms of aggressive mimicry maintenance in natural populations of coral reef fish. The model we studied is the juvenile bluestreaked cleaner wrasse, an active cleaner which is conspicuously coloured, with electric blue lateral stripes on a black body. The mimic is the bluestriped fangblenny (*Plagiotremus rhinorhynchos*), which in its mimetic colour form is identical in colouration to juvenile cleaners (Kuwanara 1981). Non-mimetic forms (i.e. brown, olive or orange, with blue stripes) of *P. rhinorhynchos* are also known, but these are not usually found in association with juvenile cleaner wrasses (Moland & Jones 2004; Côté & Cheney 2005) and were therefore not considered within this study. It has already been shown for this and other reef fishes that the abundance of aggressive mimics is correlated with the abundance of models and that mimics are usually rarer than models (Eagle & Jones 2004; Moland & Jones 2004). Here, we focussed instead on the attack success of mimics, a correlate of fitness, and predicted that it should increase as (i) mimics become rarer compared to their juvenile cleanerfish model, (ii) ectoparasite loads of clients increase and (iii) more potential victims are present on the reef. We tested these predictions correlatively with field observations of foraging activity of cleaner mimics on reefs varying in mimic and model densities, client ectoparasite loads and reef fish densities.

2. MATERIAL AND METHODS

(a) Study sites

Our study was conducted near Hoga Island, in the Wakatobi National Marine Park, southeast Sulawesi, Indonesia, from 25 June to 4 September 2004. We used four study reefs (Kaledupa, Hoga Buoy 2, Sampela and Pak Kasim's), each separated by more than 1 km. These reefs varied in profile, but all had high coral cover, fish biodiversity and abundance.

(b) Behavioural observations

We observed juvenile bluestreaked cleaner wrasses (total length <6 cm) and bluestriped fangblennies in mimetic colour *in situ* using SCUBA at depths of 2–18 m. At each site we haphazardly located 10–15 cleaner mimics. For each mimic, we recorded the abundance of all juvenile cleaner wrasses and mimetic fangblennies within a 5×5 m quadrat centred on each fangblenny's location at first sighting. The excellent visibility (20+ m) made double-counting improbable. We then conducted a 15 min observation of each mimic on three different days, marking the centre of each mimic's home range to facilitate relocation. We recorded the number of attempted attacks (i.e. mimic darted towards a potential victims) and whether each attack was successful (i.e. clear contact made with the victim's body with a jolt by the victim). We also estimated the time fangblennies spent within 30 cm of the nearest juvenile cleaner wrasse, because when in close proximity to their models, fangblennies encounter more

potential victims and have an increased attack rate (Côté & Cheney 2004).

We estimated the abundance of potential clients/victims at each site with five 5 min point counts at 5 and 10 m depths, noting the number and species of fishes within or passing through a 3×3 m quadrat. To quantify the densities of models and mimics on a reef-wide scale, we counted the number of juvenile cleaner wrasse and fangblennies in 10 quadrats (5×5 m) per site.

(c) Ectoparasite assessment

We quantified ectoparasite loads on three to six individuals of nine common client species at each site, following the non-destructive method of Grutter (1995a) (*Zebrafish* *scopas*, *Ctenochaetus striatus*, *Acanthurus pyroferus*, *Pomacentrus brachialis*, *Amblyglyphidodon curacao*, *Chromis ternatensis*, *Amblyglyphidodon leucogaster*, *Chaetodon lunulatus* and *Scolopsis bilineatus*). Clients were herded into a net, caught with hand nets and individually placed into sealable plastic bags filled with seawater. In the laboratory, each fish was anaesthetized with 1 ml of 95% ethanol/5% clove oil solution l⁻¹ of seawater. The fishes were then placed into individual freshwater baths for 10 min and brushed gently with a paintbrush to remove ectoparasites. After recovering in seawater, the fishes were released at their capture point. Survival rate was 95.2% (137/144). All fluids from each fish were filtered using a 100 µm plankton mesh to recover ectoparasites. Ectoparasites were preserved in 70% ethanol, then identified and counted using a binocular microscope.

