# Does melanism influence the diet of the mountain stone weta *Hemideina maori* (Orthoptera: Anostostomatidae)?

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**Abstract:** Melanic and non-melanic mountain stone weta *Hemideina maori* (Orthoptera: Anostostomatidae) from the Rock and Pillar Range exhibit differential rates of melanotic encapsulation, a response to the presence of pathogens within the body. Because pathogens normally enter the body via food contamination, we hypothesised that dietary differences between the colour morphs might exist. We used faecal pellet analysis to determine the diet of the mountain stone weta. We then tested for differences between the two colour morphs in overall diet composition and the proportional contribution of invertebrate fragments to faecal pellets. The number of invertebrate fragments to the diet did not differ between the two colour morphs. Only when the plant species that were eaten infrequently were given greater weight in an Analysis of Similarity (ANSOM) did significant differences emerge in the diet of the two colour morphs. Hence, there was a weak but significant difference in the overall composition of their diets, but the ecological significance of this difference is unclear. Experimental studies could be used to determine if the dietary differences shown reflect true feeding preferences, or were due to small-scale differences in food availability or stochastic variation.

Key words: dietary analysis; cuticle colouration; colour morphs; wetas.

## Introduction

New Zealand's invertebrate fauna is poorly known, especially considering the importance of invertebrates to community functioning (Patrick, 1994). In particular, knowledge of the basic ecology, including diet, of many species is lacking (Patrick, 1994). For example, despite their prominent status within the fauna of New Zealand, there have been no published studies of the diet of weta (Orthoptera), or of intraspecific variation in their diet. It is known that they are omnivorous, feeding on live and decaying matter (Gibbs, 1998). Intraspecific dietary differences are known from a range of invertebrates, including orthopterans (e.g. le Gall *et al.*, 1998), and need to be taken into account when determining the overall habitat requirements of a species.

The mountain stone weta (*Hemideina maori* Pictet and Saussure) lives on the drier eastern mountain ranges of the South Island of New Zealand (Gibbs, 1998; 2001). Individuals within the species vary in the degree of cuticular melanisation, a feature best known from the Rock and Pillar Range, Central Otago. Here weta are coloured black (tergites alternating black and dark purple-brown with a dark purple-brown ventral surface) or yellow (alternating black and yellow tergites with a creamy yellow ventral surface completely devoid of speckling; King *et al.*, 1996). The black and yellow morphs on the Rock and Pillar Range are genetically distinct, and are thought to have diverged 2.0–2.1 m.y.a. (King *et al.*, 1996; 2003). Their distributions are largely allopatric, but where sympatric they are able to hybridise, producing a continuum of phenotypes (King *et al.*, 1996; 2003).

Recently, it has been shown experimentally that non-melanic (yellow) weta on the Rock and Pillar Range exhibit higher rates of melanotic encapsulation than melanic (black) weta (Robb *et al.*, 2003). Melanotic encapsulation (the isolation of potentially harmful foreign bodies within an insect by encapsulation in melanin) is a response to internal pathogens, most of which are ingested with contaminated food (Boman and Hultmark, 1987). Hence different melanotic encapsulation rates between the colour morphs may reflect dietary differences between them. Therefore the aim of this study was to use faecal pellet analysis to determine if there are differences in the diet composition of melanic and non-melanic *Hemideina maori* from the Rock and Pillar Range, Central Otago. Methods

The study took place in the central area of the Rock and Pillar Range (45°28' S, 170°02' E). For a description of the vegetation, climatic and physical features of the Rock and Pillar Range, see Bliss and Mark (1974). On the Rock and Pillar Range, weta are found under rocks on or surrounding large schist outcrops, known as tors (Leisnham and Jamieson, 2002). This study centred on five of these tors at which black and yellow weta were both found. The tors range in altitude from 1240 to 1350 m a.s.l. and are surrounded by lightly grazed alpine grassland, herbfield, cushionfield and shrubs.

