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Body temperature of the parasitic wasp *Pimpla turionellae* (Hymenoptera) during host location by vibrational sounding

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Abstract. The pupal parasitoid *Pimpla turionellae* (L.) uses self-produced vibrations transmitted on the plant substrate, so-called vibrational sounding, to locate immobile concealed pupal hosts. The wasps are able to use vibrational sounding reliably over a broad range of ambient temperatures and even show an increased signal frequency and intensity at low temperatures. The present study investigates how control of body temperature in the wasps by endothermic mechanisms may facilitate host location under changing thermal environments. Insect body temperature is measured with real-time IR thermography on plant-stem models at temperature treatments of 10, 18, 26 and 30 °C, whereas behaviour is recorded with respect to vibrational host location. The results reveal a low-level endothermy that likely interferes with vibrational sound production because it occurs only in nonsearching females. At the lowest temperature of 10 °C, the thoracic temperature is 1.15 °C warmer than the ambient surface temperature whereas, at the high temperatures of 26 and 30 ° C, the wasps cool down their thorax by 0.29 and 0.47 °C, respectively, and their head by 0.45 and 0.61 °C below ambient surface temperature. By contrast, regardless of ambient temperature, searching females always have a slightly elevated body temperature of at most 0.30 °C above the ambient surface temperature. Behavioural observations indicate that searching females interrupt host location more frequently at suboptimal temperatures, presumably due to the requirements of thermoregulation. It is assumed that both mechanisms, producing vibrations for host location and low-level endothermy, are located in the thorax. Endothermy by thoracic muscle work probably disturbs signal structure of vibrational sounding, so the processes cannot be used at the same time.

Key words. Behaviour, echolocation, host location, parasitoid, thermoregulation, vibrational sounding.

Introduction

Nearly all biological processes are influenced by temperature and, the smaller an organism is, the more its body temperature is influenced by ambient conditions (Willmer *et al.*, 2000). Insects are mostly ectotherms and possess physiological and behavioural traits that are well adapted to the temperatures of their habitats (Heinrich, 1993; Willmer *et al.*, 2000; Kroder *et al.*, 2006). There are two potential strategies in insects with respect to changes in ambient temperature: either they are thermoconformers, so that their body temperature fluctuates with ambient temperature, or they are thermoregulators, and can adjust their body temperature to some extend independently of ambient conditions (Sanborn, 2005). Several ectotherms, for example, elevate their body temperature by basking (i.e. absorbing solar radiation) or microhabitat selection, but also avoid overheating by selecting cooler sites (Heinrich, 1993; Kührt *et al.*, 2005; Samietz *et al.*, 2005). In particular, several hymenopteran species are able to raise and maintain

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their body temperature above the environmental temperature without external heat sources (Esch, 1988; Heinrich, 1993; Stabentheiner *et al.*, 2003a). Such a temporary endothermy is referred to as heterothermy and is well studied in honeybees and bumblebees (Heinrich, 1981; Stabentheiner *et al.*, 2003b), but is also found in other insect taxa such as Homoptera (Sanborn, 2001) and Lepidoptera (Esch, 1988).

Temperature also affects the ability of insects to produce and receive airborne and substrate-borne vibrations (Sanborn, 2005). In parasitoids, vibrational cues are known to be used during host location (Meyhöfer *et al.*, 1997; Broad & Quicke, 2000). Host location represents an essential element of reproduction and the efficiency of the mechanisms employed is vital in a parasitoid's life cycle. Vibrational sounding is a specific mechanosensory mechanism used in several ichneumonid and orrussid parasitoids to locate concealed hosts (Wäckers *et al.*, 1998; Broad & Quicke, 2000; Kroder *et al.*, 2006). Similar to the principles of echolocation, female wasps transmit self-produced vibrations onto the substrate and receive the reflected signals, allowing them to locate and recognize their potential hosts (Otten *et al.*, 2002; Fischer *et al.*, 2003, 2004). The subgenual organs as mechanoreceptors receive the vibrations (Otten *et al.*, 2002), and it is assumed that the vibratory signals originate from muscles in the insect's body. The temperate pupal parasitoid *Pimpla turionellae* (L.) use vibrational sounding successfully over a temperature range of 8–28 °C (Samietz *et al.*, 2006). Behavioural assays confirm that the performance of vibrations by the female is less at higher and lower ranges of habitat temperatures (Samietz *et al.*, 2006).

