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Thermal ecology of western tent caterpillars *Malacosoma californicum pluviale* and infection by nucleopolyhedrovirus

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Abstract. 1. Western tent caterpillars hatch in the early spring when temperatures are cool and variable. They compensate for sub-optimal air temperatures by basking in the sun.

2. Tent caterpillars have cyclic population dynamics and infection by nucleopolyhedrovirus (NPV) often occurs in populations at high density.

3. To determine whether climatic variation might influence viral infection, the environmental determinants of larval body temperature and the effects of temperature on growth and development rates and larval susceptibility to NPV were examined.

4. In the field, larval body temperature was determined by ambient temperature, irradiance, and larval stage. The relationship between larval body temperature and ambient temperature was curvilinear, a property consistent with, but not necessarily limited to, behaviourally thermoregulating organisms.

5. Larvae were reared at seven temperatures between 18 and 36 °C. Larval growth and development increased linearly with temperature to 30 °C, increased at a lower rate to 33 °C, then decreased to 36 °C. Pupal weights were highest for larvae reared between 27 and 30 °C.

6. The pathogenicity (LD_{50}) of NPV was not influenced by temperature, but the time to death of infected larvae declined asymptotically as temperature increased.

7. Taking into account larval growth, the theoretical yield of the virus increased significantly between 18 and 21 °C then decreased slightly as temperatures increased to 36 °C.

8. Control and infected larvae showed no difference in temperature preference on a thermal gradient. The modes of temperature preference were similar to those for optimal growth and asymptotic body temperatures measured in the field on sunny days.

9. Warmer temperatures attained by basking may increase the number of infection cycles in sunny springs but do not protect larvae from viral infection.

Key words. Basking, forest caterpillars, host–pathogen interactions, temperature, thermoregulation, viral infection.

Introduction

Western tent caterpillars *Malacosoma californicum pluviale* (Dyar) (Lepidoptera: Lasiocampidae) undergo cyclical population dynamics with outbreaks occurring every 8–11 years in south-western British Columbia, Canada (Myers, 1993, 2000), and nucleopolyhedrovirus (NPV) plays an important role in driving these outbreak dynamics (Kukan & Myers, 1999). Various authors have noted in other species that larval susceptibility to NPV is sensitive to temperature (Stairs & Milligan, 1979; Kobayashi *et al.*, 1981; Johnson *et al.*, 1982; Mohamed *et al.*, 1985; Reichenbach, 1985; Ribeiro & Pavan, 1994). Long-term population and climatic data suggest that climate may also play an important role in the interaction between western

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tent caterpillars and NPV. There is a positive relationship between NPV prevalence and hours of sunlight during May, the month of maximum larval development (Fig. 1).

The observed relationship between sunlight and disease prevalence appears at first to be counter-intuitive. Ultraviolet (UV) radiation breaks down NPV (Benz, 1987) and other authors have shown that transmission of NPV is lower at the sunny forest edge than in the shaded interior (Roland & Kaupp, 1995); however clouds do not block UV radiation as completely as a forest canopy (Kuchinke & Nunez, 1999) and other factors, such as temperature, could be more important than UV.

Tent caterpillars bask gregariously in the sun and thus increase their body temperature above ambient levels (Knapp & Casey, 1986). Temperature in turn has an important influence on the physiology and ecology of insects (Heinrich, 1981, 1993). This influence extends to various processes including growth (Petersen *et al.*, 2000), development (Gilbert & Raworth, 1996), and susceptibility to pathogens (Caruthers *et al.*, 1992; Ribeiro & Pavan, 1994; Thomas & Jenkins, 1997). Although increasing body temperature is likely to be beneficial to tent caterpillars in terms of growth and development, higher temperatures could be detrimental if they promote the process of viral replication. Furthermore, by reducing variance in body temperature, basking may reduce variance between individuals in disease susceptibility. Heterogeneity in disease susceptibility has a stabilising effect on disease dynamics (Dwyer *et al.*, 1997, 2000). If certain environmental conditions, such as sunlight, are more conducive to basking, climate could influence the variance in larval susceptibility to disease and thus the stability of disease dynamics.

