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Why clean and be cleaned? The importance of client ectoparasites and mucus in a marine cleaning symbiosis

Received: 22 September 2000 / Revised: 13 July 2001 / Accepted: 24 July 2001 / Published online: 5 September 2001
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Abstract The preferences exhibited by cleaner fishes for particular client species and the high variability in rates at which various clients visit cleaning stations have remained largely unexplained. In this study, we assessed the relative importance of client ectoparasite load and mucus characteristics for the behaviour of cleaning gobies, *Elacatinus* spp, and their fish clients on a Barbadian fringing reef. Client species with high ectoparasite loads visited cleaning stations more often than less parasitised species. This effect was independent of body size. Frequency of visits to cleaning stations was not related to client mucus characteristics. These results suggest that the main motivation for clients to interact with cleaners is ectoparasite removal. Cleaners did not preferentially clean clients with higher ectoparasite load or better mucus, nor did they spend more time inspecting such clients. The interests of cleaners and clients therefore appear to be inconsistent. This may be due to the generally low rate of ectoparasitism on Barbadian fish compared to fish of other regions. Cleaning gobies fed at a lower rate on client species with higher loads of gnathiid isopod larvae, which may be explained if cleaners switch from eating ectoparasites to other items, such as mucus, on clients with few ectoparasites. Our estimates of caloric and protein content of fish mucus suggest that it may be as valuable a food source per unit weight as ectoparasites. However, no data are available to compare the value of each item per unit feeding time. The fact that clients with few ectoparasites still visit cleaners, albeit at a low rate, suggests that the cost of mucus removal may be

low, compared to the benefit of incidental parasite removal. Thus, the outcome of cleaning interactions may remain positive, even in areas characterised by naturally low parasitism on clients.

Keywords Cleaning gobies · Cleaning symbiosis · *Elacatinus* · Ectoparasites · Fish mucus · *Gnathia*

Introduction

Cleaning symbioses among coral reef fishes are ubiquitous interspecific interactions. During a cleaning encounter, small fishes such as cleaning gobies and cleaning wrasses remove ectoparasites, mucus and scales (Youngbluth 1968; Losey 1974; Grutter 1997) from the body surface of apparently co-operating fishes known as clients (Poulin and Grutter 1996). Cleaner fishes generally occupy traditional sites known as cleaning stations (Losey 1972; Potts 1973) which client fishes visit to solicit cleaning. Clients often adopt an immobile, stereotyped pose at cleaning stations (Hobson 1971; Losey 1972) and, while posing behaviour appears to increase the chances of being cleaned (Côté et al. 1998), it does not guarantee that cleaning will occur (Losey 1974; Arnal and Côté 1998; Côté et al. 1998). The duration of inspection of the client's body surface can also be extremely variable (Arnal and Côté 1998; Grutter and Poulin 1998). Cleaner fishes may thus exhibit preferences for particular client species (Losey 1972; Grutter 1995a; Wicksten 1995, 1998; Arnal et al. 2000), which have yet to be explained. Similarly, clients visiting cleaning stations do so at highly variable rates (Arnal and Côté 1998; Grutter and Poulin 1998; Wicksten 1998). Elucidating the reasons for such preferences and differences is fundamental for understanding cleaning associations.

Ectoparasites, such as gnathiid isopod larvae, form a large part of the food items ingested by cleaner fishes (Losey 1974; Grutter 1997, 1999a; Arnal and Côté 2000), and cleaners can have a significant impact on client ectoparasite size and abundance (Gorlick et al. 1987;

Communicated by M. Abrahams

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Grutter 1999b; Cheney and Côté 2001). Client ectoparasite load may therefore be an important factor determining both cleaner preferences for clients as well as the tendency for some clients to visit cleaning stations. Gorlick (1984) showed, in laboratory experiments, that the Hawaiian cleaner wrasse, *Labroides phthiophagus*, prefers to clean parasitised individuals when given a choice between parasitised and non-parasitised clients. In the field, the cleaner wrasse, *L. dimidiatus*, inspects larger individuals of any given species more often and for longer than smaller ones (Grutter 1995a; see also Arnal et al. 2000 for a similar result for cleaning gobies, *Elacatinus* spp., choosing among client species), and larger clients are expected to have heavier ectoparasite loads, especially of gnathiid larvae (Poulin 2000). Poulin (1993) found that larger client species tended to be found in the vicinity of cleaning stations more often than smaller clients. However, most inter-specific studies using correlates of ectoparasitism, such as body size and social behaviour, have failed to explain either cleaner fish preferences or differences in client willingness to visit cleaners (Côté et al. 1998; Grutter and Poulin 1998; Arnal et al. 2000).

