Frequency and distribution of melanistic morphs in coexisting population of nine clownfish species in Papua New Guinea

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Abstract Coral reef fishes exhibit extreme diversity in colouration, with many species being recognised as having colour polymorphism. While melanin-based colouration is known to occur in clownfishes (Pomacentridae), little is known about the frequency at which melanistic morphs occur in natural populations. The aim of this study was to assess the frequency of melanistic morphs in a coexisting population of nine clownfishes and to identify ecological covariates associated with morph distribution. In the Kavieng lagoonal system of Papua New Guinea (2°36′S, 150°46′E), melanistic morphs were found in *Amphiprion chrysopterus* (56 % of individuals), *A. clarkii* (29 %), *A. percula* (20 %), and *A. polymnus* (25 %) populations. *A. leucokranos*, *A. melanopus*, *A. perideraion*, *A. sandaracinos*, and *Premnas biaculeatus* populations were also surveyed, but were found to be without melanistic morphs. Unbiased recursive partitioning analyses identified a suite of interacting and conditional ecological factors encompassing social rank, host anemone species, and location effects as the primary factors predicting the distribution of melanistic morphs. Melanistic morphs were generally associated with host anemones from the genus *Stichodactyla* and with fish having a high social rank. The lack of a distinct melanistic morph locational 'hot spot' common to all species, despite locational coexistence, suggests that causative factors of location effects were different among species. Our results highlight the complexity of clownfish polymorphism associated with melanism, with multiple non-exclusive potential explanations identified for future investigation.

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Introduction

Extreme diversity in colouration among coral reef fishes extends to some species exhibiting colour polymorphism (Fautin and Allen 1997; Planes and Doherty 1997; Bernardi et al. 2002; Messmer et al. 2005). Species may exhibit colour morph variation both within and among populations, and even within individuals over time (Messmer et al. 2005; Cortesi et al. 2015). The evolutionary processes favouring the persistence of colour polymorphisms within populations have received considerable attention and have been attributed to a consequence of developmental plasticity, in which the trajectories of developing organisms diverge under the influence of environmental and/or genetic

cues. Colour morphs can also represent differences in traits other than appearance. Colouration is often the most obvious phenotypic expression of a suite of differences in morphological, physiological, and behavioural traits between morphs that may otherwise go unnoticed (McKinnon and Pierotti 2010; McLean and Stuart-Fox 2014). These trait complexes represent fitness optima and may allow multiple morphs to coexist in a population by balancing phenotype frequencies (Maynard-Smith 1966; Sinervo and Lively 1996).

Colour polymorphism has been noted to occur in clownfishes (Pomacentridae) at multiple scales. Geographical variation in colouration is common among widely distributed species, such as *Amphiprion clarkii* (Moyer 1976; Bell et al. 1982; Fautin and Allen 1997) and *A. melanopus* (Drew et al. 2008). Sympatric variation is also reported in some populations where sexual dichromatism and ontogenetic differences govern colouration (Moyer 1976; Fautin and Allen 1997). These variations are inclusive of a reduction of stripes as juvenile fish mature (Fautin and Allen 1997) and changes in caudal fin colouration for some species (Moyer 1976). The occurrence of melanism, which comprises a blackening of the body pigmentation, is also reported to occur within localised populations of multiple clownfish species, contributing to polymorphism (Allen 1972, 1973; Moyer 1976; Fautin and Allen 1997). This darkening of the body pigmentation can extend to the anal, pelvic, and dorsal fins, but does not impact on the prominent white stripe patterns. Transplant studies suggest there is capacity for environmental cues to regulate the expression of melanistic characteristics (Allen 1972, 1973) though production of melanistic clownfishes through culture coupled with genetic differences among clownfish colour morphs (Litsios et al. 2014) is also suggestive of genetic regulation to some degree. The occurrence of melanism within natural clownfish populations has been attributed to host anemone species (Allen 1972, 1973; Moyer 1976; Fautin and Allen 1997), social dominance (Moyer 1976), and local variations in habitat (Bell et al. 1982), suggestive of a synergy between host anemone and social interactions. The extent to which these attributes influence the distribution of melanistic morphs within populations of clownfish has yet to be fully explored, with past studies being limited by sample size and restricted to study of *A. clarkii*.

It is surprising that the study of colour polymorphism in clownfishes has received little attention to date despite extractive activities, such as the marine aquarium trade, selectively targeting morphs in some instances (unpublished data; Wabnitz et al. 2003). Establishing baseline frequencies at which melanistic morphs occur and their associated ecological variables also serves as a prerequisite to assessing whether certain morphs are under disproportional selective pressures. The importance of habitat and social interactions as predictors of the distribution of morphs can also help inform fishery management agencies in conserving and managing these resources.

