THE INFLUENCE OF ATMOSPHERIC HUMIDITY ON THE THERMAL DEATH POINT OF A NUMBER OF INSECTS

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(With Five Text-figures.)

This work is an attempt to investigate the effects of various atmospheric humidities on the death points of insects at high temperatures. A number of species of insects were used, and an attempt made to correlate the results with those of previous workers.

The apparatus used was similar in essentials to that previously used by Beattie (1928) and by Buxton (1931 b). It is shown in Fig. 1. The whole apparatus was submerged in a tank of water heated electrically, and thermostatically controlled to within a fifth of a degree centigrade. Compressed air blown through the water kept it well stirred. The insects were placed in the Woolfe's bottle A, which was kept well submerged below the water. It was found that if only the necks of the bottle were above the surface, the air in A might be several degrees cooler than the water. The total submergence of A made it more difficult to introduce the insects, and the method is unsuitable for very large insects. A broad glass tube, B, was introduced into the middle neck of A. A small bag of bolting silk, D, containing the insects, was put down the tube B on a thin glass rod. The humidity in A was controlled by means of a layer of an appropriate mixture of sulphuric acid and water at the bottom, and fresh air could be blown in through C, another Woolfe's bottle containing pumice and the same acid as A^1 . At the outlet, E, there was a piece of capillary glass tube to prevent undue backward diffusion.

This method of introducing the insects in small silk bags has certain advantages. There is no need to anaesthetise the insects, and they are in intimate contact with the air. At first it seemed difficult to get the insects into the bags, but Fig. 2 shows how this is easily accomplished. F is a corked tube, the cork having two holes. In one hole there is a tube, G, leading to a suction pump. At the other hole there is a tube through the cork also. On its inner end, the small silk bag is fastened, and to the other end is attached a rubber tube and another piece of glass tubing, H. The insects are sucked up H into D. This method is not suitable for blow-flies, which

¹ The method of preparing mixtures of sulphuric acid and water to control atmospheric humidity is described by Buxton (1931 b).

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are caught and pushed head first into the bags. As soon as their heads and thoraxes are inside, the blow-flies lie almost still, any slight movements they make only sending them farther down the narrow bag.

The main apparatus was frequently checked. An accurate thermometer was put down B to make sure that the temperature in A was the same as in the tank. The acid in A was changed every week; the specific gravity of the old acid was tested, but was never found to have changed appreciably. Four pairs of Woolfe's bottles (each pair with a different humidity) were inserted in parallel in a meccano frame. This ensured that the parallel experiments at different humidities and one temperature were strictly comparable.

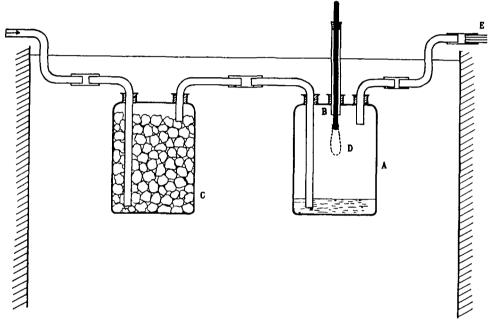


Fig. 1. Apparatus in which insects were exposed.

The following insects were investigated:

SIPHONAPTERA.

Xenopsylla cheopis Roths. (tropical rat flea). Unfed adults, under 24 hours old. Large larvae.

ANOPLURA.

Pediculus humanus corporis de G. (human body louse). Adults.

DIPTERA.

Lucilia sericata Mg. (blow-fly). Adults.

COLEOPTERA.

Tenebrio molitor L. (meal-worm). Larvae: (1) small, under 30 mg. (2) large, over 100 mg.

The insects listed above were all taken in batches of five individuals and put into the silk bags, and subjected to various temperatures and humidities for periods of 1 hour and of 24 hours. Relative humidities of 0, 30, 60, and 90 per cent. were used. It was found impracticable to use air with 95 and 100 per cent. relative humidity, as under these conditions condensation took place on the insects and upset the experiments.

The limiting temperature which the insects survived for particular times and humidities was surprisingly sharply defined. There was seldom found to be a range of more than 1° C. between the temperature which gave a 100 per cent. fatality, and that at which they all lived unharmed.

