

# The evolution of colour polymorphism in British winter-active Lepidoptera in response to search image use by avian predators

JAMIE C. WEIR *Institute of Evolutionary Biology, University of Edinburgh, Edinburgh, UK*

## Keywords:

abundance;  
apostatic selection;  
birds;  
colouration;  
crypsis;  
Lepidoptera;  
polymorphism;  
predation;  
search image.

## Abstract

Phenotypic polymorphism in cryptic species is widespread. This may evolve in response to search image use by predators exerting negative frequency-dependent selection on intraspecific colour morphs, ‘apostatic selection’. Evidence exists to indicate search image formation by predators and apostatic selection operating on wild prey populations, though not to demonstrate search image use directly resulting in apostatic selection. The present study attempted to address this deficiency, using British Lepidoptera active in winter as a model system. It has been proposed that the typically polymorphic wing colouration of these species represents an anti-search image adaptation against birds. To test (a) for search image-driven apostatic selection, dimorphic populations of artificial moth-like models were established in woodland at varying relative morph frequencies and exposed to predation by natural populations of birds. In addition, to test (b) whether abundance and degree of polymorphism are correlated across British winter-active moths, as predicted where search image use drives apostatic selection, a series of phylogenetic comparative analyses were conducted. There was a positive relationship between artificial morph frequency and probability of predation, consistent with birds utilizing search images and exerting apostatic selection. Abundance and degree of polymorphism were found to be positively correlated across British Lepidoptera active in winter, though not across all taxonomic groups analysed. This evidence is consistent with polymorphism in this group having evolved in response to search image-driven apostatic selection and supports the viability of this mechanism as a means by which phenotypic and genetic variation may be maintained in natural populations.

## Introduction

Among the diverse functions of animal colouration (e.g. Wallace, 1878, 1889; Poulton, 1890; Beddard, 1892; Thayer, 1909; Cott, 1940; Fogden & Fogden, 1974; Roulin, 2004; Caro, 2005; Trullas *et al.*, 2007; Stevens & Ruxton, 2012; Jamie, 2017), crypsis represents a widespread antipredator adaptation (Ruxton *et al.*, 2004). Paradoxically however, although natural selection should eliminate variation, many apparently cryptic species also display considerable levels of stable

colour polymorphism (e.g. Nabours *et al.*, 1933; Common, 1954; Kettlewell, 1973; Whiteley *et al.*, 1997; Johannesson & Butlin, 2017).

Cryptic yet polymorphic species possess either distinct morphs matching different patches within coarse-grained, discontinuous environments (Levene, 1953; Merilaita *et al.*, 1999; Bond, 2007; for specific examples, see, e.g. Norris & Lowe, 1964; Kettlewell, 1973; Edmunds, 1976; Sandoval, 1994; Hoekstra *et al.*, 2005); or, many morphs all bearing an approximate resemblance to the same background (Bond, 2007). Common in invertebrates (e.g. Nabours *et al.*, 1933; Whiteley *et al.*, 1997; Franks & Oxford, 2009; Oxford, 2009), many species among the latter have evolved dozens of co-occurring colour morphs in so-called exuberant polymorphisms, although the extent to which these

*Correspondence:* Jamie Conor Weir, 32 Paul Drive, Airth, Falkirk, Stirlingshire FK2 8LA, UK.  
Tel.: +44 1324 832 701;  
e-mail: Jamie.Weir@ed-alumni.net

polymorphic forms are truly discrete is unclear in many instances (Oxford, 2009).

Colour polymorphism in a coarse-grained environment can therefore be understood as a cryptic resemblance to a polymorphic background. In contrast, polymorphism in species presented to predators against relatively fine-grained, homogeneous backgrounds may evolve due to predators utilizing search images while foraging (Clarke, 1962). This may exert negative frequency-dependent selection on intraspecific colour morphs, 'apostatic selection', thus encouraging the evolution of phenotypic polymorphisms (Poulton, 1890; Tinbergen, 1960; Clarke, 1962). This mechanism consists of three distinct hypotheses: (i) predators employ search images while foraging; (ii) this results in apostatic selection on morphs in prey populations; and that (iii) apostatic selection actively maintains polymorphisms in wild populations.

Theoretically, search images are a means of optimizing foraging (Karpestam *et al.*, 2014) given the finite limit of the brain to neurologically process visual information (Desimone & Duncan, 1995; Dukas, 2002). This would otherwise result in reduced prey detection due to predators dividing their limited attentional capacity (Tinbergen, 1960; Clarke, 1962; Dukas & Ellner, 1993; Dukas & Kamil, 2000, 2001). Indeed, prey presented to predators simultaneously in polymorphic populations suffer lower mortality than monomorphic populations (Knill & Allen, 1995; Glanville & Allen, 1997; Wennersten & Forsman, 2009; Karpestam *et al.*, 2014). Although predatory attentional limits may alone be sufficient to maintain prey polymorphisms (Karpestam *et al.*, 2014), this is unlikely as prey are typically encountered sequentially during natural foraging (Bond, 2007).

Work attempting to demonstrate search image use has shown that many predators may be trained by repeated exposure to natural or artificial prey, rendering them subsequently reluctant to take novel prey (Beukema, 1968; Coppinger, 1969, 1970; Murdoch, 1969; Morrell & Turner, 1970; Ware, 1971; Shettleworth, 1972; Allen, 1973). Furthermore, predators often exhibit an improved ability to discern and distinguish particular prey after sequential encounters (de Ruiter, 1952; Kettlewell, 1955, 1973; Dawkins, 1971; Den Boer, 1970; Blough, 1989, 1991, 1992; Bond & Riley, 1991; Langley, 1996). Though not strictly evidence of search image formation (Krebs, 1973), this suggests an ability to learn to recognize prey based on a suite of specific characters. Attempts at directly demonstrating search image use have typically involved captive predators (Den Boer, 1970; Bond, 1983), artificial prey (Allen & Clarke, 1968; Murton, 1971; Allen, 1973; Cooper, 1984), or combinations of both (Croze, 1970; Dawkins, 1971; Pietrewicz & Kamil, 1979; reviewed in Punzalan *et al.*, 2005; reviewed in Bond, 2007).

From the perspective of prey populations, considerable evidence exists to demonstrate the occurrence of

negative frequency-dependent selection exerted by captive predators (Murdoch *et al.*, 1975; Tucker, 1991), wild predators in semi-natural settings (Majerus, 1978, 1980, 1998) or in wild populations of cryptic polymorphic species (Clarke, 1962; Moment, 1962; Owen & Weigert, 1962; Owen, 1965; reviewed in Clarke, 1969; Allen, 1973; Harvey *et al.*, 1975; Sargent, 1978; Atkinson & Warwick, 1983; Allen *et al.*, 1988; reviewed in Allen, 1988; Reid, 1987; Oxford & Gillespie, 1998; Oxford, 2009; Johannesson & Butlin, 2017). Therefore, the hypotheses of (i) search image formation and (iii) the occurrence of apostatic selection in natural populations of cryptic, polymorphic species are relatively well supported, although evidence is lacking to show (ii) search image use driving apostatic selection.

More recent experiments, attempting to correct this deficiency, used captive birds preying artificial computer-generated moth-like targets. It was found that the abundance of several distinct morphs would oscillate over the generations in a negative frequency-dependent manner, suggestive of apostatic selection resulting from search image use (Bond & Kamil, 1998, 2002). Furthermore, in line with theoretical expectations based on search image use (Bond, 2007), moth populations placed against complex backgrounds evolved many cryptic morphs over simulated generations, whereas those on a homogeneous background did not (Bond & Kamil, 2006). However, while compelling, this work was carried out under highly artificial conditions making it difficult to generalize the results with regard to natural populations.

Nonetheless, certain characteristics of a prey morph or species can affect whether it is adaptive for a predator to utilize search images and, consequently, whether apostatic selection occurs (Tinbergen, 1960; Clarke, 1962; reviewed in Bond, 2007). Therefore, the condition of such characteristics may be indicative of the action of apostatic selection. Modelling has suggested that the crypticity of prey; the existence or degree of disparity in crypsis between the constituent morphs in a prey polymorphism; and elements of predator psychology, such as the reinforcement effect of success with the current 'image', or the range of possible morph specialization, all affect the persistence of prey polymorphisms maintained by frequency-dependent selection (Merilaita, 2006). Furthermore, experimental studies using captive predators have shown that levels of prey crypticity (Bond & Kamil, 1998, 2002) and the complexity and structure of the environmental background (Bond & Kamil, 2006) influence both the formation of search images and the strength of the resultant apostatic effect.

The frequency with which predators encounter a prey species is also predicted to influence the occurrence and strength of apostatic selection (Clarke, 1962). It would be unprofitable to form a search image of a rare species, based on a chance encounter, given the

low probability of a subsequent encounter. Hence, it is expected that, among cryptic species, those which are most abundant ought also to be the most polymorphic for they are most exposed to the effects of search image use by predators, and are therefore most likely to evolve polymorphism in response. Limited anecdotal evidence suggests that the most common species in some taxa are particularly polymorphic (e.g. Lepidoptera, see Kettlewell, 1973; Majerus, 1998). Although it has been argued that species of intermediate abundance, or those utilized as prey at an intermediate level, ought to be subject to the greatest effect of search image use (Clarke, 1962), computer simulations have demonstrated that polymorphism evolves far more readily under apostatic selection in larger populations (Merilaita, 2006). Interestingly, this result has been supported by comparative analyses examining predator polymorphism in birds of prey (Fowlie & Kruger, 2003). However, the relationship between degree of polymorphism and abundance in those species assumed to be regularly exposed to apostatic selection by predators, such as small, cryptically coloured invertebrates, is unknown.

The Lepidoptera of the Palaearctic are a well-studied, diverse and experimentally tractable taxonomic group in which colour polymorphism occurs commonly (Ford, 1964; Robinson, 1971; Howarth, 1973; Russwurm, 1978; Harmer, 2000), including among those nocturnal species active during the winter months (Kettlewell, 1973). In Britain, of the 70 species predominantly active between October and April, over 15 display half a dozen or more distinct, common, co-occurring colour morphs (Tutt, 1891–92; Turner, 1925–26; Skinner, 2009). The adult stage of these species rely principally upon the cryptic resemblance of their wing colouration to their environment to avoid detection by predators (Ford, 1955). Kettlewell (1973) was first to suggest, with some subsequent support (Majerus, 1998), that these cryptic polymorphisms evolved due to apostatic selection arising through the use of search images by birds.