Although vibratory or acoustic signals in ectotherms are usually slower and less intense at low temperatures, vibration measurements by laser Doppler vibrometry in *P. turionellae* reveal faintly increasing carrier frequency and intensity at lower temperatures (Kroder *et al.*, 2007). Such an increase indicates a higher muscle activity and an efficient production mechanism at low temperatures. This could be accomplished by maintaining the body temperature independent from ambient temperature as in thermoregulators. Accordingly, *P. turionellae* may use temporary endothermy by metabolic heating to perform vibrational sounding successfully. The present study examines whether parasitoid females are able to raise or lower their body temperature in relation to ambient thermal conditions and thereby optimize host location by vibrational sounding. Using real-time infrared (IR) thermography, insect body surface temperatures are measured on the head, thorax and abdomen without any intrusive contact. During thermography measurements, the behaviour of the test females is observed and recorded with respect to general activity and host searching performance.

Materials and methods

Parasitoid rearing

The laboratory strain of *P. turionellae* was reared on pupae of *Galleria mellonella* (Lepidoptera). Adults were confined in Plexiglas containers (25 × 25 × 25 cm) fed with honey and water and were allowed to mate. The wasps were kept at 15 °C, 70% relative humidity and under an LD 16 : 8 h photoperiod. Host pupae were offered three times a week for oviposition and host feeding. Parasitized pupae were stored at 24 °C, 60% relative humidity and a under an LD 16 : 8 h photoperiod of until emergence of adults.

Experimental set-up

The body temperature of *P. turionellae* females was measured in four temperature treatments at 10 °C ($n = 15$), 18 °C ($n = 17$), 26 °C ($n = 15$) and 30 °C ($n = 15$). The experiments were performed under fluorescent light and controlled conditions in a climatic exposure test cabinet (Ehret KLT/04, Emmendingen, Germany). The test females were aged 1–4 weeks and observed individually in a plexiglass box (196 × 88 × 80 mm) within the cabinet. Because metabolic heating capacity depends on body size, the body weight of each female was determined with a high-precision balance (Mettler AT261 Delta Range, Göttingen, Germany; precision 0.01 mg). A data logger (Almemo 2590-8; Ahlborn, Holzkirchen, Germany) recorded ambient air temperature and relative humidity.

