

Titti Bohlin

Stop bugging me!

Diversity in appearance of warning coloration



Stop bugging me!
Diversity in appearance
of warning coloration

Titti Bohlin

Environmental and Marine Biology
Department of Biosciences
Åbo Akademi University
Åbo, Finland, 2013

Supervisors

Docent Sami Merilaita
Environmental and Marine Biology
Åbo Akademi University
Biocity, Artillerigatan 6
FI-20520 Åbo, FINLAND

Professor Birgitta S Tullberg
Department of Zoology
Stockholm University
SE-106 91 Stockholm, SWEDEN

Reviewers

Dr Mike Speed
Institute of Integrative Biology, Biosciences Building
University of Liverpool
Crown Street, Liverpool L69 7ZB, UNITED KINGDOM

Docent Carita Lindstedt-Kareksela
Department of Biological and Environmental Science
Centre of Excellence in Biological Interactions
P.O. Box 35
FI-40014 University of Jyväskylä, FINLAND

Opponent

Dr Candy Rowe
Institute of Neuroscience
Newcastle University
Framlington Place
Newcastle upon Tyne
NE2 4HH, UNITED KINGDOM

Author's address

Environmental and Marine Biology
Åbo Akademi University
Biocity, Artillerigatan 6
FI-20520 Åbo, FINLAND
e-mail: Titti.Bohlin@abo.fi

ISBN 978-952-12-2947-3
Painosalama Oy – Turku, Finland 2013

“I always felt that insects are the general rule,
and everything else is a special case.”

Paul Bystrak

Abstract

In nature, many animals use body coloration to communicate with each other. For example, colorations can be used as signals between individuals of the same species, but also to recognise individuals of other species, and if they may comprise a threat or not. Many animals use protective coloration to avoid predation. The two most common strategies of protective coloration are camouflage and aposematism. Camouflaged animals have coloration that minimises detection, usually by matching colours or structures in the background. Aposematic animals, on the other hand, signal to predators that they are defended. The defence can be physical structures, such as spikes and hairs, or chemical compounds that make the animal distasteful or even deadly toxic. In order for the warning signal to be effective, the predator has to recognise it as such. Studies have shown that birds for example, that are important visual predators on insects, learn to recognise and avoid unpalatable prey faster if they contrast the background or have large internal contrasts. Typical examples of aposematic species have conspicuous colours like yellow, orange or red, often in combination with black.

My thesis focuses on the appearance and function of aposematic colour patterns. Even though researchers have studied aposematism for over a century, there is still a lot we do not know about the phenomenon. For example, as it is crucial that the predators recognise a warning signal, aposematic colorations should assumingly evolve homogeneously and be selected for maximal conspicuousness. Instead, there is an extensive variation of colours and patterns among warning colorations, and it is not uncommon to find typical cryptic colours, such as green and brown in aposematic colour patterns. One hypothesis to this variation is that an aposematic coloration does not have to be maximally signalling in order to be effective, instead it is sufficient to have distinct features that can be easily distinguished from edible prey. To be maximally conspicuous is *one* way to achieve this, but not the only way. Another hypothesis is that aposematic prey that do not exhibit maximal conspicuousness can exploit both camouflage and aposematism in a distance-dependent fashion, by being signalling when seen close up but camouflaged at a distance. Many prey animals also make use of both strategies by shifting colour at different ecological conditions such as seasonal variations, fluctuations in food resources or between life stages. Yet another explanation for the variation may be that prey animals are usually exposed to several predator species that vary in visual perception and tolerance towards various toxins.

The aim with this thesis is, by studying their functions, to understand why aposematic warning signals vary in appearance, specifically in the level of conspicuousness, and if warning coloration can be combined with camouflage.

In paper **I**, I investigated if the colour pattern of the aposematic larva of the Apollo butterfly (*Parnassius apollo*) can switch function with viewing distance, and be signalling at close range but camouflaged at a distance, by comparing detection time between different colour variants and distances. The results show that the natural coloration has a dual distance-dependent function. Moreover, the study shows that an aposematic coloration does not have to be selected for maximal conspicuousness. A prey animal can optimise its coloration primarily by avoiding detection, but also by investing in a secondary defence, which presence can be signalled if detected.

In paper **II**, I studied how easily detected the coloration of the firebug (*Pyrrhocoris apterus*), a typical aposematic species, is at different distances against different natural backgrounds, by comparing detection time between different colour variants. Here, I found no distance-dependent switch in function. Instead, the results show that the coloration of the firebug is selected for maximal conspicuousness. One explanation for this is that the firebug is more mobile than the butterfly larva in study **I**, and movement is often incompatible with efficient camouflage.

In paper **III**, I investigated if a seasonal related colour change in the chemically defended striated shieldbug (*Graphosoma lineatum*) is an adaptation to optimise a protective coloration by shifting from camouflage to aposematism between two seasons. The results confirm the hypothesis that the coloration expressed in the late summer has a camouflage function, blending in with the background. Further, I investigated if the internal pattern as such increased the effectiveness of the camouflage. Again, the results are in accordance with the hypothesis, as the patterned coloration was more difficult to detect than colorations lacking an internal pattern. This study shows how an aposematic species can optimise its defence by shifting from camouflage to aposematism, but in a different fashion than studied in paper **I**.

The aim with study **IV** was to study the selection on aposematic signals by identifying characteristics that are common for colorations of aposematic species, and that distinguish them from colorations of other species. I compared contrast, pattern element size and colour proportion between a group of defended species and a group of undefended species. In contrast to my prediction, the results show no significant differences between the two groups in any of the analyses. One explanation for the non-significant results could be that there are no universal characteristics common for aposematic species. Instead, the selection pressures acting on defended species vary, and therefore affect their appearance differently. Another explanation is that all defended species may not have been selected for a conspicuous aposematic warning coloration.

Taken together, my thesis shows that having a conspicuous warning coloration is not the only way to be aposematic. Also, aposematism and camouflage is not two mutually exclusive opposites, as there are prey species that exploit both strategies. It is also important to understand that prey animals are exposed to various selection pressures and trade-offs that affect their appearance, and determines what an optimal coloration is for each species or environment. In conclusion, I hold that the variation among warning colorations is larger and coloration properties that have been considered as archetypically aposematic may not be as widespread and representative as previously assumed.

Sammanfattning

I naturen använder sig många djur av färgteckningar för att kommunicera med varandra. Färgteckningar används exempelvis som signaler mellan individer inom samma art, men också för att känna igen andra arter och om de utgör ett möjligt hot eller ej. Många djur använder sig av skyddsfärger för att undvika att bli upptäckta. De två vanligaste strategierna inom skyddsfärg är kamouflage och aposematism. Kamouflerade djur har färgteckningar som minimerar deras upptäckt, ofta genom att smälta in i bakgrundens färger och strukturer. Djur som är aposematiska däremot signalerar till predatorer att de har någon form av försvar och därför är oätliga. Försvaret kan bestå av fysiska strukturer som taggar eller nässelhår, eller av kemiska ämnen som gör dem osmakliga eller rentav dödligt giftiga. För att en varningfärg ska vara effektiv måste predatorn känna igen den som just en sådan. Studier har visat att till exempel fåglar, som är viktiga visuella predatorer på insekter, fortare lär sig att känna igen och undvika ett oätligt byte om det kontrasterar starkt mot bakgrunden eller har stora internt kontrasterande fält. Typiska exempel av aposematiska arter har iögonfallande färgkombinationer med till exempel gult, orange eller rött i kombination med svart. Min avhandling fokuserar på utseende och funktionen av aposematiska färgteckningar.

Trots att forskare har studerat aposematism i över ett sekel, så är det fortfarande mycket man inte har fått klarhet i. Till exempel, eftersom det är viktigt att predatorer känner igen en varningssignal så borde aposematiska färgteckningar utvecklas ganska likartat och selekteras för att vara maximalt iögonfallande, men istället finns det en enorm variation i mönster och färg. Många aposematiska färgteckningar innehåller också grönt och brunt, som anses vara typiska kamouflagefärger. En hypotes till denna variation är att en aposematisk färgteckning inte alltid behöver vara maximalt iögonfallande för att vara effektiv, utan det räcker med att ha distinkta särdrag som lätt kan urskiljas från ätliga byten. Att vara maximalt iögonfallande är *ett* sätt att uppnå detta, men inte en nödvändighet. En annan hypotes är att aposematiska bytesdjur som inte är maximalt iögonfallande kan utnyttja både kamouflage och aposematism genom att till exempel vara signalerande på nära håll men kamouflerad på avstånd. Många bytesdjur utnyttjar också båda strategierna genom att skifta färg vid olika ekologiska förhållanden, till exempel mellan årstider, vid skiftningar i födotillgång eller mellan olika livscykelstadier. Variationen kan också förklaras med att byten ofta exponeras för flera olika arter av predatorer som skiljer sig i visuell perception och i tolerans mot olika försvar.

Syftet med den här avhandlingen är att genom att studera dess funktion, förstå varför aposematiska varningssignaler varierar i utseende, speciellt i graden av iögonfallande och om varningssignaler kan kombineras med kamouflage.

I studie I undersökte jag om färgteckningen hos den aposematiska apollofjärilslarven (*Parnassius apollo*) skiftar i funktion med avståndet och är signalerande på nära håll men kamouflerad på längre avstånd, genom att jämföra tiden till upptäckt mellan olika färgvarianter och avstånd. Resultaten visar att den naturliga färgteckningen har en avståndsberoende dubbel funktion. Studien visar också att en aposematisk färgteckning inte behöver vara selekterad för att vara maximalt iögonfallande. Ett bytesdjur kan alltså optimera sin färgteckning genom att i första hand undgå upptäckt men också investera i ett försvar som kan signaleras vid upptäckt.

I studie II studerade jag hur lättupptäckt färgteckningen på eldlusen (*Pyrrhocoris apterus*), en typisk aposematisk art, är vid olika avstånd och mot olika naturliga bakgrunder genom att jämföra upptäckstid mellan olika färgvarianter. Resultaten visar inte på någon avstånds-beroende dubbel funktion i det här fallet, utan istället verkar eldlusens färgteckning vara selekterad för maximalt iögonfallande. Det kan bero på att eldlusen är rörligare än fjärilslarven i studie I, och rörelse är ofta oförenligt med ett effektivt kamouflage.

