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SIGNAL FUNCTION OF WING COLOUR IN A POLYMORPHIC DAMSELFLY, *MNAIS COSTALIS* SELYS (ZYGOPTERA: CALOPTERYGIDAE)

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Mnais costalis ♂♂ exist in 2 forms specialised for the demands of 2 distinct strategies, territorial fighters and non-territorial sneaks, which give approximately equal fitness payoffs. Territorial ♂♂ have orange wings, whereas typical non-territorial ♂♂ are clear-winged. By simulating agonistic encounters between ♂♂ it is shown that the 2 morphs showed distinct responses to the signal from orange wings: territorial orange-winged ♂♂ always tried to enter contests, while clear-winged ♂♂ always avoided them. On the other hand, the 2 morphs showed similar responses to the signal from clear wings: both morphs tried to attack models. Also presented are 'painted clear models' which were clear-winged ♂♂ whose wings had been painted orange, and both morphs responded as if they were orange-winged models. These observations indicate that ♂♂ discriminate between fighter and sneaker morphs using the colour of wings, and shows different styles of agonistic responses toward fighter and sneaker morphs. It is likely that non-territorial sneaks may gain an advantage from non-signalling because clear wings increase crypsis on another ♂ territory, increasing their success in stealing copulations. No indication was found that clear-winged ♂♂ are ♀ mimics, or that having clear wings reduced the level of aggression directed towards them by territorial orange-winged ♂♂.

INTRODUCTION

Attempts to understand signal function, and the information transmitted by a particular trait, often involve correlating variation in the expression of a trait with other known fitness parameters (e.g. QVARNSTRÖM et al., 2000). Exper-

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imental studies have manipulated the level of expression of the trait and then recorded the effect of the manipulation has on the reproductive success or fitness of its bearer. An alternative approach is to study systems in which some individuals within the population do not signal at all. Non-signallers may be unable to pay the high social costs elicited by bearing a signal. Pigmented areas of plumage ('badges of status') are used to this effect in Harris' sparrows (*Zonotrichia querula*), enabling some males to avoid costly aggression at feeding sites (ROHWER, 1975). In these cases 'non-signallers' are likely to be inferior individuals. However, in some other species not signalling may be adaptive because it increases the success of an alternative mating strategy. For example, in crickets (*Gryllus integer*) signalling males call to attract females, and non-signalling males silently search for females (CADE, 1979). In genetically polymorphic ruff *Philomachus pugnax* dark necked resident males strongly repel other dark necked males, but tolerate the paler submissive satellite males (LANK et al., 1995; WIDEMO, 1998) which enables satellite males more opportunity to obtain sneak copulations.

In the polymorphic *Mnais costalis* (for local variation of the polymorphism, see HAYASHI et al., 2004), males eclose as one of two morphs, which we call 'fighters' and 'sneaks' (TSUBAKI et al., 1997). The development of the two morphs is genetically controlled (TSUBAKI, 2003). Fighters are territorial, defending submerged pieces of dead wood into which females oviposit, and possess large sexually selected orange patches on their wings and a red pterostigma at the wing tip. In contrast, sneak males are non-territorial males, have no orange wing patch (although they do have the same red pterostigma), are smaller than territorial males (TSUBAKI et al., 1997), and obtain most of their reproductive success by 'stealing copulations' on other males' territories. Females' wings are also clear, but are distinguished from those of non-territorial males by the presence of a white pterostigma (HOOPER et al., 1999). In addition to the behavioural differences between the two morphs, previous studies have also shown physiological differences resulting from a selective trade-off: fighter males have evolved specialisations typically found in territorial odonate species, while sneak males have evolved specialisations typical of non-territorial odonate species (PLAISTOW & TSUBAKI, 2000). The associated costs and benefits of such different specialisation are likely to account for differences in reproductive lifespan, which results in the two strategies having equal fitness (TSUBAKI et al., 1997).

