

## Does client size affect cleaner fish choice of client? An empirical test using client fish models

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To determine whether the choice of client fishes in the cleaner fish *Labroides dimidiatus* was influenced by client size, cleaner fish were given a choice of equal amount of food spread on large and small client redfin butterflyfish *Chaetodon trifasciatus* models. All large models received bites from cleaners compared to 27% for small models. Seventy-nine per cent of cleaners took their first bite from the large fish model. The results suggest that client size may affect cleaner fish choice.

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In order to obtain food from client fishes, in the form of ectoparasites and other material found on the surfaces of fishes, cleaner fish *Labroides dimidiatus* (Valenciennes) interact with a large number of fishes. On the Great Barrier Reef, for example, individual cleaners inspect a mean 2297 fishes per day (Grutter, 1996). Yet what determines who gets cleaned and how often is still little understood. This is, in part, because both the client and the cleaner can initiate a cleaning bout (Losey, 1971). Teasing out what factors affect the outcome of cleaning interactions, therefore, is difficult.

It has generally been assumed that a positive relationship between client size and cleaning rate indicates a mutualistic relationship as larger fishes, also usually more parasitized (Grutter & Poulin, 1998a; Poulin, 2000), should seek cleaners more and be preferred by cleaners over clients with few parasites. Such a relationship has been examined in various ways (Poulin, 1993; Grutter, 1995; Grutter & Poulin, 1998b). Demonstration of a positive relationship between size and cleaning behaviour using observations of cleaner fish behaviour, however, was only recently done, with cleaner fish *L. dimidiatus* inspecting larger client

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species for longer (Bansemer *et al.*, 2002). This pattern was sustained even after the effect of client phylogeny was taken into account. A laboratory experiment using *Labroides phthirophagus* Randall showed that they preferred larger clients over small ones when offered a choice (Gorlick, 1978). The same species was used thus the comparison was not confounded by the taxonomy of the client. Because such studies do not control for client behaviour, which could potentially vary with size, the potential effect of the client's behaviour on the outcome of the interaction cannot be discounted. For example, client posing, which involves stereotyped posturing, is known to increase the likelihood that cleaners are cleaned (Côté *et al.*, 1998; Bansemer *et al.*, 2002). Thus, if large clients are older, and thus potentially more experienced at posing than small clients, their behaviour may be more attractive to cleaners and so they may be cleaned more simply because they are better posers.

To control for the effect of client behaviour on cleaner fish behaviour *L. dimidiatus* were trained to feed off trays (Bshary & Grutter, 2002), then offered food spread on two sizes of immobile models of the clients *Chaetodon trifasciatus* Park, and their behaviour recorded. *Chaetodon trifasciatus* was selected as the client because the deep, compressed body shape of this species (Randall *et al.*, 1997) was reasonably well represented by a laminated photograph on a thin plexiglass plate. Equal amounts (mass and area) of food were provided to control for the effect of food quantity available per model. *Chaetodon trifasciatus* are cleaned by *L. dimidiatus*; out of the 111 species cleaned by *L. dimidiatus* over 23.3 h of observations (Bansemer *et al.*, 2002), *C. trifasciatus*, was ranked 66 in how frequently it was cleaned (C. Bansemer & A.S. Grutter, unpubl. data).

Adult cleaner fish *L. dimidiatus* ( $n = 19$ ) were trained to feed on mashed prawns and fish flakes spread on Plexiglass plates (Bshary & Grutter, 2002) as part of another study examining their feeding preferences (Grutter & Bshary, 2004). Fish were held in captivity from 4 to 9 March 2003 when they were collected to when they were tested on 19 to 20 April 2003. They were tested by offering each a choice of a small (6.8 cm total length,  $L_T$ ) and a large (15.5 cm  $L_T$ ) client fish model with equal amounts of food on each. Both sizes represented sizes that occur in the wild with the larger model similar in size to the maximum size (15 cm  $L_T$ ) observed in the wild (Randall *et al.*, 1997). Models were laminated colour photographs of *C. trifasciatus*, made using the same digital image scanned from a single printed photograph and re-sized to obtain the two sizes using the software Photoshop 6.0. The photographs were printed on Epson Photo Paper S041140 using an Epson Stylus Photo 870 printer. Two replicates of the models were used. Models were attached to a clear Plexiglass sheet (34 × 18 cm) with the small and large models 5 and 4 cm from the bottom of the sheet, respectively, and 9 cm apart. Each model had 0.2 g of mashed prawn spread over a 2 × 2 cm area in the centre of the fish. The sheet was placed into the aquarium and cleaner fish behaviour recorded using a Sony Hi8 TRV89E video camera. A different cleaner fish was used in each trial. Two trials where the cleaner fish did not leave its shelter were omitted. The side of sheet that the large model was placed was random but balanced ( $n = 10$  on the right side and  $n = 9$  on the left side). The number of bites taken from each cleaner fish was counted for 3 min from when the cleaner took the first bite, usually within

15 s of placing the sheet in the aquarium. Cleaner fish had no prior experience with the models.