(d) Statistical analyses

The ratio of the number of models to mimics was calculated for each 25 m² area around each mimic sighted. The number of strike attempts and the proportion of successful attacks 15 min⁻¹ were averaged across the three observations made of each fangblenny. There was no difference between the total numbers of clients/victims at 5 and 10 m for any of the sites (all *t*-tests: *t*<1.9, d.f.>7, *p*>0.08); therefore counts from each depth were combined within site.

We examined the relationship between mimic attack success and model : mimic ratio at two spatial scales. First, for the small scale experienced by individual mimics, we used a general linear model (GLM) analysis (Type III SS, SPSS v. 11.0) with the proportion of successful attacks as the dependent variable, and model : mimic ratio, mean number of strike attempts and time spent with model as covariates and site as a factor. Five fishes did not strike in any of the three 15 min observations and were omitted from the analyses.

Second, for the larger reef-wide scale, we related intersite differences in fangblenny success to relative densities of models and mimics on each site using a Jonckheere–Terpstra Trend test (Jonckheere 1954), with ordinal ranking of sites being determined *a priori* by the reef-wide model : mimic ratios obtained from 5×5 m quadrats. We also used Jonckheere–Terpstra Trend tests to examine the relationships between fangblenny attack success and both mean client ectoparasite loads and mean densities of clients, derived from point count data. In both cases, the analysis could only be carried out at the reef scale, to match the scales at which the fish abundance and ectoparasite data were collected, thus precluding the use of GLMs.

3. RESULTS

(a) Does cleaner mimic attack success increase as mimics become rarer?

The densities of cleaner wrasse and cleaner mimics varied significantly among sites (figure 1; models: $\chi^2=14.8$, d.f.=3, $p=0.002$; mimics: Kruskal–Wallis $\chi^2=8.04$, d.f.=3, $p=0.045$). At all sites, the density of cleaner wrasses exceeded the density of mimics. The ratio of cleaner wrasse models to mimics also differed significantly among sites (ANOVA $F_{3,46}=4.12$, $p=0.01$).

The mean rate of attempted attacks per fangblenny ranged from between 0 and 20.1 attacks 15 min^{-1} . At the small spatial scale of mimic home ranges, attack success was frequency-dependent. As the ratio of models : mimics increased, so did the proportion of successful attacks by mimics (figure 2; $F_{1,44}=30.4$, $p<0.001$). In addition to the model : mimic ratio, variation in attack success was predicted by site ($F_{3,44}=7.82$, $p<0.001$) and positively related to the mean number of attempted attacks ($F_{1,44}=14.8$, $p<0.001$). However, time spent with models was not a significant predictor ($F_{1,44}=0.77$, $p=0.38$). At the larger reef scale, the proportion of successful attacks also increased as the overall ratio of models : mimics increased (figure 3a; Jonckheere–Terpstra Trend Test, $p=0.01$). The proportion of successful attacks did not vary with model density (Jonckheere–Terpstra Trend Test, $p=0.24$); however, it did decrease as mimic density decreased (Jonckheere–Terpstra Trend Test, $p=0.01$).

(b) Does cleaner mimic attack success increase with client ectoparasite load?

There were significant differences in the number of ectoparasites per client among sites (mean rank: Hoga=1.39, Sampela=2.67, Kaledupa=2.83, Pak Kasim's=3.11; Friedman's test: $\chi^2=9.65$, d.f.=3, $p=0.02$). Mimic attack success among sites was not correlated with site-specific mean ectoparasite loads on client fish (Jonckheere–Terpstra Trend Test, $p=0.21$).

(c) Does cleaner mimic attack success increase with the density of potential victims?