Females of the parasitoid attempt innately to insert their ovipositor into various hollow rounded substrates. A paper cylinder (length 55 mm, diameter 8 mm) made of airmail paper (45 g m^{-2}; ELCO Atlantic Clipper Air Mail, Allschwil, Switzerland) containing a cigarette filter (15 × 8 mm; Gizeh, Gummersbach, Germany) as solid section was used to imitate the hidden host pupa. This experimental approach excluded chemical and visual cues from mechanosensory cues for host location. Hence, directed orientation could be attributed to vibrational sounding alone. Such plant-stem models have been used successfully in a number of studies on host location in these insects (Wäckers *et al.*, 1998; Fischer *et al.*, 2001, 2003; Otten *et al.*, 2001). The model was fixed horizontally on the black backboard and black bottom of the box. Because of the rather small mass of the insects (37.5 ± 6.6 mg; overall mean ± SD, $n = 62$) their body temperature was measured thermographically (Stabentheiner & Schmaranzer, 1987). An IR thermography camera sensitive at wavelengths of 7–12 μm (ThermaCam SC2000 NTS; Flir Systems, Danderyd, Sweden) was focused directly on the parasitoid during the trials from a distance of 20 cm. The camera transformed the thermal radiation emitted from the insects' surface into electrical impulses, which were amplified to produce the so-called thermogrammes consisting of images showing temperature in shades of grey or in colour steps (Stabentheiner & Schmaranzer, 1987; Stabentheiner *et al.*, 2003a, b). IR images were stored digitally at a rate of 5 Hz on a DOLCH FlexPac computer (Kontron, Echingen/München, Germany). A self-constructed (A. Stabentheiner) reference source with a preset temperature of approximately 2 °C above ambient temperature was placed 0.5 cm above the right end of the model. The surface temperature of the reference source was measured to the nearest 0.3 °C by the data logger, and by the IR camera. The difference between both measurements was used to calibrate the IR camera and enabled an absolute thermographic measurement accuracy greater than 0.7 °C. Differences between body parts and ambient surface temperature were determined with a resolution of 0.1 °C. Temperature measurements were untertaken with a self-written Excel (VBA) macro (Microsoft Corp., Redmond, Washington) that controlled the IR analysis software (Agema Research 2.1, Flir Systems) and extracted temperature and humidity data from the logger files. The surface temperature of the head, thorax and abdomen of each female was measured, as well as the surface temperature at two different points on the paper roll model as ambient surface temperature. Thermographic temperature measurements were calibrated using an IR emissivity of 0.97 of the wasp cuticle (Stabentheiner & Schmaranzer, 1987; Kovac & Stabentheiner, 1999), 0.92 of the paper surface and 0.91 of the reference source. Temperature analysis was performed in searching females at three different times after at least 30 s of a certain behaviour (i.e. antennating or probing; defined below), and on images with pausing females at one point in time after at least 3 min of that behaviour.

Behaviour on plant-stem model

Simultaneously with the IR thermography, the behaviour of the searching wasps was recorded using the software 'The Observer 3.0' (Noldus Information Technology, Wageningen, the Netherlands). Observations of host location were divided into five behavioural states. It was noted whether or not the wasps fanned their wings because fanning probably corresponds with endothermic muscle shivering. The five behavioural states have been defined previously by Fischer *et al.* (2004) for the ichneumonid wasp, *Xanthopimpla stemmator*:

> *Antennating*: a pronounced surface antennation of the searching individual. Vibrational sounding is indicated by this behavioural state during which the females transmit pulses via the antennae and receive reflected signals through subgenual organs in their tibiae (Henaut & Guerdoux, 1982; Otten *et al.*, 2001).
>
> *Probing*: the female bends the abdomen to place the ovipositor tip on the substrate surface when she is standing still or walking slowly. This behavioural state occurs often alternately with antennating and surface antennation is usually continued during probing.
>
> *Insertion*: the ovipositor is inserted into the plant-stem model. This behavioural response indicates host location (Meyhöfer *et al.*, 1997; Samietz *et al.*, 2006) and occurs usually subsequent to probing.
>
> *Cleaning*: cleaning of wings or hind legs. This behaviour interrupts the typical searching behaviour on the plant-stem model (i.e. antennating, probing and insertion).
>
> *Pausing*: no visible activity at all.

Wing fanning was recorded simultaneously. It appears frequently at slow or high speed during the three behavioural states of searching as well as during cleaning, but not during pausing. The recordings were carried out for a maximum of 10 min but were discontinued sooner if the individual left the model for more than 1 min or remained in the behavioural state of pausing for more than 4 min.

Data analysis

For statistical comparisons, the differences between the insects' temperature of head, thorax and abdomen, and the average ambient surface temperature was calculated at each measuring point (ΔT_H, ΔT_T and ΔT_A, respectively). Systematic errors were eliminated because all of these parameters were measured in the same thermographical image at each time.

