Table 2. Results of hybridization of pRNA isolated from human sarcomas with MMTV $[^{3}\mathrm{H}]$DNA and AMV$[^{3}\mathrm{H}]$DNA. No positive reactions occurred. Method and data analysis are the same as in Table 1.

Product DNA	Diagnosis	RNA (count/ 10 min)	(Count/ 10 min) per σ
MMTV	Liposarcoma (1458)	0	0
MMTV	Osteogenic sarcoma (23–2)	20	0.38
AMV	Osteogenic sarcoma (23–5)	75	1.42
AMV	Fibrosarcoma (1448)	146	2.76
AMV	Liposarcoma (1441)	85	1.61
AMV	Liposarcoma (1458)	144	2.73

and human leukemias (*4*). RNA from human breast carcinoma is unrelated to the RNA of the RLV but is homologous to the RNA of the virus that causes breast tumors in mice (*2*). On the other hand, RNA from human leukemic cells is unrelated to that of the breast tumor-inducing virus, but instead shows a unique homology to the virus that causes leukemia in mice. Our studies demonstrate that human sarcomas contain RNA's that show the same sort of relatedness as do the RNA's from human leukemias; their RNA is homologous to an animal leukemogenic agent but not to that of an animal mammary tumor virus. Each of these investigations lends support to the specificity and meaningfulness of the positive responses observed in the others. Finally, the specificity pattern observed with the RNA's from human neoplasias and the viral DNA's mirrors precisely what has been observed with the analogous diseases in the animal models. Whatever these results may ultimately mean for a viral etiology of human cancer they do suggest a remarkable similarity in the specific virus-related information found in corresponding tumors of mice and men.

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8 December 1971 ■

Temperature Regulation in the Bumblebee Bombus vagans: A Field Study

Abstract. Bombus vagans *forages for nectar at 5°C in shade and at 31°C in sunshine. The production of heat while the bumblebee is on flowers, at ambient temperatures below 24°C, helps to maintain a thoracic temperature that is near the minimum for flight between flowers. However, at ambient temperatures above 24°C the thoracic temperature is no longer regulated at 32° to 33°C and rises.*

Bumblebees occur in boreal and arctic regions where diurnal temperatures fluctuate and overcast skies may prevail for the entire day. They rely for food nearly exclusively on nectar and pollen throughout their life cycle and use a portion of their caloric intake to regulate nest temperature (*1*). Food must be constantly available during brood rearing. The fact that bumblebees are able to forage at low ambient temperatures (T_A), at which no other bees are able to fly (*2*), should, in part, compensate for the small reserves of food in the nest [see (*3*)].

Like some other insects, *Bombus vagans* F. Smith has a relatively high thoracic temperature (T_{Th}) before initiating flight. Warm-up before flight in bumblebees (*4*), as in honeybees (*5*), involves rapid contractions (shivering) of the flight muscles. A layer of hair covering the body reduces the rate of heat loss by nearly one half (*6*) and thus reduces the energetic cost of maintaining a large difference between T_{Th} and T_A. However, to my knowledge, regulation of thoracic temperature has not previously been demonstrated in bumblebees.

Bumblebees land on each flower they visit and could either cease heat production and cool down or continue expending energy and maintain a high T_{Th} during these intervals. I report here that the workers of *B. vagans* (mean body weight, 120 mg) regulate their thoracic temperature when foraging in the field from *Epilobium angustifolium* L. (fireweed). The thoracic temperature is maintained at about 32° to 33°C by "warm-up" after the bees land on the flowers.

All measurements were made in the field near Farmington, Maine, during July and August, 1970 and 1971. The bees were grasped from the flowers between gloved thumb and forefinger, and a thermistor (*7*) was then quickly inserted from the ventral side to the approximate center of the thorax and into the abdomen from the posterior. Temperatures were read to the nearest 0.2°C from a telethermometer (Yellow Springs) within 4 seconds after the bee was grasped. Nectar volumes were measured in 2-μl calibrated capillary tubes, and sugar concentrations were determined with a pocket refractometer (Bellingham and Stanley). Ambient temperatures were measured to the nearest degree Celsius in the shade near the flowers.

Few *B. vagans* were foraging at 5°C (the coldest observed dawn), but many were active in shade at 15°C and in noon sunshine at 31°C. The mean T_{Th} of bees in shade at a T_A of 9° to 24°C was 32° to 33°C (Fig. 1). This T_{Th}, being slightly lower than that of bumblebees in continuous flight at these T_A's (*8*), is about 3° to 4°C above the minimum T_{Th} and at least 9° to 10°C below the maximum T_{Th} at which flight is possible (*9*). The conspicuous independence of T_{Th} from T_A implies that T_{Th} is "regulated." However, at a T_A of 26° to 31°C T_{Th} increased nearly directly with T_A, an indication of a lack of regulation of T_{Th} during foraging at this upper range of T_A where measurements were made. Stabilization of T_{Th} at high T_A, which would indicate temperature regulation through heat loss [see (*10*)] or reduction of heat production [see (*11*)], was not observed as T_{Th} rose from 33°C (Fig. 1) and approached 38°C (in shade) and 40°C (in sunshine).