Cleaner fish took more bites from large client fish models than small ones (Wilcoxon signed rank test,  $n = 19$ ,  $P < 0.001$ ) (Fig. 1). All large models received bites, whereas only 27% of the small ones received bites. Of those models that did receive bites, the median number of bites (10% quartile/90% quartile) taken from large models was 15 (5/28) and 4 (2/21) for small models. Seventy-nine per cent of cleaners took their first bites from large models. Cleaners fed by either taking small bites or by using a 'swiping' motion. They often maintained very close contact with the model with their bodies parallel to the client, nibbled on the model as they would on a live fish, and occasionally touched the fish with their pelvic fins. Cleaners only searched the models and never the areas outside the models.

Cleaner fish chose large client fish models over small ones when given a choice, despite the amount of food on each model being equal. As the models were immobile, the cleaner's choice of model was independent of the 'behaviour' of the model. This suggests that size of client may influence the initial choice of a client by the cleaner *L. dimidiatus*.

Initially selecting clients based on their size should be an efficient method of choice. Most ectoparasites on many clients are small (0.14 to 2.7 mm) (Grutter, 1994) and many, for example benedeniine monogeneans, are cryptic (Whittington & Kearns, 1990; Whittington, 1996). Thus, cleaner fish may not easily detect most parasites on clients before approaching them. Fish size, which is a correlate of parasite load (Grutter, 1995; Grutter & Poulin, 1998a), however can easily be determined from a distance. Mucus, a preferred food item in laboratory tests (Grutter & Bshary, 2003), although not correlated with body size among species (Arnal *et al.*, 2001), is probably also correlated with client size (body surface) within a species.

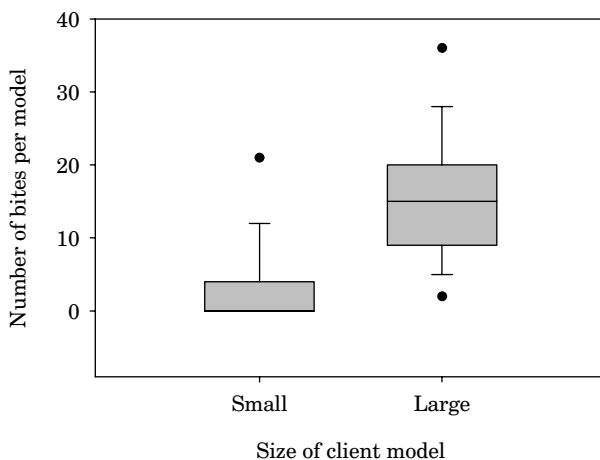


FIG. 1. Box and whisker plot of number of bites per 3 min taken by the cleaner fish *Labroides dimidiatus* on the client fish model *Chaetodon trifasciatus*. The centre line denotes the median value, the box encloses the inner two quartiles, error bars indicate the 90th and 10th percentiles and ● indicate outliers.

Other factors, in addition to client size, also probably play a role in client choice, such as a taxonomic preference for clients. For example, cleaners selectively feed on some parasites (Grutter, 1997; Arnal & Côté, 2000) and parasite composition and load is known to vary among client species (Grutter, 1994). Thus, some cleaner fishes may prefer some species based on their parasite composition. Whether clients are residents or 'visiting' fish species also affects the interactions between cleaners and clients (Bshary & Schaffer, 2002).

Although cleaner fish were offered the same amount of food per model they still chose the larger model. Thus, it was not the actual food that initially attracted them to the model. The models, however, were only available to the cleaners for 3 min. This was insufficient time for them to eat all the food and so consequently most cleaners spent most of their time with the larger model. This duration may have been too short for cleaners to learn that the amount of food on the small models was the same as on the large ones. For example, when offered similar amounts of mucus but from different fish, cleaner fish initially were not able to differentiate between the two but did on the second trial (Grutter & Bshary, 2004).

Cleaners appeared to treat models as they would real clients, with their feeding behaviour resembling that of cleaners feeding in the wild (A.S. Grutter, pers. obs.). Models of cleaner fishes have proved useful in previous studies on client cleaning behaviour (Losey, 1971; 1972; Losey & Margules, 1974) with tests revealing that the clients responded similarly to models and living cleaner fishes (Losey, 1977). Similar comparisons, however, had not been done using client models until now. The present results suggest that models of clients may be useful for studying the behaviour of cleaner fishes towards clients while controlling for client behaviour.

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