The temperature and humidity relations of various stages in the life history of some calliphorine flies

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THE TEMPERATURE AND HUMIDITY RELATIONS OF VARIOUS STAGES IN THE LIFE HISTORY OF SOME CALLIPHORINE FLIES

By

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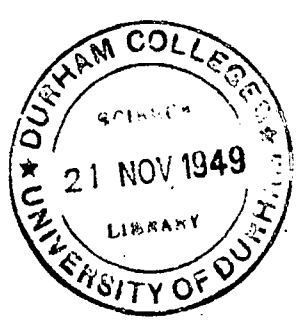
-being a thesis presented in candidature for the Degree of Doctor of Philosophy in the University of Durham, 1949.

The work described in this thesis was carried out under the direction of J. B. Cragg, M.Sc., at the Science Laboratories of the Durham Colleges in the University of Durham (July 1946 - Sept. 1948). Preliminary experiments to the work described in Part I only of the thesis were carried out at the Zoology Laboratory, University of Cambridge (Oct. 1945 - April 1946).



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GENERAL INTRODUCTION

The work herein described is confined to the study of the eggs of blowflies. No other stages in the life-histories of blowflies were studied. The investigation was carried out both by means of laboratory experiments, and by observations and experiments in the field, using sheep kept under normal British farming conditions. The effects of humidity on the survival of blowfly eggs was singled out for special attention, but the effects of temperature could not of course be ignored because of its close inter-relation with humidity. Published work by previous investigators had indicated that the eggs of the main blowfly species causing sheep myiasis in Britain, i.e. Lucilia sericata (Mg.), were very susceptible to desiccation, were almost always laid in a very dry environment when laid in sheep fleeces, and hence suffered very great mortality. No work had been done on the humidity relations of the eggs of the other species of blowflies of secondary importance as sheep myiasis producers in Britain.

The first stage of the work (Part I) was one of reinvestigating the humidity relations of Lucilia sericata (Mg.) eggs, giving particular attention to their minimum humidity requirements, and to the effects of humidity fluctuations on their survival. Later, the work was 'taken into the field' and a study of humidity conditions in sheep fleeces carried out (Part III) on more extensive lines than hitherto, particular attention being given to the occurrence of humidity fluctuations in the fleece atmosphere. The choice of the cobalt-chloride, cobalt-thiocyanate paper techniques and their modifications, facilitated this study. During the first summer of field work several laboratory laid L.sericata eggs were placed in the fleeces of living sheep, and their

fate recorded, under humidity conditions about which at least something was known by means of simultaneous cobalt-chloride paper humidity readings. Following this, the laboratory work was extended to include the study of the humidity requirements of the eggs of five other blowfly species (Part II) which sometimes cause sheep myiasis in Britain, and subsequently the survival of the eggs of these species in sheep fleeces, was tested on lines similar to parallel experiments with L.sericata eggs. This work (Part III) involving the placing of laboratory laid egg batches in sheep fleeces and recording their fate, was not carried out on as large a scale as would be desired. The experiments must therefore be considered merely as preliminary in nature.

As would be expected, several side-lines developed out of the work, one of which was the detection and study of sweating in sheep. This factor was considered to be potentially important in sometimes providing suitably humid conditions in sheep fleeces, for the development and hatching of blowfly eggs and for subsequent myiasis development. Here again, the work (Part IV) must be considered merely as being preliminary to further work.

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PART I

LABORATORY STUDIES ON THE EGG OF LUCILIA SERICATA (Mg.)

INTRODUCTION

The laboratory studies of Evans (1934) on the humidity and temperature relations of L.sericata eggs showed that they were illadapted to survive at low humidities. He found that the minimum humidity for development was above 18mm. sat. def., equivalent, at 37°C - the assumed temperature prevailing near the skin of sheep, to about 62% R.H. Yet, Davies & Hobson (1935) found that low humidities of 40-55% R.H. were of common occurrence in the fleeces of living sheep, where L.sericata eggs may be laid. Macleod (1940) obtained similar results, and concluded that a state of low humidity normally prevailed in the fleece atmosphere. Further, Davies & Hobson re-emphasized Evan's conclusion that the eggs of L.sericata were very susceptible to desiccation, and expressed the opinion that a steady humidity of over 90% R.H. for 14 hr. was necessary to ensure the hatching of eggs (accomplished in 8 hr. at 37°C) and the establishment of myiasis. The present laboratory work was undertaken to repeat Evan's experiments, and accompanied a field study of humidity conditions in sheep fleeces using methods not hitherto employed for this purpose (Part III). In the laboratory work attention was given to the effects of humidity fluctuations on L.sericata eggs, since field work indicated that considerable fluctuations in R.H. occurred in the fleece atmosphere over fairly short periods of time, in British summer weather.

A study has also been made of the structure of the egg-shell, and its water-proofing mechanism, on the lines of the work of Beament (1946 a and b) on the egg of the bug Rhodnius prolixus. Such information promised to be of

interest in relation to conditions in the natural environment of the blowfly eggs.

I. GENERAL MORPHOLOGY OF THE EGG-SHELL

(i) Microscopic structure

Methods. The shell of the ovarian egg about two days before laying was studied, when, as far as could be seen in sections, it had reached its final thickness and shape. At this stage the follicle cells had undergone almost complete necrosis, being represented merely by their flattened nuclei and greatly reduced cytoplasm, pressed flat between the follicular membrane and the contained full sized egg. In this condition the follicle cells could no longer add secreted material to the shell. The chorion then had the same external morphological features, such as the hexagonal imprints of the follicle cells and the fully developed longitudinal hatching pleats (see Sikes & Wigglesworth, 1931) as are found in the chorion of the laid egg. Eggs were removed as whole ovaries from flies, fixed in Alcoholic Bouin, and paraffin sections made. Evidence on the nature of the shell components at this stage was compared with that on the shell of laid eggs, which were not fixed but embedded in paraffin after preliminary dehydration and clearing.

Microscopic structure of chorion. In sections the chorion of the ovarian eggs about two days before laying appeared identical in structure with that of laid eggs. About 5.0 microns thick over the main part of the shell, it was thickened to about 10.0 microns in the region of the hatching pleats and at the edges of the circular area surrounding the external micropyle (Fig. 1b & c) (see Weismann, 1863).

Staining and other chemical tests showed that the chorion was composed of two main layers with a row of dark bodies forming parts of the boundary between them (Fig.2). The

outer layer, about 3.0 microns thick, was readily stained, whilst the inner layer, less readily stained, was 2.0 microns thick over most of the shell. Where the chorion was thickened, for example at the edges of the circular micropylar area, most of the increase in thickness was due to an

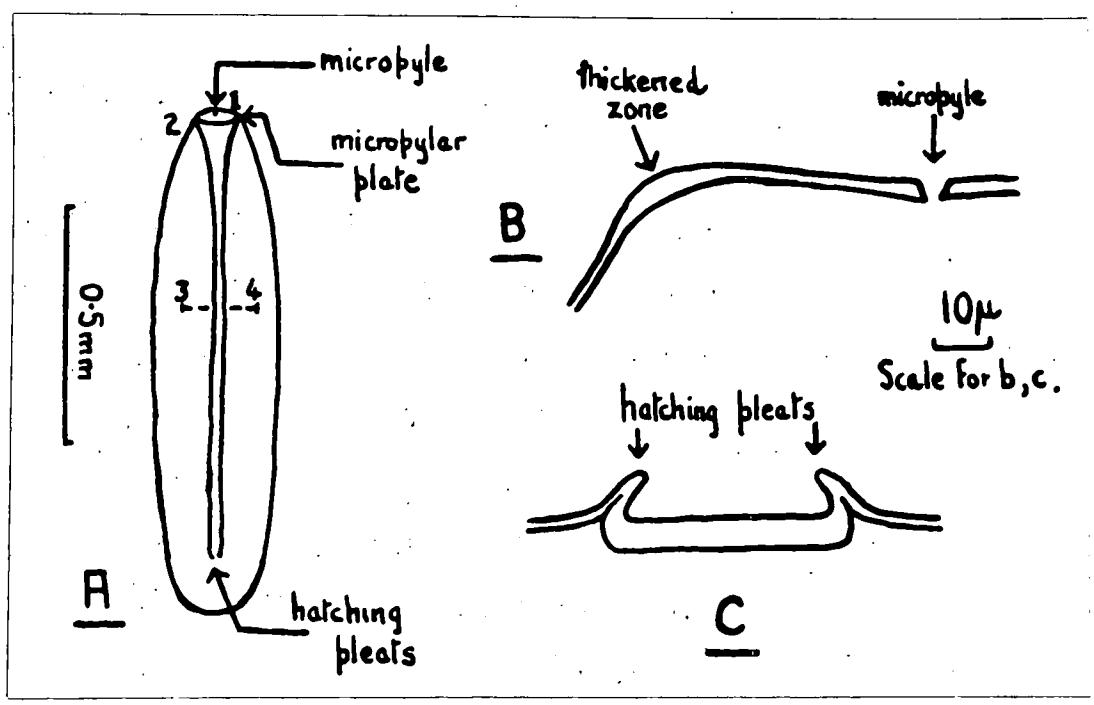

FIG.1. A, external features L. sericata egg; B, Section of chorion from 1 to 2 in A; C, T.S. chorion pleats from 3 to 4 in A.

expansion of this inner layer, the outer layer remaining about the same thickness over all parts of the shell. The dark bodies embedded in the shell were only about 2.0 microns in diameter. Further details of their structure and nature and of the vertical divisions apparently dividing the chorion into columnar elements could not be made out owing to their small size, even under a 1/12 microscope objective.

This columnar structure of the chorion of L.sericata eggs (Fig.2) agreed with the description given for the eggs of other Tachinidae by Pantel (1913).

The outer chorion layer, both in the ovarian and laid egg was stained weakly by acid Fuchsin, pale brown by Ehrlich's haematoxylin, pale blue after Mallory's triple staining, and pale green after Light Green. The inner

chorion layer was not stained by these substances at all. Both layers, however, stained deeply in basic Fuchsin, gave a positive xanthoproteic test, and negative results both with ninhydrin and warm Millon's reagent. It was evident therefore that both layers contained protein, but differences in their staining properties indicated that they were not of identical composition. The reduced affinity of the inner chorion layer was noteworthy and contrasted with the relative ease of staining the outer layer with some of the histological stains commonly employed.

The chorion as a whole when placed in the form of shell fragments in cold concentrated nitric and hydrochloric acid did not dissolve nor appear to have been changed macroscopically at all, beyond becoming rather softened, even when left in for 30 min. The chorion slowly dissolved in warm ($40^{\circ}C$) concentrated nitric acid and dissolved completely and rapidly in saturated caustic potash solution at $150^{\circ}C$, thus showing it to be completely non-chitinous. Its resistance to cold concentrated acids however indicated that it was composed of surprisingly resistant protein. The protein of insect egg-shells in general appears to be surprisingly resistant to strong mineral acids. Many of the protein layers of the Rhodnius egg shell were found to be acid resistant by Beament (1946 b). The chorion of the Culex pipiens egg is also acid resistant (Sir S. R. Christophers, in conversation). It appears also to be a general feature of insect eggs that the chorion is completely non-chitinous (Beament, 1946 b, Slifer, 1937, 1938). The general term 'chorionin' has been used for the resistant protein of insect egg shells. The elucidation of the complex layered structure of the egg of Rhodnius by Beament, each chorion layer differing in chemical composition, has however

rendered the term 'chorionin' rather inadequate.

When sections of shells were immersed overnight at room temperature in a saturated aqueous solution of p-benzoquinone, the outer chorion layer and the row of dark bodies (Fig.2) became tanned to a deep brown colour over the whole of the egg shell, whilst the inner layer remained colourless. This result was obtained both with ovarian eggs and laid eggs. It is concluded therefore that the protein of the inner layer was already tanned or, alternatively, so modified that tanning by p-benzoquinone could not occur - and that the

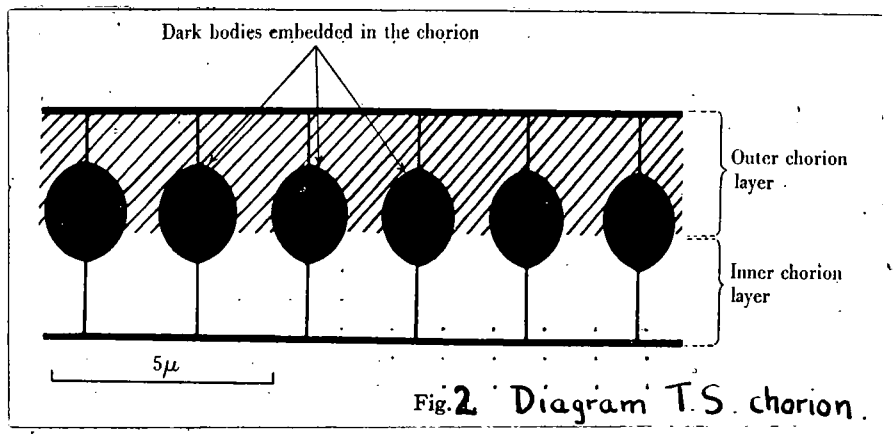

Fig.2. Diagram T.S. chorion.

outer chorion layer was largely composed of protein normally susceptible to tanning agents, and thus untanned in the original egg shell, both before and after laying.

