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David Goulson · Jenny. S. Cory

Responses of *Mamestra brassicae* (Lepidoptera: Noctuidae) to crowding: interactions with disease resistance, colour phase and growth

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Abstract This study examines phenotypic plasticity in relation to rearing density in larvae of the moth, *Mamestra brassicae*. Larval phase, growth rate, weight at moulting and susceptibility to disease were quantified when reared at five densities. Larvae develop more quickly, but attain a smaller size and are more susceptible to disease, when reared at high than at intermediate densities. They also exhibit a higher degree of melanisation than larvae reared at intermediate densities, or singly. A review of the literature suggests that a switch to a rapidly developing dark phase at high densities is a widespread phenomenon within the Lepidoptera. Rapid development at the expense of attaining a large size, and increased melanisation, are interpreted as adaptive responses to reach pupation before food supplies are depleted, as is likely when larval density is high. High susceptibility to viral infection at high density may be a result of physiological stress associated with rapid development, or due to a shift in allocation of resources from resistance to development: larvae that developed quickly were more susceptible to infection. Larvae reared singly appeared to be less fit than larvae reared at intermediate densities: they exhibited many of the characteristics of larvae reared at high density, particularly low weight, a right-hand skew in their weight frequency distribution, and high susceptibility to disease. I hypothesise that expression of resistance may be phenotypically plastic with regard to environment. Contact with other larvae may, up to a point, stimulate both growth and resistance to infection, for the risk of infection will increase with the density of conspecifics.

D. Goulson(✉)[1] · J.S. Cory
Ecology and Biocontrol Group,
NERC Institute of Virology and Environmental Microbiology,
Mansfield Rd, Oxford, OX1 3SR, UK

Present address:
[1] Department of Biology, University of Southampton,
Biomedical Sciences Building, Bassett Crescent East,
Southampton SO16 7PX,UK
Fax: +44-1703-594269

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Introduction

Density effects on the morphology and development rate of lepidopteran larvae are well documented, particularly the phenomenon of phase polymorphism. Numerous species from diverse taxonomic groups exhibit a switch to a darker (melanised) larval phase when reared at high densities. The difference between larval phases involves more than simply colour. Dark phase larvae may have higher activity, respiration rate and growth rate, consume more food, and may be more tolerant of starvation or unpalatable food, but have lower larval, pupal and adult size (Iwao 1963; Shibizaki 1969; Shibizaki and Ito 1969; Hodjat 1970; Fescemyer and Hammond 1986). The adults resulting from dark phase and pale phase larvae are indistinguishable, but adults from dark phase larvae exhibit greater variability in flight activity in *Spodoptera exempta* (Parker and Gatehouse 1985).

To date, phase polymorphism has been described in five species in the genus *Spodoptera* (Faure 1943 a,b; Matthée 1947; Hodjat 1970; Tojo 1991), three *Orthosia* species, *Autographa gamma* and *Lacanobia oleracea* (Long 1953), *Mythimna separata* (Kunimi and Yamada 1990), *Agrotis ipsilon* (Noctuidae) (Sappington et al. 1992), *Pavonia pavonia* (Saturnidae) (Long 1953), *Cephonodes hylas* (Sphingidae) (Sasakawa and Yamazaki 1967), *Alsophila pometaria* (Futuyma et al. 1981; Mitter and Futuyma 1983), *Ennomos subsignarius* (Geometridae) (Drooz 1966) and *Zeiraphera diniana* (Tortricidae) (Day and Baltensweiler 1972; Baltensweiler et al. 1977). The phenomenon has often been compared with the gregarious and solitary phases exhibited by locusts, since both are dependant on rearing density (reviewed in Rhoades 1985).

Many of the lepidopteran species which exhibit phase polymorphism are important agricultural pests. Baculoviruses are frequently used as biological control agents

against Lepidoptera, yet our understanding of the genetic and environmental components of susceptibility to baculovirus infection remains poor (reviewed in Anderson and May 1981; Myers 1988). Environmental effects in particular have received little attention. It seems intuitively likely that the stresses induced by increasing density may increase susceptibility (Tanada 1964), and stress induced susceptibility has been proposed as a potential factor influencing population cycles of forest insects (Myers 1988), yet most studies have failed to find any such response. Crowding and food shortage may even decrease susceptibility to infection (Benz 1987; Kunimi and Yamada 1990). However, food quality, which may vary with larval density, can directly affect susceptibility (Keating et al. 1990). If larval susceptibility does vary according to density there are important implications for studies of insect - baculovirus ecology, and particularly for theoretical models concerned with population dynamics.