Ectoparasites alone are clearly not the answer, thus focussing on other items ingested by cleaners from their clients may be necessary. Fish mucus has been recorded (Gorlick 1980) or suggested (Youngbluth 1968; Grutter 1997) in the gut content of many cleaning species. In addition, Gorlick (1980) showed that for a small number ($n=5$) of client species, there was qualitative agreement between cleaning wrasse (*L. phthiophagus*) preference for specific clients and the quantity and energetic value of the clients' mucus. The importance of client mucus to cleaner fishes has since remained unexplored. This is unfortunate, since mucus may be a more reliable food source for cleaner fishes than ectoparasites, which are known to fluctuate seasonally in size and abundance (Grutter 1994). Moreover, mucus may be an important source of glycoproteins (Nakagawa et al. 1988; Shephard 1994).

In this study, we assessed for the first time the relative importance of client ectoparasites and mucus characteristics for cleaner and client behaviour. We focussed on the cleaning symbiosis between cleaning gobies (genus *Elacatinus*) and their reef fish clients on a Barbadian fringing reef. While cleaning gobies co-occur with a number of other cleaning organisms, including other fish species and shrimp, they are by far the most active cleaners at this location (personal observations). Specifically, we asked how variation in client mucus and ectoparasites affects (1) the willingness of clients to visit cleaning stations, (2) the tendency for clients to adopt solicitation poses to be cleaned, (3) which clients cleaning gobies choose to inspect, (4) the duration of inspection and (5) the feeding rate of cleaning gobies on each client species. By linking behavioural observations to a direct assessment of ectoparasite load and mucus quality, we hoped to provide the clearest picture yet of the factors governing variability in a marine cleaning symbiosis.

Methods

Behavioural observations of cleaning interactions

Behavioural observations were carried out at 12 cleaning stations on a fringing reef of the Barbados Marine Reserve on the west coast of Barbados (13°10' N, 59°30' W), West Indies, between February and June 1996. All observations were made using SCUBA at depths of 3.5–6 m. Two species of cleaning gobies, *Elacatinus evelynae* (incorrectly identified as *E. genie* in Arnal and Côté 1998) and *E. prochilos*, were present on the reef. Given that both have similar behaviour and habitat (Colin 1975), we did not try to distinguish between them during the observations. Several cleaning gobies were captured at the end of the observations and identified. This revealed that cleaning stations were always occupied by gobies of a single species, but the distribution of the cleaning stations operated by each species overlapped completely on the study site (Arnal and Côté 1998). Furthermore, no differences between goby species were found in the number or species composition of their clients (Arnal 1996; Arnal and Côté 1998).

Observations were carried out between 0830 and 1530 hours, which is the most active period for *Elacatinus* spp. (personal observations). A single diver recorded all data directly on underwater plastic slates, after a preliminary delay of 5 min to allow the cleaning gobies to become used to the presence of the diver. Diving occurred only when the weather and currents resulted in a minimum visibility of 5 m. A total of 25 h of observation of 12 cleaning stations were made. All 25 client species observed at cleaning stations during this study were recorded in the first 8 h of observations, suggesting that our total sampling time was adequate. Each cleaning station was observed for 10 min, twice per week for a total of 150 observations periods. The number and species of clients visiting each cleaning station were recorded. A visit was recorded when a fish approached and remained within 15 cm of a cleaning station for at least 5 s. For each visit, we noted whether the visiting fish posed, whether it was inspected by gobies, the inspection duration, and the number of bites taken by cleaning gobies on each client's body.

