Intraspecific experiments demonstrated the presence of a pre-attachment pheromone in the species Dermacentor variabilis (Say), Dermacentor andersoni (Stiles), Dermacentor parumapertus (Neumann) and Haemaphysalis leporispalustris (Packard) (table 1). The species Amblyomma maculatum Koch and Amblyomma cajennense (Fabricius) tested in the same series did not aggregate within the 1-h period. Within the limits of our data it appears that when assembly behavior is present the ability to produce or respond to the pheromone is not sex-determined.

In a parallel control series (males and females tested in dishes with untreated discs only) no significant evidence of assembly appeared with the exception of the species *D. andersoni*. 'Clumping' of these ticks is so strong that aggrega-

- 1 We thank Dr J. Keirans, Rocky Mountain Laboratory, Hamilton, Montana, for providing ticks, Dr Hallie Bundy, Chemistry Department, Mount St. Mary's College, Los Angeles, California, for helpful assistance and Dr R. VandeHey, St. Norbert College, DePere, Wisconsin for review of the manuscript.
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tion occurred within the h but in various sectors. However, when pheromone was present in a specific sector aggregation invariably occurred in that sector.

Interspecific experiments demonstrated that *D. andersoni* and *A. americanum* recognized each other's pheromone and that *A. americanum* recognized that of *H. leporispalustris* (table 2). Our earlier studies indicated heterologous response between *H. dromedarii* and *Hyalomma asiaticum*<sup>11</sup>. To determine whether the tick's excretory products might function as aggregation factors we challenged *D. andersoni* males with guanine<sup>12</sup> in quantities of  $10^{-5}$ ,  $10^{-4}$ , and  $10^{-3}$  g and with hemin<sup>13</sup> in quantities of  $10^{-7}$ ,  $10^{-6}$ ,  $10^{-5}$ ,  $10^{-4}$ ,  $10^{-3}$  g in 20 µl of 10% NH<sub>4</sub>OH solution. Results were inconsistent but further work seems indicated.

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## Influence of a meal on skin temperatures estimated from quantitative IR-thermography

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Summary. In young men at 25 °C, quantitative IR-thermography showed that fasting values of skin temperatures over suspected areas of brown adipose tissue (BAT) were higher than where no BAT is thought to occur. However, at 30 min and again at 60-90 min after a meal of 2.5 MJ, the magnitude of the increase in skin temperature was similar in areas with or without suspected BAT. In conclusion, either thermography was unable to detect the activation of BAT, or the meal did not stimulate heat production in the sites of suspected BAT.

It has been shown previously in man that the sympathomimetic agent ephedrine causes an increase in skin temperature on the neck and upper back1. These locations correspond to the sites where brown adipose tissue has been found to exist in infants<sup>2</sup> and adults<sup>3</sup> and it was therefore suggested that the findings could be interpreted as evidence that noradrenaline stimulates the production of heat by brown adipose tissue in adult man. Feeding causes an increase in both metabolic rate and the circulating concentration of noradrenaline in adult humans<sup>4</sup> and young pigs<sup>5</sup>. Since small amounts of brown adipose tissue occur in both these species<sup>3,6</sup>, it is therefore possible that part of the extra heat production associated with feeding is due to the stimulation of this tissue. Further evidence which lends support to this idea is that the  $\beta$ -blocker propranolol can reduce the heat production caused by diet1,7. The aim of the present investigation was therefore to determine the extent to which any changes in skin temperature after a meal could be correlated with the probable presence of brown adipose tissue. Quantitative IR-thermography was used, since preliminary studies with temperature sensors attached to the skin had shown variations in recorded temperature which were probably due to differences in adhesion of the probes. These variations introduced errors which could have masked any small changes in skin temperature of the order of 0.5 °C.

Materials and methods. Five healthy men aged  $21\pm0.5$  (SEM) years, volunteered to take part in the investigation. They were all familiarized with the procedures before measurements were made. Their mean  $(\pm$  SEM) height and weight were  $1.77\pm0.013$  m and  $69.2\pm0.35$  kg. Skinfold thicknesses for triceps, biceps, subscapular and suprailiac sites were  $6.0\pm0.78$ ,  $3.5\pm0.48$ ,  $8.6\pm1.22$  and  $7.1\pm0.70$  mm respectively. The subjects were also well-matched for habitual physical activity; they were all students who climbed in their spare time. Dietary recall revealed that their food intakes were also similar.

On the day of experiment the subjects were transported to the unit by car and arrived at 09.00 h after an overnight fast. The measurement room was maintained at an ambient temperature of 25 °C, since earlier studies had indicated this to be within the thermally neutral zone of lightly clad adults to be a comfortable temperature for sedentary subjects<sup>8</sup>. The subjects wore trousers, shoes and socks, were seated on chairs (without leaning against the back) and reported that they felt comfortable throughout the period of measurement.