I studie III undersökte jag om säsongrelaterad färgväxling hos den kemiskt försvarade strimlusen (*Graphosoma lineatum*) kan vara en anpassning till att optimera sin skyddsfärg genom att skifta från kamouflage till aposematism mellan två säsonger. Resultaten bekräftar hypotesen att färgteckningen som uttrycks på sensommaren har en kamouflerande funktion då den smälter väl in i bakgrunden. Vidare undersökte jag om själva mönstret i färgteckningen ytterligare förstärker kamouflaget. Återigen bekräftar resultaten hypotesen, då färgteckningen med internt mönster var svårare att upptäcka än enfärgade färgteckningar utan mönster. Den här studien visar hur en art kan optimera sitt försvar genom att skifta från kamouflage till aposematism under sin livscykel.

Syftet med studie IV var att studera selektionen på aposematiska signaler genom att försöka identifiera karaktärer i färgteckningen hos aposematiska arter som är gemensamma för dessa och som skiljer dem från andra arter. Jag jämförde kontrast, komponentstorlek och färgproportioner mellan en grupp insekter med försvar och en grupp insekter utan försvar. I motsats till förväntningen så visar resultaten inte på några signifikanta skillnader mellan de två grupperna i någon av analyserna. Resultaten kan bero på att det inte finns några universella karaktärer som är gemensamma för aposematiska signaler, utan att selektionen som försvarade arter utsätts för varierar, och därmed påverkar deras utseende olika. Det kan också betyda att alla arter med ett försvar inte har selekterats för en starkt iögonfallande varningsfärg.

Sammantaget så visar min avhandling att en starkt iögonfallande varningsfärg inte är det enda sättet att signalera aposematism på. Aposematism och kamouflage är inte

heller två oförenliga motsatser då det finns arter som utnyttjar båda strategierna. Det är också viktigt att förstå att bytesdjur utsätts för många olika selektionstryck och avvägningar som påverkar utseendet och avgör vad som är den optimala färgteckningen för varje enskild art eller miljö. Min avhandling visar också fördelar med att använda bildbaserade studier och analyser för att studera vissa aspekter av skyddsfärger som är svårt att göra med levande byten och predatorer. Slutligen vidhåller jag att variationen inom varningsfärger är större och att de egenskaper hos färgteckningar som har ansetts vara arketypiskt aposematiska kanske inte är så utbredda och representativa som tidigare antagits.

Table of contents

List of original papers.....	2
1. INTRODUCTION.....	3
1.1. Aposematism.....	3
1.2. The evolution of aposematism.....	4
1.3. Distinctiveness or conspicuousness.....	5
1.4. Trade-offs in aposematic coloration.....	5
1.5. Distance-dependent function.....	6
1.6. Seasonal polyphenism.....	7
1.7. Aims of the thesis.....	7
2. METHODS.....	8
2.1. General approach.....	8
2.2. Study species.....	9
2.2.1. <i>Parnassius apollo</i>	9
2.2.2. <i>Pyrrhocoris apterus</i>	9
2.2.3. <i>Graphosoma lineatum</i>	10
2.2.4. Selection of defended and undefended species.....	11
2.3. Photography.....	12
2.4. Image manipulation.....	13
2.5. Photograph treatment.....	13
2.6. Detection experiments.....	15
2.7. Human predators.....	15
2.8. Image analyses.....	16
3. STATISTICAL ANALYSES.....	17
4. RESULTS AND DISCUSSION.....	18
4.1. Paper I.....	18
4.2. Paper II.....	19
4.3. Paper III.....	20
4.4. Paper IV.....	22
5. CONCLUSIONS.....	23
Acknowledgements.....	26
References.....	28
Original papers (I-IV).....	33

List of original papers

The thesis is based on the following papers, which are referred to by their Roman numerals in the text

- I** **Bohlin T**, Tullberg BS & Merilaita S (2008) The effect of signal appearance and distance on detection risk in an aposematic butterfly larva (*Parnassius apollo*). *Animal Behaviour* 76: 577-58

- II** **Bohlin T**, Gamberale-Stille G, Merilaita S, Exnerová A, Štys P & Tullberg BS (2012) The detectability of the colour pattern in the aposematic firebug, *Pyrrhocoris apterus*: an image-based experiment with human ‘predators’. *Biological Journal of Linnean Society* 105: 806-81

- III** Tullberg BS, Gamberale-Stille G, **Bohlin T** & Merilaita S (2008) Seasonal ontogenetic colour plasticity in the adult striated shieldbug *Graphosoma lineatum* (Heteroptera) and its effect on detectability. *Behavioral Ecology and Sociobiology* 62: 1389-1396

- IV** **Bohlin T** & Merilaita S (2013) What is an aposematic signal? A comparative study on colour pattern in defended and undefended insects. *Manuscript*.

The original papers have been reprinted with the kind permission of Elsevier (paper **I**), Linnean Society of London (paper **II**), and Springer (paper **III**).

1. INTRODUCTION

1.1. Aposematism

In nature, many animals use coloration to communicate with each other. Colour pattern can be involved in various functions, such as recognition of conspecifics, attraction of potential partners, regulation of body temperature, protection from UV radiation and recognition of other species as 'friend or foe' (Cott 1940; Schaefer 2010). Prey animals constantly have to avoid predation, and as many predators are visually oriented hunters, body coloration is commonly used for protective purpose. The two most widespread strategies of protective coloration are camouflage that prey use to avoid being detected or recognised by predators, usually by blending into the background, and warning or aposematic coloration, in which prey signal to potential predators that they are defended (Cott 1940; Ruxton et al. 2004).

Already the early evolutionary biologists were intrigued by the appearance and function of animal coloration. Charles Darwin hypothesised that bright, conspicuous colour patterns and ornamentations found in many species were the result of sexual selection, and thus adaptations that increase an individual's success in mating competition (Darwin 1859). However, he was puzzled by the occurrence of spectacular and bright coloration in many butterfly larvae, as larvae are sexually immature, and therefore their coloration is obviously not related to mating. He shared his dilemma in a letter to his fellow naturalist Alfred Russell Wallace who suggested that in some species conspicuous coloration may have a warning function, such that they are used by defended prey animals to signal their unprofitability to potential predators:

"...protected by a disagreeable taste or odour, it would be a positive advantage to them never to be mistaken for any of the palatable caterpillars (...) Any gaudy & conspicuous colour therefore, that would plainly distinguish them from the brown & green eatable caterpillars, would enable birds to recognise them easily as a kind not fit for food..."

(Wallace 1867)

The phenomenon was later coined aposematism (greek: apo= away, sema= sign) by Poulton (1890), and refers to prey that combine a warning signal with a secondary defence. The secondary defence can be a chemical, such as a toxic or noxious compound, a distasteful or nauseously smelling compound, or physical, such as spines or irritating hairs (Cott 1940; Edmunds 1974).

1.2 The evolution of aposematism

The evolutionary origin of aposematic coloration has intrigued scientists over the years as it seems to involve a paradox: The first conspicuously signalling mutant ought to suffer increased predation risk as it is easy to spot, and at the same time it cannot gain any extra protection from the signal as the predators lack any previous experience, and do not associate the signal with the defence. Therefore the conspicuous mutants should have been likely to become extinct before their genes could have spread in a population (Ruxton et al. 2004). On the other hand, if the defence evolved prior to the signal, predators would not have been able to distinguish the first defended mutants from palatable prey, and the defence should have disappeared due to the extra cost it involved. However, aposematism is a widespread phenomenon, indicating that there must be solutions to the apparent paradox. Nowadays there are several hypotheses explaining, at least partially, how aposematism could arise. For example, aposematic butterfly larvae can very well survive a bird attack because the bird can reject a larva before killing it due to its distastefulness (Järvi et al. 1981; Wiklund & Järvi 1982). Thus, an increased attack rate on rare defended mutants does not necessarily lead to their extinction before the predators have learnt to associate the defence with the warning signal. In addition, if there is already a high opportunity cost involved in a cryptic strategy of a species, then there may be only low, if any, elevated cost involved in the evolution of an aposematic strategy. An exceedingly high cost of crypsis can occur in visually heterogeneous habitats where highly efficient camouflage can be impossible to achieve or for highly active prey, as movement typically constrains efficient camouflage (Merilaita & Tullberg 2005). Also, to develop an aposematic strategy in an environment that constraint crypsis allows an increase in environmental exploitation (Speed & Ruxton 2005a). Moreover, many prey species have visible defences, such as spines or hair which may facilitate avoidance learning without the need of a predator tasting the prey (Speed & Ruxton 2005b).

Conspicuousness seems to be a universal trait in aposematic coloration, and it has often been assumed that aposematic signals are selected for maximal conspicuousness to warn off predators more efficiently. For example, it has been shown that the larger a warning signal is (Forsman & Merilaita 1999), or the more contrasting a prey is to the background (Gittleman & Harvey 1980), the more rapidly the predators learn to avoid distasteful prey. Accordingly, the aversion towards warning coloration may be a response that predators have learnt due to their encounters with conspicuous prey that have had aversive qualities, but it may also be an innate response, present even in naïve predators towards certain colours. In addition, conspicuous defended prey not only involve a more rapid

avoidance learning rate in predators than cryptic defended prey do, strong signals also facilitate recognition (Guilford 1986; Osorio et al. 1999) and prevent predators from soon forgetting about the negative experience (Kraemer 1984; Roper & Redston 1987). Hence, an innate or a quickly learnt aversion towards prey with a certain colour or defence can enable such traits to evolve and increase in frequency in a population. Taken together, all these factors mentioned above may have been fundamental for the initial evolution of aposematic coloration.

1.3 Distinctiveness or conspicuousness

Even though many aposematic prey may be maximally (or at least highly) conspicuous, it seems that this is not always the case. Instead, if one scrutinises and compares different aposematic species, they seem to exhibit varying levels of conspicuousness (Darst et al. 2006), and many times typical aposematic colours can be found in combination with typical cryptic colours (e.g. Endler & Mappes 2004). As a matter of fact, when Wallace (1889) proposed how warning coloration works, he suggested that the key feature of a warning signal is that it makes the defended prey easy to distinguish from undefended, typically cryptic prey species. Although conspicuousness provides one way to achieve distinctiveness, it is also possible to think of other ways that do not require maximisation of conspicuousness. For example, in a field experiment, Wüster et al. (2004) demonstrated how the assumingly cryptic but distinct zig-zag markings on the European viper (*Vipera berus*) had an aversive effect on natural predators that avoided plasticine models of vipers regardless of detectability against the background. An aposematic coloration may also be distinct but less than maximally signalling as prey species often are exposed to several predator species varying in toxic tolerance, behaviour and visual perception (Endler 1988; Exnerová et al. 2003; Endler & Mappes 2004). Taken together, this implies that an increase in conspicuousness does not always translate to a corresponding decrease in the risk of being attacked in aposematic prey. This opens several interesting questions regarding the evolution and appearance of warning signals.