Significant deviation of insect body temperature from the ambient surface temperature was determined by one-sample *t*-test. Effects of different temperature treatments and body weight on ΔT_H, ΔT_T and ΔT_A of searching and pausing females were tested using analysis of covariance (ANCOVA) with multiple comparisons applying the Sidak post-hoc test. Subsequently, temperatures of searching and pausing females were compared at each temperature by ANCOVA.

Regarding the observed searching behaviour on the plant-stem model, the transition frequencies between the five behavioural states were counted using a state-lag sequential analysis (The Oberserver, 1996). The transition probabilities (i.e. transition frequency in relation to the total number of transitions) from antennating to pausing, from antennating to probing and from probing to antennating were tested for differences between the temperature treatments using Fisher's exact test with adjusted *P*-values by the Hommel method (R Development Core Team, 2005).

The time spent in each behaviour (antennating, probing, insertion, pausing and cleaning) was divided by the total duration of searching behaviour to test the thermal dependence of performing mechanosensory host location in the parasitoid. These relative durations of searching states were tested between the temperatures using the nonparametric Kruskal–Wallis rank sum test and subsequently compared pairwise by the Mann–Whitney *U*-test with adjusted *P*-values by the Hommel method (R Development Core Team, 2005). The thermal dependence of wing fanning was also tested by comparing the relative duration of this behaviour between the temperatures using the Kruskal–Wallis rank sum test. Subsequent pairwise comparisons were undertaken using the Mann–Whitney *U*-test with adjusted *P*-values by the Hommel method.

All statistical analyses were conducted using the statistical computation language R (R Development Core Team, 2005).

Results

Body temperature of the parasitoids

A total of 62 females were used in the temperature treatments at 10, 18, 26 and 30 °C; of these, 42 females were observed for their body temperatures during searching activity and 20 females were pausing.

The head and thoracic temperatures of the pausing wasps were significantly higher than the ambient surface temperature at 10 °C and significantly lower at 26 and 30 °C (Fig. 1). The difference between body temperatures and ambient surface temperature was not significant at 18 °C (one-sample *t*-test; head: $t_4 = 1.11$, $P = 0.33$; thorax: $t_4 = 1.38$, $P = 0.24$; abdomen: $t_4 = -0.984$, $P = 0.38$). Similarly, no significant difference was noted between abdominal and ambient surface temperature in all temperature treatments (one-sample *t* test; 10 °C: $t_4 = 0.606$, $P = 0.58$; 26 °C: $t_4 = -2.45$, $P = 0.07$; 30 °C: $t_4 = 1.83$, $P = 0.14$).

The effect of the different environmental temperatures was significant on the head and thoracic temperature of the pausing wasps (ANCOVA, factor; head: $F_{3,20} = 7.57$, $P = 0.003$; thorax: $F_{3,20} = 6.79$, $P = 0.004$). In the subsequent multiple comparisons, ΔT_H and ΔT_T at 10 °C differed significantly from ΔT_H and ΔT_T at 26 and 30 °C (Sidak post-hoc tests; ΔT_H, 10–26 °C: $P = 0.018$; ΔT_H, 10–30 °C: $P = 0.004$; ΔT_T, 10–26 °C: $P = 0.025$; ΔT_T, 10–30 °C: $P = 0.008$). The abdomen temperature did not differ significantly between the treatments (ANCOVA factor; abdomen: $F_{3,42} = 1.15$, $P = 0.36$).

The body weight of the pausing parasitoids (mean ± SD, 39.1 ± 2.5 mg; minimum: 33.8 mg; maximum: 43.1 mg; $n = 20$) did not affect the three body temperatures measured (ANCOVA covariate: not significant in all treatments).