The aim of this study was to assess the frequency and distribution of melanistic morphs in a coexisting population of nine clownfish species within the Kavieng lagoonal system in Papua New Guinea (PNG) with respect to ecological variables. We specifically determined whether host anemone species, anemone size, surrounding substrata, location at two spatial scales, depth, total fish inhabiting the anemone, presence of other cohabiting species, total conspecifics inhabiting the anemone, and social rank within the anemone were associated with melanistic morphs.

Materials and methods

Population surveys

Natural populations of clownfishes were assessed within the Kavieng lagoonal system of New Ireland Province, PNG (2° 36′S, 150° 46′E), during October 2013. At the time of the study, this location had no history of exploitation of clownfishes for the marine aquarium trade or research, suggesting a virgin population. The Kavieng lagoonal system comprises the Kavieng Harbor (Fig. 1) and the larger Kavieng Lagoon (ca. 18 km long and 12 km wide) through which three distinct island chains extend, parallel to its seaward edge. Within the lagoon, depth rarely exceeds 20 m between islands, with a maximum depth inside the lagoon of approximately 30 m. Outside the lagoon, the seafloor gradually slopes into the Pacific Ocean reaching a depth of 200 m approximately 7 km from the outer island chain. Most of the coral reef development is restricted to fringing reefs along the shorelines of islands and the main landmass. North-easterly winds produce large swells, which render the barrier island (Fig. 1) reefs highly exposed to these oceanic influences. The mid-lagoon island chain (Fig. 1) experiences intermediate conditions, with the nearshore reefs (Fig. 1) being heavily sheltered by comparison.

Water clarity improves, and fluctations in salinity decrease, at sites further offshore within the Kavieng lagoonal system. Freshwater run-off from the main land mass can lower the salinity and increase the turbidity of nearshore waters after substantial rainfall. Similarly, the community compositions of reef-associated organisms vary between inner and outer island reefs, with species diversity increasing with seaward progression through the lagoon (Hamilton et al. 2009).

Given the scarcity of the target species, GPS-tracked surface swims were conducted between 1000 and 1500 h over a cumulative 50 km of shallow-water habitat. All
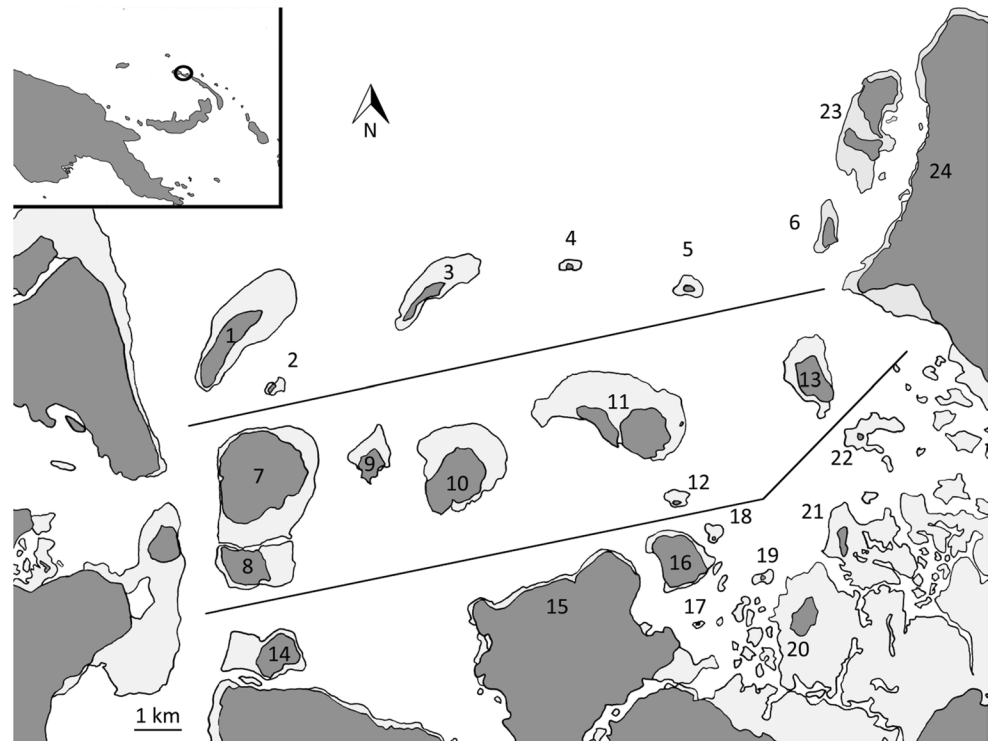
surface swims were conducted along 2- and 5-m depth contours simultaneously, with personnel using snorkel surveying 2.5 m to either side of the isobath for the presence of the target species. When found, the host anemone and inhabiting clownfish were identified to species and notes were made regarding whether individual fish could be best described as non-melanistic or melanistic morphs (Online Resource). This was done using visual observation from a distance; once data were recorded for all visible individuals, the host anemone was measured across the largest and smallest axis of its oral disc and gently probed with a blunted rod to identify the presence of any cryptic fish not initially visible. For the host anemone, *E. quadricolor*, which sometimes forms colonial aggregations, the colony was considered as a single anemone and axis measurements were taken of the colony. For other anemone species, fishes were rarely (5.2 % of all anemones) observed to occupy multiple anemones during the period of observation. Where such instances were observed, the anemone in which the fishes spent the majority of time in, or in the proximity of, was considered the host anemone for the purposes of this study. The absence of observed migration of fishes between anemones was taken to signify anemones and their fishes as separate entities. Fish within a given anemone were assigned a social rank score based on relative size and behaviour. The dominating substratum within a 2 m radius of the host anemone was recorded by visual approximation. Substrata were classified as fine, loose sediments (including sand and silt), loose rubble (including