Speed to kill (Johnson *et al.*, 1982), pathogenicity, and yield per host (Ribeiro & Pavan, 1994) are three important viral parameters that may be influenced by temperature. To explore the potential influence of temperature on disease dynamics of western tent caterpillars and NPV, larvae were reared in the laboratory at seven constant temperatures ranging from 18 to 36 °C. At each of these temperatures, the pathogenicity (LD_{50}) of NPV was measured, together with its incubation time, and the growth and development rates of tent caterpillar larvae. Theoretical pathogen yield per host was estimated based on speed to kill and larval growth rates. The results are related to the temperature preference of control and infected larvae in a thermal gradient and to the influence of climatic variables on larval body temperature.

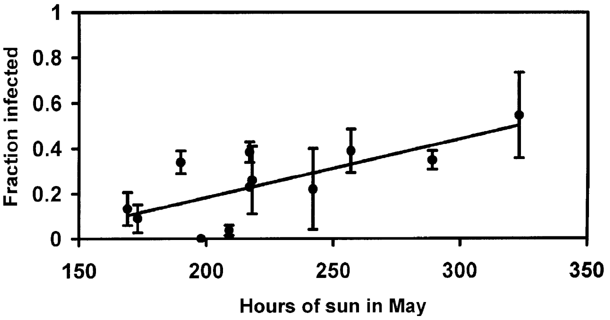

Fig. 1. The relationship between sunlight and NPV prevalence (± SE) for 12 years of data on four populations of western tent caterpillar in British Columbia (prevalence = −33.58 + 0.26 × hours of sun in May, $r^2 = 0.5$, $P < 0.01$). Prevalence data were taken from Kukan and Myers (1999) and Myers (2000). Hours of sunlight are from the Environment Canada weather station at Vancouver International Airport.

Materials and methods

Field determinants of larval body temperature

Field observations on body temperatures of a total of 96 western tent caterpillar larvae were collected on eight occasions between 12 May and 16 June 2000 at three sites in the vicinity of Vancouver, British Columbia, Canada (49°11′50″N, 123°10′57″W): Westham Island, the University of British Columbia Campus, and Cypress Mountain. To avoid pseudo-replication, only one larva per group was measured. On each occasion, ambient temperature (T_a), larval body temperature (T_b), irradiance, the developmental stage of the larva, and the size of the group with which it was associated were recorded.

Ambient and body temperatures (°C) were measured using a digital thermometer with a thermistor on a wire probe (Fisher Scientific, Vancouver, British Columbia). Ambient temperature was measured in the open, 2 m above the ground and 1 m away from the focal larva. Larval body temperature was measured by placing the thermistor against the dorsal side of the larva. Knapp and Casey (1986) found no difference between this technique and the more intrusive technique of stabbing the larva with a thermistor encased within a hypodermic needle. Irradiance (photon μmol $m^{-2} s^{-1}$) was measured using a photometer (LI-COR, LI-189, Li-Cor, Lincoln, Nebraska) on the most direct path of natural light to the larva. A hand-held ruler was used to estimate the average diameter of a larval cluster and thus to estimate group size area (cm^2).

Multiple linear regression with backwards elimination was used to determine which independent variables influenced larval body temperature significantly. It was ensured that the assumptions of homogeneous variance and normality of residuals were met. The shape of the function between ambient temperature and body temperature was determined by a cubic spline with normal errors, a form of non-parametric regression that makes no *a priori* assumptions about the shape of the function (Schluter, 1988). All cubic splines (see below) were generated using glmsWIN 1.0 (Schluter, 2000). Standard errors and confidence intervals were estimated from 1000 bootstrap replications. Confidence intervals were used to evaluate the significance of the relationship. Results for all cubic splines are reported with figure legends stating: *n*, the number of individual observations; λ, the smoothing parameter; and the effective number of parameters, which is a description of the complexity of the function (Schluter, 2000). λ was chosen in order to minimise the generalised cross-validation score. This value of λ approximately minimises the sum of squared deviations between the estimated and true functions (Schluter, 1988).

Egg mass collection and larval rearing

Larvae for all experiments were obtained from egg masses collected in February and March 2000 and 2001 at Westham Island or at the University of British Columbia campus. The egg masses were stored at 4 °C for a maximum of 3 months. To eliminate any potential contamination with NPV, egg masses were washed in a 6% solution of sodium hypochlorite for approximately 2 min, until most of the spumaline coat was dissolved (Fitzgerald, 1995). They were then rinsed thoroughly with running water, set out to dry, and left to hatch at room temperature in 300 ml paper cups with plastic lids. The time interval between removal from the refrigerator and hatching was ≈5 days.