Searching females showed body temperatures above ambient temperature in all temperature treatments (Fig. 1). Temperatures of all three body parts (i.e. head, thorax and abdomen) were significantly higher at 18, 26 and 30 °C. At 10 °C, the thorax temperature, but not the head and abdomen temperatures, were also significantly different from ambient temperature (one sample *t*-test: head: $t_{9,10} = 2.09$, $P = 0.066$; abdomen: $t_{9,10} = 2.05$, $P = 0.070$). Absolute values of differences between female body and environment temperatures were not significantly affected by the temperature level of the environment (ANCOVA, factor: not significant in all treatments), nor were they influenced by the body weight of the insects (ANCOVA, covariate: not significant in all treatments). Head, thoracic and abdomen temperatures (mean ± SD) of searching females differed from ambient surface temperature by 0.21 ± 0.07 °C, 0.30 ± 0.09 °C and 0.17 ± 0.06 °C, respectively, averaged over all four treatments.

As a consequence, head and thoracic temperatures differed significantly between searching and pausing females at 10, 26 and 30 °C (ANCOVA, factor; 10 °C, head: $F_{1,15} = 5.35$, $P = 0.039$; 10 °C, thorax: $F_{1,15} = 7.39$, $P = 0.019$; 26 °C, head: $F_{1,15} = 67.9$, $P < 0.001$; 26 °C, thorax: $F_{1,15} = 29.2$, $P < 0.001$; 30 °C, head: $F_{1,15} = 34.1$, $P < 0.001$; 30 °C, thorax: $F_{1,15} = 27.9$, $P < 0.001$), and abdomen temperature differed clearly at 26 °C (ANCOVA, factor; abdomen: $F_{1,15} = 12.4$, $P = 0.004$). The significant difference in the abdomen temperature between these different behavioural activities was marginal at 18 °C (ANCOVA, factor; abdomen: $F_{1,17} = 4.68$, $P = 0.048$), whereas head and thoracic temperatures between searching and pausing females did not differ at this temperature (ANCOVA, factor; head: $F_{1,17} = 0.440$, $P = 0.52$; thorax: $F_{1,17} = 0.899$, $P = 0.36$). No significantly different abdomen temperatures between searching and pausing wasps were found at 10 and 30 °C (ANCOVA, factor; 10 °C: $F_{1,15} = 0.030$, $P = 0.87$; 30 °C: $F_{1,15} = 2.41$, $P = 0.15$). Body weight of searching females (mean ± SD, 37.3 ± 6.6 mg; minimum: 25.9 mg; maximum: 51.4 mg; $n = 42$) did not affect the three body temperatures measured (ANCOVA, covariate: not significant in all treatments).

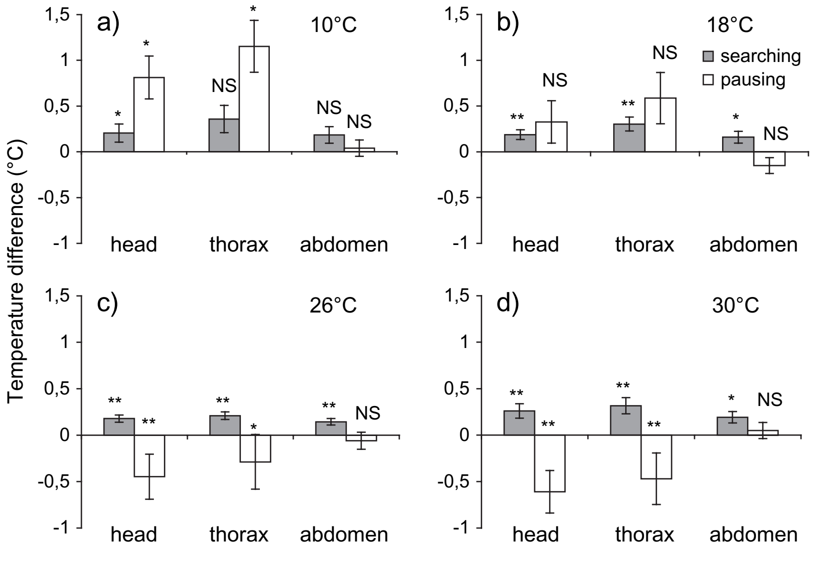

Fig. 1. Thermal deviation from ambient surface temperature of head, thorax and abdomen of searching (grey) and pausing females (white) at (a) 10 °C, (b) 18 °C, (c) 26 °C and (d) 30 °C. Columns and error bars indicate the mean ± SD. Significant difference from ambient surface temperature tested by one sample *t*-test (*$P < 0.05$, **$P < 0.01$; NS, not significant).