Using the British winter-active Lepidoptera as a model system, this study aimed to test the theory that search image use can result in apostatic selection, leading to the evolution of colour polymorphisms in natural populations. To test (a) whether birds exert apostatic selection on populations of winter-active Lepidoptera, artificial moth-like models mimicking these species were placed in dimorphic populations under natural conditions and exposed to predation. The frequency at which various colour morphs occurred was varied between populations and the rate of predation of either form by wild birds was recorded, to detect the presence of apostatic selection resulting from search image use. Under search image use, predators bias their search based upon a range of morph-specific characters (Tinbergen, 1960; Clarke, 1962), this 'image' being strengthened or degraded based on the nature of

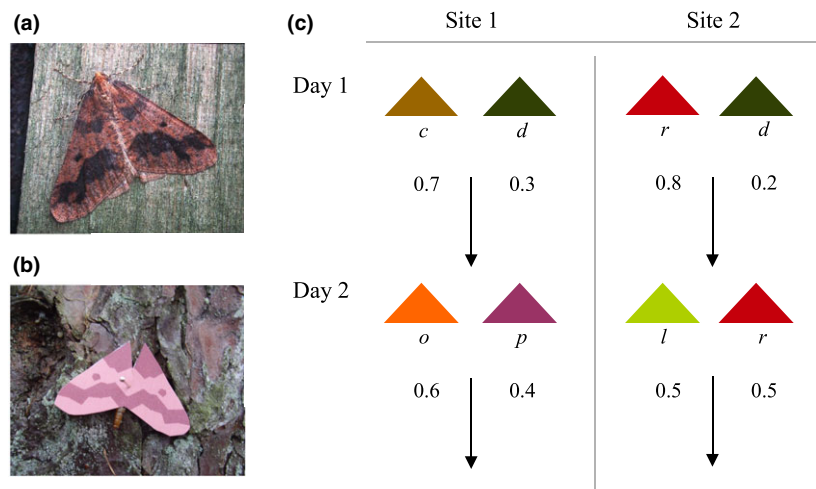
subsequent prey encounters (reviewed in Bond, 2007). As the disparity in morph frequency increases, the profitability of hunting to the image of the common morph is correspondingly greater. A sigmoid relationship is therefore expected between morph abundance and the proportion of those predated belonging to that morph, in contrast with approximately linear direct proportionality where predation is random (Mook *et al.*, 1960; Tinbergen, 1960). That is to say, a positive relationship is predicted when we consider the probability than an individual of a given morph will be predated against morph abundance. In addition, to test (b) for a positive correlation between the abundance of a species and degree of polymorphism, as predicted where phenotypic polymorphism evolves as a result of search image-driven apostatic selection (Clarke, 1962), a series of phylogenetic comparative analyses were conducted across a range of British winter-active and non-winter-active Lepidopteran taxa.

## Materials and methods

### (a) Do birds exert apostatic selection on populations of artificial winter-active moths?

Artificial moth-like prey formed targets for bird predation, based upon those used by Cuthill *et al.* (2005). To recreate a naturalistic predator–prey system, the external design of the models was based upon the wing pattern and colouration of *Erannis defoliaria* Clerck (mottled umber moth), a species active from October to December (Fig. 1a; Skinner, 2009). To create an initial moth model which was cryptically coloured, a photograph of the bark of a Scots pine *Pinus sylvestris* L., a common tree species in the study area, was taken. From this, two major blocks of colour (visible to the human eye) were identified, the pale brown bark and dark brown shade of the fissures in the bark. Using Microsoft Paint, 10 readings of the RGB values in each of these colour blocks were taken. The mean ( $\pm$ standard deviation) value of each colour component obtained from the bark ( $R = 172 \pm 27.3$ ,  $G = 145 \pm 31.2$ ,  $B = 117 \pm 40.2$ ) were used as the values for the ground colouration of the model, and those from the fissures ( $R = 121 \pm 40.1$ ,  $G = 99 \pm 37.6$ ,  $B = 74 \pm 37.5$ ) were used for the transverse lines and discal spot (Morph *c*).

To simulate a natural polymorphism in a cryptic species, six additional morphs were generated by individually modifying the R, G or B values of both the ground colouration and other wing markings by  $\pm 25$  units (Morph *o* [orange]  $R + 25$ ; Morph *d* [dark green]  $R - 25$ ; Morph *l* [light green]  $G + 25$ ; Morph *r* [red]  $G - 25$ ; Morph *p* [purple]  $B + 25$ ; Morph *t* [tan]  $B - 25$ ). The morphology of the wing pattern was kept constant, only the colour being modified (Fig. 2). However, because birds have a different visual system, they may have perceived the degree of difference between



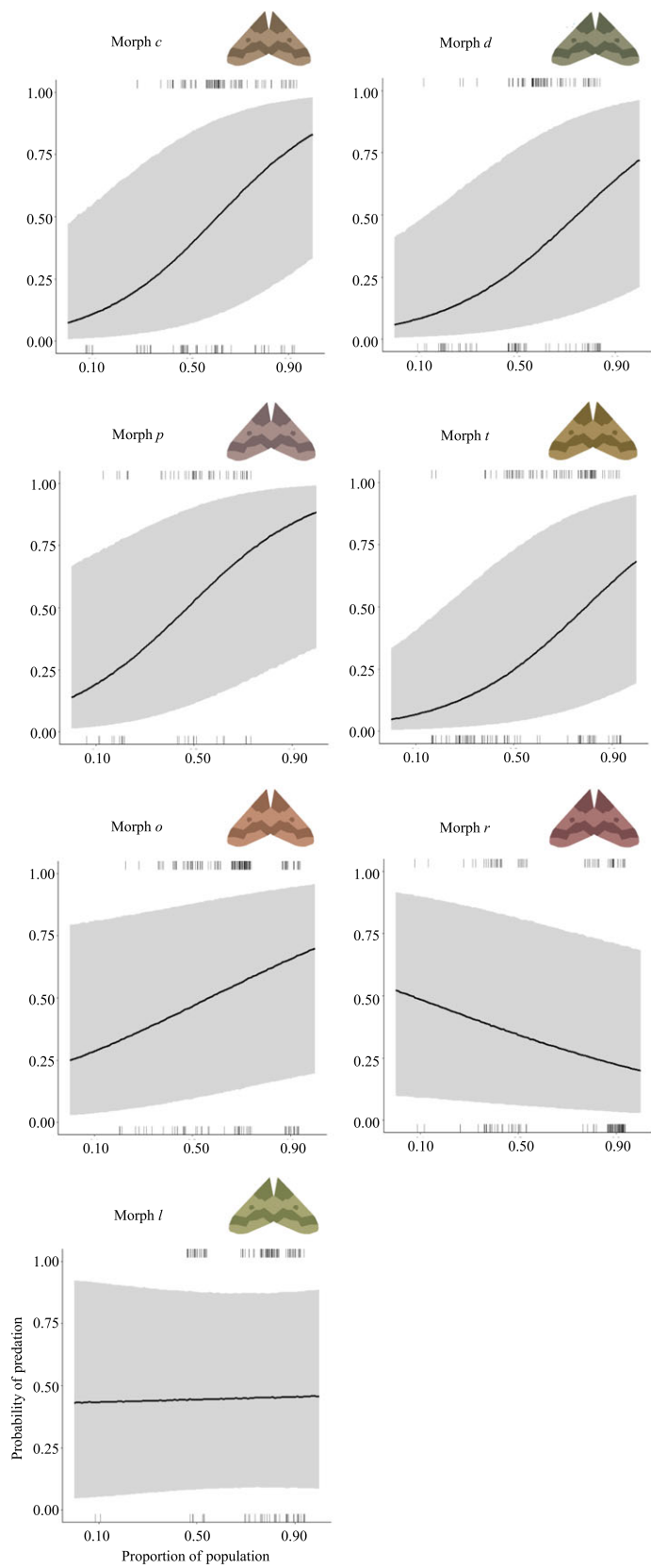
**Fig. 1** Design of field experiments. (a) Models were based upon the wing pattern of *Erannis defoliaria* (mottled umber moth). Image © Mr Nick Greatorex-Davies. (b) Models were pinned to the bark of trees to imitate natural nocturnal Lepidoptera, with a dead mealworm attached to the pin beneath the artificial wings to provide an edible body component for predators. Photograph shows morph *r*. (c) At each of the 10 woodland sites, models were pinned to trees in dimorphic populations of 20 individuals at varying relative frequencies. Models were left in place for 24 h, and the numbers of either morph predated were recorded. The old population was then replaced and the procedure repeated. An example set-up at two sites over 2 days is shown, with morph colour code and relative proportion in the population given.

morphs as being variable, despite this quantitatively fixed modification in colour from a human perspective. In reality, this is not a problem – testing the existence of search images only requires that the morphs be different, such that hunting to the image of one morph interferes with the ability to detect another visually distinct morph, and there is no evidence that any two here were regarded as visually identical by birds. It should be stressed that these models were not intended as accurate reproductions of a particular species of Lepidoptera – in which case ultraviolet components of wing colour patterns (e.g. Meyer-Rochow, 1991; Brunton, 1998; Lyytinen *et al.*, 2004), visible to birds (Bennett & Cuthill, 1994), would have to be reproduced – but rather to be sufficiently realistic that birds would treat them as a novel species of prey, and consequently display natural predatory behaviour.

Devilla Forest (NS9587 and NS9687), a Forestry Commission property in Fife, United Kingdom, was selected as the study area. This mixed woodland contains stands of dense, commercial coniferous planting as well as large areas of open *P. sylvestris* woodland with a deciduous understorey, composed primarily of *Betula* (birch). Other parts of the woodland consist of stands of deciduous trees such as *Fagus* (beech) and *Quercus* (oak). Most winter-active Lepidoptera utilize deciduous trees as larval food-plants (Skinner, 2009), and hence this area represented a potential natural habitat of the species mimicked by the models. Resident bird populations may therefore be used to predate similar prey items during winter.

Field work was carried out in the week beginning 26 December 2016. Within the study area, 10 small experimental sites were selected. At each site an artificial dimorphic population of 20 individual moth models was set-up. The models were printed onto Toughprint waterproof paper using an HP Envy 4500 inkjet printer and pinned to trees at an approximate height of 2 m from the ground. Beneath the ‘wings’ of each model a dead mealworm (frozen and thawed) was attached to the pin to act as an edible body component (Fig. 1b). Models were pinned individually to trees at approximately 2 m intervals in a clump.

The frequency of either colour morph in each dimorphic population was varied. Ten populations were set up over the experimental period with two morphs occurring at one of five levels of relative frequency, defined as a proportion of the population (0.1/0.9, 0.2/0.8, 0.3/0.7, 0.4/0.6, and 0.5/0.5). Each population was randomly assigned two different colour morphs (one to occur at each proportion), an experimental site in the woodland, and a date on which to be set-up (Table 1). Populations were set-up at approximately 11 am each day and left in place for 24 h, after which the sites were revisited and the number of models predated was recorded (Fig. 1c). Where the mealworm was entirely removed it was assumed to have been predated by a bird. This could be distinguished from predation by a slug or snail, which left a slime trail, and spiders which sucked the bodily fluids from the mealworm. The experiment was conducted in winter partly to avoid the problem of ants which may also remove prey entirely



**Fig. 2** Variation in the probability of an individual of each morph being predated with changing proportion in the population. Plots show predicted probability of predation based on the fit of the minimal model, with 95% confidence intervals of the model prediction. Rug plots illustrate actual attack data on the models at various levels of frequency in the populations (0/survived; 1/predated). All colour morphs used in the experiment are shown. Most morphs display a positive relationship, as expected where search image use results in apostatic selection. However, the relationships displayed by morphs *l* and *r* deviate significantly from that of the reference level (morph *c*).