dead coral and rock pieces), solid rock (including sedimentary and igneous), and live coral (including Scleractinia and Alcyonacea). The GPS coordinates of each anemone's location were taken at the surface (Garmin 72H).

Data analysis

Host anemone size was calculated from the elliptical area of the oral disc/colony using the largest and smallest axis measurements taken in the field. We conducted unbiased recursive partitioning using the ctree function in the R package *partykit* (Hothorn and Zeileis 2015) to determine whether primary host anemone species (Table 1), anemone size, surrounding substrata, zonation within the lagoonal system (i.e. nearshore, mid-lagoon, barrier island, Kavieng Harbor), island/reef location, depth, total number of fish inhabiting the anemone, total number of conspecifics inhabiting the anemone, presence of other cohabiting species, and social rank significantly predicted the distribution of melanistic morphs. This analysis relies on a permutation-based significance test to select variables and avoids the bias of some recursive partitioning methods that tend to select variables with many possible splits or many missing values (Hothorn et al. 2006). This statistical approach identified the best predictors of melanistic morph distribution, but remains limited in the capacity to elucidate causation as predictive factors may be confounded by correlations with other significant predictors. Ecological variables predicting melanistic morphs were determined for *A. chrysopterus*, *A.

Table 1 Host utilisation by clownfishes and the frequency of melanistic morphs in the Kavieng lagoonal system, Papua New Guinea

Fish	Anemone species % of total number of host anemones surveyed (total number surveyed)								
	Total (n = 2253)	Eq 34 % (n = 765)[a]	Ha <1 % (n = 7)	Hc 33 % (n = 747)	Hm 13 % (n = 290)	Md <1 % (n = 2)	Sg 6 % (n = 139)	Sh 1 % (n = 29)	Sm 12 % (n = 274)
Amphiprion chrysopterus	182 (no. of fish) 56 % (melanistic)			**109** **40 %**					**73** **79 %**
A. clarkii	2162 29 %	1 0 %	18 17 %	**1612** **9 %**	1 0 %		4 0 %	23 22 %	**503** **93 %**
A. leucokranos	57 0 %			**26** **0 %**					**31** **0 %**
A. melanopus	1428 0 %	**1364** **0 %**		62 0 %			2 0 %		
A. percula	1068 20 %				**792** **2 %**		**275** **70 %**		1 100 %
A. perideraion	274 0 %			1 0 %	**270** **0 %**		3 0 %		
A. polymnus	106 25 %			1 100 %				**105** **25 %**	
A. sandaracinos	321 0 %			4 0 %					**317** **0 %**
Premnas biaculeatus	1343 0 %	**1340** **0 %**		1 0 %		2 0 %			

Anemones surveyed are abbreviated as follows: *Entacmaea quadricolor* (Eq), *Heteractis aurora* (Ha), *H. crispa* (Hc), *H. magnifica* (Hm), *Macrodactyla doreensis* (Md), *Stichodactyla gigantea* (Sg), *S. haddoni* (Sh), *S. mertensii* (Sm).

Bold values indicate 'primary' host anemone species hosting more than 5 % of a given clownfish's population within the lagoonal system

[a] Anemones of this species coexisting as colonies were considered as n = 1

clarkii, *A. percula*, and *A. polymnus* independently. Given the large samples sizes of *A. clarkii* (n = 2162) and *A. percula* (n = 1068), node size was controlled by implementing a minimum node size of 30; without this restriction, many nodes result, each comprising a small sample size and rendering interpretation uncertain. No node control was implemented for *A. chrysopterus* and *A. polymnus*, because sample sizes were considerably smaller. In all cases, only significant (P ≤ 0.05, with Bonferroni's correction) splits were considered.

Results

A total of 6941 clownfish individuals were observed from nine species, while 2253 host anemones were observed from eight species. Melanistic morphs were apparent within the localised population of clownfishes, but were not ubiquitous to all species of clownfish within the Kavieng lagoonal system. Melanistic morphs were observed to occur in only four (*A. chrysopterus*, *A. clarkii*, *A. percula*, and *A. polymnus*) of the nine species present (Table 1).