Assessment of client ectoparasites

Eight of the commonest fish species on the reef were chosen to represent a wide variation in frequency of use of cleaning stations, as estimated from the behavioural observations described above. Fish collection took place in April 1999 on the reef where behavioural observations were carried out. Five individuals per species were sampled, which appears sufficient to characterise ectoparasite abundance on a species at a given place and time (e.g. Grutter 1994). Fish collection and parasite extraction methods followed Grutter (1995b). Using SCUBA, two divers drove target fish into a 3×1.5 m barrier net with a 15-mm mesh. Fish were then captured with handnets, and each individual was placed immediately in a sealable plastic bag with as little water as possible. Fish died quickly from lack of oxygen. In the laboratory, fish were placed in glass crystallising dishes, along with the contents of the bag. The plastic bag was rinsed with seawater and all liquids were kept for later filtration. Fish were soaked in 0.4% chlorobutanol (Sigma) for 90 min to dislodge ectoparasites, after which fish body surface and gills were rinsed thoroughly with seawater in a wash bottle. All liquids were filtered on filter paper (60 µm mesh). Filter and filtered material were preserved in 10% formaldehyde diluted in seawater. These samples were examined under a binocular microscope (×250–500), and crustacean ectoparasites, as well as non-parasitic crustaceans, were isolated for identification and enumeration. We did not record other ectoparasites such as monogeneans since they have not been found in the diet of *Elacatinus* gobies (Arnal and Côté 2000; P. Sikkil, personal communication) and are exceedingly rare on the clients of cleaning gobies in Barbados (P. Molloy and I.M. Côté, unpublished data). We thus report, for each of eight client species, the mean numbers of gnathiid isopod larvae (*Gnathia* spp), caligid copepods (*Caligus* spp), and other parasitic copepods (including *Bomolochus* spp, *Ergasilus* spp, and unidenti-

fed copepod larvae), and the mean number of all non-parasitic copepods (including harpacticoids, cyclopoids and calanoids) per individual client. The majority of these non-parasitic crustaceans were benthic-living copepods, making it unlikely that they originated from the seawater trapped in the sampling bags.

Assessment of client mucus load and quality

Client fish ($n=15$ species) were collected for mucus analysis at the same time and location as samples for ectoparasite extraction. Fish ($n=1-3$ individuals per species) were driven into a barrier net by divers as before, but this time individual fish were placed into large sealable plastic bags full of seawater. Fish were transported alive to the laboratory where they were maintained in aerated tanks with running seawater. Within 24 h (usually less than 12 h) of capture, individual fish were killed humanely by decerebration. To assess mucus load and quality of each fish, we followed the method of Gorlick (1980). Each fish was placed in a freshwater rinse for 5 s to remove excess seawater from the body surface, and was then submerged for 60 s in a bath containing tap water heated to 50°C. This temperature was sufficient to denaturise and coagulate the surface mucus, which turned a milky-white colour. Each fish was then suspended by the jaw over a beaker, and mucus was gently scraped off the fish with a scalpel, taking care not to remove epiderm. Surface mucus was collected in 5-ml tubes. The water/mucus mixture was then dried to constant weight in a drying oven at 70°C and weighed to the nearest milligram. We measured each fish (SL, cm) and determined its surface area (cm²) by wrapping the left side of the body, all fins spread, with aluminium foil to conform to body curvature, and cutting out the outline of the body. The pectoral fin was removed from the body and its outline was obtained separately. The weights of both outlines were then combined, doubled to represent total fish area and converted to cm² using a weight/area relationship established with pieces of foil of known area. Dried mucus weight (DW) was thus expressed in mg/cm² of fish. Finally, the gross chemical composition of dried mucus was estimated with a CHN analyser (Service Central d'Analyse, CNRS) to obtain percentages of carbon (C%), nitrogen (N%) and ash (ash%). Protein content of client mucus was estimated according to Holland et al. (1991): protein (%DW)=6.25×N%. Direct calorimetric measurement was not possible so we used the substitute presented by Gorlick (1980). He approximated calorific content of client mucus as: calories/g DW=1351+106(C%)–21.1(ash%).