**Table 1** Attack data on dimorphic artificial moth populations.

Morph 1		Morph 2												
Proportion of population	Frequency in population	Colour morph	Frequency predated	Proportion of those predated	Proportion of population	Frequency in population	Colour morph	Frequency predated	Proportion of those predated	Date	Site	Temp. (°C)	Cloud Cover (est. in Oktas)	Proximity to path (est. nearest 10 m)
<b>0.1</b>	2	c	0	<b>0.00</b>	<b>0.9</b>	18	r	0	<b>0.00</b>	29/12/16	1	9	6	10
		p	0	<b>0.00</b>			t	8	<b>1.00</b>	27/12/16	2	7	8	30
		d	0	<b>0.00</b>			r	9	<b>1.00</b>	29/12/16	3	9	6	30
		p	1	<b>0.08</b>			o	11	<b>0.92</b>	30/12/16	4	12	7	20
		c	0	<b>0.00</b>			l	13	<b>1.00</b>	30/12/16	5	12	7	20
		l	0	<b>0.00</b>			o	10	<b>1.00</b>	29/12/16	6	9	6	20
		c	0	<b>0.00</b>			r	4	<b>1.00</b>	31/12/16	8	12	7	10
		d	1	<b>0.14</b>			r	6	<b>0.86</b>	30/12/16	9	12	7	20
		r	0	<b>0.00</b>			l	7	<b>1.00</b>	27/12/16	10	7	8	10
		r	2	<b>0.14</b>			c	12	<b>0.86</b>	31/12/16	10	12	7	20
<b>0.2</b>	4	t	0	<b>0.00</b>	<b>0.8</b>	16	l	13	<b>1.00</b>	30/12/16	1	12	7	20
		t	0	<b>0.00</b>			d	10	<b>1.00</b>	29/12/16	2	9	6	10
		t	2	<b>0.13</b>			l	14	<b>0.88</b>	30/12/16	2	12	7	30
		p	1	<b>0.17</b>			d	5	<b>0.83</b>	27/12/16	3	7	8	30
		o	1	<b>0.09</b>			l	10	<b>0.91</b>	31/12/16	5	12	7	20
		p	0	<b>0.00</b>			b	9	<b>1.00</b>	27/12/16	6	7	8	20
		p	4	<b>0.24</b>			t	13	<b>0.76</b>	30/12/16	7	12	7	10
		d	0	<b>0.00</b>			r	11	<b>1.00</b>	28/12/16	8	8	8	10
		d	0	<b>0.00</b>			t	10	<b>1.00</b>	28/12/16	9	8	8	20
		d	0	<b>0.00</b>			t	8	<b>1.00</b>	29/12/16	10	9	6	20
<b>0.3</b>	6	d	2	<b>0.13</b>	<b>0.7</b>	14	o	14	<b>0.88</b>	27/12/16	1	7	8	30
		o	1	<b>0.09</b>			d	10	<b>0.91</b>	28/12/16	1	8	8	20
		r	3	<b>0.19</b>			t	13	<b>0.81</b>	31/12/16	1	12	7	20
		t	0	<b>0.00</b>			o	10	<b>1.00</b>	27/12/16	4	7	8	30
		t	0	<b>0.00</b>			o	12	<b>1.00</b>	29/12/16	4	9	6	10
		d	1	<b>0.11</b>			o	8	<b>0.89</b>	31/12/16	4	12	7	20
		t	0	<b>0.00</b>			c	13	<b>1.00</b>	28/12/16	6	8	8	20
		t	0	<b>0.00</b>			p	11	<b>1.00</b>	30/12/16	6	12	7	20
		c	0	<b>0.00</b>			l	9	<b>1.00</b>	31/12/16	7	12	7	10
		c	2	<b>0.15</b>			o	11	<b>0.85</b>	30/12/16	8	12	7	10

**Table 1** (Continued)

Morph 1		Morph 2												
Proportion of population	Frequency in population	Colour morph	Frequency predated	Proportion of those predated	Proportion of population	Frequency in population	Colour morph	Frequency predated	Proportion of those predated	Date	Site	Temp. (°C)	Cloud Cover (est. in Oktas)	Proximity to path (est. nearest 10 m)
<b>0.4</b>	8	<i>t</i>	5	<b>0.36</b>	<b>0.6</b>	12	<i>o</i>	9	<b>0.64</b>	28/12/16	2	8	8	20
		<i>r</i>	3	<b>0.25</b>			<i>p</i>	9	<b>0.75</b>	31/12/16	2	12	7	20
		<i>r</i>	2	<b>0.17</b>			<i>c</i>	10	<b>0.83</b>	31/12/16	3	12	7	30
		<i>r</i>	4	<b>0.25</b>			<i>c</i>	12	<b>0.75</b>	28/12/16	4	8	8	20
		<i>o</i>	5	<b>0.33</b>			<i>t</i>	10	<b>0.67</b>	27/12/16	5	7	8	30
		<i>p</i>	6	<b>0.33</b>			<i>d</i>	12	<b>0.67</b>	29/12/16	5	9	6	10
		<i>c</i>	7	<b>0.39</b>			<i>d</i>	11	<b>0.61</b>	31/12/16	6	12	7	20
		<i>r</i>	8	<b>0.42</b>			<i>d</i>	11	<b>0.58</b>	27/12/16	7	7	8	10
		<i>t</i>	0	<b>0.00</b>			<i>c</i>	4	<b>1.00</b>	29/12/16	8	9	6	10
		<i>o</i>	6	<b>0.38</b>			<i>c</i>	10	<b>0.63</b>	27/12/16	9	7	8	20
<b>0.5</b>	10	<i>d</i>	5	<b>0.42</b>	<b>0.5</b>	10	<i>l</i>	7	<b>0.58</b>	28/12/16	3	8	8	10
		<i>t</i>	7	<b>0.50</b>			<i>l</i>	7	<b>0.50</b>	30/12/16	3	12	7	10
		<i>l</i>	10	<b>0.56</b>			<i>d</i>	8	<b>0.44</b>	28/12/16	5	8	8	20
		<i>t</i>	7	<b>0.58</b>			<i>c</i>	5	<b>0.42</b>	28/12/16	7	8	8	20
		<i>p</i>	8	<b>0.47</b>			<i>o</i>	9	<b>0.53</b>	29/12/16	7	9	6	30
		<i>d</i>	1	<b>1.00</b>			<i>r</i>	0	<b>0.00</b>	27/12/16	8	7	8	30
		<i>r</i>	8	<b>0.57</b>			<i>p</i>	6	<b>0.43</b>	29/12/16	9	9	6	20
		<i>c</i>	0	<b>0.00</b>			<i>d</i>	0	<b>0.00</b>	31/12/16	9	12	7	20
		<i>c</i>	9	<b>0.50</b>			<i>o</i>	9	<b>0.50</b>	28/12/16	10	8	8	10
		<i>o</i>	3	<b>0.33</b>			<i>t</i>	6	<b>0.67</b>	30/12/16	10	12	7	20

Populations consisted of 20 individual moth models of two different colour morphs. Each morph was presented at a given proportion of the population. The table shows proportion and frequency of occurrence, as well as the frequency of each morph predated and the proportion of those predated which were of that morph. Where predation is random, the proportion at which a morph occurs ought to equal the proportion of those predated which are of that morph. Where predators utilize search images, it is expected that morphs occurring commonly ought to be overpredated and rare morphs underpredated – that is, as a proportion of those predated, in excess and below their respective proportion occurrence in the population. The proportion of a morph in the population and the proportion of those predated which were of that morph are emboldened to facilitate comparison. Field measurements of cloud cover, temperature and estimated proximity to a hiking path are all given.

but are not active at this time of year. In each of these cases, the individual was recorded as having survived. After the 24 h period, all models at the site were then removed and replaced with the next scheduled population, typically using the same trees. Each day at approximately 10 am temperature was recorded to the nearest degree using an analogue alcohol thermometer, and cloud cover estimated in oktas. It was hypothesized that cloud cover would affect light levels and potentially the efficiency of hunting, or the ability of birds to notice particular colour morphs, hence influencing their survival. Equally, variation in temperature could be expected to influence predator activity and therefore morph survival. At each site, the proximity to a path was also estimated to account for potential disturbance by walkers.

Individual moths were scored as having either survived or been predated (0/1) after the 24 h period. The results were analysed using a binomial generalized linear mixed model in R (R Core Team, 2017) using the lme4 package (Bates *et al.*, 2015). Predation was modelled as the binary response variable. The starting model included colour morph of the individual, the frequency at which that morph occurred as a proportion of the initial population, temperature, cloud cover and proximity to a path as fixed effects, with date and site as random effects. An interaction between colour morph and the proportion at which that morph occurred was tested for, to determine whether the probability of predation with changing frequency differed significantly across morphs. Model simplification was conducted by successive removal of terms and interactions from the model followed by evaluation of the significance of the model fit using likelihood ratio tests (LRT). The merTools (Knowles & Frederick, 2016) package was used to plot the minimal model for each colour morph with confidence intervals. To do so, a new data frame was generated with all values of morph proportion in the population, at intervals of 0.01, holding date and site constant at arbitrarily selected values for each colour morph. Predicted values of probability of predation were generated for each, across the range of proportion levels, based on the modelled relationship. Confidence intervals (95%) were calculated by generating a simulated distribution of the random and fixed effects then estimating the values of the response variable across this range, therefore including variation across all model parameters. Graphs were plotted using ggplot2 (Wickham, 2009).

### **(b) Is abundance positively correlated with degree of polymorphism in winter-active moths?**

The number of named and described intraspecific taxa assigned to each species of British Lepidoptera was used as a measure of the degree of polymorphism of that species. Two monographic treatments of intraspecific

variation were referred to for this information (Tutt, 1891–92; Turner, 1925–26), each of which only consider a selection of the British fauna approximately equivalent to the modern Noctuidae and Geometridae, respectively (Kloet & Hincks, 1972). Taxonomy and nomenclature below species level has been an area of considerable debate (e.g. Edwards, 1954) and variants which occur as part of a balanced polymorphism, of the kind which might be maintained by apostatic selection, represent only one of a range of within-species groups to which names and descriptions are typically ascribed (reviewed in Weir, 2016). In some groups, names have been applied to morphs which represent merely arbitrary stages in a continuously varying trait. However, Lepidoptera as a group have been the subject of considerable research for at least two centuries, with particular emphasis on variation in their colouration. Thus our knowledge of this is fairly well advanced and we are sufficiently familiar with the range of variation displayed by each species to understand which are likely true, discrete polymorphisms. The intraspecific taxa described by Tutt (1891–92), which he referred to as ‘varieties’, do indeed primarily represent true polymorphic forms which co-occur throughout the distribution of a species. However, Turner (1925–26) lists only ‘races’ (geographical variants, see Weir, 2016) and ‘aberrations’, the latter encompassing both the participant forms in balanced polymorphisms and named but very rare developmental and genetic mutants. Both are therefore likely to give a reliable estimation of the degree of polymorphic variability of a given species, although data obtained from Turner (1925–26) may have over-estimated this and should be treated with caution. Winter-active species occur within a range of higher taxonomic groups and data for these species were obtained from whichever source was appropriate.

The absolute recorded abundances of all British species belonging to the families Geometridae and Noctuidae, and all winter-active species, were obtained from the Rothamsted Research National Light Trap Network. The Network uses standardized light traps consisting of a 200 W tungsten bulb operated nightly in long-running, fixed locations (Fry & Waring, 2001). Data were obtained for the 5 year period from 2008 to 2012, inclusive, from only those 63 traps operated continuously throughout this period and within the territory of the United Kingdom. It was assumed that the relative abundance of each species in the sample recorded by the Rothamsted traps was equal to that in the wild.