Eight of the nine clownfish species were found to associate with multiple anemone species, the exception being

A. polymnus, which associated exclusively with *S. haddoni* on loose sediment substrata (Table 1). For all species of clownfish observed, only one (*P. biaculeatus*, *A. melanopus*, *A. perideraion*, *A. polymnus*, *A. sandaracinos*) or two (*A. clarkii*, *A. chrysopterus*, *A. leucokranos*, *A. percula*) host anemone species accounted for more than 5 % of a clownfish's population within the lagoonal system. Those anemone species contributing to a minority of a clownfish's population (<5 %) were not considered primary hosts for the species.

Distribution of melanistic morphs

The selected ecological variables were capable of predicting the occurrence of melanistic morphs with a high degree of accuracy for *A. chrysopterus* (81.3 % correct), *A. clarkii* (91.0 % correct), *A. percula* (91.5 % correct), and *A. polymnus* (89.5 % correct) with a unique suite of predictor variables for each species (Figs. 2, 3, 4, 5).

For *A. polymnus*, the likelihood of occurrence of melanistic morphs was initially associated with a fish's social rank within a given anemone (P < 0.001): the largest fish within an anemone (i.e. social rank 1) was more likely to be melanistic than fish with social ranks ≥2 (Fig. 2). Further
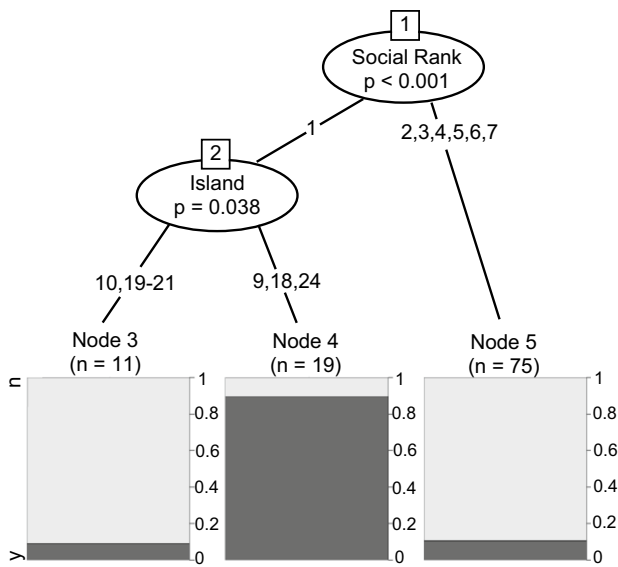
Fig. 2 Conditional inference tree for ecological variables predicting the distribution of melanistic morphs within the *A. polymnus* population. *Dark shading* indicates the proportion of melanistic individuals at each terminal node. Island numbers refer to reefs in Fig. 1

separation was based on island location, but only for the largest fish within an anemone ($P = 0.038$). This split showed a clustering of non-melanistic *A. polymnus* towards the Kavieng Lagoon's interior (islands 19–21) with melanistic morphs generally occurring closer to the lagoonal system periphery (islands 9, 18, 24 in Fig. 1).

Social rank was also the primary determinant of the occurrence of melanism in the *A. chrysopterus* model, with the largest fish within an anemone being more likely to be melanistic than fish of social ranks ≥2 ($P < 0.001$; Fig. 3). Where the largest fish occupied an anemone with >2 total fish, they were more likely to be melanistic (97.4 % of fish in this node were melanistic) by comparison with those on anemones housing fewer fish ($P > 0.015$). For fish of social ranks ≥2, anemone species defined the next split, differentiating between fish inhabiting *H. crispa* and those inhabiting *S. mertensii*; the latter were more likely to be melanistic ($P < 0.001$). A further division of fish occupying *S. mertensii* showed there to be a critical anemone size where anemones >1220 cm² had a higher proportion of melanistic fish (100 % melanistic) by comparison with those inhabiting smaller anemones (50 % melanistic, $P = 0.045$).