The orange wing colour in the territorial morph, unlike in most other calopterygids (SIVA-JOTHY, 2000) varies with nutrient levels and age, suggesting that orange wings are costly to produce and maintain (HOOPER et al., 1999). Thus one explanation for why the smaller clear-winged males do not produce the orange signal is that they are avoiding the energetic cost required to do so. Morph fitness is equal (TSUBAKI et al., 1997), and selection pressures are different (PLAISTOW & TSUBAKI, 2000), suggesting that non-signalling may provide

additional selective benefits to clear-winged males. There are three ways non-territorial sneaks may gain an advantage from non-signalling: (1) clear wings increase crypsis on another males' territory, (2) clear wings enable non-territorial males to mimic females, and/or (3) clear wings induce less aggression from territorial orange-winged males. These hypotheses are not mutually exclusive, however they can be tested separately because they are related to the aggressive responses occurring at different stages of a behavioural sequence: the first hypothesis is concerned with the detection of conspecifics flying nearby, the second is concerned with morph recognition after detection, and the third is a behavioural decision made after morph recognition. In this study we made field observations to assess the first hypothesis, and we measured the response of the two morphs to control and experimental models of each morph (or a female), in order to determine the function of the orange wing-spot and test the second and the third hypotheses.

METHODS

STUDY SITE AND FIELD OBSERVATIONS. — Insects were observed at a mountain woodland stream in Gozenyama, Ibaraki Prefecture, Japan (36°33'N, 140°17'E) in 1998. In both years all adults present in, and entering, a 20 m stretch were captured and individually marked with enamel pens. Left hind wing length was taken as a measure of body size.

Between May 24 and July 4 1998, we made daily focal studies of all individuals in this stretch of river, from 1200 hr to 1500 hr. We recorded the territorial status of each male, and scored the number of copulations each secured. Individuals seen on only one day were excluded from the analyses. Mating success for each individual is presented as the average number of copulations obtained per day.

MODEL EXPERIMENTS. — We gauged the response of different morphs to different signals by artificially flying dead males mounted on wire into orange-winged males' territories, or into the airspace of perched clear-winged males. Dead males are hereafter referred to as 'models'. Mounting the models on wires allowed us to simulate natural flight patterns by vibrating the wire. In 1997, we used three types of model. 'Real orange-winged models' were simply dead orange-winged males mounted on wire. 'Painted clear-winged models' were clear-winged males whose wings had been painted orange with a felt-tipped artist's pen (Copic). The third type, 'control clear-winged models', were dead clear-winged males. We 'flew' the models at the subject male for 30s and recorded the amount of time he attacked the model. If the subject male escalated during the encounter, which in *M. costalis* amounts to sharp vertical jumps (NOMAKUCHI et al., 1984), we similarly escalated the 'fight' of the model. All males along the 50m stretch of river were subjected to 30s trials with each model, in a random order. Each male was subjected to the same model only once but was tested with three different models unless it disappeared (a few clear-winged males disappeared after the first or the second tests). We allowed at least 15 minutes between successive trials on the same subject male, and in this experiment males were individually marked with a series of coloured dots on the abdomen.

An additional experiment assessed the response of orange-winged males resident on territories to a dead female model, presented mounted on wire in a similar way to the male models.

RESULTS

DAILY MATING SUCCESS

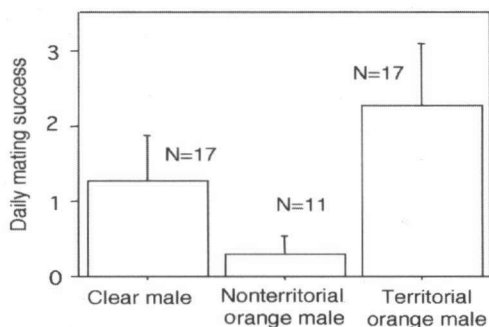


Fig. 1. Average daily mating success of territorial, non-territorial orange-winged males, and clear-winged male.