Before use, larvae were kept in the laboratory at room temperature and supplied daily with a diet of field-collected red alder leaves. Leaves were washed with a 10% bleach solution and rinsed thoroughly with water. Leaves were kept in floral water picks to maintain their turgor. Cups were cleaned of faecal pellets and dry or decaying leaves as needed.

Growth, development, and pupal size

To measure growth and development rates, 20 larvae were reared from the third stadium to pupation at each of the following temperatures: 18, 21, 24, 27, 30, 33, and 36 °C. Larvae were reared individually in 200 ml cups and fed red alder leaves in water picks. They were weighed prior to being placed in the growth chambers, again 5–6 days later, and at pupation. Larvae were checked daily for pupation. Growth rate for each individual larva was measured between the first two weighings as:

$$\mathrm{gr} = \ln\left(\mathrm{weight}_{\mathrm{final}}/\mathrm{weight}_{\mathrm{initial}}\right)\,\mathrm{time}^{-1} \tag{1}$$

Development rate was measured as the reciprocal of days between the third stadium and pupation. Individual growth, development, and pupal weight curves were fitted as functions of temperature using cubic splines with normal errors (Schluter, 1988, 2000). Sex was used as a categorical covariate for developmental rates and pupal weights. Sex was used as a covariate for growth rate because not all larvae survived to pupation, the first stage at which they can be sexed easily.

Larval temperature preference

Larval temperature preference was measured as indexed by body temperature on a radiant-light thermal gradient in spring 2000 and on a dark thermal gradient in spring 2001. The light gradient consisted of a cardboard platform (50 × 58 cm) with a 60-W light bulb in the centre. Larvae were inoculated with a virus dose of 10 000 occlusion bodies 4 days before testing. Prior to placing them on the thermal gradient, larvae were held at room temperature and fed leaves every morning. During the 4 days post inoculation, 18 infected larvae and 17 control larvae were placed individually at an arbitrary location on the gradient. Larvae were allowed to roam in the area for 5 min, after which their body temperature (T_b) was measured using a thermocouple connected to a digital thermometer. As with field measurements, the thermocouple was placed against the dorsal side of the larvae until the temperature stabilised (≈2 s). The order in which larvae were placed on the gradient was determined randomly and larvae were measured only once. Air temperatures on the surface of this gradient ranged from 20 to 58 °C.

The dark thermal gradient consisted of a metal tray with a hot plate under one end and an ice pack under the other. The tray was covered with foil to block out light but with enough space to allow larvae to move. This design removes the confounding effect of light preference inherent in the previous experiment. Thirty-two larvae that had been inoculated with 10 000 viral occlusion bodies and 32 controls were placed individually at the centre of the gradient and left for 10 min before their body temperature was measured as described above. Air temperatures on the surface of this gradient ranged from 19 to 61 °C. Differences in the temperature preference of infected vs. uninfected larvae were tested for using *t*-tests.

Effect of temperature on larval–virus interactions

The effect of temperature on virus pathogenicity was measured with an analysis of the lethal dose 50 (LD_{50}). Third-stadium larvae were reared at seven temperatures (18, 21, 24, 27, 30, 33, and 36 °C). At each temperature, larvae were fed one of seven doses of NPV (± SE): 0 (controls), 236 (±52), 1462 (±260), 2863 (±203), 5025 (±335), 11 175 (±295), and 35 188 (±3358) occlusion bodies. These doses are based on quantifications of dilutions that were produced to increase approximately exponentially from one level to the next.

Virus dilutions were prepared from a mixed stock of frozen larvae killed by virus in the laboratory the previous year. Occlusion bodies were isolated through two series of centrifugation: slow (1000 r.p.m. for 30 s) to remove non-virus particles, and fast (14 000 r.p.m. for 20 min) to form the virus pellet. The pellet was resuspended and subsequent dilutions were quantified using a haemocytometer.

Each temperature and dose combination was replicated twice. A replicate consisted of eight to 10 larvae from different egg masses. Larvae were infected upon reaching the third stadium. Larvae were starved for 24 h before being fed 5 μl of dH_2O with virus solution on a leaf disc. Larvae that failed to eat >75% of their disc were not used. Larvae were inoculated individually but subsequently reared gregariously; because of the gregarious nature of the species,

Table 1. ANOVA summary for model of environmental factors influencing western tent caterpillar larval body temperature (model $r^2 = 0.82$).

