Barnett CA, Bateson M, Rowe C. Better the devil you know: Avian predators find variation in prey toxicity aversive. Biology Letters 2014, 10(11), 1-4.

Copyright:
© 2014 The Authors. Published by the Royal Society under the terms of the Creative Commons Attribution License http://creativecommons.org/licenses/by/4.0/, which permits unrestricted use, provided the original author and source are credited.

DOI link to article:
http://dx.doi.org/10.1098/rsbl.2014.0533

Date deposited:
03/08/2015

This work is licensed under a Creative Commons Attribution 4.0 International License

Newcastle University ePrints - eprint.ncl.ac.uk
Better the devil you know: avian predators find variation in prey toxicity aversive

Craig A. Barnett†, Melissa Bateson and Candy Rowe

Centre for Behaviour and Evolution, Institute of Neuroscience, Newcastle University, Newcastle upon Tyne NE2 4HH, UK

MB, 0000-0002-0861-0191; CR, 0000-0001-5379-843X

Toxic prey that signal their defences to predators using conspicuous warning signals are called ‘aposematic’. Predators learn about the toxic content of aposematic prey and reduce their attacks on them. However, through regulating their toxin intake, predators will include aposematic prey in their diets when the benefits of gaining the nutrients they contain outweigh the costs of ingesting the prey’s toxins. Predators face a problem when managing their toxin intake: prey sharing the same warning signal often vary in their toxicities. Given that predators should avoid uncertainty when managing their toxin intake, we tested whether European starlings (Sturnus vulgaris) preferred to eat fixed-defence prey (where all prey contained a 2% quinine solution) to mixed-defence prey (where half the prey contained a 4% quinine solution and the other half contained only water). Our results support the idea that predators should be more ‘risk-averse’ when foraging on variably defended prey and suggest that variation in toxicity levels could be a form of defence.

1. Introduction

Many species, both plants and animals, defend themselves with harmful toxins in order to reduce the chances that they are eaten [1,2]. Toxic insects often advertise their defences to potential predators using warning signals, such as conspicuous colour patterns, sounds and odours [3]. This anti-predator defence strategy is known as ‘aposematism’ [4] and is effective because predators can readily learn to associate the warning signals with the ingestion of toxins. As a consequence, predators lower their attack rates on aposematic prey in order to reduce and regulate their intake of potentially harmful toxins as they forage [5,6].

One problem for predators regulating their intake of toxins is that individuals within an aposematic prey species often vary in their toxicity [7,8]. ‘Automimicry’ is characterized by the presence of non-toxic individuals (‘automimics’) in a population of otherwise aposematic prey (‘automodels’) [9,10]. Explaining this variability has been a long-standing theoretical challenge in evolutionary biology. The problem is that if possessing toxins is costly, for example, reducing growth or fecundity [11,12], then undefended individuals will benefit from not paying those costs, while at the same time benefitting from the aposematic defence generated by more toxic individuals. This then leads to increasing numbers of automimics in the population, diluting the model’s defence and increasing the costs of being conspicuous. Potential solutions to this problem have largely involved the role of secreted defences, which allow predators to taste and selectively reject individuals according to their toxicity [10,13,14]. However, not all aposematic insects have inducible chemical defences that are produced upon attack [15], and alternative mechanisms to explain variability in defences in these species are required.

© 2014 The Authors. Published by the Royal Society under the terms of the Creative Commons Attribution License http://creativecommons.org/licenses/by/4.0/, which permits unrestricted use, provided the original author and source are credited.
There are a number of explanations for the variation among prey in their chemical defences, which revolve around individual prey responses to environmental heterogeneity (e.g. host plant availability, predator presence) or intrinsic factors (e.g. sex differences, changes across the lifespan) [16]. Here, we propose a radically different solution to this problem: that the ways in which predators avoid toxic prey can actively promote variability in defence levels among prey individuals [17]. Such variability among prey in their chemical defence could be adaptive if predators find variably defended prey (which we term mixed-defence prey) more aversive (e.g. by making it more difficult for predators to manage their toxin burdens [18]). Therefore, it could benefit a female insect to increase the variability in toxicity in her offspring (e.g. by laying eggs on host plants that vary in their toxicity [19]) in order to reduce predation of them. In our experiment, we investigated birds’ behaviour towards two populations of insect prey that had the same mean levels of toxin, but differed in whether their defences were variable or not. We tested whether or not variation in toxicity is indeed more aversive to predators, and hence, if predator decisions can promote automimicry.