After 30 min of equilibration the baseline thermogram was taken at time 0. An A.G.A. 680 Thermovision System was used, at a fixed distance from the subject, to take a picture of the whole back from the hairline to the iliac crests. After taking the baseline thermogram, the test meal was drunk, within 5 min. It consisted of 150 g natural flavor Complan (Glaxo-Farley Foods Ltd, Plymouth, Devon, England) mixed in 350 ml water, and contained 2.5 MJ (600 kcal). A further 10 thermograms were then taken at 15-min intervals over the next 2.5 h, starting at 15 min after the baseline measurement.

Visual inspection of each thermogram gave an indication of differences in skin temperature over particular areas of the back, and of changes with time. A black body was fixed as a control, at a temperature of 31 °C. The range of temperature covered by each thermogram was 5 °C, with a midpoint of 32.75 °C. Ten different colors were used, each representing 0.5 °C. This visual approach could only be used to make approximations and therefore a more detailed quantitative analysis was made by computer.

Accurate estimates of skin temperature over defined areas of the body were obtained by interfacing the Thermovision System to an Apple II microcomputer and using Thermoteknics software. Three sites of the body were chosen: the back of the neck, the interscapular region at the center of the back, and the left shoulder. The 1st 2 areas were chosen

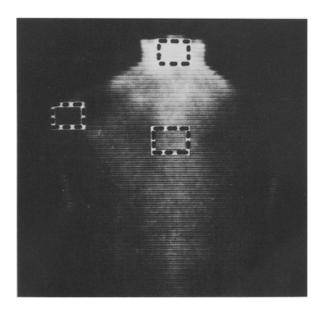




Figure 1. A greytone thermogram from 1 subject showing the 3 small areas of skin ([...]), each of 100 pixels, over which temperatures were calculated as shown in figure 2. This thermogram was taken 15 min after the meal and shows that of the 3 sites the back of the neck was hottest and the left shoulder coolest. This finding is the same as that obtained before the meal, after an overnight fast.

as suspected areas of brown adipose tissue, while the 3rd was chosen as a control area where no such tissue is thought to exist. Mean skin temperatures at the 3 sites were obtained over fixed areas of both 520 pixels (large area) and 100 pixels (small area). Figure 1 shows the 3 small 100 pixel squares drawn on a thermogram over the defined areas of the back for one of the subjects. The areas were sited over the thermograms using anatomically defined co-ordinates which were standardized for each subject. Paired t-tests were used to analyze the results in terms of changes in temperature over the defined areas, compared with the baseline value at time 0. Results for the large and small areas were similar and therefore only those for the small areas are presented in detail.

Results and discussion. Figure 2 shows the mean values with SEM for skin temperatures over the small areas of the neck, interscapular site and shoulder. It shows clearly that for the 3 sites the baseline value of skin temperature after an overnight fast was highest on the back of the neck and lowest on the left shoulder. Mean values  $\pm$  SEM for the small areas were  $33.6 \pm 0.25$ ,  $33.1 \pm 0.19$  and  $32.4 \pm 0.27$  °C for the neck, interscapular region and shoulder respectively. For the large areas, the corresponding values were  $33.5 \pm 0.19$ ,  $33.1 \pm 0.21$  and  $32.6 \pm 0.25$  °C. Thus quantitative IR-thermography revealed that during fasting the suspected areas of brown adipose tissue were warmer than the other areas of the back. 15 min after the meal there were no changes in skin temperatures from the baseline values. Figure 1 was taken at this time, and shows that the differences in skin temperature between the sites were maintained. However, although these differences were maintained throughout the period of measurement, figure 2 shows that by 30 min after the meal there were small but statistically significant increases in skin temperature from time 0 for 2 of the 3 sites: one with and one without suspected brown fat. On the neck and shoulder the mean increases  $\pm$  SEM were  $0.4 \pm 0.04$  (p < 0.001)

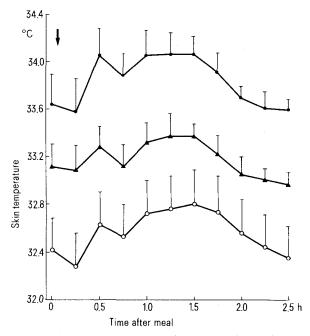


Figure 2. Skin temperatures estimated by quantitative IR-thermography before and after a meal ( $\downarrow$ ) of 2.5 MJ. Measurements were made over 3 areas of skin, each of 100 pixels: back of the neck ( $\bullet$ ), interscapular region ( $\blacktriangle$ ), and tip of the left shoulder ( $\bigcirc$ ). Time 0 was at 09.30 h. Values are means  $\pm$  SEM for 5 young men.