Source	d.f.	Sum of squares	Mean square	*F* ratio	*P*(*F*)
Regression	3	4493.1277	1497.71	135.3090	<0.0001
Irradiance	1	772.9947		69.8354	<0.0001
Ambient temperature	1	1036.3937		93.6319	<0.0001
Stadium	1	144.9263		13.0932	0.0005
Error	92	1018.3306	11.07		
Total	95	5511.4583			

it is difficult to successfully rear larvae individually in the laboratory, particularly during earlier stages of development. Larvae were monitored for virus mortality and fed daily a diet of red alder leaves in floral water picks.

Due to the delay between host infection and death, virus mortality occurs in cycles. The first cycle is due to the experimental inoculation, whereas later cycles are due to infection from the release of infective occlusion bodies at the death of the initially infected larvae. LD_{50}s were determined from the mortality that occurred during the first disease cycle. The end of the first disease cycle was defined as the day on which mortality decreased to a minimum before increasing again to begin the second cycle. Indispensable mortality, the difference between mortality at each dose and control mortality, was transformed in each replicate to probit values using:

$$\text{probit} = \ln\left(\frac{\text{mortality}}{1 - \text{mortality}}\right) \tag{2}$$

Probit mortality was then regressed against $\log_{10}(\text{dose})$. The LD_{50} is the dose at which the predicted probit is 0 (i.e. mortality = survival). Ninety-five per cent confidence intervals were calculated around the predicted LD_{50} for each temperature following the method of Zar (1996):

$$\mathrm{CI} = \bar{x} + \frac{b(-\bar{y})}{b^2 - t^2 s_b{}^2} \pm \frac{t}{b^2 - t^2 s_b{}^2} \sqrt{s_{y \cdot x}{}^2 \left[\frac{\bar{y}^2}{\sum x^2} + (b^2 - t^2 s_b{}^2)\left(1 + \frac{1}{n}\right)\right]} \tag{3}$$

where x is dose, y is probit mortality, b is the slope of the relationship, t is $t_{0.05(2)\nu}$, s_b is the standard error of the slope, $s_{y \cdot x}$ is the standard error of the estimate, and n is the number of replicates. For each replicate, the mean time to death was calculated from viral infection (incubation time) during the first disease cycle. A cubic spline was fitted with normal errors to virus incubation time as a function of temperature (Schluter, 1988, 2000).

Theoretical yield was determined by using predicted growth rate (gr) and time to death (td) in the following equation:

$$y = e^{\mathrm{gr} \times \mathrm{td}} \tag{4}$$

This assumes that infected larvae at each temperature grow at the same relative rate as healthy larvae, that virus volume is proportional to larval size, and that it grows exponentially over time in each tissue. This measure may not be accurate if larvae die at the same viral load, regardless of temperature.

Results

Field determinants of larval body temperature

In the field, all variables measured, except group size, influenced larval body temperature significantly (Table 1). As ambient temperature, irradiance, and larval stadium increased, so did larval body temperature. The effect of stadium was not confounded by time of the growing season as various developmental stages were observed during each excursion. Larval body temperature was as high as 21 °C above ambient. There was a sigmoid relationship between body temperature and ambient temperature (Fig. 2). As expected for a behaviourally thermoregulating organism, body temperature plateaus as higher ambient temperatures are reached (Blanford *et al.*, 1998; Blanford & Thomas, 2000).

Growth, development, and pupal size

Larval growth rate increased curvilinearly with temperature (Fig. 3a). Development rates increased linearly between 18 and 30 °C, increased at a lower rate to 33 °C, then

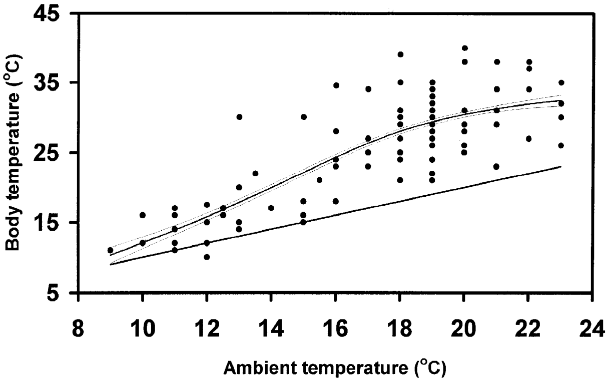

Fig. 2. Predicted larval body temperature as a function of ambient temperature (±95% CI) fitted with a cubic spline ($n = 96$, $\lambda = 1.4$, effective number of parameters = 3.9). The solid straight line is where body temperature equals ambient temperature.