2. Material and methods

Our experiment used an established protocol where European starlings (Sturnus vulgaris) were sequentially given single mealworms (Tenebrio molitor) that varied in their defence levels [20–22]. Seven male wild-caught starlings were individually caged in the laboratory (see the electronic supplementary material for full details of capture and housing; see also [20–22]). Initially, birds were trained to flip white paper lids off sequentially presented Petri dishes, each containing a mealworm. We then gave a series of daily sessions where in each session they received: six undefended prey (all injected with 0.02 ml of water); six ‘fixed-defence’ prey (all injected with 0.02 ml of 2% quinine sulfate solution) and six ‘mixed-defence’ prey (three injected with 0.02 ml of water and three injected with 0.02 ml of 4% quinine sulfate). Therefore, the two defended prey types contained the same mean amount of toxin, but differed in the variation around the mean. The three prey types had distinguishable coloured lids (colours counterbalanced across individuals). Once birds had learned to discriminate between undefended and defended prey, we conducted 10 further sessions on 10 consecutive days. We recorded which mealworms were eaten, allowing us to calculate the proportion of fixed and mixed prey eaten at each presentation number in the sequence (i.e. from 1 to 18) for each bird across the 10 test sessions. We used generalized additive mixed models (GAMMs) to compare the ingestion of fixed and mixed prey within a session (see the electronic supplementary material for further details).

3. Results

Birds were less likely to eat a mixed-defence prey than a fixed-defence prey (GAMM: $\chi^2 = 7.6114, p = 0.0058$, figure 1), and the probability that they ate either type of defended mealworm decreased with increasing presentation number within a session ($\chi^2 = 89.862, p < 0.0001$, figure 1). The interaction between prey type and presentation number within the daily session was not significant ($\chi^2 = 1.3477, p = 0.2457$). The difference between the fixed and mixed prey was not due to birds ingesting more quinine following attacks on the mixed prey. There was no difference in the numbers of 4% quinine-injected and water-injected mixed-defence mealworms eaten (GAMM: $\chi^2 = 0.4809, p = 0.488$; see the electronic supplementary material for further details), meaning that the mean defence level of the mixed-defence prey experienced by the birds was approximately 2%. Therefore, the difference in mean intake of toxin across individuals from mixed-defence and fixed-defence prey was negligible and so could not explain the differences in preferences.

4. Discussion

While the probability of eating a toxin prey decreased with presentation number in a session, due to either the improving nutritional state of the predator or an increasing toxin burden [5,6,21,22], birds were less likely to eat a mixed-defence prey compared with a fixed-defence prey at any point during an experimental session. This clearly shows that birds found mixed-defence prey more aversive than fixed-defence prey, and could use preys’ visual signals to reduce their intake of prey with more variable defences. This means that predatory attacks on a toxic prey population are affected not just by the mean toxin content of a prey population [23–25], but also by how variable it is around that mean. Therefore, the foraging decisions of avian predators could promote variation in toxicity in the wild, leading to the intriguing possibility that automimicry itself could be a form of defence.

Our results suggest that predators’ behaviour could promote variability in toxicity in aposematic species where predators cannot discriminate between variably defended individuals prior to ingestion, particularly where selective advantages can accrue through living in close proximity to kin [13,15]. For example, a gravid female insect could benefit by laying her eggs on a range of local host plants, or if she has a fixed amount of toxin to distribute among her eggs, differentially provisioning her brood with defensive compounds [19,26]. If her undefended offspring (which have high fecundity because of their lack of investment in toxins [12]) gain sufficient protection from their more toxic brood-mates, the overall fitness of her brood could increase relative to each offspring having a fixed amount of toxin. Of course, whether increasing variability would be a better strategy would depend on the strength of predators’ aversion towards variably defended prey in the wild, along with other factors,
such as the availability of other undefended and toxic prey in the environment, and how the costs associated with reduced fecundity change with increasing toxicity. However, it remains a possibility; understanding whether predators’ reduced willingness to eat prey with variable defences affects investment in defences across natural prey populations is certainly an intriguing avenue for future research.