Statistical analysis

For each client species, we considered five behavioural variables. For variables expressed as rates, we obtained residuals from the regression of the numerator on the denominator, to avoid the problems inherent to ratios. Clients' tendency to visit was measured as the total number of visits to cleaning stations over 25 h of observation. Although visit rate is often related to client abundance on the reef (Grutter and Poulin 1998; Arnal et al. 2000), this was not the case for our small sample ($r^2=0.03$, $F_{1,9}=0.30$, $P=0.60$, abundance data being derived from Rakitin and Kramer 1996), thereby removing the need to control for abundance. The tendency to perform solicitation poses was defined as the residuals of the regression of number of solicitation poses versus number of visits to cleaning stations ($n=15$, $r^2=0.99$, $P<0.0001$). Similarly, cleaner preference, i.e. their willingness to inspect specific clients, was estimated as the residuals of the relationship between the number of inspection events versus the number of visits by these client species ($n=15$, $r^2=0.92$, $P<0.0001$). Inspection duration was corrected for the number of inspections ($n=15$, $r^2=0.78$, $P<0.0001$) on each client species. Finally, the residuals of the relationship between number of bites by cleaners and inspection duration ($n=15$, $r^2=0.98$, $P<0.0001$) represented a measure of cleaner feeding activity on each client species. The number of visits and solicitation poses by clients, inspections by cleaners, cleaning duration and number of bites were $\log(x)$ - or $\log(x+1)$ -transformed prior to the regression analyses above.

Table 1 Measures of client behaviour (visits and poses), cleaning goby cleaning activity (inspections and bites), and mucus characteristics (mean±SE) for client species visiting 12 focal cleaning stations over 25 h. Mucus quality index was obtained by averaging the ranking of mucus protein and mucus calories (see Methods)

Client species	Total number of visits	Total number of poses	Total number of inspections by cleaners	Total inspection duration (s)	Total no. of bites by cleaners	Mucus load (g dry weight/cm ² of client body surface) (×10 ³)	Mucus protein (% dry weight)	Mucus calories (cal/mg dry weight)	Mucus quality index
<i>Acanthurus bahianus</i>	13	10	7	59	2	0.23±0.03	61.04±1.02	4.68±0.49	11.5
<i>Acanthurus coeruleus</i>	46	42	26	303	15	0.23±0.05	62.97±2.44	5.34±0.09	13.0
<i>Autostomus maculatus</i>	14	13	8	345	27	0.09±0.02	46.04±4.44	3.56±0.11	6.5
<i>Chromis multilineata</i>	988	975	952	9801	614	0.41±0.18	44.99±0.41	3.52±0.09	5.5
<i>Clepticus parrae</i>	27	27	17	69	2	0.36±0.18	61.70±17.8	3.74±0.09	10.5
<i>Haemulon carbonarium</i>	11	10	2	136	3	0.06±0.04	46.35±2.88	3.97±0.21	9.5
<i>Haemulon chrysargyreum</i>	39	34	3	17	1	0.14±0.04	38.15±2.00	3.20±0.10	1.5
<i>Haemulon flavolineatum</i>	93	92	17	241	3	0.39±0.17	42.18±0.97	3.58±0.11	5.5
<i>Microspathodon chrysurus</i>	34	33	20	203	4	0.22±0.04	44.13±0.49	3.74±0.15	7.5
<i>Mulloidichthys maritimus</i>	148	131	128	879	28	0.14±0.04	49.77±6.65	3.73±0.25	9.0
<i>Myripristis jacobus</i>	11	7	4	25	0	0.19±0.05	40.34±1.84	2.33±0.11	2.0
<i>Scarus taeniopterus</i>	12	10	4	197	7	0.29±0.03	72.78±0.97	5.64±0.10	14.5
<i>Sparisoma viride</i>	9	6	1	12	0	0.40±0.23	71.42±1.98	5.67±0.19	14.5
<i>Stegastes dorsoponicans</i>	99	95	70	760	20	0.48±0.03	48.80±3.24	3.49±0.16	6.5
<i>Stegastes partitus</i>	16	13	7	31	1	0.10±0.02	39.40±2.43	3.27±0.01	2.5

Mucus protein content and caloric value were significantly correlated ($n=15$, $r^2=0.86$, $P<0.001$). To remove the problem of intercorrelations among mucus characteristics, we generated an index of client mucus quality by averaging the ranks obtained for protein content and caloric value for each client species (Table 1). This mucus quality index was not correlated with mucus load ($n=15$, $r^2=0.06$, $P=0.38$).