Figure 1 shows the average daily mating success of territorial orange-winged males, non-territorial orange-winged males and clear-winged males. The average mating success of territorial males (2.27/day/male) was higher than non-territorial orange-winged males (0.28/day/male) or clear-winged males (1.25/day/male) (Tab. I). Non-territorial clear-winged males showed higher mating success than non-territorial orange-winged males.

MODEL EXPERIMENTS

We performed an ANOVA with model type (control clear, painted clear and real orange) and morph type (orange- or clear-winged male) as independent variables. This enabled us to determine the basis for the interaction. There was a highly significant difference in fight time with respect to model type (Fig. 2): fight times

Table I

Result of ANOVA and Fisher's PLSD test for daily mating success of three types of males (orange-winged territorial, orange-winged nonterritorial and clear-winged nonterritorial)

ANOVA

	DF	SS	MS	F	P
Type	2	27.009	13.505	8.716	0.0007
Residual	42	65.073	1.549		

Fisher's PLSD

	Mean diff.	Critical diff.	P
Clear, Nonterritorial orange	0.974	0.972	0.0496
Clear, Territorial orange	-1.016	0.862	0.0220
Territorial, Nonterritorial orange	-1.989	0.972	0.0002

were longer when the control clear-winged models were used ($F_{2,55} = 12.65, p < 0.0001$). There was also a significant difference in fight time with respect to morph type: orange-winged males fought more than clear-winged males ($F_{1,55} = 8.11, p = 0.006$). And there was a significant interaction between model type and morph type, indicating differences in fight time of each morph according to model type: clear-winged males mainly fought the clear-winged model, whereas orange-winged males fought all model types ($F_{2,55} = 5.75, p = 0.005$).

There was no significant difference in fight time according to whether the model was a real orange-winged male or a clear-winged male painted orange (Fig. 2, Orange male, paired t test, $t = 0.124$, $df = 9$, n.s.; Clear male, $t = 0.533$, $df = 18$, n.s.), suggesting that there was no perceived difference between the natural orange colour and that of the pen, and that the orange wing colour was the signal which elicited the fight response.

RESPONSE OF MALES TO FEMALE MODELS

In the experiment we conducted using a dead female model, orange-winged males always ignored the model (20/20 times).

DISCUSSION

There was a behavioural difference between males of each morph in their response to the models: orange-winged males fought any model but clear-winged males only fought models of clear-winged males. Clear-winged males invariably fled when presented with an orange model. When presented to an orange-winged

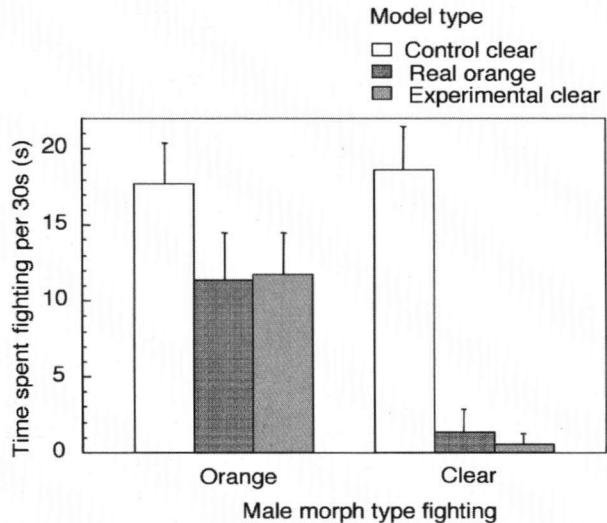


Fig. 2. Time that males of each morph spent fighting each of three models that were artificially flown at them. There was no significant difference in fight time by orange-winged males with respect to model type, but there was for clear-winged males: clear-winged males mainly fought against clear-winged models.

male, a clear-winged male model painted orange elicited the same fight response as did a real orange-winged male. Display of an orange wing probably signifies 'I am a fighter' to conspecifics, whilst the general appearance of a clear-winged male (the red pterostigma and the blue body colour) is alone enough to elicit the fight response from both morphs. The orange wing modifies the response (fight or flight) of the clear-winged morph.