Both phase polymorphism and the host-baculovirus interaction have been extensively studied in the cabbage moth, *Mamestra brassicae* (L.), but previously no attempt has been made to assess how rearing density and larval phase affect susceptibility to infection. *M. brassicae* is a widespread and abundant species throughout Europe. The larvae are highly polyphagous, but are found in particular on *Brassica* crops were they are a pest of moderate importance. Eggs are laid in batches of up to approximately 200, so that although the larvae are not gregarious they exhibit a highly contagious distribution. On *Brassica* crops larvae tend to feed near the heart of the plant, where they can attain high densities (D. Goulson, unpublished work). Dark phase larvae can be induced by high densities (Hirata 1957a,b, 1962; Kazimirova 1992), and by low humidity or low temperature (Goulson 1994). Dark phase larvae absorb radiant heat more efficiently and thus maintain a higher internal temperature than pale larvae, so that induction of the dark phase at low temperatures may be adaptive (Goulson 1994). The polymorphism has a genetic component. Estimates of additive heritability (h^2), based on regression of brood means against mid-parental values, were 0.237 ± 0.07 (SD) for fourth instars and 0.421 ± 0.10 (SD) for fifth-instar larvae (Goulson 1994). The development of melanisation is mediated, among other factors, by the frequency of contact between larvae (Kazimirova 1992). Dark phase can be induced by artificial mechanical disturbance in *E. subsignarius* (Geometridae) (Drooz 1966) and *C. hylas* (Sasakawa 1973). Phase polymorphism is known to be under endocrine control in the noctuid moths *Mythimna separata* (Ogura 1975) and *Spodoptera litura* (Tojo et al. 1985).

The induction of dark phase larvae at high density is presumed to be an adaptive response to intraspecific competition for food. High density also increases the risk of infection from conspecifics, particularly in later instars (Hochberg 1991a). Gregarious Lepidoptera tend to exhibit a disproportionate increase in resistance to baculovirus disease as they age, compared to solitary species (Hochberg 1991a). A parallel, but intraspecific association between development of resistance and larval density has been described in *M. separata*, resistance increasing in proportion to density (Kunimi and Yamada 1990). Kunimi and Yamada (1990) also report a corresponding increase in melanisation of larvae reared at high density, although no statistically significant relationship was found between larval colour and resistance.

This study quantifies phenotypic plasticity in response to rearing density in *Mamestra brassicae*. I examine the effects of density on susceptibility to infection with virus, on phase variation, and on weight and growth rates, and also examine whether colour phases differ in susceptibility.

Materials and methods

Insect material for this study was obtained from female moths trapped at light near Winchester, Hampshire in 1991. Offspring have since been reared continuously in captivity on artificial diet (Hunter et al. 1984). Batches of offspring used in experiments were from approximately 80 adults caged together. All experiments were conducted at 24°C (±1°).

The virus strain used was a multiply enveloped nuclear polyhedrosis virus of *M. brassicae* (MbNPV) originally obtained from A. Gröner (BBA, Darmstadt, Germany) in 1976. It originated from an epizootic in an insect culture in Darmstadt in 1973. It has since become known as the Oxford isolate, and has been extensively studied both in terms of biological activity and host range (Evans 1981, 1983; Evans et al. 1981; Doyle et al. 1990) and biochemical characteristics (Brown et al. 1981; Possee and Kelly 1988).

Effects of rearing density

Neonate larvae were reared to fifth instar in plastic pots (4 cm diameter × 3.5 cm high) containing artificial diet, at densities of 1, 2, 4, 10 and 20 larvae per pot. In each density treatment 100 larvae were used and the experiment was replicated three times, using 1,500 larvae in total. Larvae were inspected every 24 h, and food replenished as necessary (no additional food was required at densities 1, 2 and 4). The addition of fresh food for larvae reared at densities 10 and 20 may contribute to differences between these larvae and larvae reared at lower densities. Frass was allowed to accumulate in the rearing pots. When larvae reached the fifth instar they were removed. The development time from neonate to fifth instar was recorded for each larva to the nearest day. The degree of melanisation of fifth-instar larvae was also recorded on a scale of 1 (pale) to 4 (dark) after Goulson (1994). Larvae were then individually administered virus by inoculating small plugs of diet with 1 µl of virus solution, so that all of the dose was consumed within 24 h. The dose used was c. 2.56×10^5 PIBs (Polyhedral Inclusion Bodies) per larva, close to the LD_{50} for fifth-instar larvae (c. 2.38×10^5 PIBs per larva, Evans 1981). Larvae which did not consume all of the plug were discarded. After inoculation larvae were reared individually in the dark, and examined daily until death or pupation. When the cause of death was in doubt, cadavers were smeared and stained with Giemsa, and examined under a light microscope (× 1000) for the presence of polyhedral inclusion bodies. If this test was negative larvae were excluded from further analysis. As controls, a further 20 larvae per density/replicate combination were reared as above, but not inoculated with virus.