To take into consideration phylogenetic relatedness among client species, we used the principal coordinate analysis method (PCoA) (Diniz-Filho et al. 1998; Legendre and Legendre 1998). This method allowed us to obtain a Euclidean representation (the principal coordinates in a Cartesian coordinate system) of a phylogeny presented as a distance matrix. A major difference between this method and the conventional paired-contrasts method (Felsenstein 1985) is that with the former, phylogenetic relatedness among species can be used as a continuous independent variable, thus the sample size is the number of species in the study. This represents a significant advantage in studies involving relatively small sample sizes, such as ours. The phylogeny of the 15 client fishes was inferred from the taxonomy of Lecointre (1994) and Nelson (1994) (see Arnal et al. 2000). We obtained a 15×15 distance matrix (Euclidian distances) using the software PAUP*4.0 (Swofford 1999). Using this distance matrix, we performed a PCoA using DistPCoA software (Legendre and Anderson 1999) and generated 15 eigenvalues and the principal co-ordinates (eigenvectors) corresponding to positive eigenvalues. To obtain the best possible Euclidean approximation of the original distances, we selected eigenvalues using a broken-stick model (Diniz-Filho et al. 1998; Legendre and Legendre 1998). In our case, we selected only the first of the 15 eigenvalues, which represented 52% of the total variance in the distance matrix. This variable representing phylogenetic relatedness was then used in regression analyses.

We first assessed only the influence of mucus characteristics of 15 client species on each of the five dependent behavioural variables by including the following independent variables in multiple regressions: (1) mucus load (i.e. total dry weight) of client species, (2) mucus quality index, and (3) phylogenetic relatedness among client species (obtained from the PCoA analysis). Then, with the smaller sample of 8 species for which we obtained ectoparasite data, we first examined the inter-relationships between gnathiid and non-parasitic copepods (which are the two main crustacean prey of cleaning gobies on Barbadian reefs; Arnal and Côté 2000), client mucus characteristics, client size (mean standard length, measured on the individuals sampled), and phylogenetic relatedness among client species. Finally, we examined the influence of ectoparasite load on each of the five behavioural variables, by considering (1) the number of gnathiid isopod larvae and (2) the number of non-parasitic copepods, (3) the independent variables related to gnathiid or non-parasitic copepod loads identified above (to take into account their relative contribution) and (4) the phylogenetic relatedness among client species.

Since our data did not meet the assumption of normality, we tested the significance of our multiple regressions using a permutation method (Legendre et al. 1994; Legendre and Legendre 1998; PERMUTE! 3.4 software, P. Casgrain unpublished, avail-

able at <http://www.fas.umontreal.ca/BIOL/Casgrain/en/labo/index.html>). Multiple regressions were performed and repeated after each of 999 random permutations of the dependent variables. We used a backward stepwise procedure, dropping the least significant variable at each step until only significant variables remained in the model. All probability values are presented as one-tailed values.

Results

Influence of mucus load and quality

A total of 15 client species were included in these analyses. We found no relationship between mucus characteristics and either client tendency to visit cleaning stations, solicitation poses by clients, cleaning goby tendency to inspect certain client species, inspection effort or the biting rate of cleaning gobies on various clients. Neither mucus load nor quality was related to fish size (SL).

Influence of client ectoparasitism

Eight client species were included in these analyses. The mean number of gnathiid larvae per client species tended to increase with client standard length ($r^2=0.39$, $b=0.63$, $P=0.06$; Table 2). Gnathiid load was unrelated to mucus characteristics or to non-parasitic copepod number. The load of non-parasitic copepods was not related to mucus characteristics or to client body size. We found no relationship between fish gnathiid and non-parasitic copepod loads. Our multiple regressions therefore included gnathiid and non-parasitic copepod loads, client standard length, and phylogeny.