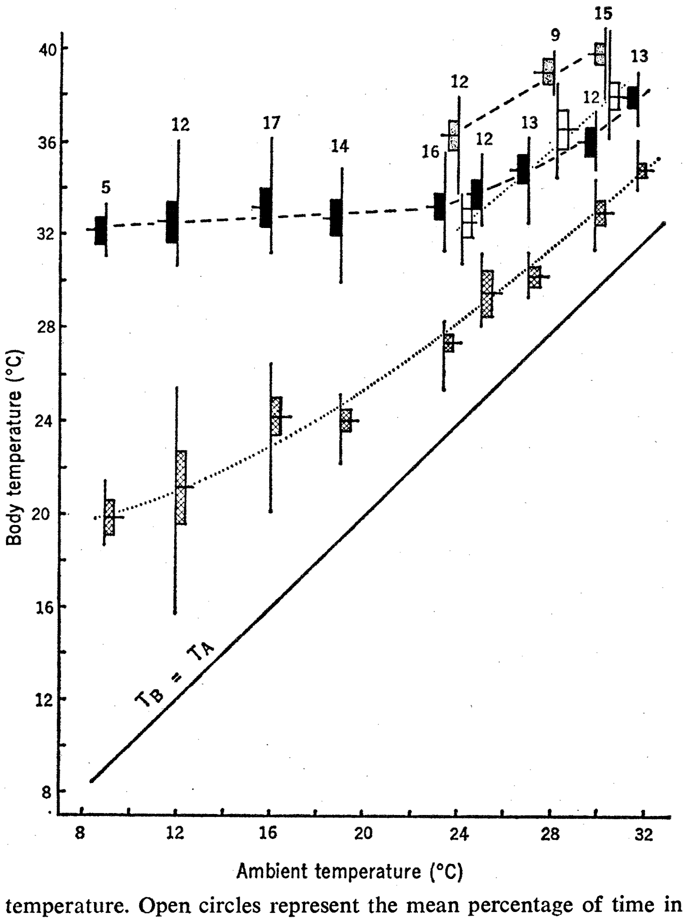
The difference between T_{Th} and T_A, both in shade and in sunshine, was relatively constant at all $T_A > 25°C$. It can therefore be inferred that (in shade) this difference resulted primarily from the endogenous heat that is the by-product of muscular activity associated with foraging and not from temperature regulation. A rate of heat production at 9°C similar to that at $T_A > 25°C$ should result in a T_{Th} of about 16° to 18°C. However, the mean T_{Th} at a T_A of 9°C was 32°C, an indication that heat in addition to that resulting exclusively as a by-product of foraging was produced at 9°C, and at the other $T_A < 25°C$.

Bumblebees could produce additional heat either by increasing the percentage of time in flight, or by generating heat specifically for temperature regulation. The first possibility can clearly be dismissed; the bees spent less time in flight and more time on each flower at the lower T_A than at the high T_A (Fig. 2). During the relatively prolonged (4 to 12 seconds) stationary periods on flowers at low T_A, T_{Th} should decline rapidly if endogenous heat is not produced to counteract heat loss [see (*6*)]. Although the reduction of mobility minimizes convective heat loss and aids in the conservation of endogenously produced heat [see (*5*)], this behavior would become energetically unproductive if only a few flowers are then visited.

The following experiment was performed to determine if, indeed, *B. vagans* produces heat during the periods during which it is stationary on flowers. Usually *B. vagans* stopped at flowers only briefly (≈ 1.5 seconds) at $T_A > 15°C$ (Fig. 2). However, the bees could be induced to remain stationary for up to 3 minutes if they were fed from flowers provided with viscous sugar syrup. As during warm-up [see (*12*)], the wings were folded dorsally during feeding and abdominal respiratory movements were rapid. After 2 minutes of feeding on syrup ($T_A = 16°$ to 20°C) the bees had a mean T_{Th} of 34.8°C (range, 30.0° to 37.4°C; $N = 26$) and a mean abdominal temperature of 26.5°C (range, 22.2° to 31.2°C). Both of these temperatures are significantly higher ($P < .05$) than those normally observed during foraging from *E. angustifolium* at 16° to 20°C. In contrast, the T_{Th} of heated dead bees dropped from 33° to about 23°C in a comparable period of time under similar conditions (*13*).