During the first run of the experiment it became apparent that larvae reared at high densities moulted to the fifth instar at a smaller size than larvae reared at lower densities. To quantify this observation larvae in replicates two and three were weighed prior to inoculation.

Removal of larvae as they reached the fifth instar inevitably meant that larvae which developed more slowly experienced reduced densities just prior to inoculation. However, as larval growth was reasonably synchronous within density treatments, most larvae reached the fifth instar at approximately the same time. Any larva which did not reach the fifth instar within 3 days of the first of its cohort was discarded.

Larval colour and susceptibility to viral disease

To examine possible associations between larval melanisation and susceptibility to MbNPV in greater detail, larval mortality was assessed in relation to colour for both fourth- and fifth-instar larvae (the two instars which exhibit the greatest degree of colour variation, Goulson 1994), and over a range of virus doses. Larvae were reared on diet at a constant density of 100 per container. Containers were ventilated clear plastic boxes (16 × 28 × 10 cm), lined with damp tissue paper, and stored in the dark. For use in experiments larvae were removed within 24 h of moulting so that they were of even age. Preliminary analysis of the density experiment suggested that susceptibility might vary with development rate. Consequently only the first batch of larvae to reach the appropriate instar (fourth or fifth) from each box were used, so that all larvae were of equal development rate.

Larvae were numbered, weighed and their colour recorded as described above. They were then individually administered virus on plugs of diet, as described above. Three doses of virus were used, 10^3, 10^4 and 10^5 PIBs per larva, and a control (distilled water). Larvae were discarded if they did not consume all of the diet plug within 24 h. Fifty larvae were used per treatment, and the experiment replicated four times for fifth-instar larvae (800 larvae in total), and twice for fourth-instar larvae (400 larvae in total).

Statistical analysis

Mortality was analysed using the Generalised Linear Interactive Modelling program (GLIM) (McCullagh and Nelder 1989) and was estimated with binomial errors, substantiated during the analysis. Rearing pots were treated as replicates (so that there were fewer replicates at higher densities) with weighting for number of larvae per pot. As the degree of overdispersion was within acceptable limits (Pearson's χ^2 divided by the residual degrees of freedom was between 1 and 3), a dispersion parameter was calculated and used to adjust the scale parameter. All factors were initially included in the model (rearing density, larval colour, weight, development time from first to fifth instar), and all two-way interactions. Factors which failed to contribute significantly were then removed. Variation in the proportion of larvae in each colour class according to rearing density was similarly estimated with binomial errors.

Inspection of residuals confirmed that larval development time was approximately normally distributed. Larval weights were square root transformed to give an approximation to normality. Skewness of larval weight frequency distributions (g_1) was calculated as described in Sokal and Rohlf (1981).

Results

Effects of rearing density on susceptibility to viral infection

Susceptibility of larvae to viral infection differed according to rearing density prior to inoculation ($\chi^2_2 = 24.1$, $P < 0.001$) (Fig. 1). Larval survival was lowest at the highest density (density 20) (23.7%), and second lowest when larvae were reared singly (density 1) (35.0%). Survival was highest at the three intermediate densities (mean 45.0%). Model simplification resulted in the three intermediate densities being grouped together, indicating that susceptibility did not differ significantly between these treatments. There were no virus deaths in control larvae.

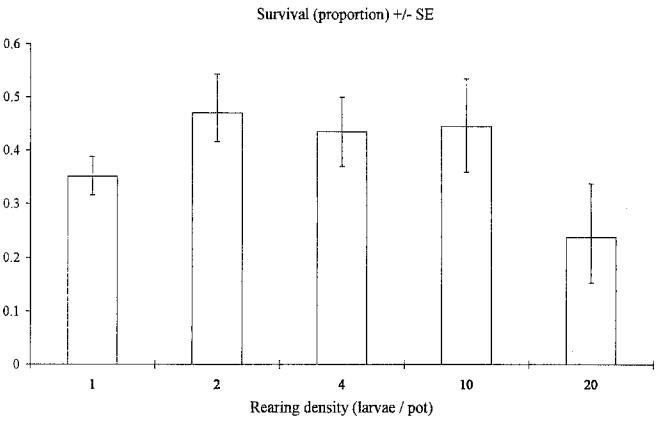

Fig. 1 Proportion of larvae surviving inoculation with virus when in the fifth instar (256,000 PIBs (Polyhedral Inclusion Bodies) per larva), following rearing from first to fifth instar at a range of densities (± SE estimated in GLIM). Numbers of larvae were 228, 236, 239, 238, and 218 for densities 1, 2, 4, 10 and 20, respectively