Since orange-winged males they have resources to defend it is not surprising that they should fight more than clear-winged males (NOMAKUCHI et al., 1984). However, this does not explain why clear-winged males were prepared to fight providing there was no orange signal. PLAISTOW & TSUBAKI (2000) showed that selection for traits influencing flight ability differs between the two morphs: compared to clear-winged males, orange-winged males have higher flight muscle ratios and invest more in the build up of muscle and stored fat. The current study supports such a differentiation by experimentally confirming that they show behavioural differences to the same stimulus. A likely explanation, therefore, for the divergent response of the morphs to a model with orange wings is that the orange wing signals the male's status as a territorial morph, and that orange wings function as an honest indicator of male fighting ability (HOOPER et al., 1999). The two morphs have evolved divergent adaptive behavioural responses to this signal.

The adaptive significance of the non-territorial morphs' 'flee' response to an orange wing signal of a fighter male is obvious, but it does not explain why the non-territorial morph has lost, or never evolved, orange wings. Since developing and maintaining an orange wing is energetically costly (HOOPER et al., 1999), it would accrue no benefits to a male that does not defend a territory and has evolved a 'non-fighting' strategy. Territorial orange-winged males attacked clear-winged males, which suggests that the benefit clear-winged males get from not signalling is not one of avoiding aggression from orange-winged males.

Orange-winged males never attempted to engage clear-winged models in the 'tandem' pre-copulatory grasp and did not show any interest in female models, suggesting that they do not confuse them for females. However orange-winged males are occasionally seen in tandem with clear-winged males in the field. In natural conditions orange-winged males do sometimes apparently mistake clear-winged males for females (WATANABE & TAGUCHI, 1990). However such homosexual tandem pairs originated usually when males grasped a clear-winged male, which had already in tandem with a female on a perch. There seems little confusion over sex when they are in air, suggesting that the hypothesis of "mimicking females" is unlikely in this species.

No direct test was made of the female mimicry and crypsis hypotheses. However, we observed non-territorial orange-winged males occasionally trying to intercept females arriving at territorial areas, but in most cases they were immediately chased away by resident males. In contrast, clear-winged males often succeeded

in grasping females before territorial males found them. The lower average mating success of non-territorial orange-winged males compared to that of clear-winged males (Fig. 1) is probably due to their conspicuous wing colour, which may preclude successful sneaky behaviour around territories.

The benefit that non-territorial males get from not developing an orange wing – aside from a reduced energy expenditure (HOOPER et al., 1999) – is probably derived from increased crypsis around orange-winged males' territories, or in approaching females. This idea is consistent with observations of the behaviour of clear-winged males on and around orange-winged males' territories (NOMAKUCHI et al., 1984; TSUBAKI et al., 1997).

HOOPER et al. (1999) showed that aspects of the pigmentation of orange wings declined with male age and were influenced by levels of nutrition, suggesting that orange wing pigmentation may honestly signal current condition. Costly traits that honestly signal an aspect of quality, such as condition, may evolve and be maintained by inter-sexual selection, intra-sexual selection, or a mixture of the two (ANDERSSON, 1994; BERGLUND et al., 1996). In territorial odonates intrasexual selection is particularly important since male reproductive success is often dependent on obtaining a territory (e.g., PLAISTOW & SIVA-JOTHY, 1996). In calopterygids contests are best described as energetic wars of attrition in which the fatter male normally wins (MARDEN & WAAGE, 1990; PLAISTOW & SIVA-JOTHY, 1996). In this study we found that territorial males always engaged in aggressive interactions if they were challenged. In *M. costalis* also, contests between orange-winged males seems to be resolved by energetic wars of attrition, however, contests between orange-winged and clear-winged males are resolved by colour signalling: this signals works in an all-or-nothing manner in this system. Further work will be required to determine whether any components of a male's wing pigmentation are used in contests between fighter males.