decreased at 36 °C (Fig. 3b). Female pupae were larger than males (Fig. 4). No females survived to pupation at 36 °C. In general, there were fewer females than males at pupation. The primary sex ratio was unknown, but the higher number of males may have been caused by differential survival. Lower female survival may have been associated with their longer developmental time and thus prolonged exposure to uncontrolled mortality factors, such as bacterial infection (Fig. 3b). Pupal weights as a function of temperature were described with a cubic spline with sex as a categorical covariate (Fig. 4). The largest pupae occurred for larvae reared between 27 and 30 °C.

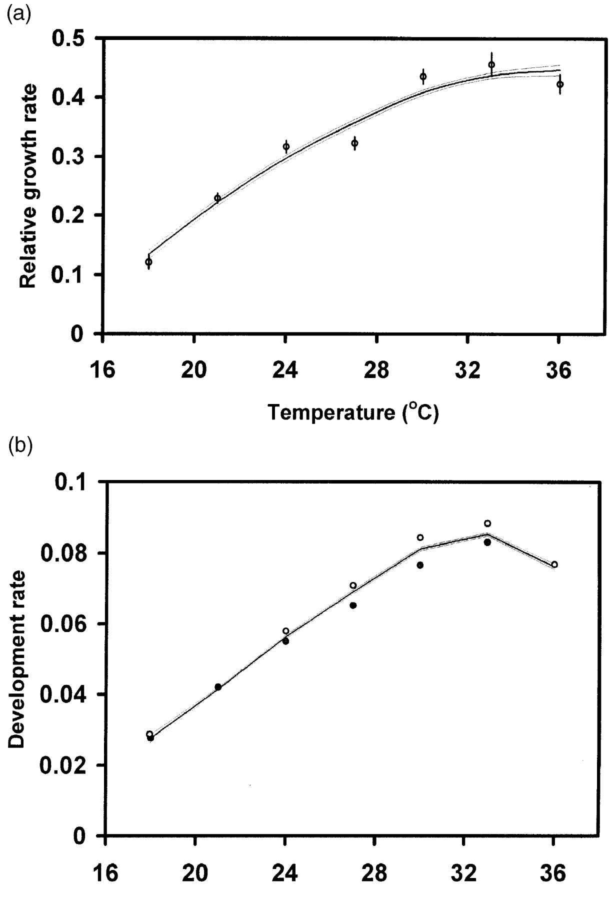

Fig. 3. (a) Larval growth and (b) development rates (mean ± 95% CI) fitted with cubic splines as a function of temperature (growth: $n = 136$, $\lambda = 3.1$, effective number of parameters = 3.4; development: $n = 89$, $\lambda = -0.6$, effective number of parameters = 5.7). ○ male development rates, ● female development rates.

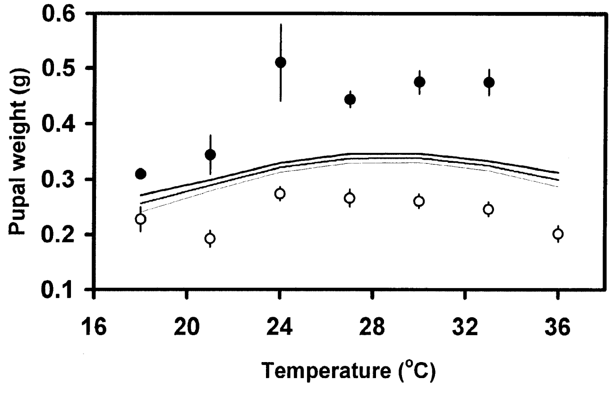

Fig. 4. Mean pupal weights (± SE) for larvae reared at seven different temperatures (○ females, ● males). The curve indicates the cubic spline fitted to the data (±95% CI) using sex as a categorical covariate ($n = 89$, $\lambda = 2.8$, effective number of parameters = 3.2).