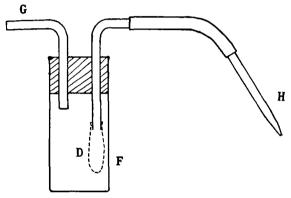


Fig. 2. Apparatus used for putting insects into the bags.

One-hour exposures.

Table I shows the results obtained with *Pediculus humanus*. It will be seen that at all humidities they were able to survive just over 46° C. The adults used in these experiments were taken not more than a week after their last moult, and had all recently fed. The deaths in the experiments were clearly caused by heat, as under favourable conditions lice will live for 3 days at least without food.

Tem- perature °C.	Relative humidity (%)				
	0	30	60	90	
47 46·8	5 dead	5 dead	5 dead	5 dead	
46·5 46·3 46·2	_	3 alive, 2 dead	2 alive, 3 dead	5 alive	
46.3	_	2 alive, 3 dead	5 alive	1 alive, 4 dead	
46.2		5 alive	5 alive	5 alive	
45 9	5 alive	-	-	I —	
45.2	5 alive	5 alive	5 alive	5 alive	
44 3	_	5 alive	5 alive	5 alive	

Table I. Results of exposing batches of five individuals of Pediculus humanus to various temperatures and humidities for periods of 1 hour.

The results obtained with adult *Xenopsylla cheopis*—unfed adults less than 24 hours old—are shown in Table II. The fleas were able to withstand only about

The Influence of Atmospheric Humidity on the Thermal Death Point 225 40.5° C. for 1 hour, and the atmospheric humidity did not appear to affect the thermal death point.

Table II. Results of exposing batches of five individuals of Xenopsylla cheopis (adults) to various temperatures and humidities for periods of 1 hour.

Temperature C.	Relative humidity (%)			
°C.	0	30	60	90
41·2 41 0 40·7 40 6 40·4	5 dead 	5 dead 5 dead 5 dead 3 alive, 2 dead 5 alive	5 dead 5 dead 5 dead 4 dead 5 alive	5 dead 5 dead 5 dead 4 dead, 1 alive 5 alive
39·8 39·4	5 alive	5 alive	5 alive 5 alive	5 alive

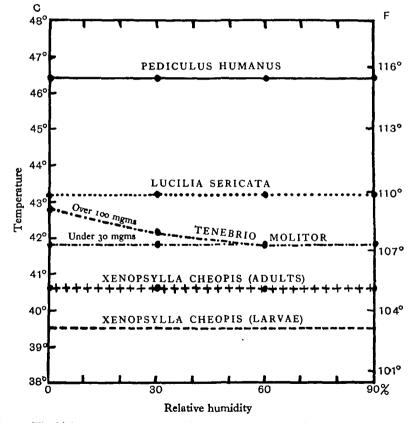


Fig. 3. The highest temperatures at which the insects can survive exposures of 1 hour, under conditions of controlled atmospheric humidity.

The results for *Lucilia*, *Tenebrio* larvae and *X. cheopis* larvae were tabulated in the same way, and Fig. 3 is a summary of the results. The curves show the highest temperatures which the insects survived for exposures of 1 hour. It will be seen

that atmospheric humidity did not alter the thermal death point of any of the insects, except the large meal-worms, which were able to stand about a degree higher temperature in dry than in moist air.

Twenty-four-hour exposures.

Xenopsylla cheopis larvae were found to be able to survive much higher temperatures for 24-hour experiments in moist than in dry air. Fig. 4 shows the results

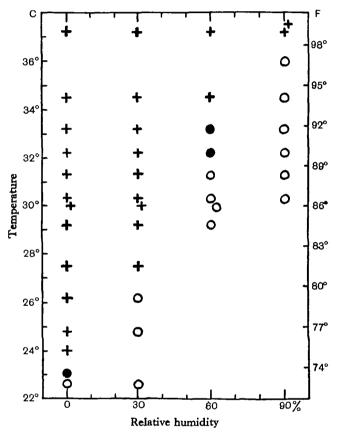


Fig. 4. Results of 24-hour exposures of larval Xenopsylla cheopis to various temperatures and atmospheric humidities. Where all died, a cross is shown; where all survived, a circle; where some died and some survived, a black dot. [At 30° C. the same humidities were used as at other temperatures.]