Host location behaviour on the plant-stem model

The wasps were observed for a total of 64.4 min at 10 °C, 26.6 min at 18 °C, 16.3 min at 26 °C and 37.2 min at 30 °C during host location behaviour (Table 1). Transitions occurred between the behavioural states antennating and probing, probing and insertion, antennating and pausing, antennating and cleaning, and between pausing and cleaning.

Once the parasitoids found the plant-stem model, they started antennating on the model immediately. At 10 °C, the wasps switched from antennating to pausing more often than at 18 °C (Fisher's exact test: $P = 0.008$). The transition probability from antennating to pausing at 26 and 30 °C did not differ significantly from the probabilities at other temperatures. The probability of wasps switching from antennating to probing and vice versa from probing to antennating showed also no significant differences between the temperatures.

The relative duration of antennating (Fig. 2) was affected by temperature (Kruskal–Wallis *H*-test: $H_3 = 10.5$, $P = 0.015$). The highest proportion of time spent in this behaviour was at 26 °C, and the clearly lowest proportion was at 30 °C (Fig. 2) and the difference was only significant between these two temperatures (Mann–Whitney *U*-test; 26–30 °C: $W = 84.5$, $P = 0.041$). Furthermore, the relative duration of probing was affected by temperature (Kruskal–Wallis *H*-test: $H_3 = 10.3$, $P = 0.016$) and the pairwise comparisons with adjusted *P*-value by Hommel showed a significantly higher proportion spent probing at 30 than at 18 °C (Mann–Whitney *U*-test: $W = 25.0$, $P = 0.0498$). Correspondingly, the highest proportion of searching time spent probing was at 30 °C and the least at 26 °C, whereas the relative duration of this state was similarly low at 10 and 18 °C (Fig. 2). The females barely inserted the ovipositor during the trials, and hence the relative duration of this behaviour was low at 18, 26 and 30 °C, and did not occur at 10 °C. Temperature treatment did not significantly affect the duration of pausing or cleaning (Kruskal–Wallis *H*-test; pausing: $H_3 = 3.15$, $P = 0.37$; cleaning: $H_3 = 5.90$, $P = 0.12$), although the mean values on relative duration of pausing were conspicuously higher at the extreme temperatures of 10 and 30 °C. This varied very much from individual to individual.

The wasps fanned their wings at all temperatures but the speed of fanning increased with increasing temperature. A large proportion of the time was used for fanning at 26 and 30 °C, whereas this activity covered less than half the time at 10 and 18 °C (Fig. 2). Furthermore, the variation in the relative duration of this activity was significant between the temperatures (Kruskal–Wallis *H*-test: $H_3 = 20.4$, $P < 0.001$), and the pairwise comparisons showed differences between 26 °C and all other temperatures, as well as between 10 and 30 °C (Mann–Whitney *U*-test; 26–10 °C: $W = 2.0$, $P < 0.001$; 26–18 °C: $W = 11.0$, $P = 0.003$; 26–30 °C: $W = 83.0$, $P = 0.032$; 10–30 °C: $W = 13.0$, $P = 0.015$).

Discussion

The IR thermography of host-searching *P. turionellae* wasps reveals a body temperature slightly above the ambient surface temperature, and independent of environmental temperature treatments. Females appear to thermoregulate their body when pausing. The body temperature of pausing females (i.e. females without any visible activity) is significantly higher than the environment at low ambient temperatures and significantly lower than the environment at high ambient temperatures. With respect to host-location behaviour, the wasps pause more often at 10 °C, whereas they search continuously at 18 °C once they start. Transitions from searching to pausing are also observed at higher ambient temperatures of 26 and 30 °C.