Predictors of melanism for *A. percula* indicate an importance of both depth and anemone species (Fig. 4). Anemones in waters deeper than 1.7 m showed melanistic morphs were more frequently encountered in *S. gigantea* than in *H. magnifica* ($P < 0.001$). For anemones ≤ 1.7 m depth, likelihood of melanism was first associated with reef location ($P < 0.001$), but then also split by anemone species, with *S. gigantea* having more melanistic morphs than with *H. magnifica* at either group of locations. The location split demonstrated a concentration of melanistic morphs at the most interior reefs (islands 16–17, 19–21 in Fig. 1) of the lagoonal system. The majority of *S. gigantea* anemones
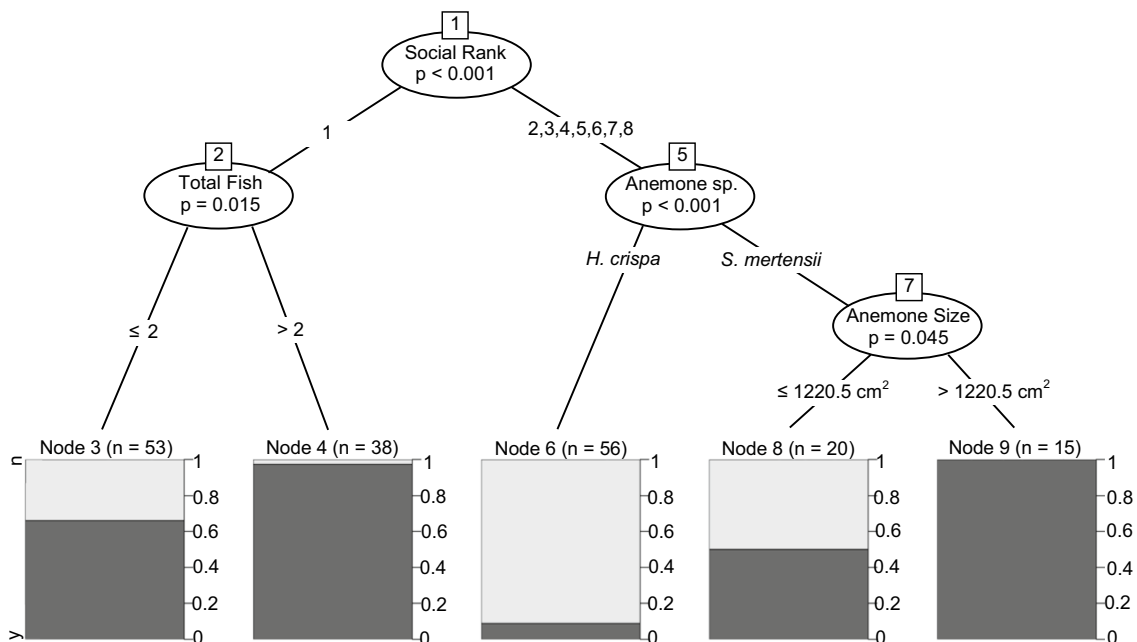