obtained. The results for the other insects worked with were plotted in the same way, and are all summarised on Fig. 5. It shows the highest survival temperatures for all the insects for 24 hours—the highest temperatures which were just not fatal to any of the insects. The curve for the *Lucilia* was the only one which was difficult to obtain. These insects frequently die in a few hours in tubes kept at room temperature, and after a number of attempts it was found impossible to obtain a 100 per cent. survival in the 24-hour experiments—no such difficulties occurred in

The Influence of Atmospheric Humidity on the Thermal Death Point 227 the 1-hour experiments. The curve for Lucilia shows, therefore, the temperature at which some at least of the flies were found to live.

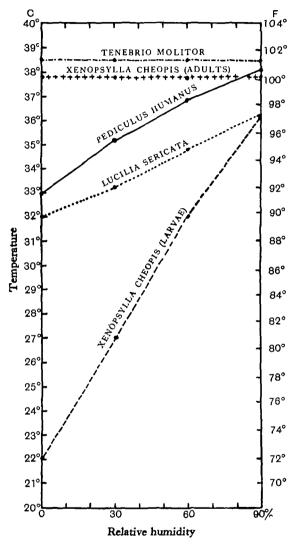


Fig. 5. The highest temperatures at which the insects can survive exposures for 24 hours, under conditions of controlled atmospheric humidity.

It will be seen that the *Tenebrio* and *X. cheopis* adults died around 38° C. regardless of humidity, and that the *Pediculus* adults also died at about that temperature in moist air, while they died at considerably lower temperatures in dry air. The death points for *Lucilia* adults and *X. cheopis* larvae, in air with a relative humidity of 90 per cent., are within a couple of degrees of the other species shown, but in dry air they die at much lower temperatures.

DISCUSSION.

Uvarov (1931) has recently summarised the work on the effects of heat and humidity in combination on the vital limits of insects. Certain workers are shown to have asserted that the humidity of the air has no influence on the resistance of insects to high temperatures, others say that a high humidity is more favourable than a low, while others express the third view that a low humidity is the more favourable. These observations cause Uvarov to state that a "simple general rule cannot be laid down."

The reason that the apparently conflicting views mentioned above have been stated is that the workers concerned have worked at different ranges of temperatures and with different species of insects. By referring to the different curves on Figs. 3 and 5, it is possible to show that all three views are correct. The atmospheric humidity has no effect on the thermal death point of the adult *X. cheopis*, whether the insect is exposed for 1 hour or 24 hours. And in all the experiments of 1 hour's duration, all the insects excluding the large meal-worms die at the same temperature in dry as in moist air. The large meal-worms find the dry air more favourable for withstanding high temperatures. In the 24-hour experiments, three of the species are able to withstand the heat best in the moist air.

It appears that these results can be fitted into a clear scheme, provided that the time factor is borne in mind. The temperature of the thermal death point of these insects depends on the length of the exposure to heat. There appear to be two different causes of death in these experiments in different parts of the temperature scale.

In the 1-hour experiments, the insects die at various temperatures, all above 39° C. The cause of these deaths seems to be the heat. Death would appear to be caused by some internal reaction of the insects, the products of which cannot be removed sufficiently fast at high temperatures. There is no definite proof of this, but many workers have shown that when insects are given a sub-lethal exposure to a high temperature, they are at first apparently quite dead, but they recover completely in the cool. If death was due to some organic injury, for instance to the central nervous system, the insects would not be able to recover completely. Also, the time which they take to recover is roughly proportional to the time they are exposed to the high temperature; this supports the view that some substance has to be removed from the body, which was produced during the exposure. The remarkable constancy in the time in which death takes place in insects at high temperatures, which makes the thermal death point for 1-hour exposures so sharply defined, also fits in with the view that death is caused by the product of some reaction which is accelerated by heat.