Larval temperature preference

There was no significant difference in the temperature preference (mean ± SE) of infected (24.8 ± 1.3 °C) and uninfected (26.2 ± 1.3 °C) larvae in a *radiant light* thermal gradient ($t = 0.80$, d.f. = 33, P = NS, LSD = 6.7 °C, Power = 0.12) or in the dark thermal gradient (26.9 ± 1.1 °C infected vs. 27.6 ± 1.1 °C uninfected, $t = 0.41$, d.f. = 62, P = NS, LSD = 4.8 °C, Power = 0.08). Power was low for both tests. The frequency distribution of larval body temperatures in the radiant light thermal gradient is shown in Fig. 5 and for the dark thermal gradient in Fig. 6. Control body temperature distributions tended to have a narrower mode than those for inoculated larvae in the dark thermal gradient. The modal temperature for control larvae was between 28 and 31 °C.

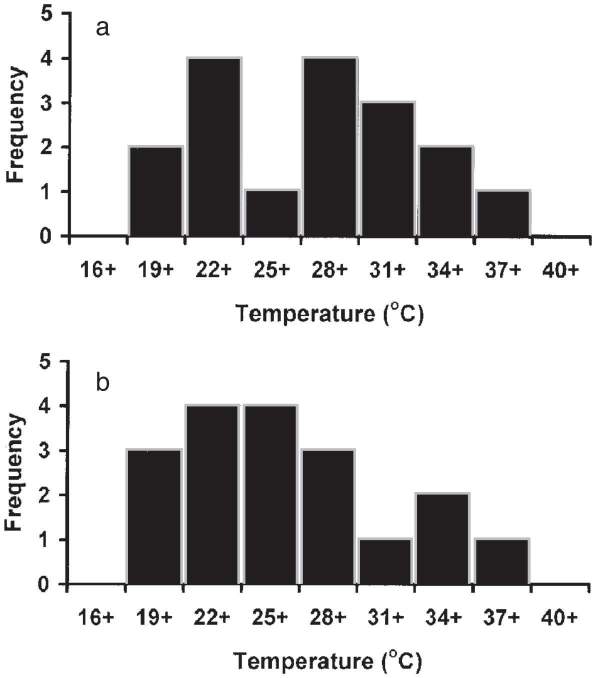

Fig. 5. The frequency distribution of body temperatures for (a) control and (b) inoculated larvae after 5 min on a radiant light thermal gradient.

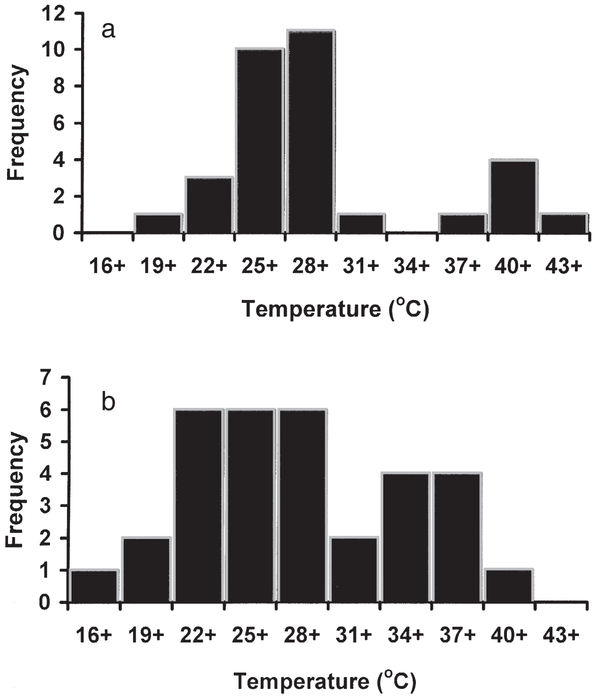

Fig. 6. The frequency distribution of body temperatures for (a) control and (b) inoculated larvae after 10 min on a dark thermal gradient.

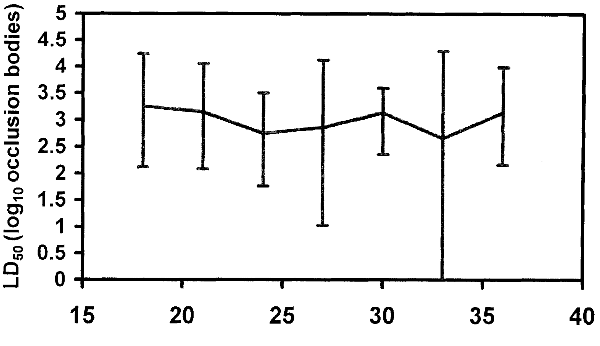

Fig. 7. LD_{50}s (±95% CI) of larvae inoculated with NPV.