The two-layered nature of the chorion of the L. sericata egg, with one layer not tanned and the other possibly tanned, may explain some of its peculiar mechanical properties. A strip of chorion removed from an egg and mounted so that one end is attached and the other free, underwent rapid curling movements when subjected to humidity changes. When a wet needle was held near it, the chorion strip curled downwards. When a warm needle was held near, it curled in the opposite direction, instantly returning to its former condition when the warm needle was removed. These movements are illustrated by Fig.3. It will be observed that the curling (Fig.3,b) is inwards in high R.H. and outwards in low R.H. in relation

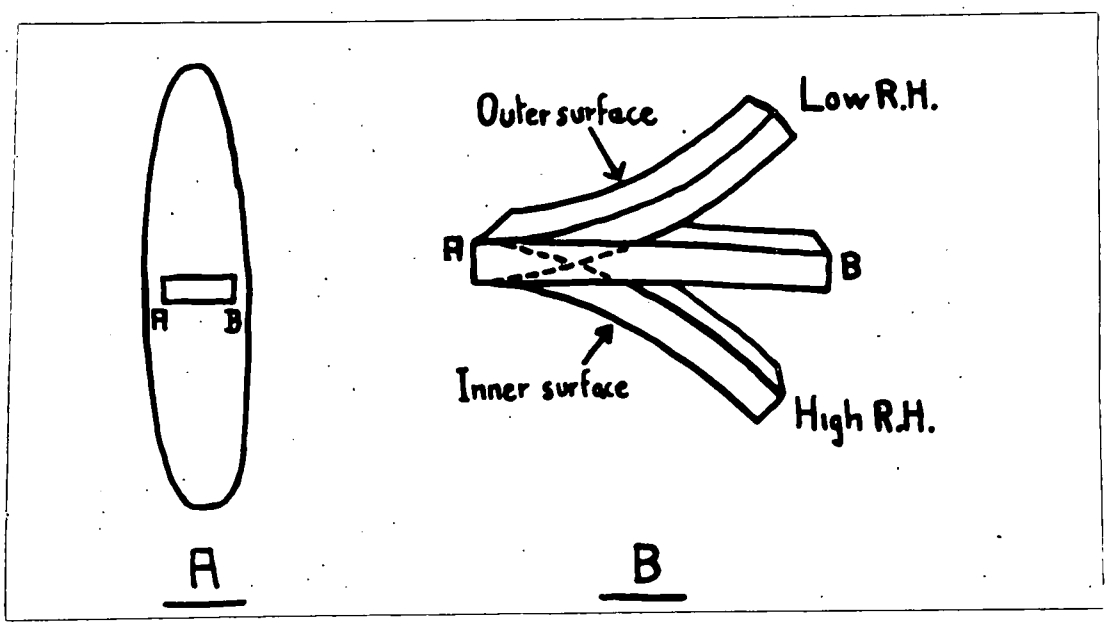

Fig. 3. Chorion curling movements; A shows orientation of chorion fragment shown in B.

to the inner and outer surface of the chorion fragment; and in addition that the curling movement occurred across the chorion strip i.e. in the transverse plane in relation to the complete egg shell. Curling in the longitudinal plane of the egg was negligible and is not represented in Fig.3(b).

These curling movements would be explained if the untanned protein of the outer chorion layer could undergo volume changes dependent on variations in air humidity, due to hygroscopicity of its protein moiety, while the inner layer, being tanned would be more rigid and possibly composed of non-hygroscopic protein. The curling movements fit in with the assumption that the outer chorion layer is hygroscopic. In low R.H. it curls in a manner consonant with a reduction in the volume, and thus a reduced water content in the outer chorion layer, and in high R.H. curls in a direction consonant with a higher moisture content and thus increased volume. This mechanical property of the chorion was when observed

an isolated phenomenon of unknown significance. Later observations on the hatching mechanism of blowfly eggs possibly throw some light on its significance (page 57).

Microscopic structure of the chorionic vitelline membrane.

The chorion of the blowfly egg closely invests an underlying membrane which encloses the ovum itself. The membrane is comparatively strong and transparent, and shows irridescent colours when isolated. The chorion can be stripped off the egg, leaving the egg retaining its original shape (Evans, 1934) within this underlying membrane, which has been termed the chorionic vitelline membrane by previous investigators (Pantel, 1913).

This membrane (hereinafter referred to as the c.v. membrane) was found in sections of ovarian and laid eggs to

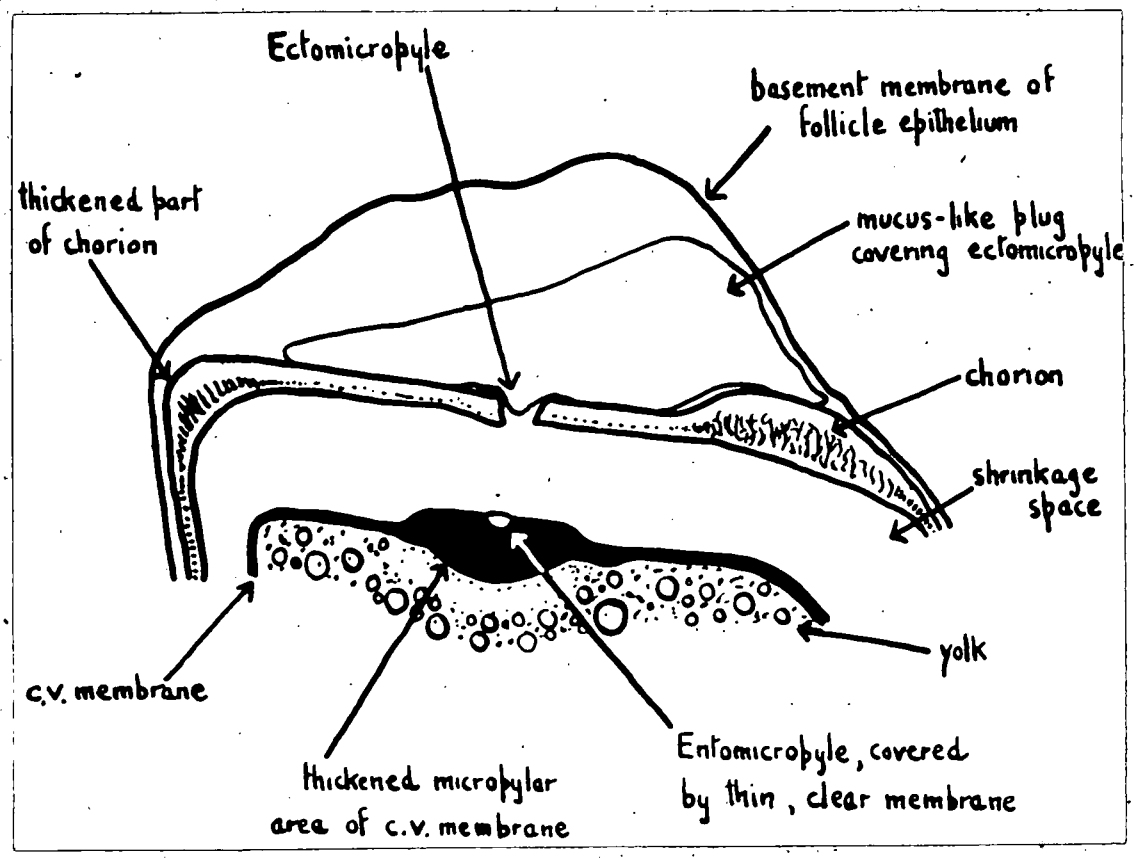

FIG. 4. L.S. through anterior end of L. sericata egg, passing through micropyles. (Ovarian egg about 2-3 days before laying.)

be about 3.0 microns thick over the general surface of the egg, but thickened to about 10.0 microns in the region of its micropyle. This micropyle in the c.v. membrane was

described by Pantel (1913) in the eggs of various Tachinidae. It has been termed the entomicropyle and is situated immediately beneath the micropyle of the chorion (ectomicropyle). The appearance of the ecto- and entomicropyle in a preparation stained with Ehrlich's Haematoxylin and Eosin is shown in Fig.4.

The entomicropyle will be seen to be a circular canal, about 3-5 microns in diameter, situated in the middle of the thickened micropylar area of the c.v. membrane. A fine membrane was clearly seen covering the outer opening of the entomicropyle (Fig. 4). ~~e.m.~~ This membrane was not stained by haematoxylin while the thickened c.v. membrane was stained to a dark brown colour.

The c.v. membrance as a whole was stained heavily by Ehrlich's haematoxylin, pale blue after Mallory's triple staining, green after Light Green, and gave a positive xanthoprotein test. It would thus seem to be of fairly easily stained protein, and in this feature resembled the outer chorion layer. It was tanned a dark brown colour in p-benzoquinone at room temperature overnight, indicating that it was composed of untanned protein. It resisted attack by concentrated nitric acid in the cold for several minutes, while on heating it dissolved more rapidly and gave off oily droplets. It dissolved rapidly in saturated caustic potash solution at 150°C. The c.v. membrane is therefore non-chitinous, and is composed mainly of protein which can be dissolved in warm nitric acid with the liberation of oily droplets.

In sections of fixed ovarian eggs, treated with 1% ammoniacal silver nitrate, the c.v. membrane gave the characteristic deep yellow colour (Lison, 1936) indicating the existence of polyphenols in it, except in the thickened micropylar region which remained pale. At the edges of

the non-stained micropylar region there appeared a row of very fine, jet black granules embedded in the membrane as though emerging from the thin part, but not continuous across the thick part (Fig. 5). This layer of silver-reducing granules

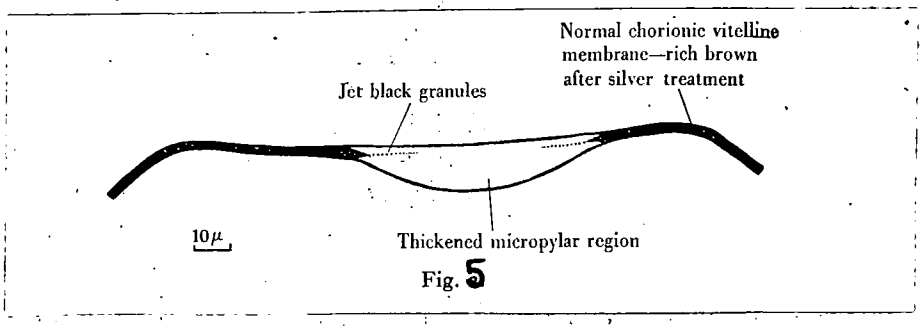

Fig. 5

may have been continued along the thin region of the c.v. membrane over the main part of the egg, but were invisible as such even under a 1/12th objective, possibly owing to the excessively fine dimensions of the membrane itself.

In the laid L.sericata egg the c.v. membrane showed no silver reducing properties. It may be mentioned here that the chorion showed no significant silver reducing properties either in the ovarian or laid egg. In some preparations of laid eggs only, the chorion sometimes showed very weak reduction of silver leading to a very pale yellow colour which was resistricted to the outer chorion layer. Since other similar preparations from sections of laid eggs failed to show this slight reduction, no significance is attached to this occasional slight silver reducing in the chorion.

The existence of considerable quantities of polyphenols in the c.v. membrane of the ovarian egg as indicated by the above observations, may be interpreted as follows. The polyphenols may be the precursors of quinones responsible for 'tanning' the inner chorion layer (Pryor, 1940 a,b). In the ovarian egg these polyphenols being situated in the c.v. membrane, are immediately beneath the inner chorion

layer. It may be that they are oxidized and utilized to tan the inner chorion layer, and are used up before they can reach the outer chorion layer. This would mean that the boundary between the untanned outer protein layer and the tanned inner layer is not clear cut, but that the degree of tanning, passing outward through the chorion, decreased quickly after about the middle of the thickness of the chorion, with completely untanned protein on the outside. In sections of chorion tanned with p-benzoquinone the boundary between tanned and untanned protein did not appear to be sharp. The absence of polyphenol in the c.v. membrane of the laid egg, indicated by its non-silver-reducing properties, may be explained by the fact that the tanning of the inner chorion layer was then complete, and that all the polyphenols had been oxidized to quinones and utilized in tanning, or removed in some other way.

(ii) Lipoid content of the egg-shell.

Ovarian egg. Eggs removed from the ovary some two days before they were due to be laid had no resistance to desiccation, although the shell as seen in sections had reached its final dimensions, and the follicle cells had undergone necrosis. Such eggs appeared similar to laid eggs except that the chorion was slightly transparent, the yellow colour of the egg yolk being visible through it. At this stage, the eggs collapsed completely through water loss if left on a dry slide for a few minutes at room temperature and humidity. They collapsed immediately in saturated sodium chloride solution at 19°C and swelled and burst in distilled water, indicating that the shell was freely permeable to water in both directions.

Fragments of chorion and c.v. membrane from these ovarian eggs with permeable shells, when placed in cold concentrated

nitric acid, either alone or saturated with potassium chlorate, showed no change within the first few minutes, but rapidly dissolved in both these media on heating. No oily droplets were formed during their disintegration. The shell of the ovarian egg did not therefore appear to contain any appreciable lipoid material.

The ovarian egg at this stage was noticed to be less turgid than the laid egg. When removed from the ovary and immediately examined within a drop of the body fluid of the parent fly, the ovarian eggs showed longitudinal wrinkles in the chorion, and appeared slightly flattened, probably by the pressure of neighbouring eggs in the ovary (see Fig. 6). When placed for a few minutes in distilled water these eggs reached the turgid sausage-shape of laid eggs, by disappearance of the chorion wrinkles until the latter was smooth. If left too long they burst as already mentioned. It appears that eggs, sometime before being laid, aquire water through their shells from the body of the parent female, either by active absorbtion by the oocyte itself, or by secretion due to the adult fly, or possibly a combination of both mechanisms.

Laid Egg. The shell had the opaque white appearance of the typical Muscid egg. The chorion, considering its thin nature (5.0 microns thick) was very strong. The water permeability of the shell was now much lower. Most eggs

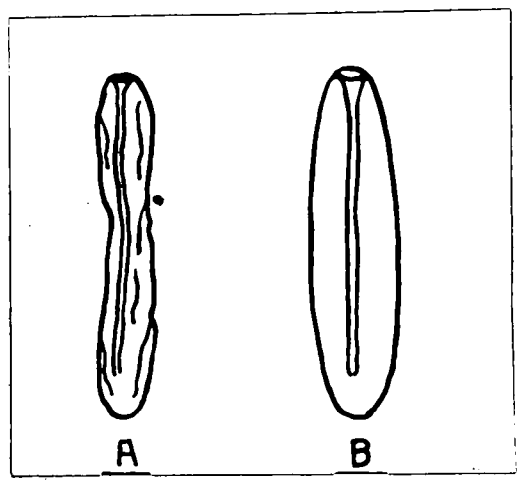

Fig. 6. A. appearance of ovarian egg, observed in blood of parent fly; B. appearance of laid egg.

could complete development whilst immersed in saturated sodium chloride solution or in concentrated picric acid at 19°C. In the ovarian egg, picric acid rapidly penetrated into the interior and fixed the oocyte. The observations on the laid egg therefore indicated that the egg shell had been proofed against the entry of large molecules such as those of picric acid, and also against the passage outward through the shell of small molecules such as water.