1.4 Trade-offs in aposematic coloration

It is important to remember that animal coloration holds several other functions beyond predator-prey interactions. For example, two highly common pigments in animals, carotenoids (Clotfelter et al. 2007) and melanin (Wilson et al. 2001), play an important part in immune defence. Melanin is also essential in thermoregulation for many animals, even if melanism may decrease the warning signal efficiency (Lindstedt et al. 2009; Hegna et al. 2013). There may also be physiological constraints in the production of certain characteristics or pigments.

For example, animals cannot synthesise carotenoids *de novo* and therefore have to sequester them from their food. Prey animals only have a limited energy resource and therefore they may have to allocate energy from other important functions to maintain aposematism (e. g. Srygley 2004; Ohsaki 2005). Hence, an optimal warning coloration may be influenced by trade-offs with other activities not related to predation.

1.5 Distance-dependent function

Many aposematic colour patterns involve bright colours such as red, orange and yellow, often in combination with black, whereas drab colours like brown, green or grey have been thought to be typical cryptic colours (Poulton 1890; Edmunds 1974). Conventionally, aposematic and cryptic coloration have been considered as the two opposite extremes of the conspicuousness continuum (Poulton 1890; Cott 1940), and therefore they also have been considered as mutually exclusive strategies. Accordingly, as pointed out above, aposematic signals have been expected to be selected for maximal conspicuousness to effectively deter predators and by that benefit from the increased detection risk it pertains. The idea that aposematism and camouflage would always be mutually exclusive has been questioned. For instance, it has been suggested that a colour pattern can have a combined function of the two apparently opposite traits. That is, an aposematic colour pattern may involve a distance-dependent dual function, by having a warning function at a close range but being cryptic when further away from the viewer (Papageorgis 1975; Tullberg et al. 2005). All objects are more difficult to detect at a distance due to the limited resolution of the eye, but if there is a

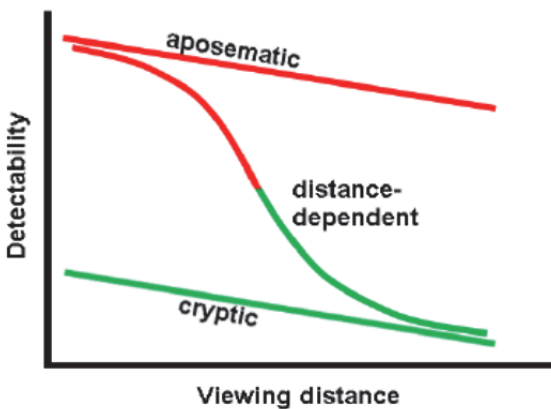


Fig 1. A schematic illustration how the decrease in detectability is much more pronounced for an aposematic coloration with a distance-dependent dual function compared to a purely aposematic or cryptic coloration.

distance-dependent switch in the function of a coloration, the change in the function of a coloration, the change in detectability with increasing distance would be much more pronounced than for a coloration that has purely a warning or a concealing function (Fig. 1).

1.6 Seasonal polyphenism

Interestingly, it is not unusual to find aposematism and camouflage at different life-stages of the same species (Booth 1990). For instance, many butterfly species are aposematic as larvae and adults but cryptic in the pupal stage (Wiklund & Sillén-Tullberg 1985), and some butterfly species even change strategy between different larval instars (Nylin et al. 2001; Grant 2007). Such ontogenetic colour change may be related to changes in behaviour of the prey species itself, as for many butterflies that have active larvae and adults, and immobile and more vulnerable pupae. It can also be a response to shifts in external conditions such as seasonal variations that can involve a switch in the predator community composition. For example, in temperate areas there may be more naïve and inexperienced predators later in the season that have not yet learnt which prey to avoid, so therefore a cryptic strategy may be more advantageous. Seasonal changes in the environment can also alter the costs and benefits of acquiring aposematism or camouflage, such as changes in the visual properties of the background the prey species are seen against (Tullberg et al. 2008; Johansen et al. 2010).

1.7 Aims of the thesis

My general aim with this thesis is to better understand how warning signals vary in their degree of conspicuousness, and if there may be different functions involved. My ultimate aim is to increase the understanding of the diversity in the appearance of warning coloration. To achieve these aims, I investigate how different colour patterns can affect the detectability of aposematic species and whether the colour patterns of the aposematic insects I have studied have all been selected for maximal signalling aposematism or if they vary in signal strength.

Although the evolution and function of aposematism have gained much attention during the last decades, there are still several important questions remaining unanswered. For instance, because there appears to be reason to question the ‘classical’ view of aposematism, according to which all aposematic species should be selected for maximised conspicuousness, optimisation of the level of conspicuousness and the appearance of warning signals is now an important topic. Even so, most studies on aposematism still focus on species possessing colours that have subjectively been classified as aposematic. Aposematic signals have traditionally been described as consisting of at least one of the typical

warning colours red, orange or yellow, often together with black. Yet, there is a large variation in appearance among aposematic signals, and many aposematic colour patterns seem to exhibit only intermediate levels of signalling. In this thesis I specifically focus on the following questions:

- 1) Can an aposematic colour pattern involve a distance-dependent switch in function, being cryptic from a distance but signalling when seen close up?
- 2) How do the spatial distribution of pattern elements affect conspicuousness of an aposematic pattern, and how important is an internal pattern as such?
- 3) Can colour polyphenism in an aposematic species be an adaptation to also benefit from a cryptic strategy?
- 4) Are there any characteristics in prey colour patterns that can be considered to be typically aposematic and that distinguish them from other prey species? If so, is it possible to identify these characteristics?

2. METHODS

2.1 General approach

In the four studies included in this thesis, I have used a digital image-based approach when investigating different aspects of aposematic coloration in insects. The use of digital photographs allows controlling for factors such as light, distance, background and predation (paper I, II & III), not feasible in the field with natural predators such as birds that are an important group of natural predators on insects. Additionally, this technique also enables me to manipulate prey coloration, and compare different colours and patterns while keeping other parameters unchanged (paper I, II, III & IV). In the studies where I compared detectability of different colour patterns against natural backgrounds, I used humans as ‘predators’ (paper I, II & III). Many studies on protective coloration have adopted this approach, and one advantage of using humans as ‘predators’ is that they are easier to instruct than natural predators. Additionally, a problem with natural predators is that they may not attack immediately after detecting the prey, or even avoid the prey altogether.

2.2 Study species

2.2.1 *Parnassius apollo* (paper I)

The Apollo butterfly, *Parnassius apollo*, is a member of the swallowtail family, Papilionidae. This group is easily recognised by its bright, assumingly aposematic coloration, and include some of the largest butterflies (Resh & Cardé 2003). *P. apollo* has a distribution ranging from mountainous areas in Spain to Southern Fennoscandia and Eastwards to Mongolia, but the population of *P. apollo* has declined, and even become extinct in many parts of Europe. The larva is sturdy and measures up to 6 cm (Fig. 2). The early instars are velvet black with fine setae, but gradually conspicuous orange protuberances develop on both sides of each body segment. The black colour is produced by melanin, the most common pigment in butterflies, and the orange colour is most likely from carotenoids (O'Toole 2002). *Parnassius apollo* is oligophagous and the larvae feed mainly on different species of *Sedum*. In Sweden *Sedum telephium*, but also *Sedum album* is a typical host plant (Nakonieczny & Kedzioriski 2005). Many *Sedum* species contain alkaloids, a group of chemical compounds that are commonly sequestered and used as a secondary defence by many insect species. However, whether *P. apollo* sequesters any substances from the host plants making it noxious is not clear, but in trials where larvae were presented to chicks, the chicks found them distasteful and avoided them (Gamberale-Stille & Tullberg, unpublished data).



Fig 2. The black and orange last instar larva of the Apollo butterfly (*Parnassius apollo*). (Photo: Birgitta S Tullberg)

2.2.2 *Pyrrhocoris apterus* (paper II)

The firebug, *Pyrrhocoris apterus* (L., 1758) (Hemiptera; Heteroptera: Pyrrhocoridae), is a ground-living and widely distributed Palearctic true bug, and is one of the few species found all the way to Siberia (Fig. 3). The red and black firebug is phytophagous and feeds primarily on seeds from mallows (Malvaceae) and linden (Tiliaceae; Socha 1993), and in Europe it can be seen in aggregations at the feet of linden trees. The red and black colour is made up by pteridines and

melanin, respectively. There are also occasional colour mutants varying from white to orange depending on the pteridine content, but with the same black pattern (Exnerová et al. 2006). The firebug also exhibit two different wing morphs, one long-winged (macropterous) and one with short wings (brachypterous), with the latter being the most common morph. However, neither wing morph is capable of flying (Socha & Zemek 2000). The firebug, as many other heteropteran species, are considered to be aposematic both as larvae and adults and combine a conspicuous red and black colour pattern with a chemical defence involving over 40 identified compounds (Farine et al. 1992). Studies have also shown that birds reject firebugs when presented to them (Exnerová et al. 2003, 2006). (For an extensive summary of the biology of *P. apterus*, see Socha (1993)).



Fig 3. The aposematic firebug (*Pyrrhocoris apterus*) with its distinct red and black colour pattern. (Photo: Alice Exnerová)

2.2.3 *Graphosoma lineatum* (paper III)

The striated shieldbug *Graphosoma lineatum* (L.1758) (Heteroptera: Pentatomidae) is a phytophagous true bug widespread throughout Europe, in Western Asia and Northern Africa. In Sweden it is usually found on host plants of the Apiaceae family, such as cow parsley (*Anthriscus sylvestris*), and also ground elder (*Aegopodium podagraria*). In Central and Southern Europe, the striated red and black shieldbug can exhibit some variations in the red pigment between individuals, from pale yellow to bright red. However, in Sweden this variation is reported to be linked to an ontogenetic colour change over the seasons within individuals. The newly moulted adult in late summer is striated in beige and black (Fig. 4), but after hibernation in the undergrowth during winter, the reproductively mature adult emerges in May and has changed to a red and black striation (Fig. 4). The beige striated phenotype appears in late summer in a background of dried umbels and stems of its host plants, whereas the red striated phenotype is usually found against a green and lush background in early summer. The black pigment is melanin and the red pigment has not been identified, but pteridines are involved in several other heteropteran species (Tullberg et al. 2008).