Both development time and larval weight were significantly altered by rearing density (see below), so interpretation of their effects on subsequent susceptibility to viral infection is difficult. However, all three factors (density, development time and weight) were concurrently significant in the model (each explained a significant proportion of variation in susceptibility between larvae). Thus details of the effect of each factor on susceptibility are included, although numerous seperate experimental manipulations would be necessary to disentangle these relationships.

Susceptibility to viral infection differed according to the rate of development of larvae from first to fifth instar (the mean rate of development per pot) ($\chi^2_1 = 25.5$, $P < 0.001$). The first larvae to reach the fifth instar (11 days) were far more susceptible to infection than larvae which moulted to the fifth instar in the following 3 days, most clearly illustrated by the survival rate of individual larvae which took 11, 12, 13 and 14 days to reach the fifth instar (Fig. 2).

The weights of larvae when newly moulted to the fifth instar affected their probability of survival when subsequently inoculated with virus. Larvae from rearing pots with a high mean weight were more likely to survive ($\chi^2_1 = 28.6$, $P < 0.001$) (weight of survivors 1.00 g ± 0.035 SE compared to 0.84 g ± 0.022 SE for larvae which died of viral infection).

Susceptibility did not vary significantly according to larval colour (mean colour per pot), although there was a trend towards reduced survival in darker larvae ($\chi^2_1 = 1.8$). Interactions between density, development time, larval weight and colour were not significant.

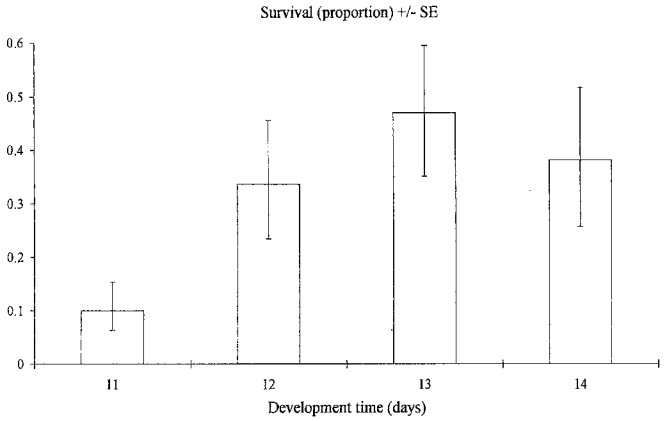

Fig. 2 Proportion of larvae surviving inoculation with virus when in the fifth instar, according to the number of days taken to develop from hatching to the fifth instar (± SE estimated in GLIM). Larvae are pooled for all rearing densities (there was no significant development time×rearing density interaction). Numbers of larvae were 60, 473, 555 and 71 for days 11, 12, 13 and 14, respectively

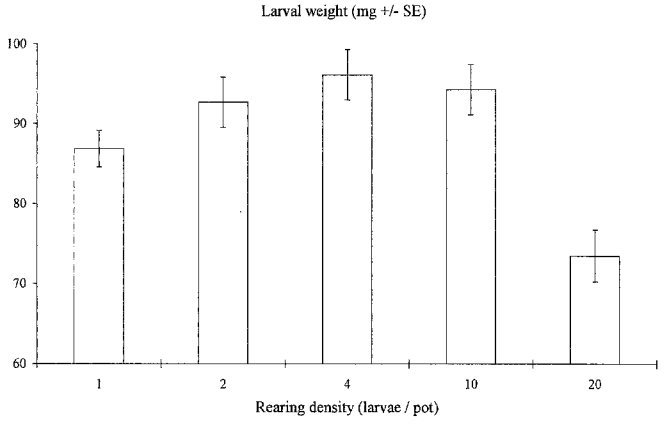

Fig. 3 Larval weight as a newly moulted fifth instar (mg ± SE), according to the density at which they were reared from hatching to fifth instar. Numbers of larvae given in Fig. 1

Effects of rearing density on larval weight, rate of development and colour

Rearing density influenced the weight at which larvae moulted to the fifth instar, the relationship mirroring that between rearing density and susceptibility: the mean weight of newly moulted larvae was lowest at the highest density (density 20), and second lowest when larvae were reared singly ($F_{2,\ 369} = 29.5$, $P < 0.001$) (Fig. 3). Mean weight was highest when larvae were reared at intermediate densities (i.e. 2, 4 and 10), model simplification resulting in these densities being grouped together. Larval weights exhibited a significant positive skew at all densities except for density four (4 larvae/pot). However, the skew was most pronounced when larvae were reared singly (Fig. 4).