The number of gnathiid ectoparasites and the number of non-parasitic copepods proved to be good predictors of client visits to cleaning stations [gnathiids: $r^2=0.92$, $b=0.59$, $P=0.003$ (Fig. 1); non-parasitic copepods: $r^2=0.92$, $b=0.97$, $P=0.001$ (Fig. 2)]. However, client tendency to pose, the likelihood of inspection by cleaners, and inspection effort were not explained by client ectoparasite load. Cleaning gobies had a higher biting rate on client species having the lowest gnathiid loads ($r^2=0.48$, $b=-0.69$, $P=0.02$). Client body size did not explain a significant amount of variance for any of the behavioural variables.

Table 2 Mean standard length (\pm SE) and numbers of crustacean ectoparasites and non-parasitic copepods for eight client fish species ($n=5$ per species)

Client species	Standard length (cm)	<i>Gnathia</i> spp.	<i>Caligus</i> spp.	Copepod larvae	<i>Bomolocus</i> spp.	<i>Ergasilus</i> spp.	Non-parasitic copepods
<i>Acanthurus bahianus</i>	12.7 \pm 0.2	0.2 \pm 0.2	0.2 \pm 0.2	0	0.2 \pm 0.2	0	0
<i>Chromis multilineata</i>	6.9 \pm 0.1	0	0.4 \pm 0.2	2 \pm 1.3	0	1.2 \pm 0.8	35 \pm 17.4
<i>Haemulon flavolineatum</i>	10.2 \pm 0.7	0.8 \pm 0.3	1.2 \pm 0.6	0.2 \pm 0.2	0	0	2 \pm 1
<i>Haemulon chrysargyreum</i>	9.8 \pm 0.3	0.2 \pm 0.2	1.6 \pm 0.9	0.2 \pm 0.2	0	0	1.2 \pm 0.3
<i>Mulloidichthys martinicus</i>	15.6 \pm 0.3	0.8 \pm 0.3	1 \pm 0.3	0	0	0	0.2 \pm 0.2
<i>Scarus taeniopterus</i>	8.5 \pm 0.3	0	0	0	0	0	0
<i>Stegastes dorsopunicans</i>	6.7 \pm 0.2	0.4 \pm 0.4	0	0.4 \pm 0.2	0	0	1.6 \pm 0.8
<i>Stegastes partitus</i>	5.7 \pm 0.1	0	0	0.6 \pm 0.4	0	0	0.2 \pm 0.2

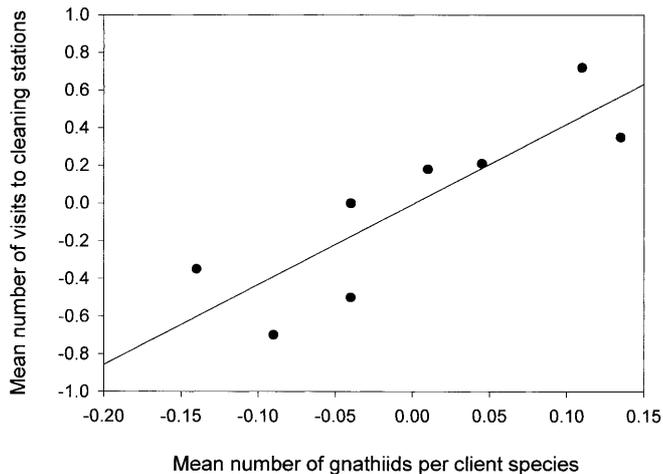


Fig. 1 Relationship between the number of visits by clients to cleaning stations and the mean number of gnathiid isopod larvae observed on clients (corrected for non-parasitic copepod number) ($n=8$ species). Variables were log-transformed prior to analysis