It is likely that the significant gradient between body temperatures and ambient temperature results from metabolic activity in the muscles. The thoracic temperature of host-searching females is generally the highest of all body parts in all treatments and, because muscle activity produces metabolic heat (Heinrich, 1981) it is likely that vibrations produced in the thorax of the wasps for mechanosensory host location are responsible for this effect (cf. Henaut, 1990; Otten *et al.*, 2002; Samietz *et al.*, 2006). It is hypothesized that the vibrations are produced by shivering muscles without any visible thoracic movement similar to that known in honeybees and bumblebees during metabolic heating (Otten *et al.*, 2002). However, direct production of vibrations by continuous muscle contractions is rather unlikely because of the independence between temperature and vibrations and the low heat production observed in the present study compared with shivering in other hymenopterans. Another option could be that a single muscle contraction elicits trains of vibrations. Such a mechanism means a lower muscle activity with a lower heat production and would be compatible with

Table 1. Transition probabilities between behavioural states during host location by vibrational sounding in four temperature treatments (i.e. number of certain transition divided by number of total transitions).

Temperature treatment (°C)	Total time (min)	Total transitions (*n*)	Antennating to pausing	Antennating to probing	Probing to antennating	Probing to insertion
10	64.4	58	0.172	0.138	0.172	0.000
18	26.6	66	0.015	0.152	0.136	0.030
26	16.3	62	0.065	0.129	0.113	0.016
30	37.2	232	0.086	0.272	0.272	0.022

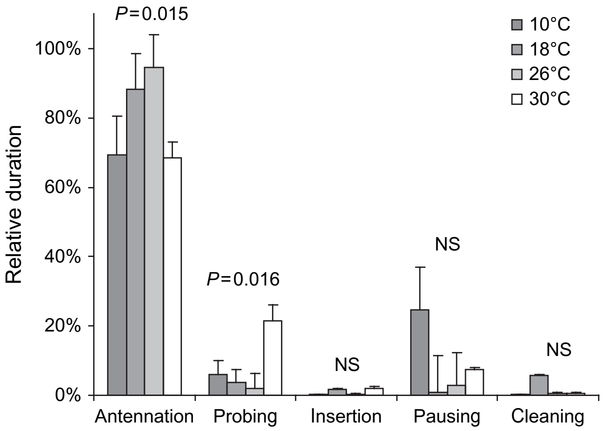

Fig. 2. The proportion of time during searching behaviour spent with antennating, probing, insertion, pausing and cleaning as percentages at 10, 18, 26 and 30°C. Columns and error bars indicate the mean ± SD. Significant temperature influence tested by the Kruskal–Wallis rank sum test (*P*-values; NS, not significant).

an independence from temperature, as observed in the present study.

In pausing wasps, the difference between body and ambient temperature depends on the environmental temperatures. In particular, the head of the insect is cooler than ambient at higher temperatures whereas, in particular, the thorax is warmer than the environment at low temperatures. No difference is found between temperatures of insect body and environment at the intermediate temperature of 18 °C. Endothermic heating in insects is typically located in the thorax whereas the head plays a more important role in cooling mechanisms such as evaporation by regurgitation (Heinrich, 1981, 1993). These findings suggest a low-level endothermy in *P. turionellae* that probably interferes with vibrational sounding because it only occurs in pausing females. This leads to the conclusion that low-level endothermy and producing vibrations for vibrational sounding are two different mechanisms, both located in the thorax. Metabolic heat production by muscular work could disturb the signal structure of simultaneously produced vibrations and would need to be discontinued for efficient use of vibrational sounding. Behavioural observations are in agreement with this hypothesis. Searching females interrupt antennating more often at 10 °C than at 18 °C. In short pausing periods, the parasitoids would be able to heat up the body sufficiently to continue with host-location behaviour.