Effect of temperature on larval–virus interactions

There was no mortality at any temperature in the controls, and no evidence that temperature influenced the pathogenicity (LD_{50}) of NPV (Fig. 7), however temperature had a strong influence on the incubation time of the virus (Fig. 8). As temperature increased, time to death decreased asymptotically. Estimates of theoretical virus yield per larva increased significantly between 18 and 21 °C, remained relatively constant, and decreased slightly, but not significantly, to 36 °C (Fig. 9).

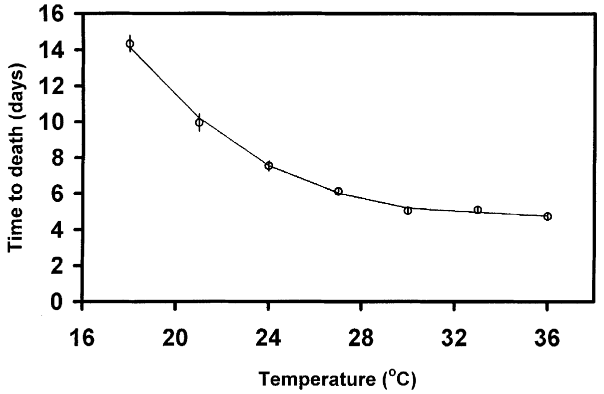

Fig. 8. Cubic spline fit of mean time to death (±SE) for larvae dying of NPV infection as a function of rearing temperature ($n = 82$, $\lambda = 0$, effective number of parameters = 5.2).

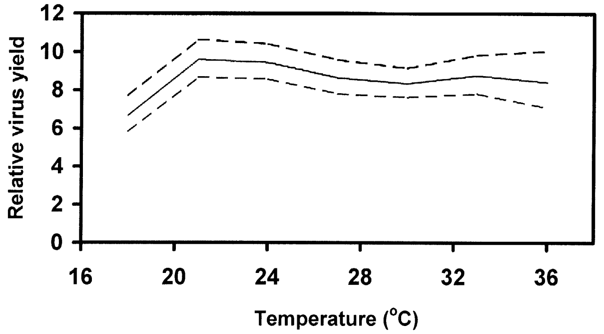

Fig. 9. Theoretical virus yield (±95% CI) as a function of temperature, relative to larval weight at the time of infection.

Discussion

The body temperature of insects is a complex function of behaviour and the abiotic environment (Knapp & Casey, 1986; Casey *et al.*, 1988; Caruthers *et al.*, 1992; Lactin & Johnson, 1998). Body temperature in turn determines the outcome of various physiological and ecological processes (Heinrich, 1981, 1993). Insects capable of thermoregulating within certain environmental constraints must compromise among these processes (Knapp & Casey, 1986). These experiments demonstrate that both the development of western tent caterpillars and their interaction with NPV are sensitive to temperature. In turn, behaviour and the environment influence larval body temperature. Therefore, both insect behaviour and the abiotic environment have implications for the outcome of population and disease dynamics.

Growth and development rates in insects tend to increase linearly with temperature within the range of temperatures experienced normally in the field (Gilbert & Raworth, 1996) and these results agree. The ratio of growth to development rates determines adult size and maximum potential reproductive output. Small changes in this ratio can result in large changes to adult size of some insects (Gilbert & Raworth, 1996). For western tent caterpillars, the optimal temperatures for maximising pupal size are between 27 and 30 °C. The modal temperatures selected by control larvae in the laboratory were within this range. The upper temperatures of this range are just below the optimal temperatures for development (Fig. 3b) and coincide closely with the temperature at which the body temperature and the ambient temperature relationship begins to reach its asymptote. The curvilinear relationship is not sufficient evidence to conclude that western tent caterpillars are thermoregulating (Hertz *et al.*, 1993), but the close match between developmental optimum and the asymptote of temperatures measured in the field is consistent with the hypothesis that larvae are thermoregulating.

Thermal sensitivity in insects can respond rapidly to natural selection (Huey *et al.*, 1991). The relatively constant pupal size of western tent caterpillars between 24 and 33 °C may indicate an adaptation to the highly variable environment in which western tent caterpillars develop. Tent caterpillars can exploit sunlight to elevate their body temperature (Knapp & Casey, 1986; Casey *et al.*, 1988; Joos *et al.*, 1988; Table 1). By elevating their body temperature when it is sunny, tent caterpillars can reduce the time to pupation while still maintaining maximal pupal size; however the body temperatures of non-basking larvae will be close to ambient temperatures, which for the months of April, May, and June reach an average daily maximum of 12.7, 16.3, and 19.3 °C (Environment Canada, 2001). Therefore, for much of the developmental period, larvae will be at sub-optimal temperatures. Overcast conditions and associated colder temperatures will increase the developmental period but apparently will not reduce pupal size greatly.