Calculations made from the measurements of T_{Th} as a function of T_A in live bees and the cooling rates of dead bees indicate [see (*14*)] that the bees should produce approximately 0.54 cal of heat per minute in the thorax in order to maintain a temperature difference of 27°C (the maximum observed) between thoracic and ambient temperatures. Such a rate of heat production in a 0.12-g bumblebee is equivalent to a respiratory rate of 56 cm^3 of oxygen per gram of body weight per hour [see (*15*)], which is close to the respiratory rate of honeybees (weight, ≈ 0.1 g) in flight (*4, 16*).

Is the energetic cost of temperature regulation justified? Each flower of *E. angustifolium*, after being screened with cheesecloth for 24 hours to exclude nectar foragers, contained a mean of 1.85 mg [see (*17*)] of sugar (range, 0.98 to 3.10 mg; $N = 19$). The 1.85 mg of sugar from one flower is sufficient fuel to allow *B. vagans* to produce the 0.54 cal of heat [see (*14*)] for 12.7 minutes (*18*). Since *B. vagans* usually collected pollen and nectar from 10 to 20 flowers of *E. angustifolium* per min-



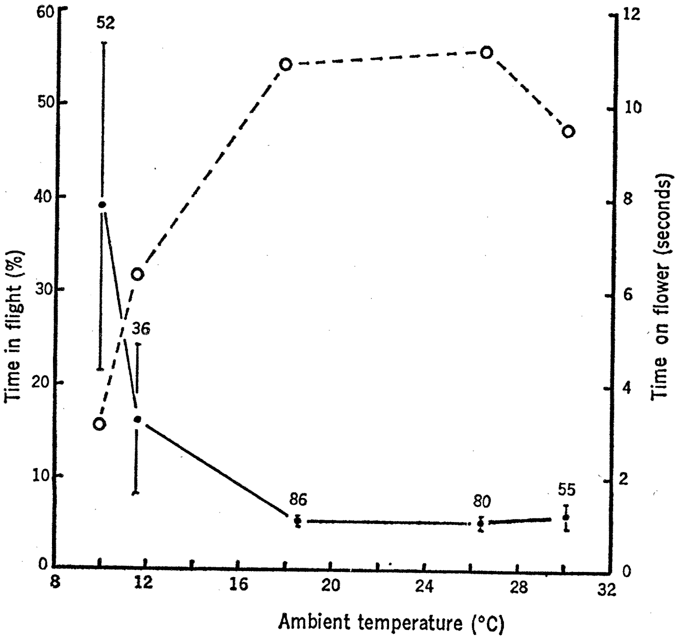


Fig. 1 (left). Body temperature (T_B) of *B. vagans* foraging on *E. angustifolium* in relation to ambient temperature (T_A). The boxes enclose the mean plus two standard errors on each side of the mean. The vertical lines indicate the range, and the numbers refer to the number of measurements made on different bees. Thoracic temperatures are indicated by solid boxes (in shade) and stippled boxes (in sunshine). Abdominal temperatures are indicated by crosshatched boxes (in shade) and open boxes (in sunshine). Fig. 2 (right). Foraging activity of *B. vagans* foraging on *E. angustifolium* in relation to ambient temperature. Open circles represent the mean percentage of time in flight, and solid circles represent the mean duration on each flower. The vertical lines indicate two standard errors on each side of the mean. Numbers indicate the number of timed observations.

ute, the calories expended for temperature regulation are easily replaced if the flowers contain their full complement of nectar.

Many bees and other insects, as well as hummingbirds, take nectar from *E. angustifolium* and thereby reduce the amounts of sugar that are available in the flowers at any one time. For example, the unscreened flowers (examined an hour after the screened) contained a mean of only 0.086 mg of sugar (range, 0.00 to 0.17 mg; $N = 51$). However, despite the relatively low sugar content of these flowers, *B. vagans* should make an energetic profit from them at low T_{A} if each bee extracts nectar from more than 1.7 flowers per minute. The above approximations suggest that the energetic cost of temperature regulation is justified in *B. vagans* foraging from *E. angustifolium* at relatively low T_{A}, especially if the bees can harvest nectar at times when other insects are excluded from the flowers.

The bumblebees are probably seldom required to actively dissipate heat from the thorax while foraging; under maximum thermal conditions observed in the field (noon sunshine at 31°C), T_{Th} tends to be no higher than 40°C (Fig. 1). Since the abdomen is often heated by solar radiation and approaches T_{Th}, and since heat loss by forced convection from the abdomen is greatly reduced when the bees are walking on flowers, the abdomen obviously could not act as an efficient heat dissipater for excess heat from the thorax during foraging, as in the sphinx moth *Manduca sexta* [see (*19*)].