Fig 4. The striated shieldbug (*Graphosoma lineatum*). The pre-hibernation beige and black morph seen against the dry late summer background (left), and post-hibernation red and black morph seen against the lush early summer background (right). (Photo: Birgitta S Tullberg (left), Aleksandra I Johansen (right)).

Both colour morphs of *G. lineatum* are chemically defended (Šanda et al. 2012) and are known to be highly distasteful to birds that quickly learn to avoid them in trials (Veselý et al. 2006; Gamberale-Stille et al. 2010). As most shieldbugs, they have scent glands with volatile secretion stored, which they can readily release when attacked (Pasteels et al. 1983), hence the common name ‘stink bugs’ for the Pentatomidae family.

2.2.4 Selection of defended and undefended insect species (paper IV)

The study included 40 insect species from three different insect orders; Lepidoptera, Coleoptera and Hemiptera (Table 1). The insect species were divided in two groups; 20 species were defended and 20 species were undefended. Among insects it is not unusual to find extremely camouflaged species and highly conspicuous species in the same taxon. However, the specific characteristics of colour pattern that distinguishes the two strategies have never been objectively identified. Here, I compared internal contrasts, mark sizes and colour proportions of colour patterns between the two groups to examine if there are any general visual characteristics that distinguish the two groups from each other. The species for each group were selected by strict criteria. I selected species that have been reported in scientific literature to be either rejected or accepted by natural predators in the field. As insects rejected by predators usually involve a secondary defence, I refer to the two groups as ‘defended’ and ‘undefended’. However, information about predation in the wild is quite sparse, so in order to increase the number of limited species, I also used information from scientific literature on the presence of chemical compounds in some species, well known to be used as a defence against predators (Table 1, paper IV).

Unpalatable	Palatable
<u>Lepidoptera</u>	<u>Lepidoptera</u>
<u>(Nymphalidae)</u>	<u>(Nymphalidae)</u>
Euphydryas phaeton	Junonia coenia
Heliconius erato	Apoforneuthes iulia
Panacea procilla	Agraulis vanillae
Acraea zetes	Euptoieta hegesia
<u>(Arctiidae)</u>	<u>(Arctiidae)</u>
Haploa lecontei	Pyrrharctia Isabella
Utetheisa ornatrix	Apantesis nais
Tyria jacobaea	Euchaetes egle
<u>(Noctuidae)</u>	<u>(Noctuidae)</u>
Lithacodia carneola	Acronicta morula
<u>(Geometridae)</u>	<u>(Geometridae)</u>
Xanthotype sospeta	Biston betularia
<u>(Sphingidae)</u>	<u>(Sphingidae)</u>
Acherontia atropos	Ceratomia catalpa
<u>Coleoptera</u>	<u>Coleoptera</u>
Chaliognathus pennsylvanicus	Agriotes lineatus
Cicindela flexuosa	Harpalus fraternus
Leptinotarsa decemlineata	Cassida rubiginosa
Diachromus germanus	Carabus coriaceus
Cionus hortulanus	Pissodes strobi
Naupactus bipes	Otiorhynchus sulcatus
Canthon cyaneus	Geotrupes mutator
<u>Hemiptera</u>	<u>Hemiptera</u>
Lygaeus equestris	Graptostethus servus
Cercopis sanguina	Cephus siccifolius
Murgantia histrionica	Gerris cinereus

Table 1. A list of the 40 selected insect species, 20 defended and 20 undefended, used to compare internal contrasts, marking sizes and colours between defended and undefended species (paper IV).

Importantly, the species were compared in phylogenetically controlled pairs, including one defended and one undefended species. This means that the species in each pair are closer related to each other than to other species used in the study from the same order.

2.3 Photography

The insect species were photographed in the field, against several different natural backgrounds in habitats where they are typically found (paper I, II & III). The insects were photographed from an approximately 45° angle so that the photograph included a clear view of the dorsal side of the insect and the rest of the image covered with the natural background. Each insect was also photographed at different distances between 20 cm to 100 cm, to simulate different viewing distances (paper I & II). The series of photographs used in the studies were selected on the basis of their high quality, and for the similarity in insect size as several parallel series of

photographs were compared. I also selected a suite of photographs with several distances as I tested the effect of viewing distance on detectability (paper I & II). The distances ranged from close to fairly long, but within the scope of making detection possible for the human subjects. All insect species in paper IV belonged to the collection of the Natural History Museum in Stockholm and were photographed there. The species were photographed with the dorsal side up at a fixed distance (47 cm). In the beginning of every photography session I first took a picture of a colour control card. The settings of the camera were fixed, but the card allowed me to control for different light conditions between sessions.

2.4 Image manipulation

To test various hypotheses in paper I, II & III, I needed to compare the natural coloration of the insect species with alternative coloration phenotypes. To create such alternative variants from the naturally coloured species, I manipulated images in image-editing software Adobe PhotoShop 7.0 (paper I; Adobe Systems Inc., San Jose, CA, U.S.A.) and the free image processing software GIMP 2.2 & 2.8 (paper II & III; GNU Free Software, Boston). For ecological relevance, only colours present on the natural insects were used. Pixels of the colour of interest were copied and used to cover the areas with the colour I wanted removed. For example, to create a plain black insect, the areas with the other colour were covered with pixels copied from the adjacent black areas, and vice versa. To compare insect sizes in the images (paper I, II & III), and to ensure that the proportions of the colours included in the manipulated bicoloured insects were same as in the naturally coloured insect (paper I & II), the insects and the colour areas of interest were measured using ImageJ 1.37, a free image processing and analysis program (Rasband, 1997–2012). Before the colour patterns of the 40 species were analysed (paper IV), I erased the background in every image and replaced it with a separate transparent layer using GIMP. This was done so that only the insects were analysed without the risk of that the background properties would confound the results. Moreover, some of the specimens were fixed with pins (paper IV), which were removed in GIMP by copying pixels from the surrounding areas and pasting them to replace the pinhead.

2.5 Photograph treatment

I conducted two consecutive experiments, in which I investigated how colour pattern, viewing distance and background affect detectability of the black and orange Apollo butterfly larva (paper I). This was done by comparing photographs of the naturally coloured larva with manipulated colour variants. In experiment 1,

a total of 12 images were presented to human subjects. Here, I used four photographs taken of the same larva at four different distances, against one background. In order to test how the natural colour pattern affects detectability, this series of four photographs were then manipulated to create two more series, one with a uniform orange larva and one with a uniform black larva at the four distances. In experiment 2 a total of 15 images were presented to human subjects. Here, I used one photograph of a larva (taken in another natural background than in experiment 1), and this photograph was then manipulated to create two alternative uniform colour variants (as in experiment 1) and also two bicoloured variants, one with the orange spots smaller than on the natural larva and one with the spots larger than on the natural larva. To simulate three viewing distances in total, I made two copies of the final series of five images, enlarged to 150% and 200%, respectively.

In paper II I investigated how detectability of the natural red and black firebug is affected by viewing distance and background compared with manipulated colour variants. Here, the subjects were presented with a total of 32 images. Four different series were created by photographing four firebugs, each against a different natural background. In each series I also created manipulated colour variants: one uniform red, one uniform black, and one bicoloured variant where the black markings were moved to the outline of the bugs but the colour proportions being unaltered. Here, I simulated two viewing distances by copying the final series with the 16 images and enlarging it to 200%.

In paper III, I conducted two consecutive experiments in which I examined the possibility that the striated shieldbug may use first camouflage and then warning coloration as it undergoes ontogenetic colour change, and if the striation as such improves a probable camouflage strategy by having a disruptive effect. For both experiments I selected two photographs of bugs with beige and black striation (referred to as 'pale striated'), similar in size and light conditions, photographed in August at two different distances. Second, I selected two comparable photographs of bugs with red and black striation (referred to as 'red striated') taken in June. When manipulating the images, pixels from the red striated bugs were pasted on the pale striated bugs in the two originally selected photographs, to create different colour variants. Hence, in experiment 1 four manipulated images were presented; two images of pale striated bugs against their natural background in August, and two images of red striated bugs against the same August background. In experiment 2, again four manipulated images were presented: one pale striated bug against the August background, and three colour variants lacking the stripes altogether. By using the two colours in the pale striated bug, we created one

uniform pale variant, one uniform black variant, and one brown variant in which the two colours were mixed.

2.6 The detection experiments

The images in paper I, II & III were presented on a 17" touch-screen (NEC AccuSync LCD 52 VM, NEC Display Solutions, Japan) to volunteered human subjects acting as 'predators'. The image presentation was conducted in a semi-dark room to avoid reflections on the screen. One volunteer was tested at the time. The subject received oral and written instructions before the experiment. The 'predator' was informed about the general purpose of the experiment, but not about the specific hypothesis. The 'predator' was instructed to search for a butterfly larva (paper I) or a bug (paper II & III), and to touch it on the screen as soon as it was detected (Fig. 5). Each image was displayed until detected but no longer than the maximum time of 120 seconds. The images were presented using a purpose written program that recorded the time to detection. In order to avoid search image formation that might have confounded the detection times, each 'predator' was only presented with one image.



Fig 5. An example of an image that the human subjects were presented on the touch screen. In this image there is a red and black firebug to be found (Here marked with a red circle). (Photo: Alice Exnerová)

2.7 Human predators

Several recent studies on predation are based on human performance as predators (e. g. Beatty et al. 2004; McGuire et al. 2006; Tullberg et al. 2005; Hall et al. 2013; references within Karpestam et al. 2013). This approach allows to control for factors independently such as predator behaviour, viewing distance and detectability, which is not possible to do in the field or when using natural predators. However, it is important to recognise that humans and for example passerine birds, important predators of many insects, differ in their visual systems. Humans have trichromatic colour vision, whereas birds have tetrachromatic colour vision (Vorobyev & Osorio 1998), and the chromatic sensitivity of

passerines extends into the UV-part of the spectrum. Hence they may perceive colours and patterns differently to humans. Nevertheless, the general properties of visual information processing in vertebrates are quite similar (Vorobyev & Osorio 1998). Importantly, spectrophotometric measures have been conducted on the species used in paper I, II & III to assure that they show no reflectance in the UV part of the spectrum.