Development rate of larvae differed according to the rearing density. Larvae developed most quickly when reared at high densities ($F_{2,562} = 5.88$, $P < 0.01$) (Fig. 5).

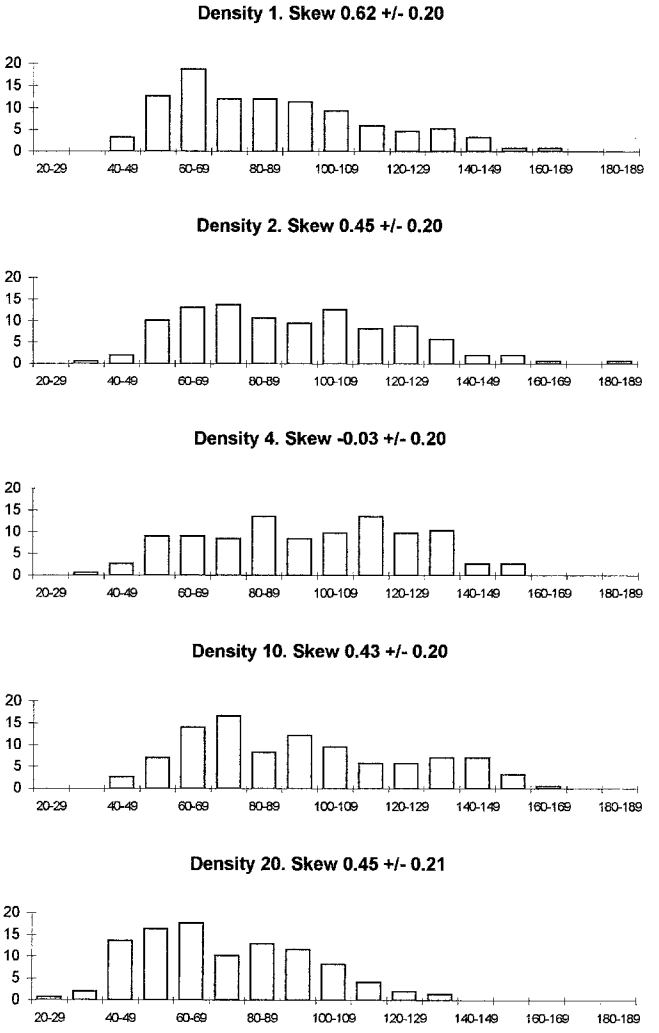

Fig. 4 Frequency distributions of larval weights according to rearing density. Skewness (g_1) calculated according to Sokal and Rohlf (1981) (± SE). Numbers of larvae given in Fig. 1

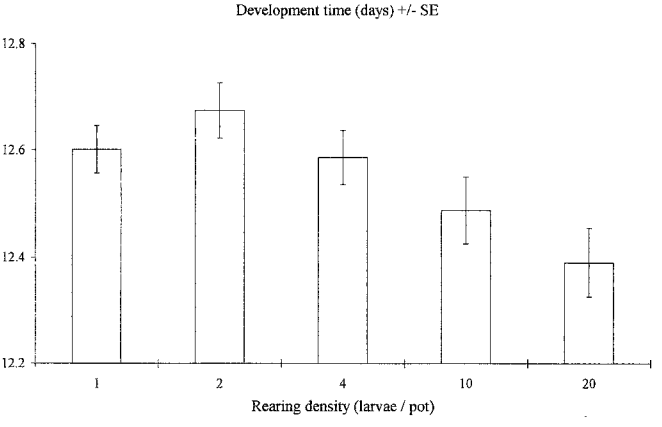

Fig. 5 Larval development time from hatching to fifth instar (± SE), when reared at a range of densities. Numbers of larvae given in Fig. 1