The proportion of successful attacks by mimics increased with total client fish density (figure 3b; Jonckheere–Terpstra Trend Test, $p<0.001$). Total client fish density did not increase with model : mimic ratio (Jonckheere–Terpstra Trend Test, $p=0.60$).

4. DISCUSSION

Aggressive cleanerfish mimics are maintained in natural populations of coral reef fishes in part by frequency-dependent foraging success. The proportion of successful attacks by the bluestriped fangblenny was higher when these mimics were relatively rare compared to their model, the juvenile cleaner wrasse. We also found that mimic success increased with the availability of potential victims on reefs. However, our prediction that cleaner mimic attack success should be related to client ectoparasite load was not supported.

The frequency-dependent attack success of cleaner mimics was evident at two spatial scales, i.e. on the small scale experienced by individual mimics and on the larger reef-wide scale. An increase in the relative frequency of mimics alters the relationship between dupe and model.

Figure 1. The density of cleaner wrasse models (black bars) and cleaner wrasse mimics (grey bars; number 25 m^{-2}) on four Indonesia reefs. Bars represent mean + s.e.

Figure 2. Proportion of successful attacks (%) by bluestriped fangblennies in relation to (log) ratio of models : mimics ($y=41.7+58.8x$). Open circles, Kaledupa; filled circles, Hoga; filled triangles, Pak Kasim's; open triangles, Sampela.

In Batesian mimicry, this alteration occurs because the predator's learnt avoidance of the model is weakened by the consumption of edible mimics (Lindström *et al.* 1997). In aggressive cleanerfish mimicry, increased mimic frequency appears to cause potential victims to become more cautious when approaching fishes with cleaner-like colours and to take evasive action sooner if all cleanerfish signals are not detected (Stummer *et al.* 2004). Ultimately, victims may learn to avoid cleaning stations near which attacks are frequent relative to cleaning events.

The success of cleaner mimics was also higher when more potential victims were available. In Batesian terms, this effect would be analogous to an increased success of mimics in populations in which more predators are present. Although this has never been considered theoretically, an increase in the pool of potential dupes should reduce the likelihood that any individual dupe will encounter a mimic. This 'dilution effect' could slow the rate at which dupes learn to avoid mimics (and models) in both Batesian and aggressive mimicry. Whereas such an effect would be advantageous for an aggressive mimic, it would not be so for a Batesian mimic which relies on predator learning for its protection.

Figure 3. Proportion of successful attacks (%) by bluespined fangblennies on four Indonesian reefs. The reefs are presented in order of (a) increasing ratio of models : mimics and (b) increasing density of reef fish clients. Bars represent mean ± s.e. Sample sizes are given in parentheses.

Contrary to our third prediction, client ectoparasite loads did not affect the attack success of mimics. Our prediction was based on the fact that clients with more ectoparasites visit cleaning stations more often (Grutter 1995b; Arnal *et al.* 2001) and derive a greater net benefit from being cleaned than less parasitized species (Cheney & Côté 2005). Clients involved in highly beneficial cleaning interactions should therefore be willing to incur relatively greater costs (e.g. attacks by mimics) to gain access to cleaning stations than clients for which the net benefits of being cleaned are marginal. Although this might be true for individual clients, the reef-wide scale of our analysis and limited sample size of reef fish species, imposed by the nature of the ectoparasite data, might have been too coarse to detect a general association between mean ectoparasite load and mimic attack success.

In summary, we found evidence that the success of aggressive cleanerfish mimics depends partly on their rarity relative to their models and partly on the availability of potential victims. The former was expected given the similarities between Batesian and aggressive mimicry, but the latter is perhaps specific to aggressive mimicry. The relative importance of these two factors in determining mimic success remains to be determined, but potential interactions may be expected. For example, sites with higher densities of potential victims may be able to support

higher relative frequencies of mimics, which can maintain a high attack success, than sites with fewer potential victims. The study of other aggressive mimicry systems should reveal whether these two mechanisms are generally responsible for the maintenance of aggressive mimics in natural populations.

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