On placing pieces of chorion or c.v. membrane from laid eggs in cold concentrated nitric acid or cold Schulze's medium (nitric acid saturated with potassium chlorate) oily droplets were slowly given off. On heating, droplets were rapidly produced and these could be stained by Sudan III. This result is in marked contrast to that obtained with the same membranes from ovarian eggs. It appears therefore that both chorion and c.v. membrane of laid eggs contain appreciable lipoid material, whilst in ovarian eggs they contain little or no such substances, although at the latter stage the follicle cells have completed their activity and undergone necrosis. No layers in the shell of laid eggs were selectively stained by either Sudan III or Sudan Black B. Treatment of the chorion and c.v. membrane in chloroform, ether or carbon tetrachloride for 12 hr. at 30°C did not visibly affect the bulk of their lipoid content . After this treatment in fat solvents, they gave off Sudan-staining droplets in warm nitric acid in quantities comparable to those given off by untreated shells. The lipoid contained in the chorion and c.v.membrane of the laid egg appeared to be bound and could not be removed by fat solvents. The relatively large quantities of oily droplets given off by the chorion in nitric acid suggests that both its layers contain lipoid incorporated into the protein structure. Moreover, this lipoid would appear to be a product of the oocyte, because it does not appear in the shell until the

follicle cells have completely degenerated. The formation of the chorion would thus appear to take place in at least two stages; the main protein structure is first laid down by the follicle cells; later lipoid material is added to the protein structure, this time by the oocyte, and becomes bound to the shell proteins. With the incorporation of this lipoid into the chorion and c.v.membrane, there occurred a marked increase in the rigidity and strength of the egg shell. In the ovarian egg fragments of chorion or c.v. membrane were very weak structures easily torn with a fine needle. In contrast fragments of chorion or c.v. membrane from the laid egg were much more rigid, and were not so easily torn. It is to be expected that lipidization of the protein of the shell would increase the rigidity of the shell. On the other hand this process of lipidization alone would not be expected to reduce markedly the shell's water permeability. It will be shown later that the bound lipoid in the shell is not itself responsible for waterproofing the egg.

The effects of fat solvents, such as ether, chloroform and carbon tetrachloride on the water permeability of intact laid eggs was gauged by comparing their behaviour in saturated sodium chloride solution at 19°C before and after treatment in a particular fat solvent. It was found that immersion in chloroform at 30°C for 12 hr produced a radical increase in the permeability of the shell, making it freely permeable to water in both directions. Laid eggs so treated in chloroform, swelled in distilled water and collapsed in strong saline solution. This indicated that some chloroform soluble material responsible for waterproofing the egg shell was removed or disorganized during the treatment. Ether and

carbon tetrachloride produced substantially similar effects. The water permeability of the eggs treated for 12 hr at 30°C in chloroform was of the same high order as that of ovarian eggs. These de-waterproofed laid eggs, however, showed a difference in behaviour in strong sodium chloride solution, to that of unwaterproofed ovarian eggs. In the former eggs, when water was withdrawn by osmosis from the egg (in strong saline) the oocyte and its enclosing c.v. membrane shrank inwards from the inner surface of the chorion which retained its original shape (Fig. 7a). In ovarian eggs,

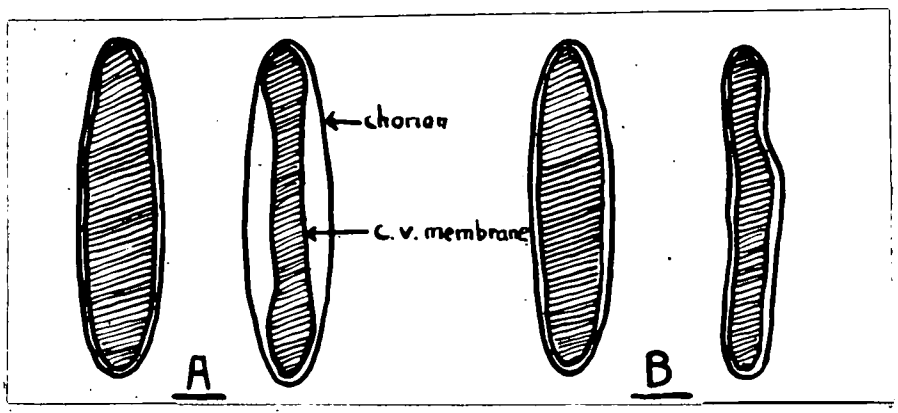

FIG. 7. Behaviour of dewaterproofed laid egg (A) and unwaterproofed ovarian egg (B) on transfer from distilled water to strong saline.

however, when placed in strong saline, both the chorion and c.v. membrane collapsed together as the oocyte lost water (Fig. 7b). These differences between ovarian and laid eggs may be explained by the greater rigidity of the lipidized chorion in the latter eggs in contract to the limp, unlipidized and hydrated chorion of the ovarian egg.

When batches of eggs, each laid by one fly, were immersed within 15 min. of laying in saturated saline at 19°C it was found that a small proportion (usually about 10%) of the eggs in each batch showed signs of waterloss by shrinkage within 15 min. The following observations suggested that these non-waterproofed eggs were those that were laid last by each fly. Eggs were dissected from flies in the process of oviposition, and their behaviour in saturated saline at 19°C compared with that of eggs already laid a few minutes earlier by the same flies. About half the eggs due to be

laid within the next few minutes, from various positions in the oviducts and vagina, showed signs of water loss after 15 min. under the above conditions - the rest were unaffected. Only about 5% of the eggs already laid by the same flies were of this non-waterproof type. The process of waterproofing the shell would thus appear to occur shortly before laying, and to be unfinished in some eggs when they are laid.

The following observations show that the waterproofing of the egg was not due to the bound lipoid which appeared in the shell shortly before laying. Eggs of the non-waterproofed type, macroscopically identical with laid eggs, were removed from the oviducts of a fly in process of ovipositing. Fragments of chorion and c.v. membrane from such eggs were placed in warm concentrated nitric acid, and droplets stained by Sudan III were given off. This showed that although the shell protein had been lipidized at this stage, the eggs were still permeable to water in both directions. The water proofing of the egg must therefore be due to some other change in the shell occurring before laying.

Occasionally laboratory culture flies laid complete batches of non-waterproof eggs with slightly transparent chorions. Such eggs shrivelled up at all humidities below saturation, and did not complete development. Gough (1946) records that the wheat bulb fly (Leptohylemyia coarctata Fall) laid a few eggs of this type when the egg batches in question contained more eggs than the average number of ovarioles in that species. In L.sericata, eggs which shrivelled up were not observed to be more frequent in large batches than in small ones, and may have been partly a result of laboratory treatment of the fly cultures.

(iii) Effect of temperature on rate of water-loss through the egg shell

Batches of eggs, weighing 20 - 30 mg. freshly laid by

a laboratory culture of flies, were teased apart on fragments of silver foil so that they formed a layer one egg thick, and were examined under a binocular to ensure that damaged eggs were not used in experiments. After storage in dry air at room temperature for 30 min. to remove all water from the outsides of the shells, they were again examined under a binocular and any damaged eggs removed. The rates of water loss from such batches when exposed to dry air at various temperatures were then measured. The foil with eggs attached was suspended in a corked flask fitted with a thermometer and containing phosphorus pentoxide as desiccant, the eggs being a standard distance above the drying agent. This apparatus was similar to that employed by Wigglesworth (1945). The flask was placed in a thermostatically controlled oven. Weighings were made by means of a torsion balance (50 mg/ 0.05 mg.). Owing to the large surface area in relation to volume, of an egg batch so spread on silver foil, water loss rates decreased rapidly at high temperatures owing to depletion of water in the eggs. Short exposures of only 15 min. at each high temperature (40 - 55°C) were therefore used, longer exposures of $\frac{3}{4}$ to $1\frac{1}{2}$ hrs. being employed at lower temperatures (20 - 38°C).

The surface areas of batches were estimated by making camera lucida drawings of eggs squashed flat under a cover-slip. By making drawings of ten such eggs picked at random from batches, and averaging the areas calculated for the eggs, a measure of the surface areas of L.sericata eggs was obtained and was 0.86 sq.mm. This figure was used for all the batches employed. In view of the variation in size of L.sericata eggs (see page 84) it is certain that these surface areas of batches were only approximations. But since large numbers of eggs were employed at each temperature (300 - 800 eggs) the variation in size was

probably similar in each lot of eggs so that errors in surface area calculations were of the same order each time. To obtain the surface area of each batch, the eggs were counted after the water loss rate at a particular temperature had been measured, and the number multiplied by the average surface area per egg given above.

By means of the above method, water loss for the eggs at various temperatures was expressed in terms of mg. water loss/sq.cm surface area/ hr. This provided data comparable to that obtained by Wigglesworth (1945) on transpiration through the insect cuticle and by Beament (1946a) on that through the shell of the Rhodnius egg. The temperature/ water loss curve obtained in this way is shown in Fig. 8.

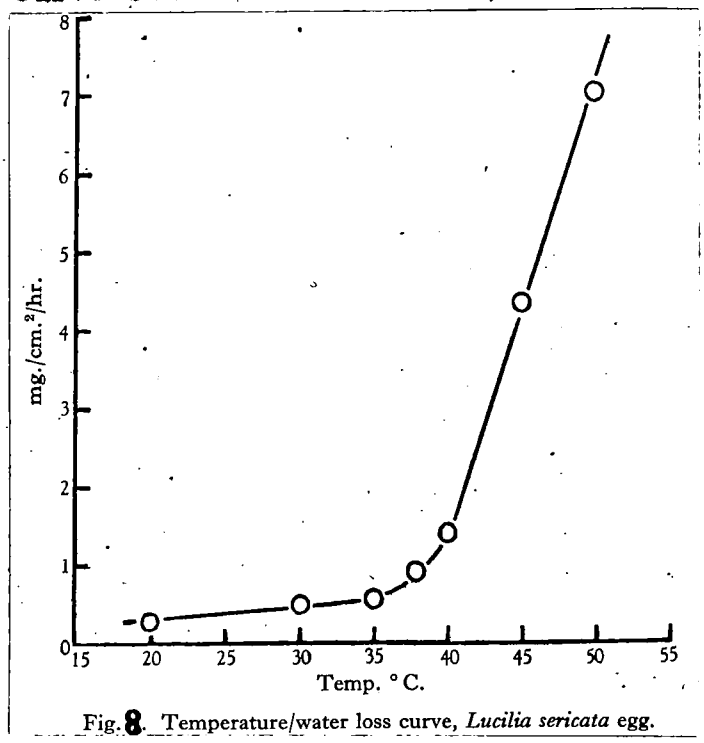

Fig. 8. Temperature/water loss curve, *Lucilia sericata* egg.

It will be seen that the rates of water loss at temperatures of 20 - 35°C were low and increased very slowly with rise in temperature within this range. But at about 38°C the water permeability of the shell increased abruptly and continued to increase much more rapidly with rise in temperature above that figure. It will be obvious from Fig. 8 that the rapid water loss at the higher temperatures necessitated

the use of separate egg batches for each new temperature. This curve (Fig. 8) with its fairly well defined 'critical temperature' above which the increase in rates of water loss with temperature is rapid, and below which it is much slower, is similar in these features to that obtained by Beament (1946 a) in comparable experiments on the Rhodnius egg, which he showed was waterproofed by a wax layer, very like that waterproofing insect cuticle. The curve is also similar in having a 'critical temperature' to those obtained by Wigglesworth (1945) in experiments on the water loss from adult, pupal and larval insects of various orders, and by Beament (1945) for the permeability of membranes covered with films of waxes extracted from insect cuticles.

It appears therefore that the Temp./water loss curve for L.sericata egg (Fig. 8) can be considered to be evidence that the egg of that species is waterproofed by a lipoid layer rather similar to that of the Rhodnius egg, and to the waterproofing layer of insect cuticle. The lipoid waterproofing layer of the L.sericata egg would thus have a 'critical temperature' in the region of 38°C.

Other features in the effects of temperature on the permeability of the L.sericata egg-shell are similar to those for insect cuticle. The rate of water loss in dry air at 30°C was found to be of the order of 0.5 mg/sq.cm/hr. (Fig.8). One batch placed in dry air at 30°C was found to lose water at the rate of between 0.4 - 0.5 mg./sq.cm./hr. This batch was then exposed to 50°C in air of high humidity, and then placed in dry air at 30°C, where it lost water at slightly over 1.0 mg./sq.cm./hr. Exposure to 40°C for 30 min. had little or no effect on the subsequent water loss rate in dry air at 30°C (0.5 mg. before exposure to 40°C, 0.6 mg. afterwards - the difference being within the likely error in successive

weighings). However, exposure to 50°C for 10 min. produced the increase noted above. High temperature appeared to cause a permanent increase in the water permeability of the shell. This was found to be the case with insect cuticle in similar experiments on Rhodnius nymphs by Wigglesworth (1945). Exposure to 50°C greatly increased the permeability of the L.sericata egg-shell in both directions. Batches of eggs desiccated at 50°C until they were heavily dimpled regained their water slowly in saturated air at 37°C, and more rapidly in saturated air at 50°C, so that the dimples in the shells disappeared. For example, a batch weighing 20 - 15 mg. was desiccated in dry air at 50°C for 40 mins. Its weight was then 9.8 mg. Placed in saturated air at 37°C it regained 1.65 mg in 3.5 hr - a regain rate of approximately 0.47 mg./hr. It was then placed in saturated air at 50°C, and regained 2.75 mg in 1.3 hr, a rate of approximately 2.1 mg./hr. In addition, these eggs eventually regained water to such an extent that they weighed about 5% more than their original weight, prior to initial desiccation, so that they appeared slightly fatter than normal laid eggs. Evans (1934) showed that eggs, partly desiccated at temperatures below the critical temperature of 38°C found in the present work, did not regain water in saturated air.