Cytochrome oxidase I (COX1) sequences for all species with known measures of both abundance and degree of polymorphism were obtained from BOLD (Ratnasingham & Hebert, 2007), a single sequence being retained as representative of each species. Only sequences for which identification had been provided by a named source were used. The occurrence of each species within the British Isles was recorded as resident,



immigrant, extinct, importation, introduction, colonist, unknown or a combination of these terms, as given by Skinner (2009). Only those species which were stated as resident, and hence which permanently inhabit and breed in Britain, were used in the comparative analyses. Annual immigrant species might be recorded in low numbers because few have migrated to the country and not because they are rare, potentially confounding any comparison of degree of polymorphism and abundance. Species with a recorded abundance of zero are problematic because this may have represented a lack of overlap between their geographical distribution and the distribution of those Rothamsted traps used in this study, rather than genuine rarity. Either it is assumed, therefore, that the light trap network is representative of the distribution of all British species, and hence an abundance of zero actually denotes rarity; or, it is assumed that if the distribution of a given species overlapped at all with the coverage of the network it would be recorded at least once in one trap across the country. Here, the second was taken to be the most conservative assumption, particularly given the often specific habitat requirements of many species (e.g. Skinner, 2009) and the sampling limitations of these light traps, in terms of distance (Fry & Waring, 2001). The numbers of species in each taxon excluded from the analyses are detailed in Tables 3 and 4; in most instances, the number of species excluded, as a proportion of those analysed, is small.

Trees were constructed of three groups of British Lepidoptera using those species for which sequences and data were available: (i) the Geometridae; (ii) the Noctuidae; and (iii) the winter-active fauna. The Geometridae and Noctuidae are the most species-rich families of larger British Lepidoptera and were analysed, along with their larger constituent subtaxa, as a form of control. These taxa contain species active across the entire year. This allowed the generality of any relationship between abundance and polymorphism to be determined, and whether this was restricted to certain taxonomic groups, with particular colouration characteristics, or a specific ecological context, in this case winter activity.

COX1 sequences were manually aligned in BioEdit (Hall, 1999). BEAST was used to infer a Bayesian time-scaled phylogenetic tree (Drummond *et al.*, 2012). A BEAST input file was generated using the alignments in BEAUti under a general time-reversible nucleotide substitution model (Tavaré, 1986) with empirical base frequencies, a gamma site heterogeneity model and a codon model partitioned for position 3 vs. 1 and 2. Trees were constrained based upon the monophyletic subfamilies and their respective dates of divergence defined in recent multi-locus higher level phylogenetic analyses (Yamamoto & Sota, 2007; Toussaint *et al.*, 2012; Reiger *et al.*, 2017). Node age priors were set as normally distributed and nontruncated. A lognormally distributed uncorrelated relaxed clock model (Drummond *et al.*,

2006) and a Yule process model of speciation (Gernhard, 2008) were used. A random starting tree was used in all analyses after several preliminary trials found that a UPGMA starting tree did not conform to the *a priori* constraints. Five runs were performed in BEAST for each group of sequences with an MCMC length of 10 million generations sampling from the posterior distribution every 1000 generations (Drummond *et al.*, 2002). LogCombiner (Drummond *et al.*, 2012) was used with a 5 million generation burn-in to combine tree and log files from all five runs in each group. Combined BEAST log files were inspected in TRACER (Rambaut *et al.*, 2014) to assess the quality of the posterior sample of trees and ensure all parameters had a satisfactory (> 200) effective sample size. For each group of taxa, the combined BEAST output tree file was used to generate a maximum clade credibility tree using TreeAnnotator (Drummond *et al.*, 2012) with a 5 million generation burn-in. The trees generated were compared to those published in larger, multi-gene studies (Yamamoto & Sota, 2007; Toussaint *et al.*, 2012; Reiger *et al.*, 2017) and conformed well to these, with no significant anomalous or unexpected results.

Phylogenetic comparative analyses were conducted in R (R Core Team, 2017) using the caper package (Orme *et al.*, 2013). To ensure normality of the residuals, abundance was  $\log_{10}$ -transformed. Phylogenetic linear regressions of the number of named intraspecific morphs against  $\log_{10}$  (Abundance) of a species were conducted, using the phylogenetic generalized least squares method (Grafen, 1989), with  $\lambda$  branch length transformations estimated by maximum likelihood. Using the maximum clade credibility tree of the winter-active species, regressions were carried out across the whole group, across those species which hibernate as adults over the coldest months of the year (e.g. *Conistra* spp., *Lithophane* spp.; Skinner, 2009), and the winter-active fauna excluding these species. This was to enable detection of any difference in the relationship between abundance and polymorphism among species adopting these differing ecological strategies. Similarly, using the trees generated for the families Noctuidae and Geometridae, respectively, regressions were carried out across these entire groups and across any of their constituent subfamilies or genera with data available for five or more species.

## Results

### (a) Do birds exert apostatic selection on populations of artificial winter-active moths?

Temperature ( $z = 0.53$ ,  $P = 0.597$ ), cloud cover ( $z = 1.11$ ,  $P = 0.268$ ), and proximity to a path ( $z = -0.90$ ,  $P = 0.369$ ) did not significantly affect the probability that an individual would be predated. A model where these terms were excluded was not significantly different

( $\chi^2_3 = 1.67$ ,  $P = 0.640$ , LRT), hence they were removed for reasons of parsimony. However, a model which included an interaction between morph and the proportion at which that morph occurred within the population was a significantly better fit to the data than one which did not ( $\chi^2_6 = 38.32$ ,  $P < 0.001$ , LRT). Removing date and site as random terms produced a model which was a significantly worse fit to the data ( $\chi^2_2 = 27.763$ ,  $P < 0.001$ , LRT). Therefore, the minimal model included the proportion at which a morph occurred in the population and morph type as fixed effects, with an interaction between the two, and date and site as random terms.

Under this model, the proportion at which a morph occurred within the population had a significant positive effect on the probability that an individual of that morph would be predated ( $z = 3.96$ ,  $P < 0.001$ ; Table 2; Fig. 2). Where predation is random, the probability of predation of any individual is expected to remain at a constant value, irrespective of the frequency of that morph in the population, because colour has no influence on the likelihood of mortality. A positive relationship, however, indicates that as a given morph became more common within a population, the probability that an individual of that morph would be predated increased. Thus, where a morph was common it was predated at a level above that expected based upon its abundance in the population and random predation (Table 1).

Probability of predation varied with colour morph (Table 2; Fig. 2), independent of the proportion at which that morph occurred. Where proportion in the population is held constant, there was no significant difference in the probability of predation between an

individual belonging to morph *c* (reference level) or morphs *d* ( $z = -0.29$ ,  $P = 0.767$ ), *o* ( $z = 1.54$ ,  $P = 0.122$ ), *p* ( $z = 0.73$ ,  $P = 0.468$ ) and *t* ( $z = -0.59$ ,  $P = 0.556$ ). However, an individual belonging to morphs *l* ( $z = 2.02$ ,  $P = 0.043$ ) and *r* ( $z = 3.19$ ,  $P = 0.001$ ) was significantly more likely to be predated. Furthermore, there was a significant negative interaction between probability of predation and the proportion at which morphs *l* ( $z = 2.49$ ,  $P = 0.013$ ) and *r* ( $z = -4.31$ ,  $P < 0.001$ ) occurred within the population, compared with the reference level interaction displayed by morph *c* (Table 2; Fig. 2). As the proportion at which morph *r* occurred increased, its probability of predation decreased approximately linearly, while that for morph *l* remained relatively constant. This suggests that particular morphs have intrinsically different likelihoods of predation due to their unique colouration, and that colouration interacts with the frequency at which a morph occurs to influence this likelihood.

In several instances where a site was revisited it was found that the artificial wings had been forcibly removed from some moth models, either by pulling them over the head of the pin or by ripping the paper. This is similar to the manner in which birds often carefully remove and disregard the wings of Lepidoptera before eating them in the wild (e.g. Collenette, 1935). Furthermore, it was evident that this damage had been inflicted by birds for clear beak impressions were visible on the wings where the model had been attacked. The beak impressions seem to indicate that the models were predated by a range of small passerines, one in particular suggesting a member of the genus *Turdus* (Collenette, 1935; Fig. S1).

**Table 2** Binomial GLMM analysis of the probability that an individual moth-like model would be predated with varying morph type and frequency in the population.

Variable	Coefficient (SE)	z-value
Prop. Pop.	4.20 (1.04)	3.96†††
Morph <i>d</i>	-0.26 (0.87)	-0.29
Morph <i>l</i>	2.25 (1.11)	2.02†
Morph <i>o</i>	1.41 (0.92)	1.54
Morph <i>p</i>	0.70 (0.97)	0.73
Morph <i>r</i>	2.61 (0.82)	3.19††
Morph <i>t</i>	-0.48 (0.81)	-0.59
Prop.Pop.*Morph <i>d</i>	-0.39 (1.47)	-0.26
Prop.Pop.*Morph <i>l</i>	-4.02 (1.61)	-2.49 †
Prop.Pop.*Morph <i>o</i>	-2.17 (1.47)	-1.48
Prop.Pop.*Morph <i>p</i>	-0.27 (1.84)	-0.15
Prop.Pop.*Morph <i>r</i>	-5.60 (1.30)	-4.31†††
Prop.Pop.*Morph <i>t</i>	-0.35 (1.31)	-0.26

Based upon minimal model with date and site as random effects. Morph frequency given as a proportion of each dimorphic population, designated 'Prop.Pop.'. Interaction effects indicated by '\*'. Morph *c* is taken as the reference level in comparisons of the effect of different morphs at varying frequencies.

† $P < 0.05$ , †† $P < 0.01$ , ††† $P < 0.001$ .

### (b) Is abundance positively correlated with degree of polymorphism in winter-active moths?

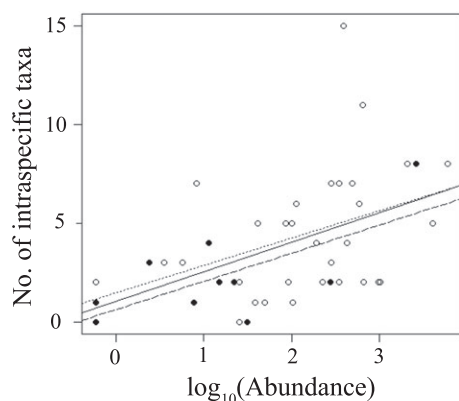
Across those species of British Lepidoptera which are winter-active (i.e. in the adult stage predominantly between October and April) there is a significant positive relationship between the abundance of a species and its degree of polymorphism ( $F_{1,39} = 11.90$ ,  $P = 0.001$ ; Table 3; Fig. 3). Similarly, among the subset of this group which are active as an adult in late autumn and early spring, but which hibernate during the coldest months, a significant positive relationship exists ( $F_{1,8} = 6.88$ ,  $P = 0.031$ ). When the remaining winter-active species are considered with this group removed the significant positive relationship holds ( $F_{1,29} = 4.97$ ,  $P = 0.031$ ). Although the coefficient of the relationship is not significantly different among these groups of taxa ( $F_{2,81} = 0.40$ ,  $P = 0.670$ , ANOVA), the model explains the highest proportion of variation in degree of polymorphism amongst those species which hibernate as adults, some 40%. The maximum likelihood estimation of phylogenetic signal (Pagel's  $\lambda$ ) suggests that covariance in these traits due to phylogeny is negligible across all groups of taxa.

**Table 3** Phylogenetically corrected regressions of abundance against degree of polymorphism in British winter-active Lepidoptera.