 $0.2 \pm 0.03$  °C (p < 0.005). The larger individual variation in the response of the interscapular site resulted in the increase of  $0.2 \pm 0.10$  °C being nonsignificant (p > 0.1). Corresponding values for the large areas over the neck, shoulder and interscapular site were  $0.4 \pm 0.04$  (p < 0.001),  $0.3 \pm 0.07$ (p < 0.025) and  $0.2 \pm 0.10$  °C (NS, p > 0.1) respectively.

Following this initial increase in temperature at 30 min, a slight decrease was next observed and this was followed by a sustained rise in temperature which reached a peak at approximately 1.5 h (fig. 2). For all 3 sites this rise represented a significant increase above the baseline value. For the neck, interscapular region and shoulder the mean temperature differences  $\pm$  SEM from time 0, for the small areas, were  $0.4 \pm 0.11$  (p < 0.025),  $0.3 \pm 0.09$  (p < 0.05) and  $0.4 \pm 0.11$  (p < 0.025) °C. Corresponding values for the 3 large areas were  $0.3 \pm 0.08$  (p < 0.05),  $0.3 \pm 0.07$  (p < 0.025) and  $0.4 \pm 0.09$  (p < 0.010) °C respectively.

After this 2nd peak at approximately 1.5 h the skin temperatures gradually returned to the baseline values so that by 2.5 h the mean differences  $\pm$  SEM were  $-0.05\pm0.18$ (p > 0.5),  $-0.15 \pm 0.11$  (p > 0.2) and  $-0.08 \pm 0.09$  (p > 0.4) °C for the small areas of the neck, interscapular (p > 0.2) and site and shoulder respectively.

Thus, 2 peaks in skin temperature were observed to follow the meal. It could be speculated that the 1st rapid increase was due to the stimulation of brown adipose tissue, since the increase in circulating noradrenaline occurs immediately after a meal. And, that the 2nd sustained rise was the result of the general vasodilatation which occurs when the heat produced on processing and oxidation of food is only gradually lost from the body9. However, the postulated explanation for the 1st increase in temperature seems unlikely, since there was no preferential increase in skin temperature over the suspected areas of brown fat. After a meal, the increase in skin temperature over areas of the back where brown adipose tissue is thought to exist was no

greater than over areas where no evidence has been reported for the presence of this tissue. Unless brown fat also exists on the shoulder, it must be concluded that the rises in skin temperature over the neck, interscapular region and shoulder which occurred after a meal, must have been due to increased blood flow and general vasodilatation. It is conceivable, however, that brown adipose tissue in the deep body sites was activated by the meal and that IR-thermography was unable to detect any such activation. In addition, the possibilities that changes which could be firmly attributed to brown adipose tissue occur after either a very large meal or prolonged overfeeding in man have not been eliminated 10.

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## Desensitization to histamine in the absence of external Ca++ in the guinea-pig taenia caecum

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Summary. Short-term desensitization of the contractile response of the guinea-pig taenia caecum to histamine was tested in the absence of Ca++. Desensitization was monitored both by the fall of histamine response and by the decrease of irreversible blockade by phenoxybenzamine. In Ca++-free solution with 0.2 mM EGTA, desensitization occurred as in normal physiological solution containing Ca++.

Short-term desensitization to histamine was first observed early by Barson and Gaddum1. Intestinal smooth muscle loses its sensitivity to histamine on contact exposure to histamine itself. In skeletal and cardiac muscle, Ca<sup>++</sup> plays a critical role in desensitization of cholinergic receptors<sup>2,3</sup>. Thus it is essential to study the influence of Ca++ on desensitization of smooth muscle. As the contraction of the guinea-pig taenia caecum in response to histamine is dependent solely on the influx of Ca<sup>++4</sup>, desensitization to this agonist should provide information on demand for Ca<sup>++</sup>. Kenakin and Cook<sup>5</sup> reported that after desensitization of the histamine H<sub>1</sub> response, phenoxybenzamine (POB) became less effective in irreversible blockade of histamine response than in non-desensitized muscle, indicating transient change of affinity of the receptor system.

We used this method in addition to direct measurement of the change of response after desensitization to histamine. Materials and methods. Male guinea-pigs, weighing 250-400 g, were killed instantly by cervical fracture and exsanguination. Strips of taenia caecum were suspended in a 30-ml organ bath bubbled with air at 30 °C. The bathing solution was Locke-Ringer solution composed of 154 mM NaCl, 5.63 mM KCl, 2.10 mM CaCl<sub>2</sub>, 2.10 mM MgCl<sub>2</sub>, 5.95 mM NaHCO<sub>3</sub> and 5.55 mM glucose (normal solution). In Ca<sup>++</sup>-free solution, CaCl<sub>2</sub> was omitted from the normal solution and 0.2 mM EGTA (glycol ether diaminetetraacetic acid) was added. Cumulative contractile response was recorded isotonically with a lever of circa 0.5 g load on a smoked drum. For desensitization in Ca++-free solution, after responses in the normal solution were observed, the