2.8 Image analyses

To quantify and compare the colour patterns between the defended and the undefended group of insect species in an objective manner (paper IV), I applied different pattern analysis techniques (e.g. Godfrey et al. 1987; Theodoridis & Koutroumbas 2008) with focus on differences in internal contrast, size of pattern elements and colour proportion between the two groups. All the analyses except for the colour proportion assessment were carried out on grayscale images. This means that each pixel in an image has one intensity value, and the analyses calculate different variations between pixel intensity values in each image.

To increase the biological relevance of my image data, the images were calibrated to correspond to how passerine birds would perceive them, using a bird vision model as described by Vorobyev & Osorio (1998). Birds constitute a large group of natural predators on insects, and as they are highly visually dependent predators (McFadden 1993) it is likely that they are an important selective factor on insect coloration. First, I conducted a texture analysis of the colour patterns of the insect species, using a grey-level co-occurrence matrix analysis (GLCM). This texture analysis describes various statistical properties of contrasts in an image, such as mean, variance and skewness (Haralick 1973; Gonzales et al. 2004; Ferguson 2007), which provides information about the overall structure in the images. Second, I wanted to investigate if there were any differences in pattern mark size between the defended and the undefended species. I achieved that by applying a method referred to as ‘granulometry’, which identifies the distribution of different grain sizes in an image. The image analyses were computed using purpose written programs in Matlab R2011a (Gonzales et al. 2004). Third, I measured the colour proportions on each species. I selected colours that are conventionally assumed to be typical aposematic colours (red, orange, yellow, white, blue, and black), and colors typically present in camouflage (green and brown), and have been used in earlier studies on aposematism (e. g. Sillén-Tullberg 1988; Tullberg & Hunter 1996; Higginson & Ruxton 2010). I marked each colour of interest on the images, taken from the control card, and marked all the pixels with the same values. By dividing the number of pixels for each colour with the

total number of pixels of the insect body, all the colour proportions were calculated. The colour proportions were measured using GIMP.

3. STATISTICAL ANALYSES

The data in paper I and III contained censored values, and I used the survival analysis in STATA 9.0 (StatacorpLP, College Station, TX, U. S. A). To test for differences in detection time between colorations and distances Cox regression models were used (paper I and III). In paper I, the log-rank test for trends was used to test for differences between distances within coloration, as well as for trends between the colour variants that had been ranked with respect to increasing amount of orange colour (i. e. signal size), ranging from only black to completely orange.

To meet the conditions for parametric tests in paper II, I applied a Box-Cox transformation on detection times. To test for the effect of colour pattern on distance and background on detection time a mixed-effects ANOVA was conducted. Colour pattern and distance were the factors of interest, and were treated as fixed factors, and the background was treated as a random factor. Further, to compare detection time between the natural colour pattern and the manipulated colour variants (paper II), Dunnett's test was used for a post-hoc comparison. All the analyses for paper II were carried out in Statistica 8.0 (StatSoft Inc.).

In paper IV dependent t-tests were used to compare the internal contrasts, in the cases where the conditions for homogeneous variances were met. In the cases where these conditions were not met, the non-parametric Wilcoxon paired test was conducted. Further, the results from the granulometry analysis were ran through a PCA (Principal Component Analysis) in which the number of variables were reduced and three principal components (variables) were identified. The PCA was succeeded by a MANOVA to find possible differences between the three variables identified. To investigate possible differences between colours, the non-parametric Wilcoxon paired test were conducted as the measurements did not meet the conditions for homogeneous variances. The analyses were conducted in RStudio (2011), except for the PCA and MANOVA that were conducted in SPSS (2012).

4. RESULTS AND DISCUSSION

4.1 Paper I

Here, I investigated if the natural bicoloured pattern of the Apollo butterfly larva has a distance-dependent function by being signalling when seen at a close distance and increasingly cryptic at longer viewing distances. I did this by comparing the change in detection time of the natural bicoloured larva with manipulated uniform black and uniform orange variants at different distances. As expected, I found that detection time for all the larvae increased significantly with viewing distance. On the other hand, detection time decreased with increasing size of the orange spots. The black larva was most difficult to detect, and even a small proportion of orange increased detectability considerably. Importantly, as the increase in detection time with increasing viewing distance was substantially larger for the natural coloration than for the orange coloration (Fig. 6), I propose that the natural Apollo butterfly larva involves a distance-dependent switch from warning

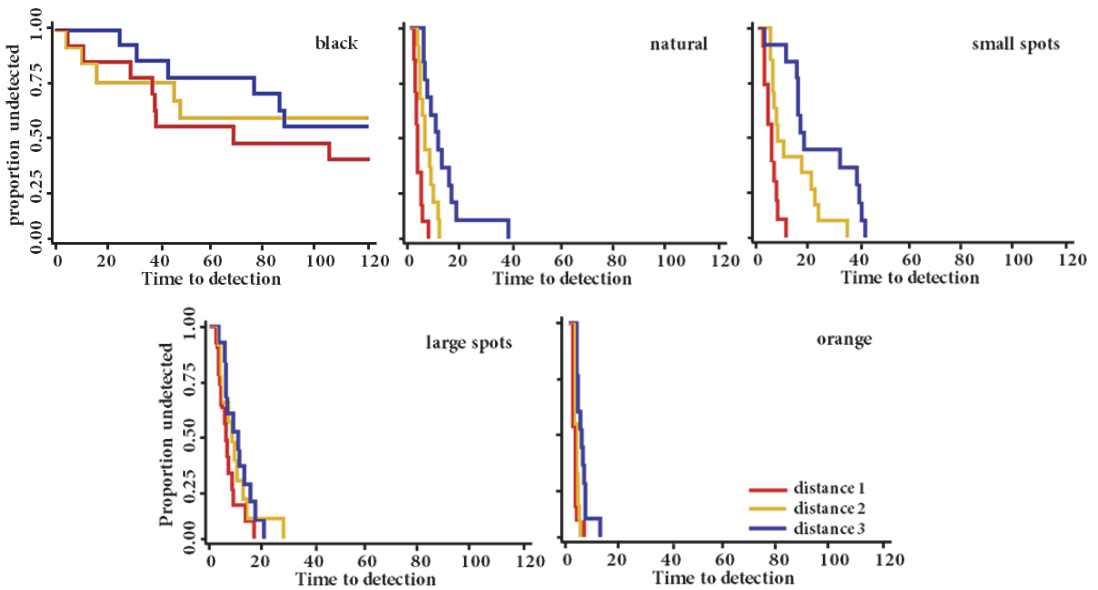


Fig 6. Detection time (s) for the natural coloration and for the four manipulations of *P. apollo* larval colorations at three distances (experiment 2, paper I). The proportion of undetected larvae illustrates the time to detection. Black ($\chi^2=1.09$, $P=0.297$), small spots ($\chi^2=23.03$, $P<0.001$), natural ($\chi^2=29.86$, $P<0.001$), large spots ($\chi^2=1.57$, $P=0.455$) and orange ($\chi^2=9.18$, $P=0.002$).

signalling to cryptic with increasing viewing distance. This dual strategy may be favourable if the cost of increased detection risk at close range is smaller than the

benefits the larva gain by being well camouflaged from a distance (Tullberg et al. 2005). The camouflage may be essential as the Apollo butterfly larva, like the larvae of many other species, are highly exposed to predation when sun-basking in the open (Stamp & Bowers 1990). Moreover, the larval stage only lasts for a few weeks and larvae may encounter naïve predators with no previous experience of unpalatable larvae (Endler & Mappes 2004). As the larva is relatively slow and soft bodied, it is vulnerable to attacks from opportunistic predators, also other invertebrates (Berenbaum et al. 1992), and therefore remaining undetected is important. By these results I have provided further insights in how an aposematic species can optimise the function of its protective coloration. By having a non-maximised warning signal, it can also make use of a cryptic strategy in a distance-dependent fashion, as suggested by Papageorgis (1975).

4.2 Paper II

Red and black is considered a typical colour combination among aposematic species and it is indeed found in several insect taxa. Here, I compared the detectability of the natural red and black firebug, *Pyrrhocoris apterus*, with digitally manipulated colour variants when seen against natural backgrounds at different viewing distances. In contrast to the results on the Apollo butterfly larva in paper I, I found that the naturally coloured firebug was significantly more conspicuous compared to the manipulated variants. Moreover, the detection time increased with distance to the same degree for all colour variants, and I found no evidence for a distance-dependent switch in the function of the natural red and black coloration of the firebug. Further, detection times of all the bugs varied between background types, and were negatively dependent on how much of the colours of the bugs were present in the background (Fig. 7). However, the natural red and black firebug was always easiest to detect, and highly conspicuous compared to the manipulated colour variants used here (Fig. 7). As the firebug is active and fast moving, a cryptic strategy that favours a less mobile lifestyle would probably not be successful (Ioannou & Krause 2009). Also, compared to the soft bodied Apollo butterfly larva in paper I, the firebug may have a hard exoskeleton and therefore better survive an attack by a predator. The results show that the detectability of the firebug varied some with background. Nevertheless, I hold that the firebug *P. apterus* is selected for high conspicuousness and that the internal pattern is an essential component. Further, even though I argue that many aposematic species may not be maximally signalling, this study presents an aposematic insect species that employ typical warning colours in an efficient warning signal.

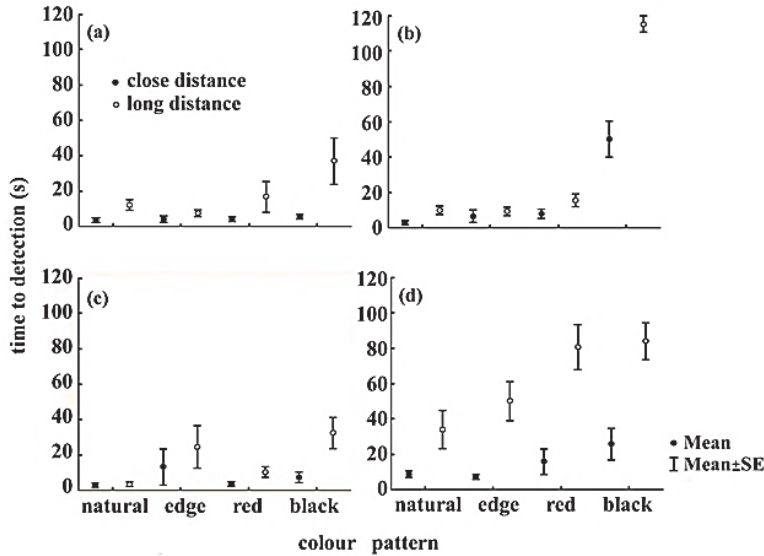


Fig 7. Mean (\pm SE) time to detection (s) of the natural and manipulated colour variants of *Pyrrhocoris apterus* against four natural backgrounds (a-d), at close distance and long distance, by human subjects in image-based experiments. (See text and Table 1 in paper II for statistical results).