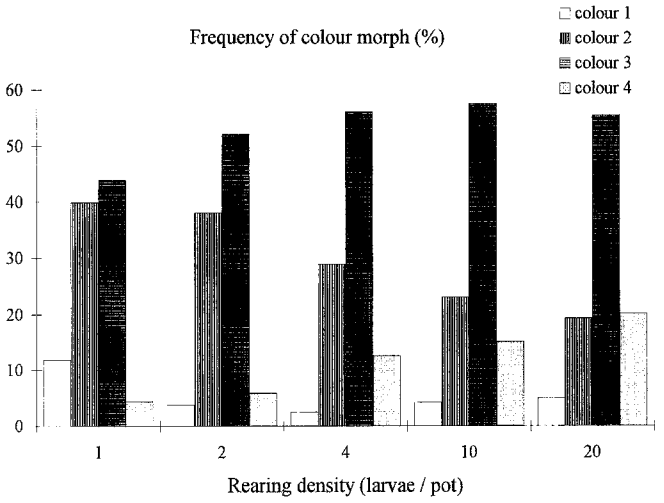

Fig. 6 Frequencies of the four colour morphs of fifth-instar larvae (%), following rearing at a range of five rearing densities. Numbers of larvae given in Fig. 1

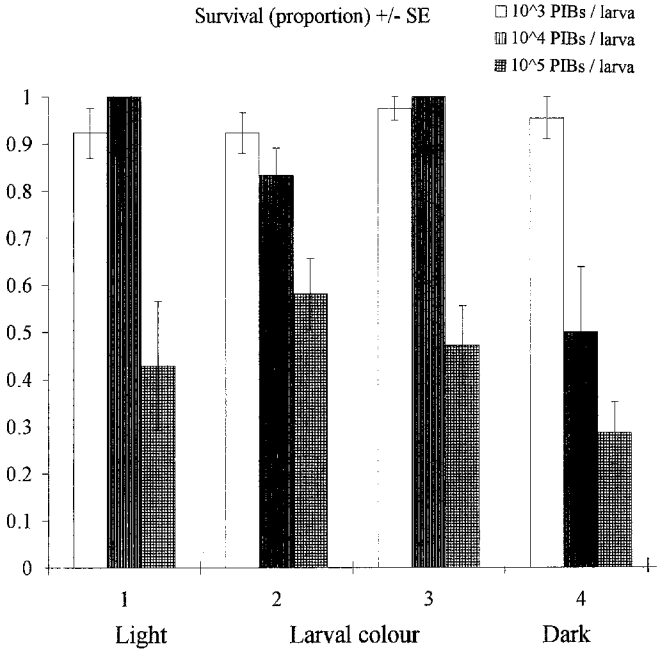

Fig. 7 Survival rates of fifth-instar larvae according to colour, following inoculation with MbNPV at three different dose levels, ± SE estimates calculated using GLIM. Mortalities not due to virus excluded; n = 151, 211, 197 and 194 for colours 1–4, respectively

Development rate did not differ significantly between densities one, two and four. Larval weight when newly moulted to the fifth instar was strongly related to the development time up to that point ($F_{2,\ 369}$ = 24.2, $P < 0.001$); larvae which developed more slowly reached a larger weight before moulting. Mean larval weights were 0.061 g ± 0.004, 0.081 g ± 0.004 and 0.103 g ± 0.004 (SE) for larvae which took 11, 12 and 13, respectively, to develop from hatching to the fifth instar. Larvae which took 14 days to reach the fifth instar are omitted due to the small sample size.

Rearing density had a marked effect on larval melanisation (mean colour per pot) ($F_{4,\ 563}$ = 32.2, $P < 0.001$) The frequency of dark larvae (colours 3 and 4) increased with rearing density, with a corresponding decrease in the frequency of pale larvae (Fig. 6). Larval colour was also related to their development time to the fifth instar, dark larvae developing more quickly, or perhaps, conversely, larvae which developed quickly tending towards greater melanisation: mean time to the fifth instar was 12.87 days ± 0.08, 12.66 ± 0.04, 12.53 ± 0.03 and 12.19 ± 0.06 (± SE) for larval colours 1, 2, 3 and 4, respectively ($F_{1,\ 563}$ = 21.0, $P < 0.001$).

Larval susceptibility to viral infection according to colour

Predictably, mortality increased with virus dose (χ^2_3 = 264, $P < 0.01$ and χ^2_3 = 23.8, $P < 0.01$, fourth and fifth instars respectively). However, in fifth-instar larvae, mortality also varied according to the degree of melanisation, (χ^2_3 = 8.08, $P < 0.01$). Lighter larvae exhibited a lower mortality than darker larvae when averaged over the range of virus doses (Fig. 7). In addition, there was a significant melanisation-dose interaction (χ^2_9 = 26.1, $P < 0.01$) indicating that the mortality-dose response differed according to the degree of melanisation of the larvae. Pale larvae (colours 1 and 2) only showed an increase in mortality at the highest dose (10^5 PIBs per larva) compared to controls, doses of 10^3 and 10^4 PIBs per larva producing no effect on survival. In contrast, dark larvae (colour 4) were more susceptible to the virus at the medium dose, exhibiting a steady increase in mortality with dose, and a survival rate at the highest dose which was lower than the other larval colours (28.6% compared to 42.9, 58.1 and 47.2% for colours 1, 2 and 3 respectively) (Fig. 7).