Sampling of faecal pellets took place approximately monthly, from December 2002 to April 2003. During monthly sampling sessions, all five tors were visited. As weta were not marked after being sampled, some individuals may have been sampled more than once. However, since there was at least one month between samples, the samples were treated as independent. Weta were found by lifting rocks, and adult and sub-adult weta were handled or contained until they defecated or until approximately 20 min had elapsed. Weta were then returned to their replaced rock. The colour of each weta (black or yellow) was recorded, as was the tor on which it was found. As black weta with slight yellow speckling, and yellow weta with slight black speckling on the underside have been shown to represent variation within the black and yellow genotypes respectively (King et al., 1996), these were included in the study as black and yellow weta. Faecal pellets were stored individually in labelled 5-ml eppendorf tubes, and then dried at room temperature and frozen until processing.

Processing of faecal pellets followed the method used by Lodge (2000) and Joyce (2002), adapted from Bhadresa (1986); see Wilson (2004) for details. After processing, samples were placed on microscope slides, dried, and mounted in glycerol jelly. The slides were labelled by number only to avoid possible biases during examination. Pellets were examined in their entirety at 40× magnification under a compound microscope, with plant cuticle fragments examined at 400× magnification. When first seen, each cuticle type was drawn, its distinguishing features noted, and the 'type' specimen's location and pellet recorded. This led to the formation of a pictorial library of all cuticle types found in the faecal pellets. As many of these as possible were identified using the Cuticle Reference Collection (CRC), a collection of cuticle fragments from plant species of the Rock and Pillar Range established by Lodge (2000) and expanded by Joyce (2002). This study made further additions to the CRC.

To score abundance, up to 50 fragments of each cuticle type were counted from each pellet. If more than 50 fragments of a species occurred, the total was then extrapolated, based on the portion of the slide that remained to be viewed. Invertebrate fragments were lumped together since most could not be identified to lower taxonomic levels. Only diet items that occurred with a frequency of 5% or more on any one tor and in one or both colour morphs were included in the statistical analyses described below.

We tested for differences in the proportion of invertebrate fragments found in black and yellow wetas' faecal pellets using the General Linear Model procedure in Minitab 8.3. Proportions were transformed using arcsine square-root to normalise the distribution (Zar, 1999), and colour was nested within tor to account for invertebrate availability. To test for differences between the colour morphs in the overall diet composition we used the two-way crossed ANOSIM (Analysis of Similarity) procedure (with tor and colour as factors) in PRIMER v5 (Clark and Gorley, 2001), with 999 permutations of the data. This analysis tests for differences between factors (in this case colour) while controlling for differences across sites (in this case tors) (Clark and Gorley, 2000), and hence differences in plant availability at the different tors were taken into account. To investigate the effects of down-weighting the more commonly eaten species, data were square-root and fourth-root transformed, similarity matrices for the ANOSIM created and the Bray-Curtis similarity measure calculated for both transformations. Similar proportions of colour morphs were sampled throughout the surveys (16-38% of samples each month were from black weta) hence no adjustment for season was included in the analyses. More females than males (51 female vs. 23 males) were sampled for the yellow morph, while the opposite was true for the black morph (9 females vs. 16 males). We had no reason to believe that male and female weta would differ in their diet composition, and because sample sizes on each tor were small, the sexes were pooled.

#### Results

A total of 99 weta faecal pellets were collected from 74 yellow weta and 25 black weta found on five different tors (Table 1). Twenty-eight plant cuticle types were identified from the pellets, as well as pollen, *Hieracium pilosella* trichomes and invertebrate parts. Of these, nine species occurred at or above the 5% threshold for inclusion in the analysis; six could be identified as plant species, one was unidentified foliose lichen and two were unidentified plant species (Table 2).

An ANOSIM of the similarity matrix using squareroot transformed data showed significant differences in the diet of weta between different tors (R = 0.281, P = 0.001), but no significant differences in the diet of

Tor	Black	Yellow	
1	7	4	
2	3	6	
3	5	7	
4	4	33	
5	6	24	
Total	25	74	

 Table 1. Number of faecal pellets of black and yellow weta collected from the study tors.