Fig. 3 Conditional inference tree for ecological variables predicting the distribution of melanistic morphs within the *A. chrysopterus* population. *Dark shading* indicates the proportion of melanistic individuals at each terminal node
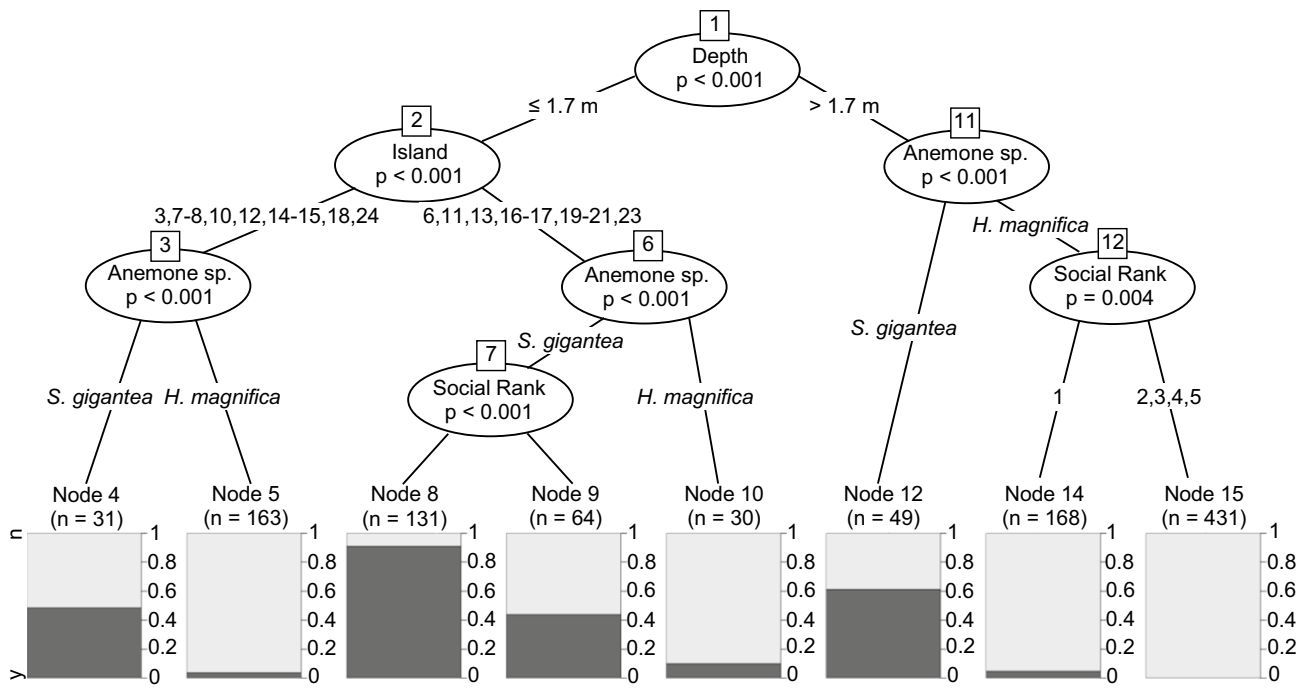
Fig. 4 Conditional inference tree for ecological variables predicting the distribution of melanistic morphs within the *A. percula* population. *Dark shading* indicates the proportion of melanistic individuals at each terminal node. Island numbers refer to reefs in Fig. 1
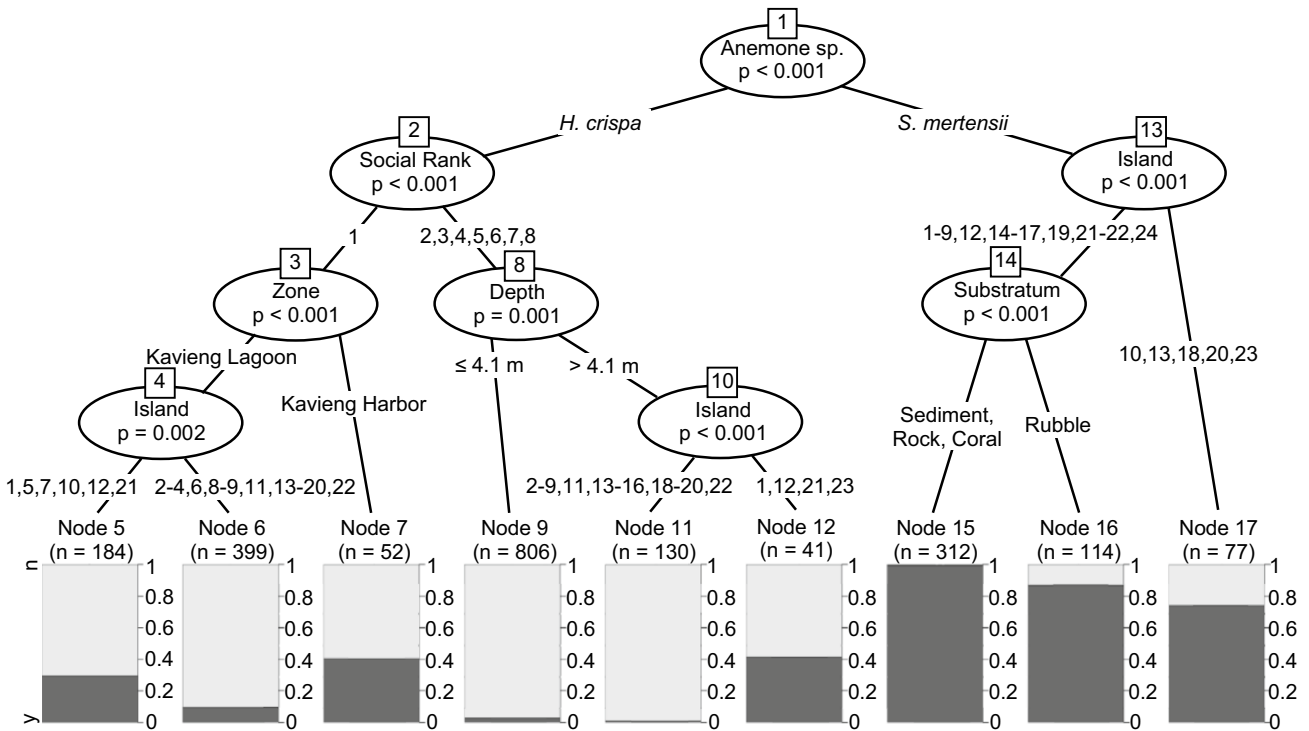

Fig. 5 Conditional inference tree for ecological variables predicting the distribution of melanistic morphs within the *A. clarkii* population. *Dark shading* indicates the proportion of melanistic individuals at each terminal node. Island numbers refer to reefs in Fig. 1. The Kavieng Lagoon zonation is inclusive of the nearshore, mid-lagoon, and barrier island zones

were found at these latter reefs, and within this anemone species, social rank was found to be a significant predictor of melanistic morph occurrence, with social ranks 1–2 more likely to be melanistic than social ranks ≥3 ($P < 0.001$).