The temperature/water loss curve (Fig.8) was found to be similar in live, and in dead eggs killed with ammonia fumes 24 hr. before use. Thus the waterproofing of the egg is not due to an active physiological mechanism.

(iv) Effect of a detergent and abrasion on shell permeability

The I.C.I. detergent C.09993 was used (see Wigglesworth, 1945). This substance is soft rather like low melting point paraffin wax. It is readily miscible with water and is itself freely permeable to water. A small quantity of

C.09993 was mixed with water to form a very thin paste. When this was applied to the outer surface of the intact c.v. membrane after the chorion had been stripped off, it caused a marked increase in the rate of water loss through this membrane in dry air at 19°C. Eggs so treated collapsed completely under the above conditions in 12 min. Untreated controls with chorion removed collapsed under similar conditions in 1hr. . This suggests that there was an emulsifiable material present on the outside of the c.v.membrane, responsible for reducing its permeability to water. Application of the detergent to the outer surface of the chorion of the intact egg also affected the rate of collapse of eggs in dry air. (Table I). These results suggest that the detergent

Table 1. *Effect of* C09993 *applied to outside of chorion, twenty eggs in each group*

	Time taken to collapse in dry air, 19° C.
Untreated	Not collapsed within 18 hr.
Whole egg-shell treated	Completely collapsed in 15 hr.
Wide end of egg treated	Shell dimpled but not collapsed in 18 hr.
Micropyle end of egg treated	Completely collapsed in 15 hr.

penetrated the chorionic micropyle (ectomicropyle) and, spreading on to the c.v.membrane, affected its water permeability.

Eggs minus the chorion when placed in strong saline showed signs of water loss within 30 min. after washing in warm ether (30°C) for 5 min. they collapsed completely in 1 - 2 min. in strong saline. On return to distilled water they regained their original shape within 10 min. and occasionally took up water to such an extent that they burst. Freshly laid eggs minus the chorion required only 5 min. washing with warm ether to become as permeable as described above. Similar eggs, killed in ammonia fumes and stored for two days required immersion for 30 min. in ether to produce the same effect. These results again suggest the existence

of a lipoid layer on the outer surface of the c.v.membrane.

The effect of abrasion on the permeability of the shell was gauged by comparing the dimpling-rates of eggs in dry air at 19°C, after drawing several times through a layer of fine alumina dust on a glass slide, with those of untreated controls. It was found that abrasion of the outside of the chorion produced no appreciable increase in rate of water loss through the shell. Eggs with chorions removed drawn through alumina dust so that the outside of the c.v. membrane was exposed to abrasion, lost water very rapidly. Such eggs became heavily dimpled within 3 min. in dry air, and completely collapsed in 10 min. by which time untreated control eggs with chorions removed had not yet lost their original fat sausage-shape. When the c.v.membrane of eggs with chorions removed was dusted, but abrasion of the membrane avoided by keeping them stationary and not moving them through the dust, no marked increase in rate of water loss occurred. This suggests that the waterproofing layer was not a mobile oil which could be absorbed by the alumina dust.

All these experiments strongly suggest that the waterproofing layer is situated between the c.v.membrane and the closely fitting chorion. When the chorion is removed damage to the waterproofing layer would be expected, so that the egg minus the chorion would become very susceptible to desiccation. Possibly some of the waterproofing lipoid is left on the outside of the c.v.membrane, as indicated by the results of abrading it, painting it with C.09993, and washing it with fat solvents. Evans (1934) records that the egg of L.sericata minus the chorion retains some slight desiccation resistance, but his statement that the chorion is responsible for the resistance of the L.sericata egg to desiccation is misleading.

(v) Site of water loss

Using the rate of dimpling of the shell in dry air a measure of water loss, occlusion of the external micropyle and/or the longitudinal hatching strip between the hatching pleats (Fig. 1a) of the chorion with a layer of cellulose paint or paraffin did not affect the rate of water loss from the egg at room temperatures; after 5 hr. under the above conditions both treated and control untreated eggs were dimpled to about the same extent. Eggs so treated completed development at appropriate humidities, showing that closing the micropyle or covering the hatching strip did not cause asphyxiation of the embryo. Thus it seems that, in effect, water loss occurred over the whole of the surface of the shell, and not through a restricted area such as the micropyle or hatching strip.

Beament (1946) has shown that the waterproofing wax layer of the Rhodnius egg is continuous over the inner openings of the micropylar canals, and points out the necessity for such an arrangement in order to prevent rapid water loss. Since in the L.sericata egg occlusion of the micropyle with waterproof paint did not appreciably reduce water loss for the egg in dry air, it would appear that in this egg also the waterproofing layer is continuous over the entomicropyle and the inner end of the shallow ectomicropyle (see Fig. 4, page 14). The clear membrane shown in this figure covering the outer opening of the entomicropyle may possibly function as a continuous substratum for the lipoid layer over the perforation.

II. EFFECTS OF EXPOSURE TO DIFFERENT HUMIDITIES

(i) Methods

Eggs were obtained from a laboratory culture of flies kept at 22 - 25°C. By careful observation, after meat was placed in the cage, it was possible to determine the time of

laying of a batch to within 5 - 10 min. Experiments were carried out at 30, 34, 37, 38, 39 and 40°C. Numerous preliminary experiments at these temperatures were carried out in electric incubators, but it was found that the temperatures within them fluctuated during an experiment about ± 0.5°C and sometimes as much as 1.0°C. Desiccators were used as humidity chambers. Owing to their thick glass walls it was not certain whether in all experiments the eggs were being subjected to exactly the right temperature. Further confirmatory experiments using large numbers of eggs were carried out using a thermostatically controlled water bath. Vaselined, ground-glass stoppered museum jars were employed as humidity chambers, and were sunk in the water-bath on a wooden shelf. Five or six such jars were kept in the bath, each jar forming a chamber at different humidity. The temperature with the jars and of the water of the bath were found to vary less than ± 0.1°C during an experiment. A 0 - 50°C thermometer was kept immersed in the bath to check water temperatures, while the water was kept in visible motion by a stirrer. Preliminary trials were made with the apparatus running with no eggs in the humidity jars. One of the jars was fitted with a thermometer so that the bulb was in the position normally occupied by the blowfly eggs. This thermometer in the jar had been carefully checked against the other thermometer immersed in the water, and selected because it showed the same reading as the other when both were immersed in the bath ½ in. apart. The jar fitted with the thermometer was kept at various positions along the shelf under water. By this means it was found that the temperature recorded within the jars at any position along the shelf was the same as that read off the water immersed thermometer. In this way the actual temperatures which the eggs were subjected were ascertained.

In both the incubator and water-bath experiments constant relative humidities of from 0 - 95% were maintained by means of sulphuric acid/water mixtures, made up in all cases to the appropriate specific gravity at 25°C according to the data of Wilson (1921). The s.g. of the acid/water mixture was checked at intervals if there was any doubt by weighing 10 cc. samples, and the R.H. within the vessels checked either by paper hygrometers (Edney hygrometers) or by cobalt chloride papers (Solomon, 1945). Trials were made with caustic potash solutions as constant R.H. solutions (Buxton, 1931) but it was found that they were not as easily made up with accuracy as were sulphuric acid mixtures, and were also more unpleasant to handle, and liable to changes within comparatively short intervals of time owing to some reaction between the solution and the glass of the humidity chambers. The advantage of caustic potash solutions owing to their power of absorbing carbon dioxide was not valid in the present experiments.

Eggs, either separated or as complete batches were incubated within the desiccators or museum jars on 3 X 1 in. slides, which could be transferred from one desiccator to another during the incubation period, with the minimum of disturbance in humidity conditions. In this way the effects of short periods at very low R.H. could be measured. Where counts of the various stages reached by the eggs at various humidities were required, the batches were always separated into their constituent eggs before incubation. Where the time of hatching, or variation in time of hatching within a group of eggs was to be determined, desiccators fitted with flat plate-glass lids were used, to facilitate observation of hatching. In all experiments a set of control eggs for each batch was allowed to develop and hatch at 100% R.H. Some delay in getting the eggs incubated after they had been laid was inevitable where the batches laid by the flies had to be

separated. This delay was always less than 30 min. and was more usually 10-20 min. During this time the eggs were at room temperatures (14 - 19°C). These temperatures are low and since the experimental temperatures were from 30 - 40°C, eggs would have undergone very little development before they were finally incubated. Free water was removed from the outsides of the eggs after they had been separated and before they were incubated, by absorbtion onto filter paper strips.

To provide data comparable to that obtained by Larsen (1943) on the eggs of dung-breeding Diptera, incubator experiments were also carried out using vessels similar to those used by her as humidity chambers. These were solid watch-glasses sealed by a glass plate vaselined around the edges, and on the underside , of which the eggs in question were attached (see Larsen, 1943, Fig.2). In the present experiments humidity in these vessels was controlled as before by sulphuric acid mixtures placed in the watch glasses so that the eggs lay within 1 cm. of the surface of the desiccant. The main disadvantage of these solid watch glasses as humidity chambers lay in the fact that only small numbers of eggs could be placed in each one, owing to the restricted volume, and the consequent danger of depletion of oxygen or accumulation of carbon dioxide during an experiment.

Eggs laid by flies from 1 - 4 weeks old were used. No significant differences were found in their humidity relations which could be attributed to differences in the age of flies. Among eggs laid by flies over 3 weeks old considerable infertility was encountered.

In general the water-bath experiments confirmed the results obtained by preliminary experiments in incubators.

(ii) Minimum humidity for development

From incubator experiments the minimum humidity for development at 37°C for L.sericata eggs was found to be about 50% R.H. (23.53 mm.sat.def). The proportion of eggs reaching the prehatching stage at this humidity varied considerably between different batches laid by separate flies, as shown in the results of fourteen experiments given below:

% eggs completing development	92	40	42	66	50	80	11
at 50% R.H., 37°C.	6	16	20	29	18	45	16

Some of these results were obtained by incubating, simultaneously in the same humidity vessel, eggs laid by different flies. Even in such circumstances the proportion of eggs completing development showed differences between batches although they had been treated identically. There must, therefore, have been considerable variation in the resistance to desiccation of eggs from different flies, as well as variation within a single batch laid by the same female.

The results of experiments in solid watch-glasses as used by Larsen (1943) were substantially similar to those in which desiccators were used as humidity chambers.

To confirm the above results, eggs were obtained from wild flies in the field by attracting them to oviposit on sheep, using the technique of Hobson (1937). Results identical with those described above were obtained with such eggs. Thus the low minimum humidity figure of 23.5 mm. sat.def. found in the present work could not have been due to using laboratory flies accidentally selected for desiccation-resistant eggs. In the circumstances it is, therefore, impossible to account for the lower figure of above 18 mm. sat.def. obtained by Evans (1934).

Experiments carried out in a constant-temperature water-bath as described above confirmed the conclusion that a proportion of L.sericata eggs could complete development at 50%

R.H., 37°C. (They were unable to hatch at this humidity - see page 38). In three experiments at 37°C (Appendix page 163) it was found that 5.6, 6.9 and 69.7% of the eggs completed development under these conditions (50% R.H). It is seen that there is again considerable variation in the proportion of eggs completing development. In one experiment (Appendix, page 163) a few eggs completed development at 45% R.H. 37°C. This is an abnormal result, and was not obtained in any other experiment, and since the proportion of eggs involved was only 6.3%, it cannot be said that L.sericata eggs can normally complete development at this humidity.

In saturated air at 37°C development was found to take 7.6 - 7.9 hr. a figure agreeing closely with that of 7.5 - 7.8 given by Wardle (1930). At 50% R.H. at the same temperature it was found that development took much longer, varying from 11.75 - 14.0 hr. This was found by incubating series of eggs at 50% R.H. 37°C and transferring them after various intervals to saturated air at the same temperature. When some of the eggs hatched fairly quickly after transference to saturated air it was known that they must have completed development while at 50% R.H. When they required some time in saturated air to hatch it was then concluded that they had not completed development within the period spent at 50% R.H. The results of one such experiment are given in Table 2. The eggs in this particular batch after about

TABLE 2

Time taken to complete development at 50% R.H. 37°C.
20 eggs in each group.

Group No.	Hours incubated at 50% RH.	Time taken to hatch after transference to 100% RH, 37°C.
1	12.5	1 egg hatch in 1.3 hr.
2	13.3	1 egg hatched after 0.2 hr.10 more within 0.5 hr, 3 more in 1.2 hr.
3	14.5	No hatch
4	15.0	1 egg hatched after 4.0 hr.

13.3 hr (Group 2) at 50% R.H. 37°C, hatched rapidly after transference to saturated air. This shows that development in L.sericata eggs was considerably retarded by low humidity. Retardation of development in the eggs of this species, caused by low humidities was also found by Evans (1934) and in the eggs of dung-breeding Diptera by Larsen (1943).

Even comparatively short exposures to 50% R.H. 37°C, retarded development of eggs which spent the rest of their incubation in saturated air. For example, an egg batch was divided into two parts. One part was incubated at 100% R.H. 37°C and hatched in 7.7 - 7.8 hr. The other part was incubated at 50% R.H. at the same temperature for 4.0 hr, and then quickly transferred to the same vessel as the other part of the batch. This second part of the batch did not hatch until about 9.0 hr. after initial incubation, showing a delay of 1.2 - 1.3 hr. in development due to exposure to 50% R.H. for 4.0 hr.

At 34°C, the minimum humidity for development was found to be about 40% R.H. (23.8 mm.sat.def) when as before a variable percentage of eggs completed development, usually between 20 - 40%. At 30°C the minimum humidity was found to be about 25 - 30% R.H. Sometimes eggs completed development at 25% R.H. at other times they failed at this humidity but succeeded at 30% R.H.