Taxa	Species analysed (Excluded, Total No.)	Coefficient (SE)	Intercept (SE)	Adjusted $r^2$	Pagel's $\lambda$ (95% CIs)	F-statistic	P-value
Winter-active (all)	41 (4, 70)	1.50 (0.44)	1.03 (1.02)	0.21	0.10 (0.00, 0.72)	$F_{1,39} = 11.90$	0.001**
Winter-active (hibernate as adult)	10 (2, 19)	1.42 (0.54)	0.62 (0.86)	0.40	0.00 (0.00, 0.88)	$F_{1,8} = 6.88$	0.031*
Winter-active (ex. hibernate as adult)	31 (2, 51)	1.39 (0.62)	1.45 (1.52)	0.12	0.12 (0.00, 0.72)	$F_{1,29} = 4.97$	0.034*

Species included are active primarily between October and April. The number of species analysed is shown in each case, with those excluded because of a recorded abundance of zero ('Excluded'), and the total number of species in that taxon occurring in the British Isles ('Total No.'). Those species active in autumn and early spring but which hibernate as adults during the coldest months are analysed separately, as are other winter-active species excluding this group. There is no significant difference in variance in abundance between those species which hibernate as adults and those which do not (Brown-Forsythe test,  $F_{1,39} = 0.45$ ,  $P = 0.510$ ). Pagel's  $\lambda$  was calculated by maximum likelihood. Abundance was  $\log_{10}$ -transformed to ensure normality.

\* $P < 0.05$ , \*\* $P < 0.01$ .



**Fig. 3** Abundance and degree of polymorphism in British winter-active Lepidoptera. Shows all species in the imaginal stage primarily from October to April, including those which hibernate as adults during the coldest months of the year (solid points) and those which do not (hollow points). Phylogenetically corrected regression lines shown for all winter-active species (solid line), only those which hibernate as adults (long dash) and winter-active species which do not hibernate as adults (short dash). A significant positive relationship was detected across all winter-active species ( $r^2 = 0.21$ ,  $F_{1,39} = 11.90$ ,  $P = 0.001$ ), those which hibernate as adults ( $r^2 = 0.40$ ,  $F_{1,8} = 6.88$ ,  $P = 0.031$ ) and winter-active species which do not hibernate as adults ( $r^2 = 0.12$ ,  $F_{1,29} = 4.97$ ,  $P = 0.034$ ). Abundance is  $\log_{10}$ -transformed to ensure normality of the residuals.

Among the other Lepidopteran taxonomic groups examined, a significant positive relationship exists between abundance and degree of polymorphism only in the subfamilies Larentiinae ( $F_{1,129} = 8.38$ ,  $P = 0.004$ ), Cucullinae ( $F_{1,31} = 7.68$ ,  $P = 0.010$ ), Hadeninae ( $F_{1,39} = 14.01$ ,  $P < 0.001$ ) and the genus *Xanthorhoe* ( $F_{1,4} = 22.39$ ,  $P = 0.009$ ; Table 4). The coefficient of the relationship was greatest in the genus *Xanthorhoe*, though was not significantly different between any of these groups ( $F_{3,207} = 1.07$ ,  $P = 0.361$ , ANOVA). The modelled linear relationship explained 81.1% of variation in polymorphism in the genus *Xanthorhoe*, more than in any

other group in which a significant relationship exists. This shows that abundance is only positively related to degree of polymorphism in some groups, suggesting that search image-driven apostatic selection only acts on certain taxonomic groups, at least with a strength sufficient to be detectable in this analysis. Phylogenetic signal varies across the taxonomic families and subfamilies. In those subfamilies for which data and sequences were available for greater than 50 species (considered reliable estimates, see Munkemüller *et al.*, 2012), the mean value of Pagel's  $\lambda$  ranged from 0.00 to 0.53, suggesting phylogenetic covariation in these traits also varies with taxonomic group (Table 4).

## Discussion

This study addresses, from two distinct approaches, Kettlewell's (1973) hypothesis that the polymorphism exhibited by British winter-active Lepidoptera is due to apostatic selection exerted by birds hunting to search images. The field experiments (a) show a significant and positive relationship between the proportion at which a particular colour morph occurred and the probability that an individual belonging to that morph would be predated. This is indicative of search image use and in contrast with the expectation of a constant relationship where individuals are predated randomly during searching (Mook *et al.*, 1960; Tinbergen, 1960). The phylogenetic comparative analyses (b) demonstrate a significant positive correlation between abundance and degree of polymorphism across those species of British Lepidoptera active during the winter months (October–April), consistent with the theoretical expectation that apostatic selection ought to be exerted most strongly on common species where predators employ searching images during foraging (Clarke, 1962; Kettlewell, 1973).

The results of part (a) cannot be explained by preference for familiar prey in predators. Typically, the acquisition of such a preference involves training predators on one prey type before introducing them to another (e.g. Beukema, 1968; Murdoch, 1969; Ware, 1971;

**Table 4** Phylogenetically corrected regressions of abundance against degree of polymorphism in the British Geometridae, Noctuidae and their larger constituent taxa.

Taxa			Species analysed	Coefficient	Intercept	Adjusted	Pagel's $\lambda$	F-statistic	P-value	
Family	Sub-family	Genus	(Excluded, Total No.)	(SE)	(SE)	$r^2$	(95% CIs)			
Geometridae			199 (24, 310)	0.34 (0.25)	3.59 (0.92)	0.00	<b>0.47 (0.19, 0.76)</b>	$F_{1,197} = 1.77$	0.185	
		Ennominae	62 (4, 90)	-0.79 (0.61)	5.51 (1.40)	0.01	<b>0.33 (0.00, 0.00)</b>	$F_{1,60} = 1.67$	0.202	
		Larentiinae	131 (10, 166)	0.75 (0.26)	2.12 (1.06)	0.05	<b>0.53 (0.14, 0.89)</b>	$F_{1,129} = 8.38$	0.004*	
			<i>Eulithis</i>	5 (0, 5)	5.44 (2.22)	-9.52 (6.41)	0.56	0.79 (0.00, 0.00)	$F_{1,3} = 6.00$	0.092
			<i>Eupithecia</i>	34 (1, 48)	0.13 (0.23)	0.57 (0.36)	0.01	0.00 (0.00, 0.47)	$F_{1,32} = 0.31$	0.583
			<i>Perizoma</i>	9 (0, 10)	0.11 (0.34)	1.64 (0.60)	0.02	0.00 (0.00, 0.00)	$F_{1,7} = 0.11$	0.747
			<i>Thera</i>	5 (0, 6)	1.19 (1.42)	1.19 (1.42)	0.19	0.00 (0.00, 0.00)	$F_{1,3} = 0.70$	0.463
			<i>Xanthorhoe</i>	6 (1, 8)	2.24 (0.47)	-1.59 (1.42)	0.81	0.00 (0.00, 0.00)	$F_{1,4} = 22.39$	0.009*
		Sterrhinae		6 (0, 40)	0.01 (1.92)	8.51 (2.37)	6.28	0.00 (0.00, 0.93)	$F_{1,4} = 2.51$	0.996
			<i>Cyclophora</i>	5 (0, 8)	-0.39 (2.20)	8.25 (2.63)	0.01	0.00 (0.00, 0.00)	$F_{1,3} = 0.03$	0.872
	Noctuidae			209 (57, 425)	0.45 (0.29)	1.82 (1.06)	0.01	<b>0.34 (0.16, 0.53)</b>	$F_{1,207} = 2.30$	0.131
		Acronictinae	11 (2, 18)	0.81 (0.88)	4.06 (0.83)	0.09	0.00 (0.00, 0.98)	$F_{1,9} = 0.86$	0.379	
			<i>Acronicta</i>	7 (2, 11)	1.11 (1.22)	3.54 (1.19)	0.14	0.46 (0.00, 0.00)	$F_{1,5} = 0.83$	0.405
		Amphipyriinae	57 (13, 107)	0.66 (0.67)	3.16 (1.18)	0.02	<b>0.00 (0.00, 0.26)</b>	$F_{1,55} = 0.99$	0.325	
			<i>Apamea</i>	12 (1, 16)	1.07 (1.18)	1.85 (2.01)	0.08	0.00 (0.00, 0.00)	$F_{1,10} = 0.81$	0.388
		Cucullinae	33 (11, 68)	1.24 (0.45)	1.74 (0.74)	0.17	0.00 (0.00, 0.00)	$F_{1,31} = 7.68$	0.010*	
		Hadeninae	41 (13, 67)	1.37 (0.37)	2.06 (0.67)	0.25	0.00 (0.00, 0.55)	$F_{1,39} = 14.01$	0.586	
			<i>Lacanobia</i>	5 (1, 7)	-0.68 (0.49)	1.82 (0.94)	0.19	1.00 (0.00, 0.00)	$F_{1,3} = 1.91$	0.261
			<i>Mythimna</i>	10 (3, 18)	0.76 (0.44)	1.49 (0.93)	0.18	0.56 (0.00, 0.00)	$F_{1,8} = 2.94$	0.125
			<i>Orthosia</i>	9 (0, 9)	0.95 (0.90)	5.38 (2.66)	0.01	1.00 (0.00, 0.00)	$F_{1,7} = 1.10$	0.329
		Hypeninae	9 (0, 19)	0.29 (0.68)	0.85 (1.42)	0.03	1.00 (0.00, 0.00)	$F_{1,7} = 0.18$	0.686	
		Noctuinae	41 (6, 64)	-0.39 (0.91)	5.59 (3.02)	0.01	0.38 (0.02, 0.69)	$F_{1,39} = 0.18$	0.674	
			<i>Agrotis</i>	7 (1, 13)	1.72 (1.62)	7.07 (2.77)	0.02	0.00 (0.00, 0.98)	$F_{1,5} = 1.13$	0.337
			<i>Noctua</i>	5 (1, 7)	3.15 (1.92)	0.49 (3.63)	0.30	0.00 (0.00, 0.00)	$F_{1,3} = 2.71$	0.198
			<i>Xestia</i>	9 (1, 11)	1.06 (0.63)	0.18 (1.48)	0.18	0.00 (0.00, 0.00)	$F_{1,7} = 2.81$	0.138
	Plusiinae	9 (4, 23)	0.53 (0.41)	0.10 (0.70)	0.08	0.00 (0.00, 0.00)	$F_{1,7} = 1.69$	0.235		

Constituent taxonomic groups (subfamilies and genera) were analysed individually where data were available for five or more species. The number of species analysed is shown in each case, with those excluded because of a recorded abundance of zero ('Excluded'), and the total number of species in that taxon occurring in the British Isles ('Total No.'). Pagel's  $\lambda$  was calculated by maximum likelihood and estimates with larger species sample sizes ( $n > 50$ ), considered more reliable (Munkemüller *et al.*, 2012), are emboldened. Abundance was  $\log_{10}$ -transformed to ensure normality.

\* $P < 0.01$ , \*\* $P < 0.001$ .

Shettleworth, 1972; Allen, 1973). Here, however, both morphs occur at different frequencies within a single population and although they will be encountered sequentially, mimicking natural conditions, there is no protracted period of training. Even if predators could acquire a preference through a few encounters only, the probability that any particular morph would be encountered initially by a predator is approximately equal to its proportion frequency within the population. Hence, an approximately constant relationship would result between proportion occurrence and probability of predation.

An alternative mechanism by which predators might consume prey in a manner superficially suggestive of search image use is by adjusting the rate at which they search the environment (Gendron & Staddon, 1983, 1984; Guilford & Dawkins, 1987). By decreasing its

search rate, and spending more time examining a single patch, a predator is more likely to perceive cryptic prey; the time required for detection increasing with increasing cryptic resemblance. However, decreasing search rate reduces the total area which may be searched in a given time. Thus, the particular rate which will optimize the number of prey detected changes with fluctuations in the relative abundance of prey of varying crypticity; for example, many less cryptic prey will favour a high search rate, reducing predation on rarer, more cryptic prey. In optimizing their search rate, predators may therefore exert negative frequency-dependent selection (Guilford & Dawkins, 1987). Implicit in this hypothesis, however, is that all equally cryptic prey will be perceptible at a particular search rate, in contrast with search image use by predators, where the utilization of an image ought to actively interfere with

perception of visually distinct prey, regardless of crypticity. The morphs used in this experiment did not differ significantly in their probability of predation, other than morphs *r* and *l*, implying approximately equal crypticity. These morphs should therefore have been equally noticeable to a predator optimizing its search rate for their level of crypsis. Thus, they would have been predated in approximate proportion to their abundance, and hence it is unlikely that the observed results were due to predatory search rate adjustment.