The predictive tree for *A. clarkii* (Fig. 5) was the most complex, but demonstrated the importance of anemone species in predicting the occurrence of melanistic morphs, with more melanistic fish found on *S. mertensii* by comparison with *H. crispa* ($P < 0.001$). The 9 % of melanistic fish (Table 1) known to inhabit *H. crispa* were split based on social rank, with melanistic morphs being associated more often with fish of a social rank 1. Zonation within the lagoon, reef location, depth, and substratum were also significant predictive factors resulting in further refinement of the model (Fig. 5).

Discussion

Melanistic morphs were either present in clownfish populations at a relatively high frequency (≥ 20 %) or entirely absent. The absence of melanistic morphs within localised populations of *A. leucokranos*, *A. melanopus*, *A. perideraion*, and *A. sandaracinos* mirrors prior studies where variations in colouration are restricted to deviations in stripe patterning for these species (Allen 1972, 1973; Fautin and Allen 1997; Drew et al. 2008). In species where melanistic morphs were present, the distribution of morphs can be predicted from a suite of interacting and conditional ecological factors primarily encompassing social rank, host anemone species, and location effects. Given that melanism-based colouration is frequently implicated in social communication (Majerus 1998) and that it covaries with many other physiological, morphological, and behavioural traits (Roulin 2004), different, and possibly competing, selective pressures likely govern the distribution of melanistic morphs in populations. This explains why prior studies have implicated different causative factors, such as social dominance (Moyer 1976) and host anemone species (Allen 1972, 1973; Moyer 1976; Fautin and Allen 1997), to explain the distribution of clownfish melanistic morphs within localised populations.

The use of unbiased recursive partitioning provides new clarity to the ecological factors predicting the distribution of melanistic clownfish morphs within a localised population. The importance of *Stichodactyla* spp. anemones as championed by Allen (1972, 1973) in governing the distribution of melanistic morphs is clear in that *A. chrysopterus*, *A. clarkii*, and *A. percula* all demonstrated a higher proportion of melanistic morphs associated with *Stichodactyla* spp. compared to *Heteractis* spp. primary hosts. However, the current study dispels notions that host anemone species alone dictates the expression of melanism.

The simplest example of this is with *A. polymnus*, a species observed in the current study to primarily inhabit only a single species of anemone (*S. haddoni*). For this species, there was a clear influence of social rank as the most significant predictive factor where the dominant and, presumably, female fish (Moyer and Nakazono 1978) was more likely to be a melanistic morph. However, the influence of social rank was conditional, with a clear influence of island location. At some islands, female fish were as unlikely to be melanistic as males or sub-adults (social ranks ≥2). A further example of melanin-based colouration occurring independent of host anemone species is evidenced from *A. chrysopterus* where fish of the highest social rank (i.e. female) were more commonly melanistic regardless of host anemone species; the importance of host anemone species in the distribution of melanistic morphs was only realised for male or sub-adult fish of this species.

The relocation of individual fish between different host species of anemone, and the resulting changes in melanism morph colouration have historically been taken as evidence that the host anemone species influences the distribution of melanistic morphs (Allen 1972, 1973). However, relocating a fish from one host anemone species to another (Allen 1972), particularly into an aquarium (Allen 1973), will inevitably coincide with a change in social pressures and finer-scale ecological differences. Given the significance of social rank and location effects in influencing the distribution of melanistic morphs revealed by our study, manipulation experiments failing to control for such variables should be interpreted with caution. This should not be taken to negate the importance of host anemone species in predicting the distribution of melanistic morphs, but it reiterates that a suite of interacting and conditional ecological factors influence distribution.

While differences based on social rank may be viewed to identify with sexual dichromatism (Moyer and Nakazono 1978), we argue the association of melanism with social rank is more reflective of a need to display dominance. Research across vertebrate taxa shows aggression to be positively linked to melanism on the basis of pharmacological studies and patterns in wild populations (Ducrest et al. 2008). In comparing within-population variation of 20 vertebrate species, melanistic individuals were more aggressive than non-melanistic individuals for 18 of the studied species (Ducrest et al. 2008). Within our study, evidence associating social rank with social dominance was most pronounced with *A. chrysopterus*, where the likelihood that a female fish was melanistic was greatest when there was a high density (>2) of clownfish inhabiting the host anemone. Density-dependent melanism was also suggested for *A. clarkii* populations in Japan, where melanistic fish were observed to be socially dominant over non-melanistic fish within the same aggregation (Moyer 1976). In our study, social rank was a significant predicative variable for *A. clarkii* inhabiting *H. crispa*; however, this was not

directly attributed to fish densities within anemones but rather location effects.

With the exception of *A. chrysopterus*, all species with melanistic morphs in their populations demonstrated location effects. The causative factors of these location effects remain unresolved; however, the clustering of location effects to some of the interior-most islands of the lagoonal system suggests against random distribution. Finer-scale ecological variables (i.e. diet, turbidity, anemone density) are likely to vary between locations, and these might coincide with benefits derived from the physio-chemical properties of melanin pigments, such as resistance to solar radiation and oxidative stress (Ducrest et al. 2008). The possibility of the high self-recruitment (up to 42 %) known to occur in clownfish populations (Jones et al. 2005; Saenz-Agudelo et al. 2012) could facilitate maintenance of such pleiotropy at individual islands.