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REFERENCES

- ANDERSSON, M., 1994. *Sexual selection*. Monographs in Behavior and Ecology. Princeton, Princeton Press.
- BERGLUND, A., A. BISAZZA & A. PILASTRO, 1996. Armaments and ornaments: an evolutionary explanation of traits of dual utility. *Biol. J. Linn. Soc.* 58: 385-399.
- CADE, W.H., 1979. Alternative male reproductive strategies in field crickets. In: M.S. Blum & N.A. Blum, [Eds], *Sexual selection and reproductive competition in insects*, pp. 343-379, Academic Press, New York.

- HAYASHI, F., S. DOBATA & R. FUTAHASHI, 2004. Macro- and microscale distribution patterns of two closely related Japanese *Mnais* species inferred from nuclear ribosomal DNA, its sequences and morphology (Zygoptera: Calopterygidae). *Odonatologica* 33: 399-412.
- HOOPER, R.E., Y. TSUBAKI & M. SIVA-JOTHY, 1999. Expression of a costly, plastic secondary sexual trait is correlated with age and condition in a damselfly with two male morphs. *Physiol. Ent.* 24: 364-369.
- LANK, D.B., C.M. SMITH, O. HANOTTE, T. BURKE & F. COOKE, 1995. Genetic polymorphism for alternative mating behaviour in lekking male ruff *Philomachus pugnax*. *Nature, Lond.* 378: 59-62.
- MARDEN, J.H. & J.K. WAAGE, 1990. Escalated damselfly territorial contests and energetic wars of attrition. *Anim. Behav.* 39: 954-959.
- NOMAKUCHI, S., 1992. Male reproductive polymorphism and form-specific habitat utilization of the damselfly *Mnais pruinosa* (Zygoptera: Calopterygidae). *Ecol. Res.* 7: 87-96.
- NOMAKUCHI, S., K. HIGASHI, M. HARADA & M. MADEDA, 1984. An experimental study of the territoriality in *Mnais p. pruinosa* Selys (Zygoptera: Calopterygidae). *Odonatologica* 13: 259-267.
- PLAISTOW, S.J. & M.T. SIVA-JOTHY, 1996. Energy constraints and male mate-securing tactics in the damselfly *Calopteryx splendens xanthostoma* (Charpentier). *Proc. R. Soc. Lond. (B)* 263: 1233-1238.
- PLAISTOW, S.J. & Y. TSUBAKI, 2000. A selective trade-off for territoriality and non-territoriality in the polymorphic damselfly *Mnais costalis*. *Proc. R. Soc. Lond. (B)* 267: 969-975.
- QVARNSTROM, A., T. PART & B.C. SHELDON, 2000. Adaptive plasticity in mate preference linked to differences in reproductive effort. *Nature, Lond.* 405: 344-347.
- ROHWER, S., 1975. The social significance of avian winter plumage variability. *Evolution* 29: 593-610.
- SIVA-JOTHY, M.T., 2000. Male wing pigmentation may affect reproductive success via female choice in a calopterygid damselfly (Zygoptera). *Behaviour* 136: 1365-1377.
- TSUBAKI, Y., 2003. The genetic polymorphism linked to mate-securing strategies in the male damselfly *Mnais costalis* Selys (Odonata: Calopterygidae). *Popul. Ecol.* 45:263-266.
- TSUBAKI, Y., R.E. HOOPER & M.T. SIVA-JOTHY, 1997. Differences in adult and reproductive lifespan in the two male forms of *Mnais pruinosa costalis* (Odonata: Calopterygidae). *Res. Popul. Ecol.* 39: 149-155.
- WATANABE, M. & TAGUCHI, M. 1990. Mating tactics and male wing dimorphism in the damselfly, *Mnais pruinosa costalis* Selys (Odonata: Calopterygidae). *J. Ethol.* 8: 129-137.
- WIDEMO, F. 1998. Alternative reproductive strategies in the ruff, *Philomachus pugnax*: a mixed ESS? *Anim. Behav.* 56: 329-336.