Birds appear to have been responsible for predated the models (Fig. S1) and the consequent apostatic effects of search image use are consistent with the results of previous studies of birds hunting artificial and natural models under experimental conditions (e.g. Allen, 1973; Majerus, 1978, 1980, 1998; Tucker, 1991; Bond & Kamil, 1998, 2002, 2006). However, the present work is the first to consider the behaviour of predator populations under natural conditions, with artificial prey mimicking a wild predator-prey system. The concurrence of results suggests that search image behaviour is consistently adopted by birds and, as the models replicated the colouration, resting site selection and phenology of British winter-active Lepidoptera, it is reasonable to conclude that they utilize search images while predated this group in nature, resulting in apostatic selection on populations of these moths.

Interestingly, morphs *r* and *l* had adjusted 'G' colour components, when compared with the reference model (morph *c*), and were significantly more likely to be predated than an individual of morph *c* (Table 2). This modification may have rendered them less cryptic, and consequently more likely to be noticed and attacked. In addition, the interaction between probability of predation and increasing proportion in the population is significantly negative in morphs *r* and *l*, compared with that of morph *c*. It does not necessarily follow, however, that these relationships are negative overall, but rather negative only with reference to the significantly positive relationship of morph *c*. Hence, the relationship in morphs *r* and *l* may encompass a range of negative, neutral or mildly positive slopes (Table 2; Fig. 2).

Variation in the relationship between probability of predation and abundance in those models with a significantly higher intrinsic probability of predation is consistent with the use of search images to detect only cryptic prey, because of the costs of limited attention to predators (Karpestam *et al.*, 2014). Conspicuous prey may be perceived without a great deal of visual processing of the environment, and search images need not therefore be employed to optimize foraging. Variation in the nature of this relationship, and hence the resultant apostatic selection, suggests relative crypticity may affect the degree to which search image-driven apostatic selection acts on different morphs within a population, determining their final equilibrium frequencies (see Bond & Kamil, 1998, 2002).

The small size of the artificial populations used in these experiments also suggests that some birds are capable of forming search images more rapidly than previously considered (Majerus, 1978, 1980, 1998; Bond & Kamil, 1998, 2002, 2006). Additionally, this highlights the importance of prey density for, given the apparent speed of image adoption, species which are locally abundant may be subject to apostatic selection though rare at a larger spatial scale. Were these individuals spread evenly over a larger area the profitability of locally hunting to their image would be much reduced (Clarke, 1962). This may present implications for species which occur at high densities within relatively restricted areas, such as populations of insects grouped on individual food-plants.

The positive correlation evident in the comparative analyses (b) is predicted under search image use because, as the abundance of a species increases, it is encountered more frequently by predators in the course of searching and it becomes more profitable to hunt to an image of any constituent morph of that species (Clarke, 1962); images of morphs of rare species will be degraded and lost due to lack of subsequent encounters. Hence, apostatic selection exerted by search image use will be stronger on common species, which consequently ought to evolve more extensive phenotypic polymorphisms (Clarke, 1962; Kettlewell, 1973). This significant positive correlation is also evident when we consider those winter-active species active in late autumn and early spring but which hibernate as adults during the coldest months of the year, and those which are fully active throughout (Table 3; Fig. 3). Although this suggests that predators exert apostatic selection on species adopting these two distinct ecological strategies, it is notable that the modelled relationship between abundance and polymorphism accounts for the greatest quantity of variation among those species which hibernate. These species may be more affected by apostatic selection due to increased reliance on avoiding visual detection during their substantial dormant periods, when they are unable to flee predators. Furthermore, there is no significant difference in variance in abundance (the explanatory variable) between these two groups, which could have potentially accounted for this difference (Brown-Forsythe test,  $F_{1,39} = 0.45$ ,  $P = 0.510$ ; Table 3).

Across the other Lepidopteran taxonomic groups examined, a significant correlation was detected in only three subfamilies and a single genus (Table 4). Winter-active species belong to a range of taxa and hence this seems supportive of the hypothesis that the environmental conditions of winter are particularly conducive to the action of apostatic selection on Lepidopteran populations. Kettlewell (1973) argued that the much-reduced size of insect populations in winter would lead to higher levels of predation by birds on the few species which were active. Hence, where apostatic selection

occurs, it may be stronger on prey populations active in winter, and therefore more likely to lead to the evolution of polymorphism. The lack of such a correlation across the higher taxonomic groups – comprised of species active across a range of time periods – seems supportive of this. Search images may also be less effective when more prey species are active, and there is a greater likelihood of a searching predator encountering a wide range of visually distinct prey. Colour polymorphism in species belonging to groups where apostatic selection does not appear to operate, as indicated by a lack of correlation, may be driven by a range of other mechanisms such as heterozygote advantage or they may represent transient polymorphisms (Ford, 1954) brought about by shifting fitness optima, such as in many industrially melanic species (Kettlewell, 1973; Majerus, 1998). Indeed, although search image use may affect all species, the magnitude of its apostatic effect on prey populations will vary with their ecology (Bond, 2007).

This lack of relationship did not exist across all taxonomic groups analysed and some groups show strong correlations, despite nonsignificance; in such instances, small species sample sizes may have rendered detecting relationships difficult. Of those groups with a significant interaction, the coefficient of this relationship was highest in *Xanthorhoe*, a typical, widespread geometrid genus (Skinner, 2009), where variation in abundance explained some 81.1% of variation in degree of polymorphism (Table 4). This suggests that the polymorphism displayed by this group is almost wholly due to apostatic effects and that the magnitude of apostatic selection, presumably exerted by birds, must be great. Interestingly, of the two noctuid subfamilies which display a significant positive correlation, many species in the Cuculliinae are active either in spring (e.g. *Conistra*) or late autumn (*Agrochola*, *Xanthia*, *Polymixis*, *Aporophylla*). As such, a positive relationship may only exist because it is composed primarily of winter-active species, among which a strong correlation is evident.

The preceding discussion assumes that a correlation between abundance and polymorphism is evidence of the action of apostatic selection, and this does not necessarily follow. For instance, although mutation rates may be roughly comparable across Lepidopteran taxa, the absolute number of mutations will be greater in the larger populations of more common species (van Treuren *et al.*, 1991; Ellstrand & Elam, 1993; Frankham, 1996). Thus, even if mutations were neutral or weakly deleterious (i.e. nonlethal) it may be expected that some positive relationship would exist between population size and phenotypic variability (Ouborg *et al.*, 1991). However, in both monographic treatments of intraspecific variation utilized as sources of information, the morphs described are generally common, widespread and co-occurring. It is not typical practice to name unique mutants, and forms with limited

geographical distribution would be designated ‘races’ (Weir, 2016); these were not included in the analyses. This explanation of the correlation is therefore unlikely and, furthermore, cannot explain the absence of a correlation in the remaining groups, which too display great variation in abundance. Such between-group variation in the correlation is expected, however, given variation in ecology and environment, under the apostatic selection hypothesis (Clarke, 1962). In addition, secondary sympatry of previously locally adapted forms is unlikely to account for apparent polymorphism given the common co-occurrence of most morphs across the continent-wide distributions of these species (e.g. Tutt, 1891; Seitz, 1912, 1914). It is also possible that any positive correlation could be a result of sampling effects, where more common species, being more frequently encountered, have received greater attention from taxonomists and hence have received more intraspecific names. The intensity with which British Lepidoptera have been studied, however, with particular emphasis on variation in colouration, makes this unlikely, and within-species variation is well described even in rare taxa.

Greater abundance may also favour the evolution of more extensive polymorphisms, if it is accompanied by an increased range of habitat utilization – such species would therefore effectively inhabit a more coarse-grained environment, with a greater range of backgrounds, potentially resulting in disruptive selection on morph colouration. Similarly, if moths were able to select resting sites which optimized their cryptic resemblance, even a species inhabiting an environment with a relatively fine-grained background could evolve polymorphism in this way. However, there is no evidence to suggest more common species occur in a greater range of habitats (e.g. Skinner, 2009). The little work which has been conducted on resting site preference in Lepidoptera has tended to yield conflicting results or suggest very broad background preferences (Majerus, 1998), for example for dark or light surfaces (Kettlewell, 1973) – of a specificity not sufficient to maintain polymorphism on a fine-grained background.

Future experiments should employ highly polymorphic model populations to better mimic the natural exuberant polymorphisms which search image use has been implicated in maintaining (Franks & Oxford, 2009). In winter-active Lepidoptera, living individuals should be incorporated into a similar experimental design (e.g. using light trapped specimens) in natural resting sites, greatly increasing the confidence with which generalizations could be made regarding apostatic selection on wild populations. In addition, the robustness of comparative analyses dealing with polymorphism would be greatly improved by modern monographic treatments of variation in British and Continental species, delimiting the boundaries of intraspecific taxa and differentiating true polymorphic

forms. More generally, if the correlation between abundance and degree of polymorphism does indeed reflect the results of search image-driven apostatic selection, it ought to be observed in other groups where search image use has evolved phenotypic polymorphisms. Groups in which morph frequencies in localized populations can be easily manipulated, such as littorial molluscs (e.g. Johannesson & Butlin, 2017), present promising, tractable targets for such research.

In conclusion, birds appear to have utilized search images while preying on the artificial populations of moth-like models used in this experiment. These models were based on the colouration of a British winter-active moth and mimicked the resting site selection and phenology of such species. If it is assumed that birds exhibit comparable behaviour towards natural winter-active Lepidoptera, apostatic selection would be exerted on these populations favouring the evolution of polymorphism. In addition, a significant positive relationship was detected between the abundance of British species of Lepidoptera active in the winter months (October–April) and their degree of polymorphism, indicative of search image use by their predators. Taken together, these two independent lines of enquiry are consistent with the theoretical expectations of Kettlewell's (1973) original hypothesis—that polymorphism in British winter-active Lepidoptera evolved as a result of search image-driven apostatic selection exerted by avian predators. Furthermore, these results support the validity of search image-driven apostatic selection as a mechanism by which phenotypic and genetic variation may be maintained within natural populations more generally.

## Acknowledgments

I am deeply indebted to my supervisor Dr Matthew Bell; any merit which this work may possess owes much to him giving generously of his own time and providing advice, discussion and comment throughout. I should also like to thank the Forestry Commission for granting me permission to carry out the field experiments in Devilla Forest. I am grateful to collaborators and staff who contributed data from the Rothamsted Insect Survey, a BBSRC-supported national capability. Specifically, I would like to thank Mr Chris Shortall for his assistance in obtaining the moth abundance data utilized in the comparative analyses. The University of Edinburgh provided funding for experimental materials, and two reviewers made many constructive suggestions on the original manuscript.

## References

Allen, J.A. 1973. Apostatic selection: the response of wild passerines to artificial polymorphic prey. Ph.D. thesis, University of Edinburgh.