Unlike growth and development of larvae, the pathogenicity of NPV was insensitive to temperature within the range explored. This is inconsistent with various studies that considered different types of NPV infection. High temperatures (30 and 35 °C) induced latent NPV infections in *Neodiprion sertifer* (Geoffroy) (Hymenoptera: Diprionidae) larvae, whereas intermediate temperatures (25 °C) did not (Mohamed *et al.*, 1985). Temperatures above 38 °C inactivated non-occluded NPV in *Galleria mellonella* (Linnaeus) (Lepidoptera, Pyralidae) larvae (Stairs & Milligan, 1979). Between 17 and 37 °C, larval mortality from NPV infection in *Diatraea saccharalis* (Fabricius) (Lepidoptera: Pyralidae) increased with temperature (Ribeiro & Pavan, 1994). In silkworm *Bombyx mori* (Linnaeus) (Lepidoptera: Bombycidae), the development of NPV infections in pupae was inhibited at high temperatures (35 °C) (Kobayashi *et al.*, 1981). The LD_{50} of NPV to *Choristoneura occidentalis* Freeman (Lepidoptera: Tortricidae) larvae decreased with temperature between 15 and 30 °C (Reichenbach, 1985).

The lack of a temperature effect in this system may not be unusual. The confidence intervals for LD_{50}s in the work of Reichenbach (1985) overlap at all temperatures. Other studies only found an effect at temperatures >30 °C, which are higher than are experienced normally by the host (Stairs & Milligan, 1979; Kobayashi *et al.*, 1981; Mohamed *et al.*, 1985). In contrast, the pathogenicity of NPV to western tent caterpillars was insensitive to temperatures within the range that is experienced normally in the field.

The insensitivity of NPV LD_{50}s to temperature does not exclude the possibility that temperature may influence disease dynamics. Both the incubation time of the virus and its theoretical yield were sensitive to temperature variation, particularly over the range of 18–24 °C, which is typical for the developmental period of larvae. These observations are similar to those of Kelly and Entwistle (1988) with viral production in *Mamestra brassicae*. The time to death in their experiments was on average $\approx$3 days faster at 22.5 °C than at 20 °C but the production of virus at both of these temperatures was similar and greater than at 25, 27.5, or 30 °C. Thus small increases in larval temperatures when temperatures are relatively low could influence the rate of production of virus without reducing the amount of virus produced. In field populations, the incubation time of the virus could determine how many additional disease cycles occur within a season (Rothman, 1997). Secondary and tertiary disease cycles can infect a large proportion of the host population even when initial infection rates are low. In biological control with pathogens, secondary cycles can emulate the environmental persistence of chemical insecticides (Thomas *et al.*, 1995). Shorter virus incubation times could also mean reduced damage caused by pests, depending on how feeding rates are influenced by temperature.

No change in larval temperature preference, as indexed by body temperature on a thermal gradient, was detected in response to infection. This contrasts with the behaviour of some grasshoppers infected with fungal disease (Blanford *et al.*, 1998). In the case of western tent caterpillars, however, any change in temperature preference would not influence their susceptibility (LD_{50}) to the virus. The frequency distribution of body temperatures for a majority of caterpillars was in the range of 22–31 °C, a range that approximates both the field asymptote of $\approx$28 °C and the optimum temperature for development, 30–33 °C. These results are consistent with, but do not confirm, the hypothesis that western tent caterpillars are behavioural thermoregulators; however the results on temperature preference should be interpreted with caution, as statistical power was low and preferences were only measured during the first 4 days post inoculation. Future experiments should consider temperature preferences during the later portion of the infection period.

Further studies are necessary to determine the role of sunlight in the disease dynamics of western tent caterpillars. Based on the LD_{50} data (Fig. 7), it is unlikely that field observations of higher disease prevalence during sunnier years are caused by changes to virus pathogenicity, however an increase in the number of disease cycles during sunnier years could influence the occurrence of epizootics of NPV in field populations of western tent caterpillars. Temperature and other factors influencing the development rates of lepidopteran larvae and their pathogens may be important in determining the onset of disease epizootics.

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