No significant association was found between the degree of melanisation of fourth-instar larvae and their susceptibility to MbNPV (χ^2_3 = 0.55), although the limited variability of this instar (65.8% of experimental larvae were of colour 2) renders sample sizes small for colours 1, 3 and 4. The weight of larvae which died due to viral infection did not differ from those which survived for either instar (t = 0.41, df = 371 and t = 1.01, df = 751 for fourth and fifth instars respectively).

Discussion

Rearing density has a profound effect on larval physiology, inducing significant variation in all aspects measured in this study, namely susceptibility to disease, weight, colour and rate of development. These relationships are summarised in Table 1. The effect of density on the variables studied is not linear, with clear effects at both very high densities, and when larvae were reared singly, compared to intermediate densities.

Table 1 Summary of the effects of rearing density on susceptibility of *Mamestra brassicae* larvae to viral infection and their development rate, weight and melanisation. Rearing densitites are larvae kept singly (low density), in groups of 2, 4 or 10 per pot (medium density) and in groups of 20 per pot (high density)

	Rearing density		
	Low	Medium	High
Resistance to virus infection	medium	high	low
Development time to fifth instar	medium	high	low
Larval weight at moulting to fifth instar	medium	high	low
Larval colour in the fifth instar	pale	medium	dark

Effects at the highest density

The range of densities naturally achieved by *M. brassicae* larvae has not been examined in detail, but densities of up to 30 larvae/plant commonly occur. In field experiments larvae showed no signs of dispersal at densities of up to 50 per cabbage, and fed side-by-side in the heart of the plant until it became defoliated (D. Goulson, unpublished work). Hence the highest density studied here may not be unrealistic of the situation in the heart of a cabbage on which a batch of eggs has been laid. However, no data are available on the densities reached by *M. brassicae* larvae in natural ecosystems.

At the highest density, larvae moulted to the fifth instar at a lower mean weight, but reached the fifth instar more quickly, compared to larvae reared at medium and low densities. They also exhibited a generally higher degree of melanisation. An inverse relationship between larval density and pupal size has been observed in many species, and appears unrelated to a direct shortage of food. For example Gruys (1971) reported a decrease in pupal size with increasing density in *Bupalus pinarius* (Geometridae) in both the laboratory and field, and even when food supplies were plentiful. In the wild, high larval densities are likely to indicate an abundance of food, but may subsequently lead to depletion of food resources. Developing through the instars more rapidly, at the expense of attaining a large size, reduces the likelihood of starvation should this occur (Iwao 1963). In *M. separata* dark phase larvae have a higher metabolic rate (Shibazaki and Ito 1969). Melanisation increases the body temperature by increasing absorption of radiant energy (Goulson 1994), and may enable activity at lower ambient temperatures. Also dark larvae spend more time basking than pale larvae (Rose 1979). Casey (1993) describes light phase larvae as thermoconformers (adopting an internal temperature close to ambient) and dark phase larvae as thermoregulators (which use behavioural mechanisms to maintain an internal temperature significantly above ambient). We suggest that the switch towards a dark phase may be one of a suite of adaptive responses to intraspecific competition, facilitating greater activity in the scramble for a diminishing food resource. However, some of the observed responses to crowding, particularly the lower weight of larvae, may simply be a result of stress (mediated for example by disturbance during feeding). It would be worthwhile to examine whether larvae responses to reduced food supplies mirror those induced by crowding.

Rhoades (1985) proposes another explanation. He suggests that switches between solitary and gregarious phases in locusts, and between light and dark phases in Lepidoptera, represent a switch between different strategies of herbivory. Solitary (and pale) phases employ a stealthy strategy, avoiding stimulating plant defences by behavioural dispersion and low feeding and metabolic rates. Conversely, gregarious (and dark) phases employ an opportunistic strategy, exploiting periods of environmental stress on hostplants, and swamping plant defences by gregarious feeding and high feeding rates per individual. This explanation predicts discrete (polymorphic) variation between phases, for an intermediate strategy is not viable. *M. brassicae* exhibits continuous variation in the degree of melanisation (Goulson 1994). To my knowledge, there is no evidence that dark phase lepidopteran larvae are more gregarious than pale phase larvae. Hence we suggest that the switch to a darker, faster developing phase in Lepidoptera is best explained by intraspecific competition.