Relatively small levels of endothermic thermoregulation are also reported in other insects, such as *Andrena taraxaci* (Schmaranzer *et al.*, 1997), the mayfly *Hexagenia bilineata* (Coelho, 1999), and the beetle genus *Nicrophorus* (Merrick & Smith, 2004). The probability and intensity of endothermy is generally low in small-sized insects such as *Pimpla* because of a high surface-to-volume ratio and a rapid heat exchange with the environment (Bishop & Armbruster, 1999). Warming up by 1.15 °C and cooling down by 0.61 °C, as recorded in the present study, are probably not of much physiological influence, but endothermy could be easily combined with ectothermic mechanisms such as the absorption of solar radiation (Heinrich, 1981, 1993; Bishop & Armbruster, 1999). Such a combination of behavioural and physiological thermoregulation is demonstrated in the wasp *Sphecius speciosus*, whereby the thoracic temperature regulation depends mainly on behavioural mechanisms (Coelho, 2001). Under natural conditions, *P. turionellae* would be similarly able to thermoregulate by habitat selection or basking because the dark body would be highly absorptive with respect to solar radiation (Willmer & Unwin, 1981). These endothermic mechanisms could then be used additionally to thermoregulate additionally under sub-optimal conditions, such as low ambient temperature and for low solar radiation. This assumption is supported by the declining difference between thoracic and ambient temperature with increasing environmental temperature. The energy investment for endothermy in small animals is very high, and therefore they should reduce physiological heat production with increasing ambient temperature. Similarly, the wasp *Paravespula germanica*, an insect capable of intense endothermy (Heinrich, 1984; Kovac & Stabentheiner, 1999), adjusts endothermic heat production to solar radiation and decreases its own energy investment with increasing solar radiation (Stabentheiner *et al.*, 2004).

The accessory wing movement in *P. turionellae* observed in the present study under high temperatures could possibly be linked to 'fanning', resulting in a higher convective heat loss. Fanning is used in social insects for ventilation of the nest at high temperature conditions (Heinrich, 1993), but is not yet known to be used as a cooling mechanism for an insect's own body. A further possible cooling mechanism is increased evaporation, which occurs in many hymenopterans and other insects to avoid overheating (Heinrich, 1993). Bumblebees and honeybees, for example, repeatedly regurgitate droplets of fluid, and this is followed by a decrease in head temperature (Heinrich, 1981). A similar mechanism could operate in *P. turionellae* because the body temperature was below ambient temperature at high ambient temperature and the head especially showed the lowest temperature.

In conclusion, the parasitoid females investigated in the present study show a limited ability to raise or lower their body temperature in relation to ambient thermal conditions. Under conditions below or above an optimal temperature range, the host-searching females interrupt the searching process frequently because they are unable to perform vibrational sounding and endothermy simultaneously. Thermoregulation during host searching is, however, far from being able to maintain a constant body temperature over wide ambient temperature ranges. With respect to host location, these parasitoids are more like thermoconformers and appear to cope very well with high thermal changes during the use of vibrational sounding because the carrier frequency and the intensity of self-produced vibrations actually increase with decreasing temperature (Samietz *et al.*, 2006; Kroder *et al.*, 2007). Thermoconformers must be able to synchronize the production and receipt of vibrations for all thermally induced changes because the temperature influences the neural components for signalling and receiving independently (Sanborn, 2005). Temperature affects the mechanosensory pathway with respect to sensitivity to physical stimuli and response thresholds in insects (Coro & Perez, 1990; Franz & Ronacher, 2002). It is postulated that the wasps optimize vibrational sounding by modifying carrier frequency and intensity at changing temperatures to compensate for thermally-induced changes of the receivers. The temperature effect on the subgenual organ of parasitoids using vibrational sounding should therefore be confirmed in further experimental studies.

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