In hovering sphinx moths, heat is produced continuously at a high rate. In *M. sexta* during free and continuous flight at $T_{\mathrm{A}} > 23$°C, T_{Th} is regulated at the upper tolerable temperature level (40° to 43°C) by shunting the excess heat (via the blood) from the thorax (*19*). However, in foraging bumblebees that land on flowers, the durations of obligatory heat production are brief; the bees are in continuous flight between individual flowers for only 1 to 3 seconds. Heat production is optimal during the stationary periods, and *B. vagans* regulates T_{Th} near the lower limit of T_{Th} [see (*9*)], at which temperature flight between the scattered and calorically rewarding flowers of *E. angustifolium* is possible. However, bumblebees on dense inflorescences often have a lower T_{Th} when the caloric contents of the flowers are low, whereas under other conditions T_{Th} can be relatively high when bees are foraging at low T_{A} (*20*).

Temperature regulation in bumblebees has been treated separately in relation to food resources, foraging strategies, and floral morphology (*20*).

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8. The T_{Th} of seven *B. ternarius* returning to their nest at a T_{A} of 11° to 15°C ranged from 32.6° to 36.0°C (arithmetic mean = 34.7°C) (*B. ternarius* is nearly the same size as *B. vagans*). The T_{Th} of five bees leaving their nest during the same time ranged from 33.8° to 36.0°C (mean = 34.5°C). The bees were caught by net.
9. Bumblebees can fly clumsily with a T_{Th} of 29°C, but *B. terricola* returning to the nest at a T_{A} of 31°C had a T_{Th} of 42.5° to 43.3°C.
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12. Bumblebees that were "cold" and could walk only slowly and deliberately sometimes warmed up rapidly when touched. During warm-up the wings were folded dorsally, wing movement was not apparent, and buzzing could not be heard. However, the frequency and amplitude of abdominal respiratory movements increased rapidly throughout warm-up until flight was initiated. (In Lepidoptera, which are neurogenic rather than myogenic, the wings beat during warm-up, and ventilation of the thoracic musculature is achieved primarily by thoracic rather than by abdominal pumping.)
13. The bees were heated in warm air from a hair dryer and then allowed to cool to near T_{A} while the body temperature (T_{B}) was observed every 30 seconds. The linear regression of the decrease in T_{Th} for ten animals is described by: $\log (T_{\mathrm{Th}} - T_{\mathrm{A}}) = 1.48 - 0.27$ minutes.
14. The rate of cooling of a 40-mg thorax, having a specific heat of about 0.8 cal per gram per degree Celsius, was about 17°C per minute when its temperature was 27°C above T_{A}. In order for T_{Th} to remain at 27°C above T_{A}, the necessary heat input was therefore equal to: 17°C per minute × 0.04 g × 0.8 cal per gram per degree Celsius = 0.54 cal per minute.
15. It is assumed (i) that the amount of heat produced in the abdomen of the 0.12-g animal is negligible in comparison with that produced in the thorax and (ii) that the utilization of 1 cm^3 of O_2 yields 4.8 cal (0.54 cal per minute × 0.12 g × 60 minutes × 1 cm^3 of O_2 per 4.8 cal = 56 cm^3 of O_2 per gram per hour).
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17. The amounts of sugar were derived from nectar volumes (1.44 to 5.43 μl per flower) and nectar concentrations (52 to 80 percent). These measurements were made at 3 p.m. to 4 p.m. in the colony of flowers where the *B. vagans* of the present study were foraging. (In the morning the nectar concentrations were unusually low.)
18. The time was calculated as follows: 3.7 cal per milligram × 1.85 mg × 1 minute per 0.54 cal = 12.7 minutes.
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22 March 1971; revised 29 July 1971

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Initiation of Protein Synthesis at an Unusual Position in an Immunoglobulin Gene?

Abstract. *The amino acid sequence of urinary β_2-microglobulin has been partially determined and found to be related to the constant region of IgG immunoglobulin heavy chain. β_2-Microglobulin is present in normal individuals. Its gene may have evolved from an immunoglobulin gene by the use of an unusually located start signal for initiating synthesis of the polypeptide.*

Large deletions have been described in the genes controlling the synthesis of myeloma or myeloma-like immunoglobulins (*1*). The details of one of these deletions led Smithies *et al.* (*2*) to propose that all of the currently documented examples might be the consequence of DNA breakage and its nonhomologous repair. One of the possible outcomes of the breakage and repair events considered was that a start signal different from the normal one might be used to initiate protein synthesis in the neighborhood of a deletion. The data presented in this report suggest that this can occur.

In the course of a search for new examples of deletions in immunoglobulins and a general survey of urinary proteins of low molecular weight we determined the partial amino acid sequences of four proteins. Two were Bence Jones proteins with starch-gel electrophoretic mobilities indicating that they might be smaller than usual. The other two were not thought to be related to immunoglobulins although they had been isolated from a myeloma patient with severe renal tubular malfunction (*3*). One of these two was a retinol (vitamin A)-binding protein (*4*); the other was immunologically identical (*4*)