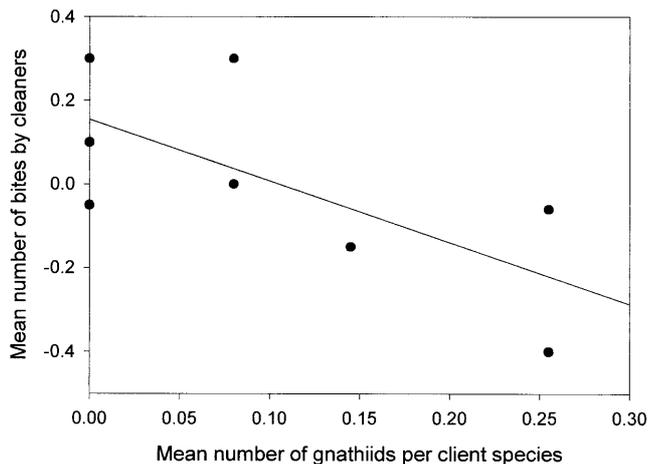


Fig. 2 Relationship between the number of bites taken by cleaning gobies on clients (corrected for duration of cleaning event) and the mean number of gnathiid isopod larvae observed on client species ($n=8$ species). Variables were log-transformed prior to analysis

Discussion

Ectoparasites appear to influence at least some aspects of client and cleaner behaviour on our Barbadian study reef. Client species hosting more gnathiid isopod larvae and non-parasitic copepods visited cleaners more often than less-parasitised species. Moreover, cleaning gobies fed at a slower rate on clients with higher gnathiid loads. However, variation in the likelihood of performing solicitation poses, cleaning specific clients and cleaning duration remain unexplained.

The link between client visits to cleaning stations and ectoparasite load, although intuitive, has long been elusive. For example, Losey (1979) found that parasitised surgeonfish, *Zebraflosoma flavescens*, in captivity visited a cleaner model more often than chemically deparasitised

counterparts, but no difference emerged between parasitised and clean butterflyfish, *Chaetodon auriga*. Body size appeared to be most important in determining proximity to cleaning stations by client species (Poulin 1993), and large-bodied species usually harbour more parasites (e.g. Poulin 2000). Grutter (1995a) found that body size, rather than ectoparasite load, was most closely correlated to the frequency of inspection of clients by *L. dimidiatus*. Note, however, that inspection frequency is not completely comparable to our measure of client visits since the former includes only visits that resulted in cleaning. It therefore reflects the mutual decisions of cleaners and clients rather than those of clients alone. In our field study, client species visited cleaning stations more often if they harboured higher loads of gnathiid ectoparasites, and this effect was independent of body size. A similar trend has been observed for clients of the Mediterranean cleaner wrasse, *Symphodus melanocercus* (C. Arnal and S. Morand, unpublished data). The importance of gnathiid isopod larvae for the diet of several species of cleaner fish has already been demonstrated (Losey 1974; Arnal and Côté 2000 for *Elacatinus* spp; Grutter 1997, 1999a for *L. dimidiatus*; Galeote and Otero 1998 for *Centrolabrus exoletus*; C. Arnal and S. Morand, unpublished data for *S. melanocercus*). Moreover, both Grutter (1999b) and Cheney and Côté (2001) have shown that cleanerfish can significantly reduce gnathiid abundance on their clients. Ectoparasite removal therefore appears to be the primary motivation of clients visiting cleaning stations on our study reef.

Interestingly, non-parasitic copepod load was also important in determining client visit rate. The role of these copepods in cleaning interactions has not yet been documented. Non-parasitic copepods are often observed in the stomachs of cleaner fish (Grutter 1997, 1999a; Arnal and Côté 2000); however, whether these are epibionts gleaned from client fish or free-living prey taken from the substratum is not clear. Our results suggest that they may be epibiotic, since our sampling method could not have captured benthic copepods from the substrate.

The tendency for clients to perform solicitation poses at cleaning stations was related neither to their ectoparasite load nor to their mucus characteristics. The former is particularly perplexing since posing is generally considered to enhance the likelihood of being cleaned (Hobson 1971; Potts 1973; Losey 1974; Côté et al. 1998). Using a theoretical cost-benefit approach, Côté et al. (1998) suggested that the relationship between posing and client ectoparasites could be variable. Depending on the rate of increase of posing benefits in relation to posing frequency, highly parasitised clients can be expected to pose either more or less than their less-parasitised counterparts. It is therefore perhaps not surprising to find no relationship between posing and ectoparasites in an interspecific correlation such as ours, in which species probably vary in how much posing increases their likelihood of being cleaned.