**Table 2.** Mean ( $\pm$  standard error) percentage of occurrence of diet items meeting the 5% threshold for black and yellow weta averaged across the five tors. Only diet items that occurred with a frequency of 5% or more on any one tor and in one or both colour morphs are included here. The statistical analysis takes into account differences across the five study tors.

	Colour morph		
Diet item	Black	Yellow	
Invertebrate	25.72 (3.2)	25.32 (5.7)	
Poa colensoi	20.90 (6.0)	26.86 (7.4)	
Polytrichum juniperum	17.50 (7.9)	19.66 (5.3)	
Lichen	14.46 (8.4)	4.76 (2.5)	
Kelleria villosa	9.04 (6.5)	6.40 (4.2)	
Celmisia viscosa	2.82 (1.2)	5.66 (3.1)	
Raoulia hectori	4.20 (1.5)	2.22 (1.2)	
Anisotame imbricata	0.84 (0.5)	6.20 (3.0)	
Sp A <sup>1</sup>	1.50 (0.7)	2.38 (1.2)	
Sp B <sup>1</sup>	1.18 (1.2)	0	

<sup>1</sup>Unidentified plant species

the colour morphs (R = 0.137, P = 0.080). Only when the data were fourth-root transformed, hence with more weight given to the lesser-eaten species in the analysis, were significant differences in diet between black and yellow weta detected (R = 0.256, P = 0.011), while the difference between tors remained significant (R = 0.263, P = 0.001). There was no difference in the average proportion of invertebrate fragments in black and yellow wetas' faecal pellets ( $F_{5.98} = 0.47$ , P =0.80).

#### Discussion

The higher number of pellets collected from yellow weta was consistent with their greater abundance within

the study areas (I. Jamieson, unpublished data). The dietary differences of weta inhabiting different tors are likely to be a reflection of differing plant availability at these tors. A significant difference in overall dietary composition between black and yellow weta was evident only after fourth-root transformation of the data. This indicates the species eaten to different degrees by the colour morphs were the diet items of lesser abundance (i.e. those given more weight in the analysis by a more severe transformation). The ecological importance of this finding is unclear, and the difference seen may be due partly to stochastic variation between the weta or their pellets. There were also no differences between black and yellow H. maori in the number of invertebrates eaten. It is not known if most of the invertebrate matter in weta faecal pellets comes from predation or scavenging, although H. maori has been observed to do both (I. Jamieson, pers. obs.). Overall, we did not find strong support for our hypothesis that differences in melanotic encapsulation rates between the colour morphs was related to major differences in their diet.

No previous studies have looked for colour-related differences in the diet of wild insect populations. Captive gregarious (dark coloured) and solitary (green coloured) desert locusts (*Schistocerca gregaria*) have been shown to eat different amounts of nutritionally unbalanced food, but the difference was likely to be a response to ecological conditions experienced by the two phases, rather than a consequence of their colour (Simpson *et al.*, 2002). Coloration may also affect insect activity levels and food intake due to thermoregulatory implications (Forsman *et al.*, 2002), although this is unlikely to apply to nocturnal weta.

Although there was no strong difference in diet composition between the two colour morphs, the cuticle fragments of only nine plant species reached the 5% frequency threshold for inclusion in the analysis. In particular, Polytrichum juniperum, Poa colensoi, Raoulia hectori, Celmisia viscosa, Anisotame imbricata and Kelleria villosa occurred more frequently in the diet of H. maori than expected based on their availability (Wilson, 2004). Of these, C. viscosa, A. imbricata, and R. hectori have unusually high lipid contents (Bliss and Mark, 1974). Hence, H. maori might have a preference for high-lipid plant species (see also Lodge, 2000; Joyce, 2002), but feeding trials would be required to determine if selective foraging is occurring. Similar experiments could also determine if the small difference shown in diet between melanic and non-melanic weta came about through chance, or were a consequence of a real difference in food preference between the two colour morphs.

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