4.3 Paper III

Here, I used an image-based approach to investigate if seasonal ontogenetic colour change is an adaptation in the aposematic striated shieldbug, *Graphosoma lineatum*, to also benefit from camouflage in the dry late summer environment. First, I compared the detectability of pale striated shieldbugs with red striated phenotypes in the dry late summer background to examine if the pale striated phenotype is more difficult to detect. Second, I wanted to test if the stripes of the pale striated phenotype increase a probable camouflage of the phenotype, by comparing its detectability with manipulated plain black and plain pale variants lacking the stripes altogether. The two plain variants were created by using the two colours of the natural pale striated phenotype. I found that the detection time was significantly longer for the pale striated phenotype than for the red striated phenotype in the late summer background, suggesting that the pale striation has a cryptic function (Fig. 8). The pale striated phenotype is also more cryptic than the red striated form to avian predators against late summer vegetation (Johansen et al. 2010). Further, the detection time was also significantly longer for the pale striated phenotype than for the manipulated non-striated phenotypes in the late summer background, suggesting that the stripes improve the camouflage of the pale striated phenotype. My results can be explained by changes in behaviour with

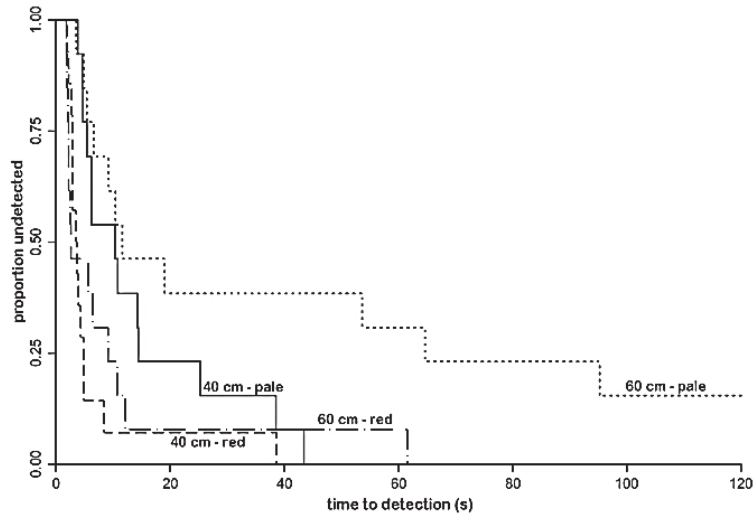


Fig 8. Detection time between the bugs in the images taken at 40 and 60 cm did not differ significantly (Cox proportional hazards model: $z = -1.71$, $p = 0.088$). The detection time was significantly longer for the pale striated bug than for the red striated bug in the 40 cm distance image (Cox proportional hazards model: $z = 2.55$, $p = 0.011$), and in the 60 cm distance image (Cox proportional hazards model: $z = 2.61$, $p = 0.009$).

development. For instance, in the spring the red striated phenotype is reproductively active, and this activity may be in conflict with a cryptic strategy. The pale striated phenotype spends more time motionless in the late summer, and a cryptic strategy may be more advantageous than an aposematic coloration that would increase the detection risk of the bug (Johansen et al. 2011). Also, the pale striated phenotype clearly benefits from its cryptic appearance against the late summer background, whereas the red striated phenotype does not match the spring environment. Another reason for the polyphenism could be that the striated shieldbug may lack the physiological ability to develop the colours needed that would allow a cryptic strategy in the lush spring environment. With these results I present another route for how an aposematic species can optimise the protective function of its body coloration. The ontogenetic colour change is most likely closely related to seasonal variations in the environment. Such seasonal plasticity allows the aposematic shieldbug to be warningly signalling in the spring and cryptic in the late summer without losing the chemical defence.

4.4 Paper IV

The main aim of study **IV** was to identify possible key characteristics that are common among aposematic species and distinguishes them from other species. I applied pattern analysis techniques on photographs of the insect species to compare possible differences in internal contrasts and marking size between the defended and the undefended group. I also compared colour proportions of colours conventionally regarded as typically aposematic and typically cryptic (Cott 1940) between the two groups. In contrast to my expectations, I found no significant differences between the defended and the undefended group in any of the measurements that were carried out on internal contrasts of the body coloration. In addition, from the analyses on marking size distribution in the colour patterns, there were no significant differences between the two groups. Last, when I compared the proportions of typical aposematic and cryptic colours between the defended and the undefended insect groups, again I found that there were no significant differences in proportion in neither of the colours between the two groups. This suggests that all defended species may not be selected for a typical aposematic coloration. Even though there is little empirical evidence (but see Wüster et al. 2004; Tullberg et al. 2005), it has several times been proposed that non-typical aposematic coloration among defended prey may be a common trait (e. g. Endler & Mappes 2004; Ruxton et al. 2009).

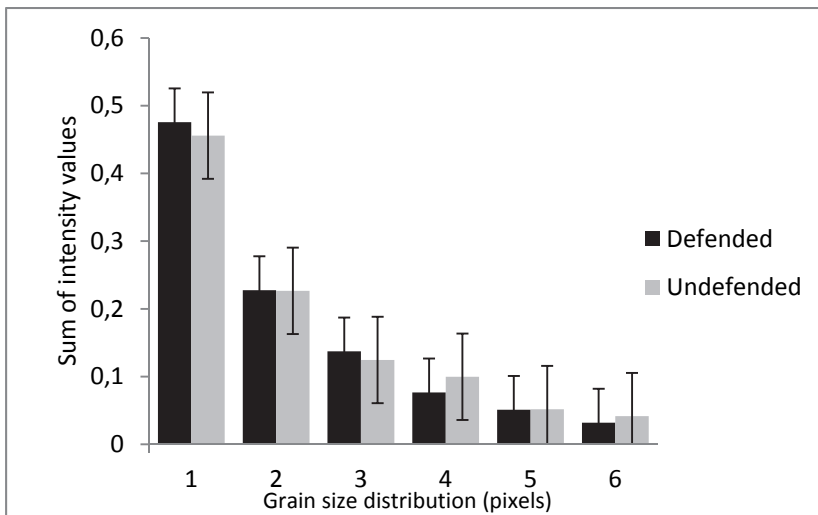


Fig 9. The histogram (mean ± SE) shows the six classes of grain size distribution in the defended insect group (black bars) and the undefended insect group (grey bars). There were no significant differences in grain size distribution between the two groups (MANOVA: $F_{3,36}=0.677$, $p=0.572$).

Many defended species may exhibit intermediate levels of warning signals rather than maximal levels, so the question is if there are any universal aposematic characteristics. Moreover, what makes an efficient warning coloration depends on the impact different ecological and physiological factors have on the colour pattern, not necessarily related to predation (e. g. Booth 1990; Ohsaki 2005; Srygley 2004; Lindstedt et al. 2009). The results imply that there is a higher diversity among warning coloration, and studies on aposematic coloration should consider a much broader spectrum of coloration than previous. Importantly, we must identify the non-typical warning colorations and take these into account to improve our knowledge about aposematism.

5. CONCLUSIONS

In this thesis, I have presented methods to investigate how factors such as colour pattern, viewing distance, and background affect detectability of aposematic coloration. These factors would be difficult to investigate and to control for in field studies, and in experiments using natural predators. I have also shown the usefulness of pattern analysis techniques in an objective analysis of colour patterns.

As can be seen by comparing the appearance of my study species (paper I, II, III & IV), aposematic colour patterns can vary a great deal between species, and some aposematic species can make use of more than one protective strategy. My results suggest that the aposematic Apollo butterfly larva makes use of crypsis at longer viewing distances, presumably to optimise the costs and benefits of detectability. This is in accordance with the findings of Tullberg et al. (2005), in which they found a probable distance-dependent function in the aposematic larva of the swallowtail *Papilio machaon*. Taken together, these studies provide strong evidence for that an aposematic colour pattern can be selected for intermediate levels of conspicuousness, and that aposematism and camouflage are not necessarily mutually exclusive

In contrast to the larvae of the Apollo butterfly and the swallowtail butterfly, the results from the study on the red and black firebug (paper II) clearly show that not all aposematic signals make use of a dual function, and that some aposematic colour patterns are selected for a high level of conspicuousness at all distances, as has been conventionally assumed. It has been suggested that the spatial arrangement of markings in internal contrasts can increase or decrease conspicuousness (Forsman & Merilaita 1999; Cuthill et al. 2005), and that markings placed on the edge of the body may have a disruptive function as they break up the outline of the animal (Thayer 1909; Cott 1940). There was a small effect of the arrangement of the black markings on the firebug, which suggests that

the natural pattern is important for an efficient warning signal (paper II). More importantly, the results show the importance of pattern as such, on detectability, as the manipulated variants lacking internal contrasts were surprisingly cryptic compared to the natural firebug. However, in recognition experiments with colour mutants of *Pyrrhocoris apterus* lacking the red colour presented to passerine birds, the black pattern alone was not sufficient to be distinguished as aposematic (Exnerová et al. 2006). This suggests that the internal contrasts play an important role in the warning signal of the firebug. Whether the detectability of the natural firebug depends on that the pattern of the firebug contrasts against the backgrounds or it is due to the internal contrasts *per se* needs further investigation.

Paper III shows yet another way an aposematic species can make use of both warning coloration and camouflage. The striated shieldbug exhibits a seasonal polyphenism and, thus, unlike the Apollo butterfly larva that uses the two strategies simultaneously but at different distances, the bug uses them temporally separately at different developmental stages. Hence, it changes colour to combine the two strategies. Ontogenetic colour change between life stages is usual among insects (Booth 1990), and one intensively studied species that exhibit polyphenism is the desert locust *Schistocerca gregaria* (e. g. Simpson et al. 1999; Sword et al. 2000). In low densities the locust is solitary, green and palatable, but in high densities the juveniles change diet and becomes chemically defended and develop aposematic black and yellow coloration. However, what is interesting in the case of the striated shieldbug is that the camouflaged phenotype is equally chemically defended as the warningly coloured phenotype (Pasteels et al. 1983; Gamberale-Stille et al. 2010). Again, this shows that a defended species has not always adopted a typical aposematic signal.