- Allen, J.A. 1988. Frequency-dependent selection by predators. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* **319**: 485–503.
- Allen, J.A. & Clarke, B. 1968. Evidence for apostatic selection by wild passerines. *Nature* **220**: 501–502.
- Allen, J.A., Raymond, D.L. & Geburtig, M.A. 1988. Wild birds prefer the familiar morph when feeding on pastry-filled shells of the landsnail *Cepaea hortensis* (Mull). *Biol. J. Linn. Soc.* **33**: 395–401.
- Atkinson, W.D. & Warwick, T. 1983. The role of selection in the colour polymorphism of *Littorina rudis* Maton and *Littorina arcana* Hannaford-Ellis (Prosobranchia: Littorinidae). *Biol. J. Linn. Soc.* **20**: 137–151.
- Bates, D., Maechler, M., Bolker, B. & Walker, S. 2015. Fitting linear mixed-effects models using lme4. *J. Stat. Softw.* **67**: 1–48.
- Beddard, F.E. 1892. *Animal Coloration: An Account of the Principal Facts and Theories Relating to the Colours and Markings of Animals*. Swan Sonnenschein and Co., London.
- Bennett, A.T.D. & Cuthill, I.C. 1994. Ultraviolet vision in birds. What is its function? *Vision Res.* **34**: 1471–1478.
- Beukema, J.J. 1968. Predation by three-spined stickleback (*Gasterosteus aculeatus* L.): the influence of hunger and experience. *Behaviour* **31**: 1–126.
- Blough, P.M. 1989. Attentional priming and visual search in pigeons. *J. Exp. Psychol. Anim. Behav. Process* **15**: 358–365.
- Blough, P.M. 1991. Selective attention and search images in pigeons. *J. Exp. Psychol. Anim. Behav. Process* **17**: 292–298.
- Blough, P.M. 1992. Detectability and choice during visual search: joint effects of sequential priming and discriminability. *Anim. Learn. Behav.* **20**: 293–300.
- Bond, A.B. 1983. Visual search and selection of natural stimuli in the pigeon: the attention threshold hypothesis. *J. Exp. Psychol. Anim. Behav. Process* **9**: 292–306.
- Bond, A.B. 2007. The evolution of color polymorphism: crypticity, searching images, and apostatic selection. *Annu. Rev. Ecol. Evol. Syst.* **38**: 489–514.
- Bond, A.B. & Kamil, A.C. 1998. Apostatic selection by blue jays produces balanced polymorphism in virtual prey. *Nature* **395**: 594–596.
- Bond, A.B. & Kamil, A.C. 2002. Visual predators select for crypticity and polymorphism in virtual prey. *Nature* **415**: 609–613.
- Bond, A.B. & Kamil, A.C. 2006. Spatial heterogeneity, predator cognition, and the evolution of color polymorphism in virtual prey. *Proc. Natl. Acad. Sci. USA* **103**: 3214–3219.
- Bond, A.B. & Riley, D.A. 1991. Searching image in the pigeon: a test of three hypothetical mechanisms. *Ethology* **87**: 203–224.
- Brunton, C.F.A. 1998. The evolution of ultraviolet patterns in European *Colias* butterflies (Lepidoptera, Pieridae): a phylogeny using mitochondrial DNA. *Heredity* **80**: 611–616.
- Caro, T. 2005. The adaptive significance of colouration in mammals. *Bioscience* **55**: 125–136.
- Clarke, B. 1962. Balanced polymorphism and the diversity of sympatric species. In: *Taxonomy and Geography* (D. Nichols, ed.), pp. 47–70. Systematics Association, Oxford.
- Clarke, B. 1969. The evidence for apostatic selection. *Heredity* **24**: 347–352.
- Collenette, C.L. 1935. Notes concerning attacks by British birds on butterflies. *Proc. Zool. Soc. Lond.* **105**: 201–217.
- Common, I.F.B. 1954. A study of the ecology of the adult bogong moth, *Agrotis infusa* (Boisd.) (Lepidoptera: Noctuidae), with special reference to its behaviour during migration and aestivation. *Aust. J. Zool.* **2**: 223–263.

- Cooper, J.M. 1984. Apostatic selection on prey that match the background. *Biol. J. Linn. Soc.* **23**: 221–228.
- Coppinger, R.F. 1969. The effect of experience and novelty on avian feeding behaviour with reference to the evolution of warning colouration in butterflies Part I: reactions of wild-caught adult blue jays to novel insects. *Behaviour* **35**: 45–60.
- Coppinger, R.F. 1970. The effect of experience and novelty on avian feeding behaviour with reference to the evolution of warning colouration in butterflies. II. Reactions of naïve birds to novel insects. *Am. Nat.* **104**: 323–337.
- Cott, H.B. 1940. *Adaptive Coloration in Animals*. Methuen and Co., Ltd., London.
- Croze, H.J. 1970. *Searching Images in Carrion Crows: Hunting Strategy in a Predator and Some Anti-Predator Devices in Camouflaged prey*. Parey, Berlin.
- Cuthill, I.C., Stevens, M., Sheppard, J., Maddocks, T., Parraga, C.A. & Troscianko, T.S. 2005. Disruptive coloration and background pattern matching. *Nature* **434**: 72–74.
- Dawkins, M. 1971. Shifts in “attention” in chicks during feeding. *Anim. Behav.* **19**: 575–582.
- Den Boer, M.H. 1970. A colour polymorphism in caterpillars of *Bupalus piniarius* (L.) (Lepidoptera: Geometridae). *Neth. J. Zool.* **21**: 61–63.
- Desimone, R. & Duncan, J. 1995. Neural mechanisms of selective and visual attention. *Annu. Rev. Neurosci.* **18**: 193–222.
- Drummond, A.J., Nicholls, G.K., Rodrigo, A.G. & Solomon, W. 2002. Estimating mutation parameters, population history and genealogy simultaneously from temporally spaced sequence data. *Genetics* **161**: 1307–1320.
- Drummond, A.J., Ho, S.Y.W., Phillips, M.J. & Rambaut, A. 2006. Relaxed phylogenetics and dating with confidence. *PLoS Biol.* **4**: e88.
- Drummond, A.J., Suchard, M.A., Xie, D. & Rambaut, A. 2012. Bayesian phylogenetics with BEAUTi and the BEAST 1.7. *Mol. Biol. Evol.* **29**: 1969–1973.
- Dukas, R. 2002. Behavioural and ecological consequences of limited attention. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* **357**: 1539–1547.
- Dukas, R. & Ellner, S. 1993. Information-processing and prey detection. *Ecology* **74**: 1337–1346.
- Dukas, R. & Kamil, A.C. 2000. The cost limited attention in blue jays. *Behav. Ecol.* **12**: 192–199.
- Dukas, R. & Kamil, A.C. 2001. Limited attention: the constraint underlying search image. *Behav. Ecol.* **12**: 192–199.
- Edmunds, M. 1976. The defensive behaviour of Ghanaian praying mantids with a discussion of territoriality. *Zool. J. Linn. Soc.* **58**: 1–37.
- Edwards, J.G. 1954. A new approach to infraspecific categories. *Syst. Biol.* **3**: 1–20.
- Ellstrand, N.C. & Elam, D.R. 1993. Population genetic consequences of small population size: implications for plant conservation. *Annu. Rev. Ecol. Syst.* **24**: 217–242.
- Fogden, M. & Fogden, P. 1974. *Animals and Their Colors*. Crown Publishers Inc., New York, NY.
- Ford, E.B. 1954. *Butterflies*. Collins, London.
- Ford, E.B. 1955. *Moths*. Collins, London.
- Ford, E.B. 1964. *Ecological Genetics*. Methuen and Co., Ltd., London.
- Fowlie, M.K. & Kruger, O. 2003. The evolution of plumage polymorphism in birds of prey and owls: the apostatic selection hypothesis revisited. *J. Evol. Biol.* **16**: 577–583.
- Frankham, R. 1996. Relationship of genetic variation to population size in wildlife. *Conserv. Biol.* **10**: 1500–1508.
- Franks, D.W. & Oxford, G.S. 2009. The evolution of exuberant visible polymorphisms. *Evolution* **63**: 2697–2706.
- Fry, R. & Waring, P. 2001. *A Guide to Moth Traps and Their Use*. Amateur Entomologists’ Society, Orpington, Kent.
- Gendron, R.P. & Staddon, J.E.R. 1983. Searching for cryptic prey: the effect of search rate. *Am. Nat.* **121**: 172–186.
- Gendron, R.P. & Staddon, J.E.R. 1984. A laboratory simulation of foraging behavior: the effect of search rate on the probability of detecting prey. *Am. Nat.* **124**: 407–415.
- Gernhard, T. 2008. The conditioned reconstructed process. *J. Theor. Biol.* **253**: 769–778.
- Glanville, P.W. & Allen, J.A. 1997. Protective polymorphism in populations of computer-simulated moth-like prey. *Oikos* **80**: 565–571.
- Grafen, A. 1989. The phylogenetic regression. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* **326**: 119–157.
- Guilford, T. & Dawkins, M.S. 1987. Search images not proven: a reappraisal of the evidence. *Anim. Behav.* **35**: 1838–1845.
- Hall, T. 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symp. Ser.* **41**: 95–98.
- Harmer, A.S. 2000. *Variation in British Butterflies*. Paphia Publishing Ltd., Lymington, Hampshire.
- Harvey, P.H., Birley, N. & Blackstock, T.H. 1975. The effect of experience on the selective behaviour of song thrushes feeding on artificial populations of *Cepaea* (Held.). *Genetica* **45**: 211–216.
- Hoekstra, H.E., Krenz, J.G. & Nachman, M.W. 2005. Local adaptation in the rock pocket mouse (*Chaetodipus intermedius*): natural selection and phylogenetic history of populations. *Heredity* **94**: 217–228.
- Howarth, T.G. 1973. *Colour Identification Guide to Butterflies of the British Isles*. Viking, London.
- Jamie, G.A. 2017. Signals, cues and the nature of mimicry. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* **284**: 20162080.
- Johannesson, K. & Butlin, R.K. 2017. What explains rare and conspicuous colours in a snail? A test of time-series data against models of drift, migration or selection. *Heredity* **118**: 21–30.
- Karpestam, E., Merilaita, S. & Forsman, A. 2014. Natural levels of colour polymorphism reduce performance of visual predators searching for camouflaged prey. *Biol. J. Linn. Soc.* **112**: 546–555.
- Kettlewell, H.B.D. 1955. Selection experiments on industrial melanism in the Lepidoptera. *Heredity* **9**: 323–342.
- Kettlewell, H.B.D. 1973. *The Evolution of Melanism: The Study of a Recurring Necessity*. Oxford University Press, Oxford.
- Kloet, G.S. & Hincks, W.D. 1972. *A Check List of British Insects: Part 2 Lepidoptera*. Royal Entomological Society, London.
- Knill, R. & Allen, J.A. 1995. Does polymorphism protect—an experiment with human predators. *Ethology* **19**: 5101–5125.
- Knowles, J.E. & Frederick, C. 2016. merTools: tools for analysing mixed effect regression models. R. package version 0.3.0. <https://CRAN.R-project.org/package=merTools>
- Krebs, J.R. 1973. Behavioural aspects of predation. In: *Perspectives in Ethology*, Vol. 1 (P.H. Klopfer & P.P.G. Bateson, eds), pp. 73–111. Plenum Press, London.
- Langley, C.M. 1996. Search images: selective attention to specific visual features of prey. *J. Exp. Psychol. Anim. Behav. Process* **22**: 152–163.