Reference to Fig. 3 shows that there is a range of nearly 7° between the temperature at which *Pediculus* adults and *Xenopsylla* larvae die in 1 hour, and the death points of the other species worked with occupy intermediate positions. The question is, Could all these insects die from the same cause or are they all killed by different reactions? Fig. 7 shows that prolonged exposure to hot, moist

air causes death to all the insects within a comparatively narrow band of temperature. This suggests that the difference of thermal death points of the insects when the exposure to heat is only 1 hour is the result of difference of speed in some harmful reaction, which is probably the same reaction in each species. The death of Xenopsylla larvae, etc., at lower temperatures in dry air is caused differently, and is discussed below.

Let us now consider the death of insects in dry air, at over 40°C. Necheles (1924) has shown that, in dry air, cockroaches are able to keep their bodies cool by evaporating water, and so stand higher temperatures than in moist air. I have repeated some of his experiments with cockroaches. The bulb of a small mercury thermometer was inserted in the vagina of a female cockroach, and the insect placed in a chamber at 45° C. In air of 90 per cent. relative humidity, the temperature inside the cockroach rose to 45° C. within 10 min., but when dry air was passed through the apparatus, the insect's temperature fell in a few minutes to 39° C. Buxton (1924) has shown that in the hot, dry air of deserts, the body temperature of certain large insects (grasshoppers, etc.) exposed to the full radiations of the sun is lower in live than in dead ones, and he had suggested that this is due to the greater evaporation from the live insects. The only insect worked with which seemed to behave in that way was the large meal-worm; it lived at 1° higher temperature in dry air than in moist. The calculations included in the appendix of this paper show that it is only possible for large insects to cool themselves by evaporation; small insects do not contain sufficient water in relation to the area of their body surface. This means that for the small insects, the results for which are shown on Fig. 3, the internal temperatures would be very near that of the hot air. The lice could not have survived by keeping their body temperature down to the temperature at which flea larvae survived—and in air of 90 per cent. relative humidity, they could not have lost much water by evaporation.

In the experiments conducted over 24 hours (Fig. 5), all the insects died below 40° C. In moist air the death points for all the species were within a narrow band of temperature, and death was presumably caused by the heat. But in dry air the Xenopsylla larvae, Lucilia and Pediculus adults, died at considerably lower temperatures. In these cases, death appears to be from another cause—desiccation. It is interesting to note that the insects which did not die at a low temperature in dry air—the meal-worms and Xenopsylla adults—have been shown by Wigglesworth (1932) to conserve their water in excretion. He has also shown that the larval Xenopsylla, which die at 23° C. in dry air in 24 hours, excrete water very wastefully. Buxton (1931 a) has shown that Rhodnius, which also conserve their water, die in 24 hours at much the same temperature as Xenopsylla adults, and the humidity does not affect the death point.

The way in which desiccation kills Xenopsylla larvae so readily is similar to the effects of dry air described by Davies (1928) on certain Collembola. These

¹ It was shown that when *Lucilia* adults and *Tenebrio* larvae were killed at 45° C. the amount of lactic acid present in their bodies was increased. This may possibly have some bearing on this point.

insects died very rapidly—in as short a time as 20 min.—at 25° C. in dry air, while the *Xenopsylla* larvae survived the same conditions for nearly 24 hours; but the phenomenon is essentially the same. The *Lucilia* and *Pediculus* adults behave in the same manner, but are rather less susceptible to desiccation, while *Tenebrio* and adult *Xenopsylla* are able to conserve their water and are unaffected by dry air in 24-hour exposures. But even they die sooner in dry air than in moist, as was shown for *Xenopsylla* (at 32° C. or lower temperatures) by Bacot and Martin (1924).

The critical temperature for all these insects seems to be between 36 and 39° C. Above this temperature, the insects die from the effects of heat. Below this temperature, they live several days at least, in moist air, and death occurs from starvation. In dry air, desiccation kills the susceptible insects at temperatures below the critical temperature. These two causes of death, together with the regulation of body temperature by insects in hot, dry air, explains why previous workers have put forward conflicting views as to the effect of atmospheric humidity on the thermal death points of insects.

SUMMARY.

An account is given of a technique suitable for exposing small insects to high temperature and air of controlled humidity. Data of survival points obtained from a number of species are given, for 1-hour and 24-hour experiments.

In the 1-hour experiments, the humidity of the air had no effect on the death point, except in the case of large meal-worms, which died at 1° C. higher in dry air than in moist. The temperature which any species can stand for 1 hour is sharply defined, but there is a range of 7° C. between the species of insects worked with.