In paper IV, I analysed and compared different visual aspects of colour pattern in defended and undefended species in order to be able to identify the key properties and common denominators of warning signals. Interestingly, my analyses did not reveal any specific properties in patterning separating the two groups. If I had subjectively selected the species included in the study based on whether they had a typical aposematic or cryptic appearance, I would very likely have found differences between the two groups. However, because I used objective criteria with respect to coloration, namely edibility to predators, the group of defended species also included species that have colour patterns that are not conventionally considered as warning signals. This again points to the direction that there appears to be a range of ways that defended species exhibit warning signals. For example, as both theoretical (Endler & Mappes 2004) and empirical studies (Wüster et al. 2004) have shown, conspicuousness is not required for an efficient warning signal as long as the signal is recognisable and distinct from

undefended prey appearance. It is possible that there are some visual differences in the colour patterns between the two groups that I did not detect with my analyses, but probably not in contrast, patch size or colour composition, which are the properties that have conventionally been expected to differ between aposematic coloration and other patterns. Instead, my study strongly suggests that there are no universal signal properties that would separate all or most aposematic colour patterns from other colour patterns. This diversity in the appearance of aposematic colour patterns may reflect differences in the exact function of warning signal between defended species, as well as differences among ecological communities in predator species composition (Endler & Mappes 2004), appearance of the habitat (Papagergios 1975; Merilaita & Tullberg 2005), or the colour patterns of the undefended species among prey communities (Lindström et al. 2004).

Collectively, my studies clearly illustrate that having a typical warning coloration is not the only way to be aposematic. Moreover, warning coloration and crypsis are not necessarily two mutually exclusive strategies. This indicates a functional versatility in how aposematic species make use of their body coloration. Importantly, prey animals are subjected to a range of selection pressures, trade-offs, and constraints that shape their colour pattern and determine the optimal appearance of warning coloration in each species. In addition, even though my studies have addressed variation in the visual properties of habitats and how that may affect the detection cost to some extent, there is still considerably more to be accomplished in that area. Finally, as paper **IV** brings to light, it remains particularly a challenge to identify the factors that influence selection imposed on warning coloration in defended prey, and to understand which features predators use to recognise defended prey.

Acknowledgements

PhD studies...what was I thinking?!!

First, I want to express my forever gratitude to my supervisors **Sami Merilaita** and **Birgitta Tullberg**, without whom this thesis would never have been a reality. Thank you **Sami**, for encouragingly guiding and leading me through the making of this thesis. Thanks also for helping me to understand the frightening computational and statistical world of ecological research, and for your constructive criticism and painstaking hours of discussions that helped me to become a better scientist. You have always taken your time and listened. And of course, thank you for showing me Cosmic Comic! Thank you **Birgitta**, for initially showing me the exciting field of aposematism, for teasing apart the many evolutionary and ecological aspects of it, and sharing your knowledge and passion for warning colours in particular, and evolution, nature and bugs in general. And also for always taking the time and lending me a friendly ear even far outside the scope of research. I also want to thank **Gabriella Gamberale-Stille** for being so smart and always up for discussions, and for making my years at Zootis much more fun and interesting, both scientifically as a colleague and with a lot of laughter besides that. But I will never offer half my banana to you again! Thanks to my co-writers in Czech Republic, **Alice Exnerová** and **Pavel Štys**, for our productive collaboration and for showing me other parts of your beautiful Prague.

I am grateful to **Mike Speed** and **Carita Lindstedt-Kareksela** for taking their time to review this thesis. I would also like to thank my thesis support group **Niklas Wahlberg** and **Carita Lindstedt-Kareksela**, for your valuable insights and for keeping an eye on me.

A special thanks to **Karin Kjernsmo**, my light in Turku! For helping me out in so many ways, and for being a hard working fellow PhD student, a dear friend and for making my time in Turku fabulous. Also, I want to thank **Cami Gustafsson** and **Lotta Kvarnemo** for making my time in Turku much more fun and interesting and **Cami** also for helping me out with my dissertation dilemma. Thanks to all the people at **Åbo Akademi** who have contributed in one way or the other – scientifically, practically or just a chat. I would also like to thank **BioInt** research school for financial support, and for the opportunities to meet all the other PhD students and researchers to exchange ideas with at various venues. A special thanks to the “**Jyväskylä gang**”. I wish I’d seen more of you!

My work would have been much harder and boring without my former roomies at Zootis, **Lina Söderlind** and **Martin Bergman**. Thank you **Lina**, for your lively and

creative spirit. We shared both goods and bads. **Martin**, you endured the years between me and Lina! However, the least noisy left the biggest empty space. Thank you for our chats about music, life and science.

I am also grateful for the **Department of Zoology** in Stockholm, for letting me have my space there, both physically and mentally, throughout this time. A big thanks to all the people at Zootis (too many to mention you all by name!) that enriched my years there in so many ways, especially all the discussions in lunch room. **Marianne Lönngren**, thanks to you I can claim that I have worked with real predators – the fierce chicks. I also have to mention **Tomas Meijer** for...well....for being you. Like all the PhD students before me, I also bow my head to **Minna, Siw, Anette, Berit** and **Ulf**. I also want to thank everyone at the **Entomology division** at **The Swedish Museum of Natural History** who helped me out with the insect collection, and for making me feel welcomed.

To my friends out in the real world: Thank you all for being supportive and patient with me even if you probably don't have a clue what I've been doing, and for giving me an occasional reality check. I hope I will meet you out there soon!

Last but not at all least, I want to thank my family. **Tony**, you've stood by me all these years in the pursuit of my PhD. Through thick and thin, as they say! My son **Bastian**, for making my agenda so much more complicated, but my life so incredibly more joyful! And **Mum** and **Dad** – you are rather responsible for who I am! Thank you for always being there and believing in me.

References

- Beatty CD, Beirincx K, Sherratt TN (2004) The evolution of müllerian mimicry in multispecies communities. *Nature* 431: 63-67
- Berenbaum MR, Moreno B, Green E (1992) Soldier bug predation on swallowtail caterpillars (Lepidoptera, Papilionidae) – circumvention of defensive chemistry. *Journal of Insect Behavior* 5: 547-553
- Booth CL (1990) Evolutionary significance of ontogenetic colour change in animals. *Biological Journal of the Linnean Society* 40: 125-163
- Clotfelter ED, Ardia DR, McGraw KJ (2007) Red fish, blue fish: trade-offs between pigmentation and immunity in *Betta splendens*. *Behavioral Ecology* 18: 1139-1145
- Cott HC (1940) Adaptive Colouration in Animals. London: Methuen
- Cuthill IC, Stevens M, Sheppard J, Maddock T, Parraga CA, Troscianko TS (2005) Disruptive coloration and background pattern matching. *Nature* 434: 72-74
- Darst CR, Cummings ME, Cannatella DC (2006) A mechanism for diversity in warning signals: conspicuousness versus toxicity in poison frogs. *Proceedings of the National Academy of Sciences, USA* 103: 5852-5857
- Darwin CMA (1859) *The Origin of Species by Means of Natural Selection*. Murray, London
- Edmunds ME (1974) *Defence in Animals: A Survey of Anti-predator Defences*. Harlow, Essex: Longman
- Endler JA (1988) Frequency-dependent predation, crypsis and aposematic coloration. *Philosophical Transactions of the Royal Society of London, Series B* 319: 505-523
- Endler JA, Mappes J (2004) Predator mixes and the conspicuousness of aposematic signals. *American Naturalist* 163:532-547
- Exnerová A, Štys P, Kristin A, Volf O, Pudil M (2003) Birds as predators of true bugs (Heteroptera) in different habitats. *Biologia (Bratislava)* 58: 253-264
- Exnerová A, Svádová K, Štys P, Barcalová S, Landová E, Prokopová M, Fuchs R, Socha R (2006) Importance of colour in the reaction of passerine predators to aposematic prey: experiments with mutants of *Pyrrhocoris apterus* (Heteroptera). *Biological Journal of the Linnean Society* 88: 143-153
- Farine JP, Bonnard O, Brossut R, Le Quere JL (1992) Chemistry of defensive secretions in nymphs and adults of fire bug, *Pyrrhocoris apterus* L. (Heteroptera, Pyrrhocoridae). *Journal of Chemical Ecology* 18: 1673-1682
- Ferguson JR (2007) *Using the Grey-level Co-occurrence Matrix to Segment and Classify Radar Imagery*. ProQuest and Learning Company
- Forsman A, Merilaita S (1999) Fearful symmetry: pattern size and asymmetry affects aposematic signal efficacy. *Evolutionary Ecology* 13: 131-140
- Gamberale-Stille G, Johansen AI, Tullberg BS (2010) Change in protective coloration in the striated shieldbug *Graphosoma lineatum* (Heteroptera: Pentatomidae): predator avoidance and generalization among different life stages. *Evolutionary Ecology* 24: 423-432
- Gittleman J L, Harvey PH (1980) Why are distasteful prey not cryptic? *Nature* 286: 149-150
- Godfrey D, Lythgoe JN, Rumball DA (1987) Zebra stripes and tiger stripes: the spatial frequency distribution of the pattern compared to that of the background is