Evidence has been given (page 24) that the waterproofing layer of the L.sericata egg has a 'critical temperature' in the region of 38°C, above which the rates of water loss through the shell increased rapidly with rise in temperature. The humidities required by eggs to complete development at 38, 39 and 40°C were therefore investigated. Water-bath experiments indicated at a high proportion of eggs could complete development at 80% R.H. 38°C (Appendix page 163-4). The result of one incubator experiment at 38 and 39°C shown

in table 3 below showed that at 38°C a high proportion of eggs could complete development at 75% R.H., but none at 70%. The same experiment (Table 3) showed that at 39°C,

Table 3

Humidity % R.H.	Percentage eggs completing development	
	38° C.	39° C.
95	90	75
80	95	10
75	95	0
70	0	0
65	0	0

some eggs completed development at 80% R.H., but none at 75%. Their ability to complete development at 80% R.H. at the same temperature was confirmed by water-bath experiments (Appendix page 164). In two experiments at 40°C (Appendix page 164-5), in one no eggs completed development below 95% R.H., while in the other small numbers of eggs completed development at 90% R.H. (4.5%), 85% R.H. (5.5%) and 80% R.H. (0.9%). The minimum humidity for development at 40°C is presumed to be about 90 - 95% R.H.

The minimum relative humidities for development of L.sericata eggs at these high temperatures may be summarized as follows:-

$$\frac{38^{\circ}C}{75} \qquad \frac{39^{\circ}C}{80} \qquad \frac{40^{\circ}C}{90\text{-}95}$$

Since at 95% R.H. 38 and 39°C, a high proportion of the eggs completed development, it is concluded that the high temperatures alone were causing some mortality apart from the effects of humidity. (In these high temperature experiments it will be seen that frequently, fewer eggs completed development at 100 than at 95% R.H. This was presumably due to condensation on the outsides of the eggs interfering with respiratory exchange. A similar phenomenon is mentioned by Larsen (1943)). At 39°C development was found to be retarded by high temperature alone, taking 9.0 - 9.2 hr. (see page 56) compared with 7.6 - 7.8 hr. at 37°C. This retardation in development does not account

for the much higher R.H. (80%) required for development at 39°C, compared with that at 37°C (50%). The product of saturation deficiency x time (hr.) for L.sericata eggs at 37 and 39°C was as follows:-

50% R.H.37°C 12.0-13.5 hr. x 23.5mm.sat.def. = 282.0 - 317.5

80% R.H.39°C 9.5-10.0 hr. x 10.5mm.sat.def. = 99.75- 105.0

Since at 95% R.H. 39°C considerable proportion of eggs completed development, this sharp fall in the desiccation resistance factor from 37 to 39°C must be due to the harmful effect of the high temperature on the lipoid water proofing layer leading to increased permeability of the shell.

In view of the fact that the waterproofing layer of the egg has a 'critical temperature' in the region of 38°C the maximum temperatures found in their natural environment - the sheep's fleece - is of interest. Burtt (1945) found that the temperature, as measured by a thermocouple, on the face and ears of a sheep was 35-37°C. In the present work temperature readings were taken by inserting a clinical thermometer into the fleeces of two sheep under summer field conditions. Sixteen readings taken against the skin at the midback averaged 38.6°C when the sheep were in the shade. When their fleeces were warmed by direct sunlight the skin temperature rose to 39.8°C (average of three readings). Temperatures at 3 cm. off the skin, within the thickness of their fleeces varied greatly, frequently being below 30°C when the sun was clouded, but rising to above 40°C in direct sunlight. These observations indicated that L.sericata eggs laid very close to the skin of the sheep may have to develop at temperatures approaching the 'critical temperature' of their waterproofing layer.

(iii) Minimum humidity for hatching.

In the preceding section it was shown that the minimum R.H. for development of L.sericata eggs depended on

temperature. Saturation deficiency would appear to be a better unit for measuring the minimum humidity than R.H. Unlike the development processes, that of hatching would appear to be independent of temperature and to depend very largely on R.H. for its operation or non-operation, since at all temperatures from 17 to 37°C, the minimum humidity for hatching was approximately the same - namely around 60% R.H. The probable explanation of this dependence of hatching on R.H. alone, is discussed later (page 71-8).

Davies and Hobson (1935) found that at 37°C L.sericata eggs required 90 - 100 % R.H. for rapid hatching. The present work amply confirmed this. With progressively lower humidities, from 90 to 60% R.H., hatching was found to be less complete, and much slower than from 90 - 100 % R.H. Not only was the time taken by each larva to extricate itself from the shell after it had succeeded in splitting it much longer at these lower humidities, but also the time taken for each larva to initially split its shell was longer and more variable, ~~(see page ——)~~. At 100% R.H. a larva was found to take usually about 0.5 - 2 min. to extricate itself from its shell after initial splitting. At 90% R.H. larvae took from 5 - 15 min. and at 60% R.H. anything from 20 - 40 min.

The variation in hatching times of the various eggs in a batch was only 10 - 20 min. at 100% R.H. At 90% R.H. it was usually about 45 - 40 min. and at 60% R.H. usually about 2 - 3 hr. Excluding infertile eggs, the percentages of eggs whichhatched decreased gradually from 90 - 100 at 90% R.H. to nil at 55% R.H. Within this range of humidities, many of the unhatched eggs contained fully developed larvae. At 60% R.H. some eggs usually hatched, while at 55% R.H. hatching sometimes occurred to a small extent, and at other times did not occur at all. At 50% R.H. no hatching occurred

except in special cases mentioned below (page 40). Example figures showing the % hatch at various humidities, and the minimum humidity for hatching are given in Table 4. (For key to column headings ("H" etc.) see Appendix page 161).

TABLE 4

Decreasing % hatch with degreasing R.H.

% R.H.	Expt.A. H.	PH.	No. of eggs	Expt. B H.	PH.	No. of eggs
70	84.8	9.1	33	-	-	-
65	48.3	29.6	27	38.8	36.9	103
60	35.8	35.8	28	12.6	57.3	111
55	3.7	81.5	27	1.5	52.7	95
50	0.0	69.7	33	0.0	6.9	58

That the progressively incomplete hatching in batches incubated in decreasing humidities from 90 - 60% R.H. was due to larvae being unable to break out through the shell, which in some way was rendered more difficult to split at low humidity, was shown by the following observation. Eggs were kept at 70% R.H. 37°C until fully developed larvae were observed moving within them. After observing their movement for 30 mins. the chorion of a few of these eggs was torn with a needle while the eggs were still in air of 70% R.H. In the eggs whose shells were torn at the front end, the larvae quickly hatched. Of the eggs whose shells were not torn, some hatched after an interval of time, other larvae failed to escape and died eventually through desiccation.

Davies & Hobson (1936) found that hatching of L.sericata eggs could be completely prevented if they were incubated at 100 % R.H. and transferred to 50% R.H. a few minuted before hatching was due. Similar experiments were carried out in the present work and confirmed this observation.

At 50% R.H. some hatching was observed in large batches of eggs incubated as laid by the flies. Forty three complete unseparated batches were incubated at this humidity. Some hatching occurred in 13 of them whilst still in air at 50% R.H. 37°C, but never more than 20% of the total eggs in each batch hatched, usually less than 10%. This hatching at 50% R.H. was presumably due to some of the eggs being protected, by close packing, against the effects of low humidity. In one large mass of approx. 1200 eggs laid by six flies, some hatched in air of 50% R.H. 37°C. It was observed that the eggs which hatched at this low humidity were usually those on the edges of the underside of the egg batch. Eggs in the middle of the batch contained fully developed larvae, no doubt unable to break out of their egg shells because the latter were cemented together into a solid mass by the dried covering of accessory gland secretion coating the eggs. All larvae which emerged at 50 - 70% R.H. 37°C were weak and shrivelled looking - the "chiton larvae" of Larsen (1943) - and died soon after hatching.

The experiments of Davies & Hobson (1935) showed that eggs incubated in saturated air to within 20 min. of hatching, did not hatch if they were transferred to air of 50% R.H. 37°C. - the fully developed larvae being imprisoned within the chorion. Further experiments on this point have shown that larvae could withstand such imprisonment for about 3 hr. Up to that time, the bulk of the larvae hatched when eggs were transferred back to saturated air. Larvae imprisoned for periods longer than 3 hr. showed increasing mortalities - a 100% mortality being reached after approximately 4 hr. imprisonment at 37°C.

(iv) Effects of variations in humidity

Immediately after laying, eggs were incubated at 37°C at humidities of 0, 30, 40 and 50% R.H. for varying periods.

They were then quickly transferred to 100% R.H. 37°C. and allowed to complete development. Mortalities produced by exposures of various lengths to these low humidities are shown in Table 5. It will be seen that at 37°C, comparatively long exposures to air of 40 and 30% R.H. are required to produce a 50% mortality. Since the R.H. of the air at t' of the sheep's fleece in summer rarely falls below 30% R.H. (Davies & Hobson (1935), Macleod (1940)), eggs in a dry fleece would therefore be expected to survive if the humidity rose rapidly during the incubation period. Consecutive readings of fleece humidity at standard points in the fleece show that such rises in fleece humidity do occur under natural conditions (Part III).

Table 5. % Mortality in eggs exposed to low R.H. immediately after laying.

Hr. exposed	0% R.H.	30% R.H.	40% R.H.	50% R.H.
1·0	10	0	—	—
1·8	18	0	—	—
3·0	14	0	0	—
3·6	78	0	0	—
4·0	100	0	0	0
4·25	100	18	0	0
5·0	—	12	0	2
6·0	—	35	4	—
7·0	—	47	52	1
8·0	—	82	58	4
8·25	—	76	—	—
9·0	—	100	100	18
9·5	—	100	100	8
12·0	—	—	—	52
13·0	—	—	—	72
14·0	—	—	—	68
14·5	—	—	—	84
14·75	—	—	—	92
15·0	—	—	—	100

It was found that eggs incubated immediately after laying in saturated air at 37°C were particularly susceptible to desiccation if subsequently incubated at lower humidities. About 30 min. in saturated air was sufficient to cause this effect. Table 6 shows the results of one experiment in which eggs from one batch were incubated in saturated air at 17, 30

Table 6 *Effect of preliminary incubation in saturated air*

Treatment	Percentage eggs completing development	Percentage eggs dying at early stages
Incubated at 100% R.H. 37° C. for 90 min.; then transferred to 55% R.H. 37° C. for rest of incubation period	10	90
Incubated at 100% R.H. 30° C. for 90 min.; then transferred to 55% R.H. 37° C. for rest of incubation period	72	28
Incubated at 100% R.H. 17° C. for 90 min.; then transferred to 55% R.H. 37° C. for rest of incubation period	85	15
Whole incubation period at 55% R.H. 37° C.	100	0

and 37°C. for 90 min. and then placed for the rest of their developmental period in 55% R.H. 37°C. Twenty eggs in each group were used. It will be seen that this 'conditioning' effect was most marked at 37°C and much less so when the period in saturated air was at 30 and 17°C. It is well known that the chorion of the Muscid egg hardens in dry air, and softens in humid air. It would appear that L.sericata eggs are laid with the chorion in the 'hard' condition, and that incubation in saturated air is needed to soften them. The conditioning of eggs by placing them in saturated air for a time may possibly be explained by this softening of the chorion. The 'hard' chorion may allow less rapid water loss than when it has been softened. Water loss through the cuticle of the wireworm was found by Wigglesworth (1945) to diminish rapidly at very low humidities, and he suggested that the lowered permeability resulted from the drying of the cuticle. In the present study it was noticed that eggs conditioned in saturated air at 37°C for 90 min. became dimpled in a shorter time than unconditioned eggs, which had been kept on the bench at about 65% R.H. for the same time, when both groups were placed in dry air. This was presumably due to the softened chorion of the 'conditioned' eggs being less resistant to dimpling than the harder chorion of the 'unconditioned' eggs. Dimpling of the shell would be expected to causedeformation of the lipoid waterproofing layer, especially at the edges of the dimple, leading to increased water loss and thus even quicker dimpling. The shells of the 'unconditioned' eggs would resist initial dimpling, and thus would lose water more slowly so that more of the eggs would complete development. (An alternative explanation for the 'conditioning effect' is given later (page 82-3)). Since softening of the chorion occurred readily in saturated air at 17° and 30°C as at 37°C., the less marked 'conditioning'

effect at the lower temperatures cannot be explained at present.

In view of this 'conditioning' effect, experiments of the following type were carried out. A single large egg batch was divided in two, and each half weighed. Onehalf (a) was 'conditioned' for 90 min. at 100% R.H. 37°C., reweighed, then placed in 0% R.H. 37°C. for 3 hr. The other half (B) was incubated on laying at 0% R.H. for 3 hr. at the same temperature. In one such experiment, batch A in 3 hr. at 0% R.H. 37°C., lost 38% of its original weight whilst batch B lost only 27% under the same conditions. This is in agreement with the higher mortalities recorded for 'conditioned' eggs.

DISCUSSION

The effects of fat solvents, a detergent, an abrasive dust, and high temperatures, on the permeability of the L.sericata egg-shell leave little doubt that it is waterproofed by a lipoid layer laid down by the oocyte and situated between the chorion and the c.v.membrane. Its waterproofing mechanism thus appears to be similar in essentials to that described by Beament (1946a) for the egg of the Hemipteran Rhodnius prolixus, and by Wigglesworth (1945) and Beament (1945) for the waterproofing mechanism of insect cuticle.

It is interesting to note that an oily layer has been found by Christophers (1945) in the Culex pipiens egg, lying between its exo- and endochorion. On this basis it would appear that the c.v.membrane of the L.sericata egg is homologous with the endochorion of the Culex pipiens egg, and the chorion of the former with the exochorion of the latter egg. Pantel (1913) states that the original fine vitelline membrane (of oocytic origin) received additional material of a gelatinous appearance from the follicle cells, so that the final membrane (i.e. c.v.membrane)

is of composite origin in Tachinid eggs.

The present work showed that bound lipoid material appeared both in the chorion and the c.v.membrane of the L.sericata egg shortly before laying, and that this lipoid alone did not waterproof the shell. Lipidized protein, some of which was in addition tanned, was found by Beament (1946b) to be an important constituent of several of the layers of the Rhodnius egg shell. Further the cuticle of insects, particularly the epicuticle has also been found to contain tanned and lipidized protein, (Wigglesworth 1933, 1947). The main function of this lipoid appearing in the L.sericata egg shell shortly before laying would appear to be to strengthen the shell prior to laying down the waterproofing layer. A thin waterproofing layer of orientated molecules would not be effective if laid down on a shell that was soft and easily deformed. Beament (1946a) showed that the waterproofing layer of the Rhodnius egg is not laid down until the shell is fairly rigid, after the follicle cells have completed the secretion of the endochorion.