In 24-hour experiments, in moist air, all the species died between 36 and 39.5° C. Their death was presumably caused by the heat. In dry air, those insects not able to conserve their water died at low temperatures—22° C. in the case of flea larvae: this was attributed to desiccation.

There seem to be two main causes of death of insects when they are killed at high temperatures:

- (1) When the temperature is over 40° C., they die from the effects of the heat.
- (2) Below 36° C. all the insects experimented with were able to survive at least 24 hours in moist air, but in dry air insects unable to conserve their water may die of desiccation.

In hot air, over 40° C., certain large insects are better able to survive in dry air, as they keep their bodies cool by evaporating water.

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¹ The work of Beattie (1928) shows that adult *Calliphora* are killed by desiccation in dry air at about 40° C. in 1 hour. For that period, 70 per cent. is the optimum humidity—in moister air the heat kills them, presumably as they cannot regulate their temperature.

REFERENCES.

BACOT, A. and MARTIN, C. J. (1924). Journ. Hyg. 23, 98.
BEATTIE, M. V. F. (1928). Bull. Ent. Res. 18, 397.
BUXTON, P. A. (1924). Proc. Roy. Soc. Lond. B, 96, 123.
—— (1931 a). Journ. Exper. Biol. 8, No. 3, 275.
—— (1931 b). Bull. Ent. Res. 22, 431.
DAVIES, M. D. (1928). Brit. Journ. Exp. Biol. 6, 79.
GOODWIN, W. H. (1914). Journ. Econ. Ent. 7, 313.
NECHELES, N. (1924). Arch. ges. Physiol. 204, 72.
PRESTON, T. (1929). Theory of Heat. London.
UVAROV, B. P. (1931). Trans. Ent. Soc. Lond. 79, part 1.
WIGGLESWORTH, V. B. (1932). Quart. Journ. Mcr. Sci. 75, 131.

APPENDIX.

This is an attempt to calculate the amount of water which must evaporate from an insect if its body is kept cooler than the air. The constants are taken from Preston's *Theory of Heat*; they cannot be taken as applying exactly to insects, but they should give some idea as to the order of the figures concerned.

If an insect in hot air cools itself by evaporation from the tracheal system, it will receive heat continuously all over its exoskeleton. This heat will penetrate into the body, but its effect will be neutralised by the heat removed from the body by evaporation of the water. The amount of heat received by the insect will depend on the difference of temperature between its body and the atmosphere, the area of the body, and the nature of the body's surface. The last of these factors will probably vary considerably, but for our purposes we can take an average figure, and assume that for every square centimetre of its surface, and for every degree centigrade which its temperature is lower than the atmosphere, an insect will absorb 0.00025 calorie per sec. (figure from Preston). This will mean roughly 1 calorie per hour.

For the insect to maintain its body at a lower temperature than the air, the heat absorbed must be got rid of, and we have assumed that this is done by evaporating water. Now at 40° C. every gram of water evaporated requires almost 600 calories. Therefore, provided that the cooling mechanism in an insect is very efficient, for every calorie of heat absorbed, about 2 mg. of water will have to be evaporated if the body is to maintain a constant temperature.

These figures can now be applied to particular insects. A cockroach weighing 1 gm. has a surface area of about 8 sq. cm. If it keeps its body 5° cooler than the air for 1 hour—if the air is at 45° C. and the insect's body temperature is 40° C.—it will absorb about 40 calories, which will evaporate 80 mg. of water, or about 8 per cent. of the insect's weight. This figure is well within the 13 per cent. of the insect's weight which Necheles found them able to evaporate.

A very large louse, weighing 3 mg., has a surface area of about 18 sq. mm. If it kept its body 5° cooler than the air for 1 hour, it would absorb nearly 1 calorie, to get rid of which about 2 mg. of water would need to be evaporated. This would be over 60 per cent. of the insect's weight, and it would be impossible for it to survive so great a loss.

It is not suggested that these figures are accurate, but they show that only larger insects, which have relatively smaller surface areas, should be able to keep their bodies cool by evaporation. It is significant that in these experiments it was only the large mealworms that survived high temperatures better in dry air than in moist.