- significant in display and crypsis. *Biological Journal of Linnean Society* 32: 427-433
- Gonzales RC, Woods RE, Eddins SL (2004) Digital Processing Using MATLAB. Prentice Hall, Upper Saddle River, NJ
- Grant JB (2007) Ontogenetic colour change and the evolution of aposematism: a case study in panic moth caterpillars. *Journal of Animal Ecology* 76: 439-447
- Guilford T (1986) How do 'Warning Colours' Work? Conspicuousness May Reduce Recognition Errors in Experienced Predators. *Animal Behaviour* 34: 286-288
- Hall JR, Cuthill IC, Baddeley R, Shohet AJ, Scott-Samuel NE (2013) Camouflage, detection and identification of moving targets. *Proceedings of the Royal Society of London, Series B* 280. Article No: 20130064
- Haralick RM, Ahanmugam K, Dinstein I (1973) Textural Features for Image Classification – *IEEE Transactions on Systems, Man and Cybernetics* 3: 610-621
- Hegna RH, Nokelainen O, Hegna JR, Mappes J (2013) To quiver or to shiver: increased melanization benefits thermoregulation, but reduces warning signal efficacy in the wood tiger moth. *Proceedings of the Royal Society of London, Series B* 280. Article No: 20122812
- Higginson AD & Ruxton GD. 2010. Optimal defensive coloration strategies during the growth period of prey. *Evolution* 64: 53-67.
- Ioannou CC, Krause J (2009) Interactions between background matching and motion during visual detection can explain why cryptic animals keep still. *Biology Letters* 5: 191-193
- Johansen AI, Tullberg BS, Gamberale-Stille G (2011) Motion level in *Graphosoma lineatum* coincides with ontogenetic change in defensive colouration. *Entomologia Experimentalis et Applicata* 141: 163-167
- Johansen AI, Exnerová A, Svadová KH, Štys P, Gamberale-Stille G, Tullberg BS (2010) Adaptive change in protective coloration in adult striated shieldbugs *Graphosoma lineatum* (Heteroptera: Pentatomidae): test of detectability of two colour forms by avian predators. *Ecological Entomology* 35: 602-610
- Järvi T, Sillén-Tullberg B, Wiklund C (1981) The cost of being aposematic. An experimental study of predation on larvae of *Papilio machaon* by the great tit *Parus Major*. *Oikos*, 36: 267-272
- Karpestam E, Merilaita S, Forsman A (2013) Detection experiments with humans implicate visual predation as a driver of colour polymorphism dynamics in pygmy grasshoppers. - *BMC Ecology* 13:17
- Kraemer PJ (1984) Forgetting of visual discriminations by pigeons. *Journal of Experimental Psychology – Animal Behavior Processes* 10: 530-542
- Lindstedt C, Lindström L, Mappes J (2009) Thermoregulation constrains effective warning signal expression. *Evolution* 63: 469-478
- Lindström L, Alatalo RV, Lyytinen A (2004) The effect of alternative prey on the dynamics of imperfect Batesian and Mullerian mimicries. *Evolution* 58: 1294-1302
- McFadden SA (1993) *The avian eye view. In: Vision, brain and behavior in birds.* (Ed. By H. P. Zeigler & H-J. Bischof), pp. 1-4. Massachusetts: The MIT Press.
- McGuire L, Van Gossum H, Beirincx K, Sherratt TN (2006) An empirical test of signal detection theory as it applies to Batesian mimicry. *Behavioural Processes* 73: 299-307
- Merilaita S, Tullberg BS (2005) Constrained camouflage facilitates the evolution of conspicuous warning coloration. *Evolution* 59: 38-45
- Nakoneczny M, Kędzierski A (2005). Feeding preferences of the Apollo butterfly (*Parnassius apollo* ssp. *frankenbergeri*) larvae inhabiting the Pieniny Mts (southern Poland). *C. R. Biologies* 328: 235-242

- Nylin S, Gamberale-Stille G, Tullberg BS (2001) Ontogeny of defense and adaptive coloration in larvae of the comma butterfly, *Polygonia c-album* (Nymphalidae). *Journal of the Lepidopterists' Society* 55: 69-73
- Ohsaki N (2005) A common mechanism explaining the evolution of female-limited and both-sex Batesian mimicry in butterflies. *Journal of Animal Ecology* 74: 728-734
- Osorio D, Jones CD, Vorobyev M (1999) Accurate memory for colour but not pattern contrast in chicks. *Current Biology* 9: 199-202
- O'Toole C (2002) *The New Encyclopedia of Insects and Their Allies*. Oxford University Press
- Papageorgis C (1975) Mimicry in Neotropical butterflies. *American Scientist* 63: 522-532
- Pasteels JM, Gregoire JC, Rowellrahier M (1983) The chemical ecology of defense in arthropods. *Annual Review of Entomology* 28: 263-289
- Poulton EB (1890) *The colours of animals: their meaning and use especially considered in the case of insects*. London: Kegan Paul
- Rasband WS (1997–2012) ImageJ. US National Institutes of Health, Bethesda, MD, USA. Available at: <http://imagej.nih.gov/ij/>
- Resh VH, Cardé RT (Eds) (2003) *Encyclopedia of Insects*. Academic Press. Elsevier Science. USA
- Roper TJ, Redston S (1987) Conspicuousness of distasteful prey affects the strength and durability of one-trial avoidance learning. *Animal Behaviour* 35: 739–747
- Ruxton GD, Sherratt TN, Speed MP (2004) *Avoiding Attack: The Evolutionary Ecology of Crypsis, Warning Signals and Mimicry*. Oxford: Oxford University Press
- Ruxton GD, Speed MP, Broom M (2009) Identifying the ecological conditions that select for intermediate levels of aposematic signalling. *Evolutionary Ecology* 23: 491-501
- Šanda M, Žáček P, Streinz L, Dračinský M, Koutek B (2012) Profiling and characterization of volatile secretions from the European stink bug *Graphosoma lineatum* (Heteroptera: Pentatomidae) by two-dimensional gas chromatography/time-of-flight mass spectrometry *Journal of Chromatography B* 881-882: 69-75
- Schaefer M (2010) Visual communication: evolution, ecology, and functional mechanisms. In: *Animal behaviour evolution and mechanisms* (Ed: Kappeler P). pp 3-28. Springer Berlin Heidelberg
- Sillén-Tullberg B (1988) Evolution of Gregariousness in Aposematic Butterfly Larvae: A phylogenetic Analysis. *Evolution* 42: 293-305
- Simpson SJ, McCaffery AR, Hägele B F (1999) A behavioural analysis of phase change in the desert locust. *Biological Reviews* 74: 461–480
- Socha R (1993) *Pyrrhocoris apterus* (Heteroptera) – an experimental model species. A review. *European Journal of Entomology* 90: 241–286
- Socha R, Zemek R (2000) Wing Movement Behavior in Long- and Short-Winged Morphs of the Flightless Bug *Pyrrhocoris apterus* L. (Heteroptera: Pyrrhocoridae) *Journal of Insect Behavior* 13: 741-750
- Speed MP, Ruxton GD (2005a) Aposematism: what should the starting point be? *Proceedings of the Royal Society of London, Series B* 272: 431-438
- Speed MP, Ruxton GD (2005b) Warning displays in spiny animals: One (more) evolutionary route to aposematism. *Evolution* 59: 2499-2508
- Srygley RB (2004) The aerodynamic costs of warning signals in palatable mimetic butterflies and their distasteful models. *Proceedings of the Royal Society of London, Series B* 271: 589-594
- Stamp NE, Bowers MD (1990) Body temperature, behavior, and growth of early-spring caterpillars (*Hemileuca lucina*:

- Saturniidae). *Journal of Lepidopterists' Society* 44: 143-155
- Sword GA, Simpson SJ, Taleb O, El Hadi M, Wilps H (2000) Density-dependent aposematism in the desert locust. *Proceedings of the Royal Society of London, Series B* 267: 63-68
- Thayer GH (1909) *Concealing Coloration in the Animal Kingdom*. Macmillan: New York
- Theodoridis S, Koutroumbas K (2008) *Pattern Recognition*, 4th Ed. Academic Press
- Tullberg BS, Hunter A (1996) Evolution of larval repellent defences gregariousness in relation to and warning coloration in tree-feeding Macrolepidoptera: a phylogenetic analysis based on independent contrasts. *Biological Journal of Linnean Society* 57: 253-276
- Tullberg BS, Gamberale-Stille G, Bohlin T, Merilaita S (2008) Seasonal ontogenetic colour plasticity in the adult striated shieldbug *Graphosoma lineatum* (Heteroptera) and its effect on detectability. *Behavioral Ecology and Sociobiology* 62: 1389-1396
- Tullberg BS, Merilaita S, Wiklund C (2005) Aposematism and crypsis combined as a result of distance dependence: functional versatility of the colour pattern in the swallowtail butterfly larva. *Proceedings of the Royal Society of London, Series B* 272: 1315-1321
- Vesely P, Veselá S, Fuchs R, Zrzavy J (2006) Are gregarious red-black shieldbugs, *Graphosoma lineatum* (Hemiptera: Pentatomidae), really aposematic? An experimental approach. *Evolutionary Ecology Research* 8:881-890
- Vorobyev M, Osorio D (1998) Receptor noise as a determinant of colour thresholds. *Proceedings of the Royal Society of London, Series B* 265: 351-358
- Wallace AR (1867) Letter from Wallace AR to Darwin C, February 24th, 1867. Darwin Correspondence Database, [http:// www.arwinproject.ac.uk/entry-5416](http://www.arwinproject.ac.uk/entry-5416), accessed on Thu Apr 18 2013
- Wallace AR (1889) *Darwinism – an exposition of the theory of natural selection with some of its applications*. London: MacMillan and Co
- Wiklund C, Järvi T (1982) Survival of distasteful insects after being attacked by naive birds: a reappraisal of the theory of aposematic coloration evolving through individual selection. *Evolution* 36: 998-1002
- Wiklund C, Sillén-Tullberg B (1985) Why distasteful butterflies have aposematic larvae and adults, but cryptic pupae: evidence from predation experiments on the monarch and European swallowtail. *Evolution* 39: 1155-1158
- Wilson K, Cotter SC, Reeson AF, Pell JK (2001) Melanism and disease resistance in insects. *Ecology Letters* 4: 637-649
- Wüster W, Allum CSE, Bjargardottir IB, Bailey KL, Dawson KJ, Guenioui J, Lewis J, McGurk J, Moore AG, Niskanen M, Pollard CP (2004) Do aposematism and Batesian mimicry require bright colours? A test, using European viper markings. *Proceedings of the Royal Society of London, Series B*, 271: 2495-2499

Titti Bohlin

Stop bugging me!

Diversity in appearance of warning coloration

This PhD-thesis investigates the variation in appearance and function of aposematic coloration in insects. It shows that warning colorations vary a great deal in design and also in detectability, and in some cases also exploit a camouflage strategy. These comparisons of different colorations increases the understanding of how viewing distance, natural backgrounds and colour pattern affect detectability of aposematic species, and how various selection pressures can form the evolution of a colour pattern.

The author recieved her MSc in Biology in June 2006 at Stockholm University where she until September 2010 worked as a researcher. Since October 2010 she has been positioned as a PhD student in environmental biology at Åbo Akademi University.



9 789521 229473 >

ISBN 978-952-12-2947-3