- Levene, H. 1953. Genetic equilibrium when more than one ecological niche is available. *Am. Nat.* **87**: 331–333.
- Lyytinen, A., Lindstrom, L. & Mappes, J. 2004. Ultraviolet reflection and predation risk in diurnal and nocturnal Lepidoptera. *Behav. Ecol.* **15**: 982–987.
- Majerus, M.E.N. 1978. The control of larval colour variation in *Phlogophora meticulosa* Linn. (Lepidoptera: Noctuidae) and some of its consequences. Ph.D. thesis, University of London.
- Majerus, M.E.N. 1980. The control of larval colour variation in the angleshades moth (*Phlogophora meticulosa* Linn.) Part II: the maintenance of the variation. *Bull. Am. Ent. Soc.* **39**: 85–89.
- Majerus, M.E.N. 1998. *Melanism: Evolution in Action*. Oxford University Press, Oxford.
- Merilaita, S. 2006. Frequency-dependent predation and maintenance of prey polymorphism. *J. Evol. Biol.* **19**: 2022–2030.
- Merilaita, S., Tuomi, J. & Jormalainen, V. 1999. Optimization of cryptic coloration in heterogeneous habitats. *Biol. J. Linn. Soc.* **67**: 151–161.
- Meyer-Rochow, V.B. 1991. Differences in ultraviolet wing patterns in the New Zealand lycaenid butterflies *Lycaena salustius*, *L. rauparaha*, and *L. feredayi* as a likely isolating mechanism. *J. R. Soc. N. Z.* **21**: 169–177.
- Moment, G.B. 1962. Reflexive selection: a possible answer to an old puzzle. *Science* **136**: 262–263.
- Mook, J.H., Mook, L.J. & Heikens, H.S. 1960. Further evidence for the role of “searching images” in the hunting behaviour of titmice. *Arch. Neerl. Zool.* **13**: 448–465.
- Morrell, G.M. & Turner, J.R.G. 1970. Experiments on mimicry I: the response of wild birds to artificial prey. *Behaviour* **36**: 116–130.
- Munkemuller, T., Lavergne, S., Bzeznik, B., Dray, S., Jombart, T., Schiffrers, K. *et al.* 2012. How to measure and test phylogenetic signal. *Methods Ecol. Evol.* **3**: 743–756.
- Murdoch, W.W. 1969. Switching in general predators: experiments on predator specificity and stability of prey populations. *Ecol. Monogr.* **39**: 335–354.
- Murdoch, W.W., Avery, S. & Smyth, M.E.B. 1975. Switching in predatory fish. *Ecology* **56**: 1094–1105.
- Murton, R.K. 1971. The significance of a specific search image in the feeding behaviour of the wood pigeon. *Behaviour* **40**: 10–42.
- Nabours, R.K., Larson, I. & Hartwig, N. 1933. Inheritance of color patterns in the grouse locust *Acrydium arenosum* Burmeister (Tettigidae). *Genetics* **18**: 159–171.
- Norris, K.S. & Lowe, C.H. 1964. An analysis of background color-matching in amphibians and reptiles. *Ecology* **45**: 565–580.
- Orme, D., Freckleton, R., Thomas, G., Petzoldt, T., Fritz, S., Isaac, N. *et al.* 2013. caper: comparative analyses of phylogenetics and evolution. R package version 0.5.2. <https://CRAN.R-project.org/package=caper>
- Ouborg, N.J., van Treuren, R. & van Damme, J.M.M. 1991. The significance of genetic erosion in the process of extinction. II. Morphological variation and fitness components in populations of varying size of *Salvia pratensis* L. and *Scabiosa columbaria* L. *Oecologia* **86**: 359–367.
- Owen, D.F. 1965. A population study of an equatorial land snail *Limnicolaria martensiana* (Achatinidae). *Proc. Zool. Soc. Lond.* **144**: 361–382.
- Owen, D.F. & Weigert, R.G. 1962. Balanced polymorphism in the meadow spittlebug, *Philaenus spumarius*. *Am. Nat.* **96**: 353–359.
- Oxford, G.S. 2009. An exuberant, undescribed colour polymorphism in *Theridion californicum* (Araneae, Theridiidae): implications for a theridiid pattern ground plan and the convergent evolution of visible morphs. *Biol. J. Linn. Soc.* **96**: 23–34.
- Oxford, G.S. & Gillespie, R.G. 1998. Evolution and ecology of spider coloration. *Annu. Rev. Entomol.* **43**: 619–643.
- Pietrewicz, A.T. & Kamil, A.C. 1979. Search image formation in the blue jay (*Cyanocitta cristata*). *Science* **136**: 262–263.
- Poulton, E.B. 1890. *The Colours of Animals: Their Meaning and Use Especially Considered in the Case of Insects*. Kegan Paul, Trench, Trübner, and Co. Ltd., London.
- Punzalan, D., Rodd, F.H. & Hughes, K.A. 2005. Perceptual processes and the maintenance of polymorphism through frequency-dependent predation. *Evol. Ecol.* **19**: 303–320.
- R Core Team. 2017. *R: A Language and Environment for Statistical Computing*. R foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org/>
- Rambaut, A., Suchard, M.A., Xie, D. & Drummond, A.J. 2014. Tracer v1.6. <http://beast.bio.ed.ac.uk/Tracer>
- Ratnasingham, S. & Hebert, P.D.N. 2007. BOLD: the barcode of life data system ([www.barcodinglife.org](http://www.barcodinglife.org)). *Mol. Ecol. Notes* **7**: 355–364.
- Reid, D.G. 1987. Natural selection for apostasy and crypsis acting on the shell colour polymorphism of a mangrove snail *Littoraria filosa* (Sowerby) (Gastropoda: Littorinidae). *Biol. J. Linn. Soc.* **30**: 1–24.
- Reiger, J.C., Mitter, C., Mitter, K., Cummings, M.P., Bazinet, A.L., Hallwachs, W. *et al.* 2017. Further progress on the phylogeny of Noctuoidea (Insecta: Lepidoptera) using an expanded gene sample. *Syst. Entomol.* **42**: 82–93.
- Robinson, R. 1971. *Lepidoptera Genetics*. Pergamon Press Ltd., Oxford.
- Roulin, A. 2004. The evolution, maintenance and adaptive function of genetic colour polymorphism in birds. *Biol. Rev.* **79**: 815–848.
- de Ruiter, L. 1952. Some experiments on the camouflage of stick caterpillars. *Behaviour* **4**: 222–232.
- Russwurm, A.D.A. 1978. *Aberrations of British Butterflies*. E. W. Classey Ltd., Faringdon, Oxfordshire.
- Ruxton, G.D., Sherratt, T.N. & Speed, M.P. 2004. *Avoiding Attack: The Evolutionary Ecology of Crypsis, Warning Signals and Mimicry*. Oxford University Press, Oxford.
- Sandoval, C.P. 1994. Differential visual predation on morphs of *Timema cristinae* (Phasmatodeae: Timemidae) and its consequences for host range. *Biol. J. Linn. Soc.* **52**: 341–356.
- Sargent, T.D. 1978. On the maintenance of stability in hindwing diversity among moths of the genus *Catocala* (Lepidoptera: Noctuidae). *Evolution* **32**: 424–434.
- Seitz, A. 1912. *The Macrolepidoptera of the World. Volume IV. The Palaearctic Geometrae*. Two volumes. Alfred Kernen, Stuttgart.
- Seitz, A. 1914. *The Macrolepidoptera of the World. Volume III. The Noctuid Moths*. Two volumes. Alfred Kernen, Stuttgart.
- Shettleworth, S.J. 1972. The role of novelty in learned avoidance of unpalatable “prey” by domestic chicks (*Gallus gallus*). *Anim. Behav.* **20**: 29–35.
- Skinner, B. 2009. *Colour Identification Guide to the Moths of the British Isles*. Apollo Books, Stenstrup, Denmark.
- Stevens, M. & Ruxton, G.D. 2012. Linking the evolution and form of warning coloration in nature. *Proc. Biol. Sci.* **279**: 417–426.
- Tavaré, S. 1986. Some probabilistic and statistical problems in the analysis of DNA sequences. *Lect. Math. Life Sci.* **17**: 57–86.
- Thayer, G.M. 1909. *Concealing-Coloration in the Animal Kingdom: An Exposition of the Laws of Disguise Through Color and Pattern*. Macmillan and Co., New York, NY.

- Tinbergen, L. 1960. The natural control of insects in pine woods I: factors influencing the intensity of predation by songbirds. *Arch. Neerl. Zool.* **13**: 265–343.
- Toussaint, E.F.A., Condamine, F.L., Kergoat, G.J., Capdevielle-Dulac, C., Barbut, J., Silvain, J. *et al.* 2012. Palaeoenvironmental shifts drove the adaptive radiation of a noctuid stem-borer tribe (Lepidoptera, Noctuidae, Apameini). *PLoS One* **7**: 1–15.
- van Treuren, R., Bijlsma, R., van Delden, W. & Ouborg, N.J. 1991. The significance of genetic erosion in the process of extinction. I. Genetic differentiation in *Salvia pratensis* and *Scabiosa columbaria* in relation to population size. *Heredity* **66**: 181–189.
- Trullas, S.C., van Wyk, J.H. & Spotila, J.R. 2007. Thermal melanism in ectotherms. *J. Therm. Biol.* **32**: 235–245.
- Tucker, G.M. 1991. Apostatic selection by song thrushes (*Turdus philomelos*) feeding on the snail *Cepaea hortensis*. *Biol. J. Linn. Soc.* **43**: 149–156.
- Turner, H.J. 1925–26. *List of the geometers of the British Isles with their named varieties*. Reprinted from *The Entomologist's Record and Journal of Variation* **36–37**.
- Tutt, J.W. 1891. *Melanism and Melanochroism in British Lepidoptera*. Swan Sonnenschein and Co., London.
- Tutt, J.W. 1891–92. *The British Noctuae and Their Varieties*. Four Volumes. Swan, Sonnenschein & Co., London.
- Wallace, A.R. 1878. *Tropical Nature, and Other Essays*. Macmillan and Co., London.
- Wallace, A.R. 1889. *Darwinism: An Exposition of the Theory of Natural Selection with Some of Its Implications*. Macmillan and Co., London.
- Ware, D.M. 1971. Predation by rainbow trout (*Salmo gairdneri*): the effects of experience. *J. Fish. Res. Board Can.* **28**: 1847–1852.
- Weir, J.C. 2016. Intraspecific taxonomy in the Lepidoptera. *Br. J. Entomol. Nat. His.* **29**: 144–154.
- Wennersten, L. & Forsman, A. 2009. Does colour polymorphism enhance survival of prey populations? *Proc. Biol. Sci.* **276**: 2187–2194.
- Whiteley, D.A.A., Owen, D.F. & Smith, D.A.S. 1997. Massive polymorphism and natural selection in *Donacilla cornea* (Poli, 1791) (Bivalvia: Mesodesmatidae). *Biol. J. Linn. Soc.* **62**: 475–494.
- Wickham, H. 2009. *ggplot2: Elegant Graphics for Data Analysis*. Springer-Verlag, New York, NY.
- Yamamoto, S. & Sota, T. 2007. Phylogeny of the Geometridae and the evolution of winter moths inferred from a simultaneous analysis of mitochondrial and nuclear genes. *Mol. Phylogenet. Evol.* **44**: 711–723.

### Supporting information

Additional Supporting Information may be found online in the supporting information section at the end of the article.

**Figure S1** Attacked moth models with evidence of bird predation.

Data deposited at Dryad: <https://doi.org/10.5061/dryad.s62ks69>

Received 18 September 2017; revised 29 March 2018; accepted 23 April 2018