The humidity experiment described in the present part showed that L.sericata eggs could withstand considerable desiccation at 37°C with some eggs completing development at 50% R.H. (23.5 mm.sat.def.). The work of Larsen (1943) showed that eggs of dung-breeding Diptera required much higher humidities for development than the eggs of L.sericata. It has been shown by previous work (Macleod 1940, Davies & Hobson 1935) and by observations described later (page 106-15) that the natural environment of the L.sericata egg - the sheep's fleece - is normally much drier than that of the eggs of dung-breeding flies.

The work of Beament (1946) showed that water loss through the waterproofed Rhodnius egg shell in dry air at 30°C occurred at about 0.1 mg./sq.cm./hr. (Beament 1946, Fig.2).

Under the same conditions water loss through the waterproofed L.sericata egg-shell was found to be in the region of 0.5 mg./sq.cm./hr. Even allowing for error in calculating the surface areas of L.sericata egg batches in the present work, it seems that the shell of the latter is more permeable to water than is that of Rhodnius. This greater permeability, coupled with the much smaller size and larger surface area/ volume ratio, account for the more humid conditions required by L.sericata eggs for development. It has been pointed out by several writers and by Wigglesworth (1937) that in assessing the effects of humidity on an insect, the length of exposure as well as the degree of humidity must be taken into account. A comparison of the desiccation resistance of the Rhodnius egg with that of L.sericata on this basis shows the following:-

i) Rhodnius (Data from Clark (1935)).
Incubation period at 32°C = 12 days (288 hrs.)
Minimum humidity for development at 32°C = 30% R.H.
(= 25.0 mm.sat.def.)

ii) L.sericata
Incubation period at 30°C. = 16.0 hr.
Minimum humidity for development at 30°C = 25% R.H.
(= 23.5 mm.sat.def.)

Thus for both species the product, saturation deficiency x time is 288 x 25.0 = 7200 for Rhodnius, and 16.0 x 23.5 = 376 for L.sericata. These are the true measures of the comparative desiccation resistance of the eggs of the two species.

The results of experiments on the humidity relations of the L.sericata egg with regard to blowfly myiasis is discussed later (page 135-8).

SUMMARY

The chorion of the L.sericata egg is shown to be composed of two main layers, both of protein; the outer can be tanned by p-benzoquinone, but the inner is apparently

already tanned. The chorion and the chorionic vitelline membrane are both lipidized shortly before the egg is laid. This makes both structures more rigid, but does not waterproof the shell. After the lipidization process is completed, a lipoid waterproofing layer is laid down by the oocyte, between the chorion and the chorionic vitelline membrane. This waterproofing layer has a critical temperature in the region of 38°C, and can be damaged by an abrasive dust, emulsified by the detergent I.C.I. C09993, and removed by chloroform at 30°C in 12 hr.

The minimum humidity for development of L.sericata eggs at 37°C has been found to be 50% R.H. At 1 - 2°C above this temperature eggs require 80 - 90% R.H. to complete development. Eggs at 37°C can withstand fairly long periods of humidities below 50% R.H., provided they have not been previously incubated insaturated air at that temperature. The latter treatment, even if continued for only 30 min., makes eggs far more susceptible to desiccation when subsequently incubated at a low humidity.

Fully developed larvae can survive imprisonment within the egg-shell for about 3 hr. at 37°C.

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PART II

LABORATORY STUDIES ON THE EGGS OF OTHER BLOWFLIES

INTRODUCTION

Experiments on the humidity and temperature relations of the eggs of five blowfly species were carried out, in order to provide data comparable with that obtained for the L.sericata egg (Part I). Observations on the waterproofing mechanisms of these eggs were also made. The species involved were as follows:-

1) Lucilia caesar (Linnaeus 1758)

2) Lucilia illustris (Meigen 1826), synonyms:- splendida (Meigen 1826), simulatrix (Pandelle 1896)

3) Protophormia terra-novae (Robineau-Desvoidy 1830), frequently known as Phormia terra-novae

4) Calliphora erythrocephala (Meigen 1826)

5) Calliphora vomitoria (Linnaeus 1758)

(see Kloet and Hincks, 1945).

While L.sericata is the chief cause of myiasis of sheep in Britain, the above fly species are of lesser importance but must be considered in any study of sheep myiasis in this country. Macleod (1943a) in his survey of the relative incidence of strike due to the above five species over most of Britain, found that L.caesar was an important striking species in Scotland, northern England and North Wales. P.terra-novae was also important in the same areas, except that it apparently did not strike sheep in North Wales. L.caesar and P.terra-novae were found to be capable of acting as 'primary' flies i.e. laying their eggs on sheep not already infested with maggots. He found that C.erythrocephala and to a lesser extent C.vomitoria larvae occur in strikes fairly generally over the country but at a very low incidence (23 C.erythrocephala and 10 C.vomitoria out of 1307 cases of strike examined). In 29 cases of strike from various parts of the country, containing Calliphora spp.

larvae, 12 contained Calliphora spp. alone, and 17 Calliphora spp. mixed with larvae of other species. Thus it seems that Calliphora spp. can sometimes act as 'primary' flies, but are more often 'secondary' flies i.e. attacking sheep already infested with maggots.

Special attention was given to the highest temperature at which the eggs of the five species could survive, since temperatures of 36 - 40°C would be expected to prevail in some regions within sheep fleeces, especially during warm weather.

The methods employed were similar to those used for the eggs of L.sericata (page 29-32).

(1) Minimum humidities for development

The results of water-bath experiments on the eggs of these species (page 47) will be found in full in the Appendix (page 161-177). The information gained by these experiments on the minimum humidities for development at various temperatures of the eggs of the five species is summarised in Table 7 (a - f), which also includes the results on L.sericata eggs for comparison.

TABLE 7

Minimum humidities for development

(a) L.CAESAR eggs

Expt.No.	Temp.°C.	Minimum %RH at which eggs completed development	% EGGS Hatching	Prehatching	Total completing development
36	37	60	0	1.8	1.8
37	37	65	0	1.2	1.2
38	38	80	0	0	0
40	38	80 (Min. RH. < 80 ?)	49.6	15.7	56.3
41	39	All RH's - no development	0	0	0
42	40.0 - 40.5	"	0	0	0

TABLE 7 (continued)

Expt.No.	Temp. °C.	Minimum RH at which eggs completed development	% Eggs Hatching	% Eggs Prehatching	% Eggs Total completing development
(b) L.ILLUSTRIS eggs					
28	37	65	0	5.5	5.5
29	37	60	0	9.4	9.4
30	37	60	0	2.2	2.2
35	37	60	0	11.5	11.5
37	37	65	0	26.5	26.5
9	38	85 (Min. R.H. < 85 ?)	10.1	8.7	18.8
31	38	80	0	5.5	5.5
32	39	85	0	9.5	9.5
39	40	All RH's - no development	0	0	0
(c) P.TERRA-NOVAE Eggs					
22	37	65	0	1.5	1.5
23	37	60	2.9	2.9	5.8
16	38.8	80 (Min. R.H. < 80 ?)	23.3	39.5	61.8
17	40	95	4.1	8.2	12.3
(d) C.ERYTHROCEPHALA eggs					
21	30	45	0	2.8	2.8
25	30	45	0	0	0
26	30	45	0	13.6	13.6
27	30	45	0	1.3	1.3
43	30	55	4.2	22.9	27.1
12	34	80	0	3.3	3.3
13	34	80	0	3.7	3.7
18	34	80	0	0	0
1	35	95	0	1.0	1.0
2	35	95	0	12.9	12.9
3	35	95	0	0	0
4	35	100	0	4.5	4.5

TABLE 7 (continued)

Expt.No	Temp.°C.	Minimum %R.H. at which eggs completed development	% EGGS		
			Hatching	Prehatching	Total completing development
5	35	100	0	0	0
11	35	100	0	0	0
6	36	All humidities - no development	0	0	0
7	36	All humidities - no development	0	0	0
		(e) C.VOMITORIA eggs			
21	30	45	0	1.4	1.4
12	34	80(Min. <80)	7.2	85.5	92.7
18	34	65	0	1.3	1.3
5	35	90	1.4	14.8	16.2
8	35	90(Min. <90)	65.5	20.7	86.2
11	35	80(Min. <80)	31.4	1.5	32.9
20	35	75	0	4.2	4.2
6	36	95	0	45.3	45.3
15	36.8	All humidities - no development	0	0	0
		(f) L.SERICATA eggs			
22	37	50	0	5.6	5.6
23	37	45	0	6.3	6.3
30	37	50	0	69.7	69.7
9	38	85(Min. <85)	94.0	0	94.0
10	38	80(Min. <80)	58.3	16.6	74.9
16	38.8	80	7.8	6.5	14.3
14	39	80(Min. <80)	24.8	18.0	42.0
33	40	80	0	0.9	0.9
34	40	95	1.2	0.6	1.8

It will be seen that there is some variation in the minimum humidity at which the eggs of one species completed development in repeated experiments at one temperature. The temperature control in the water bath was good, the thermometer

showing less than $\pm$ 0.1°C variation during an experiment, and since the eggs were incubated in thick glass vessels, the variation in results between repeated experiments is probably not due to temperature differences. The acid in the humidity vessels was changed after every four experiments. The acid mixtures for each humidity were all made up before the commencement of the water-bath experiments, so that all the changes of acid at each humidity came from the same lot kept in one bottle. The variation in the results may be due to the fact that slight differences in the precise R.H. in the humidity vessels occurred from one experiment to another. Near the minimum humidity at one temperature the total water loss for the eggs is at a delicately balanced level, small changes in which determine success or failure in completing development.

The results of the water bath experiments in general confirmed preliminary experiments carried out using electric incubators to maintain constant temperatures.

From the information gained by both water-bath and incubator experiments, the minimum humidities for development, at various temperatures, of the egg of the five species and of L.sericata eggs, may be summarised as follows (Table 8)

TABLE 8

Minimum humidities for development (% R.H.)

Temp.°C	L.sericata	L.caesar	L.illustris	C.erythrocephala	C.vomitoria	P.terranovae
30	25-30	-	-	45-50	45-50	30-35
34	40	55	-	80	65	-
35	-	-	-	95-100	80-90	-
36	-	-	-	No development	95	-
37	50	60-65	60-65	-	No development	60-65
38	75	about 80	80	-	-	-
39	80	No development	85	-	-	about 80
40	95	No development	No development	-	-	95

From the above figures it will be seen that the eggs of L.sericata consistently exhibited greater desiccation resistance than those of the five other species, so that when compared with them individually at any of the temperatures employed, the eggs of L.sericata were able to complete development at a lower humidity than those of the compared species. The considerable number of experiments carried out where the eggs of two species were obtained at the same time and incubated simultaneously in the various humidity vessels, amply confirmed this. (The results of water-bath experiments (Appendix page 163-177) are grouped under the species. Where the serial number of an experiment is the same in the case of two species, this indicates that their eggs were incubated together as stated above). At 37°C, a proportion of the eggs of L.sericata are seen to complete development at 50% R.H., while at this temperature the eggs of L.caesar, L.illustris and P.terra-novae required 60-65% R.H. At this temperature (37°C) the eggs of both Calliphora species entirely failed to complete development, but when compared with L.sericata at 30°C, they showed that they needed considerably higher R.H's to complete development (45-50%) - L.sericata eggs (25-30%). The significance of the greater desiccation resistance of L.sericata eggs in relation to sheep myiasis is discussed later (page 135).

From Table 8 the highest constant temperatures at which the eggs of the various species can complete development are seen to be as follows (Table 9). The maximum temperatures

TABLE 9

Maximum temperatures for development (°C)

L.sericata	L.caesar	L.illustris	C.erythrocephala	C.vomitoria	P.terranovae
40-41	38-39	39-40	35-36	36-36.8	40-41

for development for L.sericata and P.terra-novae eggs lie between 40 and 41°C. Melvin (1934) found that L.sericata

eggs failed to hatch at 40°C (104°F). In the two experiments on the eggs of this species carried out at that temperature, (Appendix, page 164-5, Expts. 33 and 34) in one 9.9% of the eggs hatched in 100% R.H. only, in the other no eggs hatched in 100% R.H. but 1.2% hatched in 95% R.H. The phenomenon of better hatching at 95% than at 100% R.H. at high temperatures has already been mentioned (page 36). In Melvin's (1934) experiments the eggs were kept in saturated air at each temperature. His failure to find hatching at 40°C may have been due to this reduced hatching in saturated air at high temperatures. The eggs of L.illustris were found to be able to withstand a slightly higher temperature (39°C) than those of L.caesar (38°C). The maximum temperatures for the two Calliphora spp. were considerably lower, the egg of C.vomitoria being able to complete development at 36° but not at 36.8°C, while those of C.erythrocephala needed lower temperatures, being able to complete development at 35° but not at 36°C. In all the species high humidities were required at these high temperatures in order to complete development.

(ii) Minimum Humidities for hatching

No differences in the minimum humidity at which the eggs could hatch were detected between the five species studied; the humidities they required for hatching were identical as far as could be seen with those required by L.sericata eggs. Rapid hatching at 90-100% R.H. with progressively slower and less complete hatching with decreasing humidities from 90-60%, together with occasional but very restricted hatching at 55% R.H., were found again to apply to the eggs of the five species, as to L.sericata. The dependence of hatching on R.H. and its independence of temperature was again evident. Since at high temperatures

the minimum humidities for development of the eggs of the six species studied (including L.sericata) were found to be higher than at lower temperatures, the possible minimum humidities for hatching were also higher. If eggs had been able to complete development at say 50% R.H. at the high temperatures, it is likely that the 55-60% R.H. region would have been the lowest humidity level for hatching, just as at lower temperatures. At high temperatures the total percentage eggs completing development at the minimum humidities were almost always small. For C.erythrocephala eggs the percentage and absolute numbers of eggs completing development at various humidities at 34 and 35°C, are set out in Table 10.

TABLE 10

Minimum humidities for hatching and development.
C.erythrocephala eggs. 34 and 35°C.

The lowest humidity quoted in each expt. was the minimum humidity at which development occurred in the particular expt.