The interests of cleaners and clients on our study reef are not obviously consistent, at least as far as ectoparasite removal is concerned. Cleaners did not preferentially

clean clients with high ectoparasite loads, nor did they spend more time inspecting such clients. Moreover, client body length, which is related to levels of ectoparasitism (e.g. Poulin 2000), did not influence the likelihood of being cleaned or inspection duration (Arnal et al. 2000; this study). Cleaners might prefer to clean clients on which gnathiids are more easily detected or removed, rather than those that are simply carrying more ectoparasites. Another possible reason for this lack of influence of ectoparasites on cleaner behaviour may be the very low ectoparasite loads observed on Barbadian clients. Gnathiid abundance in our study (0.8 gnathiid/fish) was nearly sevenfold lower than that observed in other regions (e.g. 5.5 gnathiids/fish at Lizard Island, GBR; Grutter 1994), and indeed few ectoparasites are found in the gut content of *E. prochilos* (Arnal and Côté 2000). Although ectoparasite abundance can vary significantly both in space and time (Grutter 1994), additional surveys of ectoparasites on clients from other reefs in other seasons and other years in Barbados confirm the generally low abundance of gnathiids found in this study (Sikkel et al. 2000; Cheney and Côté 2001; K. Cheney, personal communication). Elsewhere in the Caribbean where ectoparasite loads are higher, *Elacatinus* gobies ingest more gnathiid larvae (Losey 1974). In such areas, client ectoparasite load may generally be more important in determining cleaner preferences and inspection duration (e.g. Grutter 1995a).

Surprisingly, cleaning gobies foraged at a slower rate on clients with more gnathiids. This suggests that cleaning gobies may be switching prey in relation to ectoparasite abundance, eating mainly (although not exclusively) items other than ectoparasites at a fast rate when foraging on clients with few gnathiids. Based on gut content analyses (e.g. Arnal and Côté 2000), such alternative items include mucus and scales, which may be ingested quickly due to their higher abundance and/or lower handling time than that for ectoparasites. It follows that the energetic value of ectoparasites should be greater than that of alternative items to justify this prey switching.

At first glance, our caloric estimates for mucus do not support this idea. The caloric values of mucus found in this study (1.8–4.3 Cal/mg DW) were similar to those calculated by Gorlick (1980) for the mucus of Hawaiian fish clients (2.6–4.7 Cal/mg DW), as well as those obtained for free-living copepods (3.5–4.1 Cal/mg DW; Zhang and Uhlig 1993). Contrary to Gorlick (1980), we found that the protein content of mucus is not negligible (39.4–72.8% DW) but is comparable to that of decapod crustaceans (50–72.5% DW; Anger and Schultze 1995). However, a meaningful comparison among food items can only be obtained by comparing their energetic value per bite taken by cleaners. This information is not currently available for ectoparasites or for mucus.

Our results generate two questions of relevance to the maintenance of honesty in cleaning symbioses. First, should cleaners also take mucus from highly parasitised clients? Second, should clients with low ectoparasite loads keep visiting cleaners? Both answers rely on the

cost to clients of having mucus removed (i.e. cheating), which has yet to be measured, relative to that of ectoparasite removal. If the cost of cheating is high, cleaners should refrain from ingesting items other than ectoparasites on highly parasitised clients in order to maintain a good reputation, which will preserve the mutualistic interaction (Ferrière 1998; Nowak and Sigmund 1998). A high cost of cheating would also make it unprofitable for clients with few ectoparasites to visit cleaners, despite the fact that cleaners should occasionally ingest parasites along with mucus. The fact that such clients still visit cleaners, albeit at a low rate, suggests either that the cost of mucus removal is low compared to the benefit of incidental parasite removal or that the cost is high but that clients can adopt various behavioural strategies to control cheating. Either way, the outcome of cleaning interactions may remain positive even in areas characterised by naturally low parasitism on clients.

Acknowledgements This study was made possible by a research grant from the Fisheries Society of the British Isles. C.A. was supported by a grant from the Région Languedoc Roussillon. We thank Dr. Wayne Hunte and the staff of the Bellairs Research Institute for help and access to facilities. We are also very grateful to Dr. A. Raibault for identifying the copepods.

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