Our data also provide to our knowledge the first empirical support for the idea that predators should be less willing to eat prey with variable defences because they are uncertain of the consequences of ingesting any given individual [18]. This means that our findings are also relevant to cases of Batesian mimicry, where a palatable species copies the signal of a more toxic species. Although the Batesian mimic is parasitic on the defence of its toxic model species and dilutes its defence, the uncertainty that the visual mimic generates in relation to the signal may ameliorate these effects. This may be especially pertinent for models in the presence of another aposematic species with a mean toxin level similar to the mimicry complex. However, we cannot fully exclude the possibility that the difference in reinforcement schedules between fixed and mixed prey generated the differences in predator aversion levels between the two prey types. For example, perhaps the sudden ingestion of toxin resulting from eating a 4% quinine-injected mealworm was perceptibly worse than eating two 2% quinine-injected mealworms, leading to a stronger learned aversion. Results from a previous experiment would argue against this, as we found no evidence that eating a 3% quinine-injected mealworm was more than three times as aversive as eating a 1% quinine-injected mealworm [22]. Therefore, the aversive effects of quinine do not appear to be accelerating with increasing concentration. Moreover, our results are consistent with the literature on risk-sensitive foraging despite using different experimental methods, which indicates that animal subjects prefer rewards that are fixed in pay-off over those that are variable in pay-off [27]. However, further studies could elucidate the cognitive mechanisms underlying predators’ decisions to better understand their broader impact on the evolution of defensive strategies.

Ethics statement. All research adhered to the ASAB/ABS Guidelines for the Use of Animals in Research and was approved by the local ethical committee at Newcastle University.

Data accessibility. Data is available in Dryad (http://datadryad.org/resource/doi:10.5061/dryad.2qk02).

Acknowledgements. We thank Lin Hedgecock and Michelle Wadde for animal husbandry. We also thank John Skelhorn, the editor, and two anonymous reviewers who made many helpful comments.

Funding statement. C.A.B. was supported by a departmental studentship and an ORSAS Award, M.B. held a Royal Society University Research Fellowship and C.R. held a Royal Society Dorothy Hodgkin Fellowship during this study. The work was supported by BBSCI grants BB/D003245/1 and BB/J016446/1.