Expt. No.	°C Temp.	% R.H.	% H	% PH	Actual numbers H	Actual numbers PH	Total eggs	% development in control
12	34	95	2.0	5.6	1	3	53	63.9
		90	0.0	18.6	0	10	54	
		85	0.0	7.8	0	5	64	
		80	0.0	3.3	0	2	60	
13	34	90	1.9	11.5	1	6	52	76.1
		85	0.0	11.5	0	6	52	
		80	0.0	3.7	0	2	54	
1	35	100	1.0	1.0	1	1	100	74.2
		95	0.0	1.0	0	1	104	
.2	35	100	0.0	14.7	0	5	34	83.0
		95	0.0	12.9	0	4	31	
4	35	100	0.0	4.5	0	1	22	63.2

Figures for stages of development reached not corrected for mortality in controls.

It will be seen that the failure to hatch at say 85% R.H. (Table 10, Expt.13) may have been dur to the fact that at this humidity only six eggs out of 52 reached the prehatching stage, so that the chances of any of the larvae breaking out of the shells were small. Had larger numbers of eggs been able to complete development at 85% R.H. it is likely that some would have hatched. Since 35°C is the highest temperature at which C.erythrocephala eggs completed development, there may have been some reduction in the activity of the larvae within the shells, due to heat stupor leading to reduced chances of hatching.

That development at high constant temperatures near the upper lethal limit for the eggs, did not adversely effect the later stages of the life history was shown for C.vomitoria eggs. First instar larvae were obtained from eggs which had spent the whole of their incubation period and had hatched at 35°C 100% R.H. They were placed on meat at 25°C (a suitable temperature for larval feeding and growth), These larvae fed and moulted normally and eventually produced adult flies normal in every respect as far as could be determined by appearance and activity.

(iii) Duration of incubation period at various constant temperatures

In the course of the work the duration of the incubation period for the eggs of the various species at different temperatures were observed. Of the species studied, published information on the length of the incubation period is available for L.sericata (Wardle,1930; Melvin, 1934), and for C.erythrocephala (Scott,1934). The information gained in the present work is given in Table 11, (page 56).

The eggs were at each temperature kept in saturated air unless otherwise indicated. The figures given include the

TABLE 11

Incubation Period, hours

Const. Temp. °C	L. sericata	L. illustris	L. caesar	C. erythrocephala	C. vomitoria	P. terranovae
21	-	18·25-19·25	-	23·0-25·0	-	-
22	-	-	-	18·0-22·0	-	-
30	9·8-10·5	9·25-10·5	-	13·0-15·3	12·5-14·75	12·5-13·5
31	-	-	-	-	12·0-13·0	-
34	9·0-9·3	-	-	(16·0-16·9)†	(12·5-13·0)† 12·3-12·9	-
35	-	-	-	11·6-12·6	(14·5-15·2)† 11·5-13·0	-
36	-	8·9-9·3	-	15·0-16·5‡	-	-
37	7·6-7·9	-	8·1-8·6	-	-	9·8-10·2
38	(10·5-11·25)* 9·0-11·0	9·5-10·0	-	-	-	9·8-10·2
38.8	-	-	-	-	-	(10·6-11·5)†

†, at 95% R.H. *, at 90% R.H. ‡, temp. fluctuating 35 to 36°C.

extreme limits encountered, including at many temperatures observations on several separate batches laid by different flies. The results on the eggs of L. sericata show reasonable agreement with those obtained by Wardle (1930) and Melvin (1934).

From Table 11 it will be seen that L. sericata eggs at all temperatures at which they can be compared with the other five species, consistantly showed shorter incubation periods, except in the case of L. illustris which at 30°C had an incubation period of approximately the same length as L. sericata. At 37°C L. caesar eggs required about 0.5-1.0 hr. longer than L. sericata eggs, and Phormia more than 2.0 hr. longer to complete development. The eggs of both Calliphora spp. at 30°C required from 2.5-5.0 hr. longer than L. sericata, showing that their rate of development was considerably slower. The greater susceptibility to desiccation exhibited by the eggs of Calliphora spp., P. terra-novae and L. caesar may partly be a result of this slower development.

(iv) The hatching mechanism of blowfly eggs

In the course of measuring the eggs of various blowfly species it was observed that the eggs of all six species underwent relatively small but definite shape changes dependent on changes in the R.H. of the air in their immediate vicinity. In the first instance, these shape changes were observed when transitory R.H. changes were accidentally caused by breathing on naked eggs while they were being measured on a glass slide on the microscope stage. Experiments with eggs at controlled humidities were then carried out detailed observations on the shape changes made. The changes in shape of an egg when subjected to an R.H. change were constant in repeated observations, the extent of the changes in dimensions being very nearly the same each time, with slight variations within the range of error of the method of measuring used.

Evidence will be given in the succeeding pages that these humidity-dependent shape changes in blowfly eggs are intimately associated with their hatching mechanism - a mechanism affected and indeed governed by relative humidity (page 37-40, 53-55).

Methods. Measurements of eggs were made by means of a microscope fitted with an eyepiece micrometer scale, and a 16 mm.(x10) objective. The total magnification was x 60. The micrometer scale was calibrated and it was found that 1 micrometer division = 16.5μ

The dimensions of eggs at known controlled humidities were measured, and of individual eggs at several different humidities. Solid watch glasses sealed with flat glass lids were used as humidity chambers (page 32). Humidities were again controlled within the watchglasses by sulphuric acid/water mixtures (page 31), 100% R.H. being maintained with distilled water. The cavity of each watchglass was

filled with the appropriate acid mixture to within 5 mm. of the underside of the flat coverglass lids. Eggs were attached directly to the underside of the coverglasses so that they lay within 5 mm. of the surface of the acid, and their dimensions could then be measured through the glass lid when the whole watch glass was placed on a mechanical stage microscope. Successive measurements on several individual eggs at several R.H's were made by quickly transferring the coverglass with its attached eggs on to another solid watchglass with the appropriate acid mixture in its cavity, repeating the transfer for each R.H. All the solid watchglasses were kept covered and airtight except during the short period of a few seconds when a transfer was being made. A period of 5 min. was allowed to elapse after eggs had been transferred to a new R.H. before measurements were taken, in order to allow the egg shells to reach equilibrium with the air of the chamber, and for the correct R.H. to be restored after the disturbance. Successive length measurements made on eggs from immediately after they had been transferred to a particular R.H., until 30 min. later, showed that eggs reached their final length within 3 min. of the transfer. The shape changes brought about by humidity changes are thus seen to be very rapid. It was observed that naked eggs increased in length almost simultaneously with breathing on them. After ceasing to breathe on them, and while the moist air around them disippated in the succeeding 2-5 secs., the eggs returned to their original length within the same period.

Results. By measuring the length, breadth and various other dimensions of eggs at various humidities the shape changes in blowfly eggs caused by R.H. changes were worked out, and are summarized below:-

(a) With increasing R.H. from 0 to 100%, the eggs of all species gradually increased in length. Length

measurements for the eggs of three species belonging to the three genera studied are given in Table 12,(page 60-61). The percentage increase in length over that at 0% R.H., plotted against R.H., for single representative eggs of three species is graphed in Fig. 10 (page 62). From Table 12 it will be seen that the lengths of P.terra-novae eggs at 100% R.H. were about 4.5 - 6.6% greater than at 0% R.H., while corresponding figures for the eggs of L.sericata and C.erythrocephala were 4.5 - 5.8% and 1.0 - 2.6%, respectively.

(b) Accompanying the elongation of the eggs a reduction in their cross-sectional area occurred, mainly by inward movement of the dorsal side of each egg, but preserving a circular cross-section, (Fig.11, page 65).

(c) Simultaneously with the above shape changes, and probably forming an integral part of them, the dorsal hatching pleats and the narrow 'hatching strip' between them elongated with increasing R.H., while the hatching pleats were very slightly drawn together along the whole of their length so that the hatching strip became slightly narrower (Fig.9, see also Fig.1 (a) page 10).

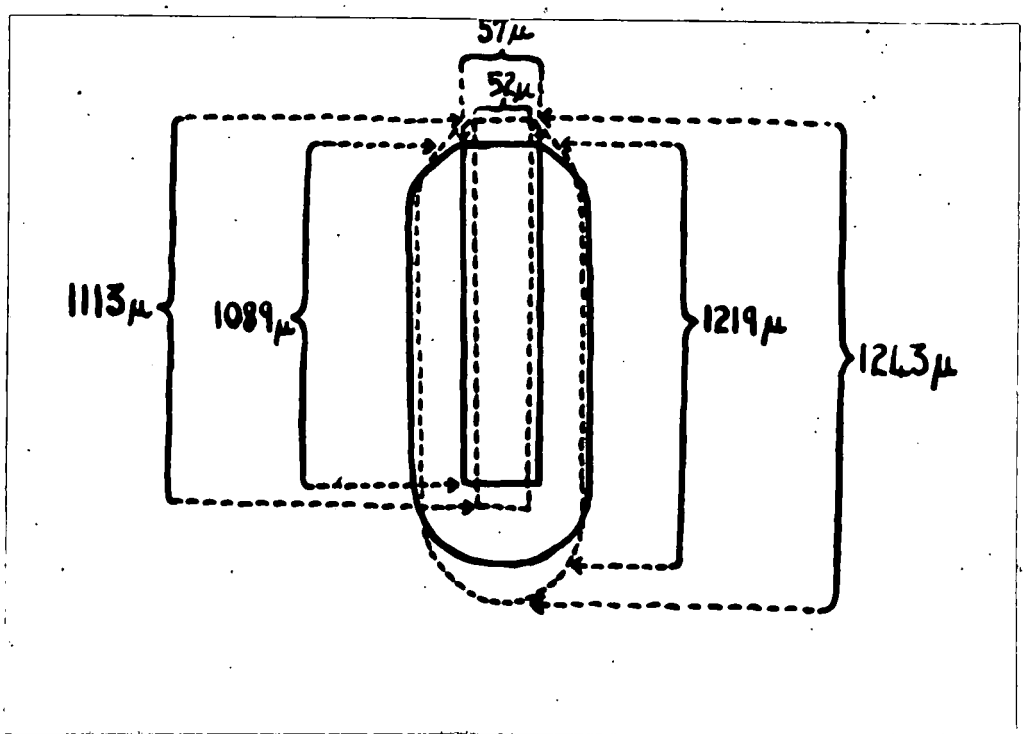

Fig. 9. Diagram of egg shape-changes; ——, low R.H.; -------, high R.H.

Fig. 9 illustrating the shape changes (a) and (b) above, shows results from one egg (L.sericata). With the R.H. change from 0 to 100% R.H., the following dimensional changes occurred:- The total length of the egg increased from 1219 to 1243μ , (shape change (a) above); the length of the hatching pleats and hatching strip between them increased

from 1089 to 1113 μ , while the width of the hatching strip, i.e. the distance between the two hatching pleats decreased from 57 to 52 μ . It should be pointed out that in

TABLE 12

Lengths of eggs at various R.H's (μ)

(a) L.SERICATA

In the case of L.sericata eggs, length measurements from 0 - 100, then back down to 0 and then up again to 100% R.H. were carried out.

	Lengths in microns					
%R.H.	Egg 1	Egg 2	Egg 3	Egg 4	Egg 5	Av.of 5
0	1245	1272	1218	1201	1211	1229
25	1256	1287	1229	1216	1226	1243
45	1267	1294	1237	1224	1237	1252
70	1279	1308	1251	1237	1249	1265
90	1292	1320	1259	1252	1267	1278
100	1307	1338	1275	1260	1282	1292
90	1290	1320	1262	1252	1269	1279
70	1279	1308	1251	1237	1254	1266
45	1267	1297	1237	1226	1241	1254
25	1260	1290	1229	1221	1234	1247
0	1241	1270	1218	1204	1204	1227
25	1254	1287	1229	1219	1221	1242
45	1264	1295	1237	1224	1230	1250
70	1272	1305	1251	1237	1245	1262
90	1285	1318	1259	1251	1257	1274
100	1303	1336	1274	1259	1277	1290
Increase in length at 100% over that at 0% RH.	62-66	66-68	56	55-59	71-78	63-65
% increase in length at 100% over that at 0% R.H. On basis of 1st series above.	4.98	5.18	4.59	4.91	5.86	5.10

(b) P.TERRA-NOVAE

0	1224	1224	1203	1170	1150	1194
25	1244	1237	1219	1183	1166	1210
45	1254	1242	1223	1188	1175	1216

TABLE 12 (continued)

Lengths in microns

%R.H.	Egg 1	Egg 2	Egg 3	Egg 4	Egg 5	Av.of 5
70	1270	1246	1234	1204	1188	1228
90	1287	1262	1254	1221	1204	1246
100	1303	1279	1270	1242	1226	1264
0	1226	1221	1204	1168	1150	1194
Increase in length at 100% over that at 0% R.H.	77-79	55-58	66-67	72-74	76	70
% increase in length at 100% over that at 0% R.H.	6.45	4.49	5.57	6.15	6.60	5.85

(c) C.ERYTHROCEPHALA

0	1452	1477	1468	1487	1503	1477
25	1462	1485	1468	1493	1508	1483
35	1465	1485	1468	1498	1510	1485
45	1467	1485	1468	1501	1513	1487
55	1468	1485	1468	1503	1516	1488
70	1470	1485	1468	1506	1518	1489
80	1470	1485	1468	1506	1518	1489
90	1472	1485	1468	1508	1520	1491
95	1477	1496	1472	1518	1523	1497
100	1485	1503	1482	1526	1531	1505
Increase in length at 100% over that at 0% R.H.	33	26	14	39	28	28
% increase in length at 100% over that at 0% R.H.	2.27	1.76	0.95	2.62	1.86	1.89

measurements of small lengths such as the width of the hatching strip amounting to about 55-60μ, considerable error was possible, with the magnification used. It was found impracticable to use a higher magnification without radically altering the apparatus and employing a complex system.