Mortality of *M. brassicae* larvae due to baculovirus infection under field conditions does not vary with instar (at a constant density of larvae and inoculum), probably because the increased dose required to kill later instars is balanced by their greater consumption of food (and contaminating PIBs) (Goulson et al. in press). Hence testing of susceptibility when in the fifth instar is of ecological relevance. Larvae reared at high density were more susceptible to infection and were darker, while at constant rearing density, dark larvae were more susceptible to infection. Also, dark larvae developed more quickly, and rapidly developing larvae were more susceptible to infection. Physiological stress associated with rapid development which occurs at high density, and which is associated with a switch to the dark phase, may pre-dispose larvae to infection. Alternatively, at high population densities the risk of food depletion may outweigh the risk of viral infection from conspecifics, so that expression of resistance is abandoned in favour of rapid development. If resistance has a metabolic cost then it may impede development: strains of *Plodia interpunctella* selected for resistance to a baculovirus exhibited slower development than susceptible strains (Boots and Begon 1993).

Effects of rearing larvae singly

Rearing larvae singly appears to mimic the effects of rearing them at high density: compared to medium densities they were smaller and more susceptible to infection. Although the difference was not significant, larvae also

developed more quickly when reared singly compared to intermediate densities. The greater susceptibility of larvae reared singly compared to intermediate densities cannot simply be because when reared singly larvae were smaller, for both size and density contributed significantly to the mortality analysis (and there was no significant interaction between the two). Decreasing susceptibility with increasing rearing density has been described in *Mythimna separata* (Kunimi and Yamada 1990). We speculate that the presence of other larvae may trigger expression of resistance: if resistance has a cost (Fuxa and Richter 1989, 1990; Boots and Begon 1993) then this cost can be reduced if resistance is only expressed when the risk of infection is high.

Most models of infectious disease population dynamics assume that transmission is directly proportional to the density of susceptible hosts (e.g. Anderson and May 1979, 1981). Although the present study does not examine transmission, the results suggest a mechanism for non-linearity, since susceptibility changes with density. Hochberg (1991b) introduces non-linearity to an Anderson and May (1981) type host-pathogen model, and predicts that increasing susceptibility with density of uninfected hosts (β increasing linearly with the density of susceptible hosts) should stabilise host-pathogen dynamics and facilitate control of the host population by the disease. Conversely, decreasing susceptibility with density, as found by Kunimi and Yamada (1990) and at low densities in this study, destabilises host-pathogen dynamics. Biologically this makes sense, for as the density of susceptible hosts increases so the pathogen becomes less efficient at infecting new hosts. Extrapolation of laboratory results to field conditions must be tentative, but increased susceptibility at high larval densities may stabilise the dynamics of the *M. brassicae*-MbNPV system. Clearly field experiments are necessary to assess this possibility.

Begon (1984) stresses the need to examine the distribution of responses to changing density; individuals within a group respond differently to competition, so that population means may not accurately represent the effects of competition. Slight differences in fitness between individuals tend to become accentuated under high levels of interspecific competition, resulting in a skewing of the distribution of variables related to fitness, noticeably size (for example Obeid et al. 1967; Wilbur and Collins 1973, reviewed in Begon 1984; Uchmanski 1985; but see also Latto 1992 for an alternative view). The commonly accepted explanation is that individuals which are more fit (for example larger) at the onset of competition are little affected by it, while individuals which are less fit suffer more, accentuating the difference. Typically the end result is a large number of unfit (small) individuals and a few large ones. However, I found no such trend. Although there was a significant positive skew at four of the five densities studied, the skew was most pronounced when larvae were reared singly. There was an almost perfectly symmetrical distribution when larvae were reared at an intermediate density (density 4) (Fig. 4).

If we assume that size is related to fitness, perhaps via future fecundity, then it appears that most solitary larvae may be disadvantaged. Possibly occasional contact with other larvae stimulates feeding, while excessive contact with other larvae, as occurred at high rearing densities, may result in significant mutual interference and hence reduced time available for feeding. A clue to the explanation for this result may be provided by truly gregarious species which are often extremely difficult to rear singly (for example *Euproctis chrysorrhoea*, J.S. Cory, unpublished work). Studies of behavioural interactions between larvae are required to assess their role in larval development.

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