It was interesting that the location effects did not reveal a melanistic morph locational 'hot spot' common to all species. The interior-most islands of the lagoonal system (islands 19–21 in Fig. 1) were identified as significant predictors of melanistic morph distribution for both *A. percula* and *A. polymnus*. While for *A. percula* these interior islands were associated with a higher proportion of melanistic phenotypes, the opposite was true for *A. polymnus*. For *A. clarkii*, which also exhibited significant location effects, the interior-most islands were divided, with contrasting associations in terms of the distribution of melanistic morphs. All three species are known to commonly inhabit nearshore areas (Elliot and Mariscal 2001; Saenz-Agudelo et al. 2012) and would likely have been exposed to similar environmental conditions at these islands. The lack of a consistent location effect on the distribution of melanistic morphs, despite locational coexistence, suggests that the finer-scale ecological variables influencing the distribution of melanistic morphs vary between species. Given the multitude of pleiotropic effects associated with the expression of melanin-based colouration (Ducrest et al. 2008), such an assumption is plausible and warrants further investigation.

We found that some of the assessed ecological covariates failed to predict the distribution of melanistic morphs. There appears to be little influence of heterospecifics inhabiting the same host anemone or of substrata on the distribution of melanistic morphs of the surveyed species. While substratum was significant for *A. clarkii*, this was only for *S. mertensii* anemones restricted to certain locations, resulting in only a small conditional effect on melanistic morph frequency. It is possible that the forced association with a host anemone for survival (Buston and García 2007) mitigates the impact of substrata as shown to explain polymorphisms of other reef fish (Munday et al. 2003). We also question the importance of depth as returned by the unbiased recursive partitioning for *A. percula*. The

importance of depth may be a product of unequal distribution of host anemone species across the critical depth value (1.7 m). Only 17.8 % of *A. percula* occupying *S. gigantea* were found at depths >1.7 m, while 75.6 % of *A. percula* occupying *H. magnifica* were found in such habitats. Thus, the importance of depth may be a proxy for the importance of host anemone species. Despite melanin pigments being shown to provide resistance to solar radiation in mammalian species (Slominski et al. 2004), the near absence of melanistic *A. percula* in shallow-water *H. magnifica* implicates the significance of depth is due to anemone distribution rather than depth alone.

Application and conclusions

Polymorphisms among coral reef fishes are known to arise for a number of reasons, including: (1) spatial or temporal variation in the relative fitness of different morphs (Planes and Doherty 1997; Randall 2005; Cortesi et al. 2015); (2) species developing different colours in response to habitats differing in depth, reef aspect, coral coverage, or associated fish assemblages (DeMartini and Donaldson 1996; Munday et al. 2003; Cortesi et al. 2015); (3) contrasting preferences for settlement habitat; (4) ontogenetic differences (Moyer 1976; Painter et al. 1999); (5) sexual dichromatism (Kodric-Brown 1998); (6) genetic differences (Drew et al. 2008, 2010); (7) different diets (Whitman et al. 2007); and (8) hybridisation (Gainsford et al. 2015). The complexity of morph variation manifests within clownfishes in that a single breeding pair of clownfish can produce, in culture, a stable range of polymorphic individuals that resemble different species (Marliave 1985). To identify the causative factors behind melanin-based colouration would require a range of manipulative field and laboratory experiments, combined with genetic evaluation. Deriving ecological associations with the frequency and distribution of polymorphic traits within a localised population is a first step in elucidating causation. It is apparent from past studies on pomacentrids and other reef fishes that melanin-based colouration can be regulated to enhance fitness under alternating environmental conditions (Frédérich et al. 2010; Cortesi et al. 2015). Further investigation into the observed location effect would help resolve why host anemone species and social rank are associated with melanistic morph distribution only at some locations.

There is a paucity of research investigating the frequency at which melanistic morphs occur in localised populations of reef fish. Understanding natural frequencies of colour morphs at localised scales is critical to monitoring the impact of exploitative activities selective of colour morphs. Colouration plays a pivotal role in selection of organisms in food fisheries and the marine aquarium trade (unpublished data; Wabnitz et al. 2003; Murray and Watson

2014), and clownfishes are among the most demanded species for the latter (Rhyne et al. 2012). Assessing the frequency of different polymorphisms in wild populations of clownfishes is a first step to ensure disproportional harvesting of select morphs is appropriately managed. Identifying ecological variables associated with the occurrence of colour morphs (whether causative or correlative) can facilitate conservation and management efforts in the face of exploitation until further research can evaluate risks from selective harvesting.

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