References

1. Adler LS. 2000 The ecological significance of toxic nectar. Oikos 91, 409–420. (doi:10.1043/j.1600-0706.2000.910301.x)
2. Foley WJ, Moore BD. 2005 Plant secondary metabolites and vertebrate herbivores from physiological regulation to ecosystem function. Curr. Opin. Plant Biol. 8, 430–435. (doi:10.1016/j.pbi.2005.05.009)
3. Rowe C, Halpin C. 2013 Why are warning displays multimodal? Behav. Ecol. Sociobiol. 67, 1425–1439. (doi:10.1007/s00265-012-1315-b)
4. Paulston EB. 1989 The colours of animals: their meaning and use especially considered in the case of insects. London, UK: Kegan Paul, Trench, Trubner and Co.
5. Skelhorn J, Rowe C. 2007 Predator’s toxin burdens influence their strategic decisions to eat toxic prey. Curr. Biol. 17, 1479–1483. (doi:10.1016/j.cub.2007.07.064)
6. Halpin CG, Skelhorn J, Rowe C. 2012 The relationship between sympatric defended species depends upon predators’ discriminatory behaviour. PLoS ONE 7, e44895. (doi:10.1371/journal.pone.0044895)
7. Pasteels JM, Grégoire J-C, Rowell-Rahier M. 1983 The chemical ecology of defense in arthropods. Annu. Rev. Entomol. 28, 263–289. (doi:10.1146/annurev.en.28.010183.001403)
8. Holloway GI, de Jong PW, Brakefield PM, de Vos H. 1991 Chemical defence in ladybird beetles (Coccinellidae). I. Distribution of coccinelline and individual variation in defence in 7-spot ladybirds (Coccinella septempunctata). Chemoecology 2, 7–14. (doi:10.1007/BF01240660)
9. Brower LP, Ryerson WN, Coppiinger LL, Glazier SC. 1968 Ecological chemistry and the palatability spectrum. Science 161, 1349–1350. (doi:10.1126/science.161.3848.1349)
10. Guilford T. 1994 ‘Go-slow’ signalling and the problem of autotomy. J. Theor. Biol. 171, 311–316. (doi:10.1006/jtbi.1994.1192)
11. Rowell-Rahier M, Pasteels JM. 1986 Economics of chemical defence in Chrysomelinae. J. Chem. Ecol. 12, 1189–1203. (doi:10.1007/BF01639004)
12. Higginson AD, Delf J, Ruxton GD, Speed MP. 2011 Growth and reproductive costs of larval defence in the aposematic lepidopteran Pieris brassicae. J. Anim. Ecol. 80, 384–392. (doi:10.1111/j.1365-2666.2010.01786.x)
13. Gambale-Stille G, Guilford T. 2004 Autocytomimicry destabilizes aposematism: predator sample-and-reject behaviour may provide a solution. Proc. R. Soc. Lond. B 271, 2621–2625. (doi:10.1098/rspb.2004.2893)
14. Skelhorn J, Rowe C. 2009 Distastefulness as an antipredator defence strategy. Ani. Behav. 78, 761–766. (doi:10.1016/j.anbehav.2009.07.006)
15. Tullberg BS, Hunter AF. 1996 Evolution of larval gregariousness in relation to repellent defences and warning coloration in tree-feeding Macrolepidoptera: a phylogenetic analysis based on independent contrasts. Biol. J. Linn. Soc. 57, 253–276. (doi:10.1111/j.1095-8322.1996.tb00312.x)
16. Speed MP, Ruxton GD, Mappes J, Sherratt TN. 2012 Why are defensively toxic species so variable? An evolutionary perspective. Biol. Rev. 87, 874–884. (doi:10.1111/j.1469-185X.2012.00328.x)
17. Skelhorn J, Rowe C. 2005 Frequency-dependent taste-rejection by avian predation may select for defence chemical polymorphisms in aposematic prey. Biol. Lett. 1, 500–503. (doi:10.1098/rsbl.2005.0359)
18. Sherratt TN. 2003 State-dependent risk-taking by predators in systems with defended prey. Oikos 103, 93–100. (doi:10.1034/j.1600-0706.2003.12576.x)
19. Malcolm SB, Brower LP. 1989 Evolutionary and ecological implications of cardemolide sequestration in the monarch butterfly. Experientia 45, 284–295. (doi:10.1007/BF01951814)
20. Skelhorn J, Rowe C. 2006 Predator avoidance learning of prey with secreted or stored defences and the evolution of insect defences. Ani. Behav. 72, 827–834. (doi:10.1016/j.anbehav.2005.12.010)
21. Barnett CA, Bateson M, Rowe C. 2007 State-dependent decision making: educated predators strategically trade off the costs and benefits of consuming aposematic prey. Behav. Ecol. 18, 645–651. (doi:10.1093/beheco/arm027)
22. Barnett CA, Skelhorn J, Bateson M, Rowe C. 2012 Educated predators make strategic decisions to eat defended prey according to their toxic content. Behav. Ecol. 23, 418–424. (doi:10.1093/beheco/arz206)
23. Ihala¨inen E, Lindström L, Mappes J. 2007 Investigating Müllerian mimicry: predator learning and variation in prey defences. J. Evol. Biol. 20, 789–791. (doi:10.1111/j.1429-9424.2006.01234.x)
24. Rowland HM, Ihala¨inen E, Lindström L, Mappes J. Speed MP. 2007 Co-mimics have a mutualistic relationship despite unequal defences. Nature 448, 64–67. (doi:10.1038/nature05999)
25. Rowland HM, Mappes J, Ruxton GD, Speed MP. 2010 Mimicry between unequally defended prey can be parasitic: evidence for quasi-Batesian mimicry. *Ecol. Lett.* **13**, 1494 – 1502. (doi:10.1111/j.1461-0248.2010.01539.x)

26. Zalucki M, Malcolm S, Paine T, Hanlon C, Brower L, Clarke A. 2001 It’s the first bites that count: survival of first-instar monarchs on milkweeds. *Austral. Ecol.* **26**, 547 – 555. (doi:10.1046/j.1442-9993.2001.01132.x)

27. Bateson M, Kacelnik A. 1998 Risk-sensitive foraging: decision making in variable environments. In Cognitive ecology: the evolutionary ecology of information processing and decision making (ed. R Dukas), pp. 297 – 341. Chicago, IL: University of Chicago Press.