Various further observations were made on these shape changes. The results are given below as numbered observations

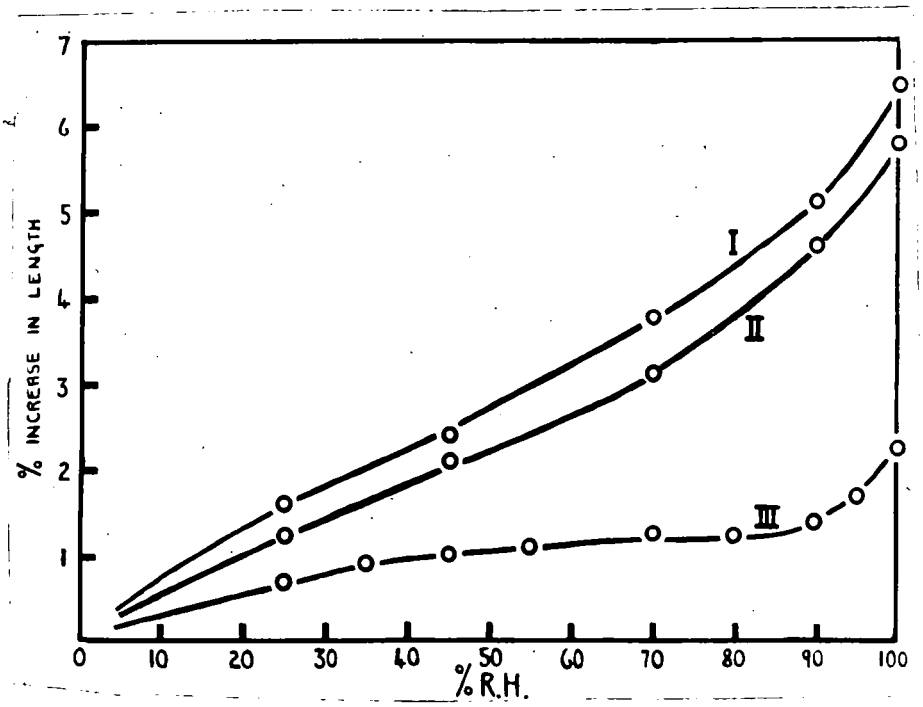

FIG.10. Increase in egg-length with rising R.H.; I, P. ferranovae; II, L. sericata; III, C. erythrocephala.

for the sake of clarity.

Observation 1. When eggs were subjected to sudden humidity changes by transferring them from an 0% R.H. chamber to one of 100%, a proportion of the egg chorions split by a longitudinal fissure running closely along the outer margin of one or other of the hatching pleats. This fissure originated at any point along the length of the hatching pleats which run more than three quarters of the lengths of eggs (see Fig. 1, page 10). Eggs which did not split during the first 0 to 100% R.H. change could be induced to do so by repeating the process for a further 2-5 times. This conclusively shows that sudden large R.H. changes cause strains to be set up in the chorion which caused it to split along one or other of two fairly constant lines. Care was taken to employ eggs at an early stage in development, so that there was no possibility of the chorion split being due to the activity of fully formed larvae within the eggs. With smaller humidity changes of 50-100 or 0-50% R.H., no rupture of the chorion occurred even after twenty repeated changes. This shows that the strain set up in the chorion is less when the R.H. change is of smaller extent, and when

the shape change is also less.

Observation 2. That the coating of accessory-gland secretion covering the outside of the chorion of the laid egg was not responsible for the shape changes was shown by removing this mucus-like covering by washing eggs in 1% sodium sulphide solution for 1 hr. Such eggs, without their coating of accessory-gland secretion, but with the chorion intact, underwent shape changes simultaneously with R.H. changes in the same way and to the same extent as eggs with the secretion covering intact.

Observation 3. That the chorion alone, and not the c.v.membrane (see page 4) or the oocyte itself or a combination of the three, was responsible for the shape changes was shown by removing the chorions from eggs (cf. Evans, 1934) which retain their shape since they are still enclosed by the intact c.v.membrane underlying the chorion. Such eggs minus the chorions, when subjected to successive R.H's from 0 to 100% underwent no detectable shape changes - their lengths were the same at 100% as at 0% R.H., as far as could be ascertained by the measuring system used. It is concluded, therefore, that the chorion alone causes the humidity dependent shape changes and that it does so by forcing the oocyte with its investing c.v.membrane to change their shape, rather like a rubber tube which is stretched would force a soft body, like a sausage within it, to change its shape. In the blowfly egg, the tendency of the chorion to alter its shape at different humidities is resisted by the liquid yolk filled egg it invests. It is probably this resistance of the egg to shape change impressed on it by the chorion that causes strains to be set up in the latter. The amount of strain is thus humidity dependent because the shape of the chorion is humidity dependent.

Observation 4. That the shape changes caused by the chorion are not due to the effect of humidity on a small specialized

area of the shell, such as the hatching strip and hatching pleats, was demonstrated in the following way. The lengths of ten eggs at 0% and at 100% R.H. were determined. The hatching strip and hatching pleats only of six of these eggs were then covered by a layer of waterproof cellulose paint in the form of a painted strip running most of the length of the eggs. On the other four eggs a similar longitudinal strip of the chorion was covered with cellulose paint, but this time on the ventral side, opposite the hatching strip, on unspecialised chorion. Thus in the former six eggs the hatching strip was covered and would not be affected by humidity changes, and in the latter the hatching strip was exposed. The eggs were again measured at 0% and 100% R.H. The increases in length at 100% over the length at 0% R.H. are given in Table 13. If the shape changes were caused

TABLE 13

% Increase in length from 0-100% R.H., before and after cellulose paint treatment (L.sericata eggs)

	Hatching strip covered						Ventral side covered			
Egg No.	1	2	3	4	5	6	7	8	9	10
Before treatment	4.8	5.4	6.1	5.0	7.2	5.0	4.6	6.4	5.5	4.7
After treatment	4.0	2.3	4.6	4.2	5.2	4.6	3.4	3.5	3.9	4.3
Decrease	0.8	3.1	1.5	0.8	2.0	0.4	1.2	2.9	1.6	0.4

by the effect of humidity on the hatching pleats and hatching strip alone, covering them with waterproof cellulose paint would inhibit shape changes when the humidity was changed. It will be seen from Table 13 that eggs so treated still elongated with rise of R.H. from 0-100%, but to a smaller extent than before treatment. This decrease in the amount of the length change is also seen in eggs where the ventral side is covered with paint leaving the hatching pleat region exposed. The smaller length increase in both groups after

cellulose paint treatment was probably due to the stiffening effect of the paint itself rendering the change of shape of the eggs more difficult to accomplish. It is thus considered that the shape changes of blowfly eggs caused by R.H.changes are not due to the properties of the chorion in the hatching pleat region alone, but rather are due to the properties of the chorion as a whole.

Observation 5. It was observed by means of width measurements of eggs, both in lateral, dorsal and ventral views that the decrease in cross-sectional area coincident with elongation of the egg is brought about mainly by inward movement of the hatching pleat or dorsal side of the egg, and less by inward movement of the ventral side opposite hatching pleats, (Fig. 11). The profile of the cross-section of an egg at 0and 100% R.H. are thus not concentric circles. In this figure are given measurement obtained from one L.sericata egg. It will be seen that the diameter was reduced from 357 at 0% R.H. to 340 at 100% R.H. Also given in the same figure on the widths of the hatching strip measured from the

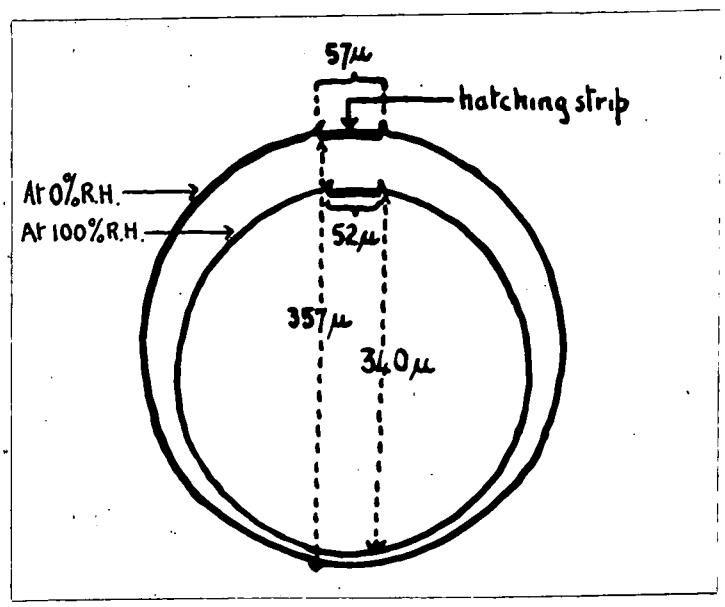

FIG. 11. T.S. L.sericata egg showing shape-changes (exaggerated). Measurements from one egg.

outer margin of each hatching pleat, at both humidities. The width of the hatching strip about half way along the egg was reduced from 57μ at 0% R.H. to 52μ at 100% R.H.

From the above information the following deduction can be made:-

Total circumference of the egg at 0% R.H. (assuming circular cross-section) = 2 x 3.1416 x 357/2 = 1121μ

Total circumference at 100% R.H. (assuming circular cross-section) = 2 x 3.1416 z 340/2 = 1068 μ

The chorion circumference as a whole must therefore contract 1121 - 1068 = 53 or a contraction of 4.72%. This contraction is that of the unspecialized chorion of the bulk of the circumference (Fig.13) plus that across the hatching strip.

The width of the hatching strip alone contracts from 57μ at 0% R.H. to 52μ at 100% R.H., or a reduction of 8.77%. Subtracting the width of the hatching strip from the circumference at 0 and 100% R.H. we get a measure of the contraction of the unspecialized chorion:-

Length unspecialized chorion circumference at 0% R.H. = 1121 - 57 = 1064μ

Length unspecialized chorion circumference at 100% R.H. = 1068 - 52 = 1016μ

Contraction of unspecialized chorion ∴ = 1064 - 1016 = 48μ or 4.51%

The fact that reduction in cross-sectional area occurs mainly by an inward movement of the dorsal or hatching pleat side of the egg and less by inward movement of the ventral side may be unimportant as far as the setting up of strains in the chorion is concerned. The explanation of this greater movement of the dorsal side seems to be that the transverse contraction of the hatching strip itself is about twice (8,77% in the example given) that of the unspecialized chorion (4.51%). There may be appreaciable errors in the measurements but it seems established that the transverse hatching strip contraction is relatively greater than that

of the unspecialized chorion.

The above conclusion is supported by the observation that in imperfect eggs (see page 84-85) in which the hatching strip and its lateral pleats are very short and only extend for about 1/10th of the length of the egg from the front end (Fig. 12) leaving the rest of the egg enclosed by unspecialized chorion, the contraction in cross-sectional area with rise in R.H. from 0 to 100%, occurred by an inward movement of the whole of the circumference, to about the same extent all round the egg. Thus in eggs with rudimentary hatching strips the cross-section of eggs at 0 and 100% R.H. were concentric circles (Fig.12) while in eggs with complete hatching strips the sectional outlines of the eggs at the two humidities were not concentric circles (Fig.11).

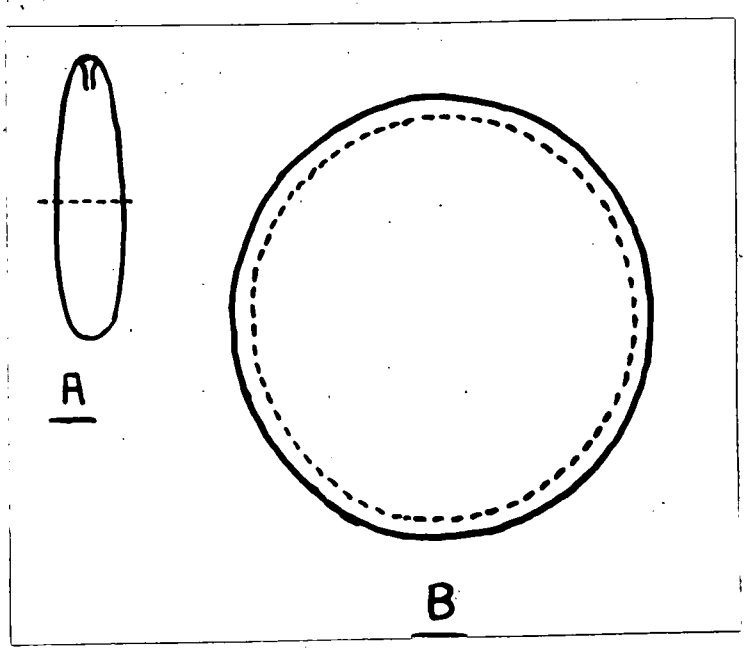

FIG.12. A shows short extent of hatching pleats and position of T.S. whose outline is given in B. In B, unbroken line = outline in low R.H.; broken line = outline at high R.H. (L.caesar egg).

Observation 6. At 0% R.H. it was observed that the hatching pleats stood nearly upright from the egg surface, and that with increasing humidity they leaned progressively inwards, towards each other so that in saturated air they lay at an angle of about 30° with the egg surface. These changes in the positions of the hatching pleats are shown in Fig. 13, as diagrammatic sections across the hatching pleats. The

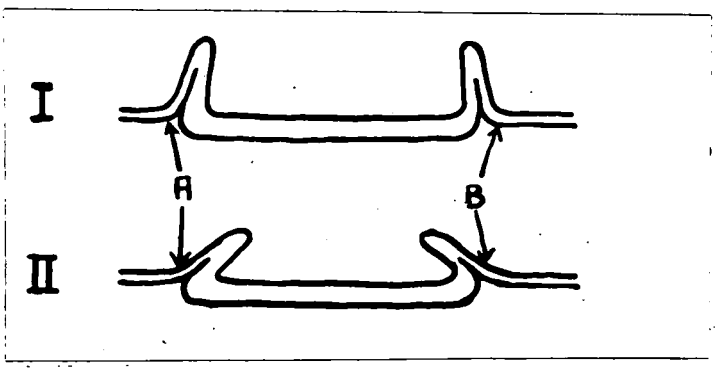

FIG. 13. Diagram of hatching-pleat movements. I, in 0% R.H.; II, in 100% R.H; Distance A-B becomes less as humidity rises.

distance from A to B in Fig. 13 was that observed to contract during humidity increases in observation 5, (page 65), being 8.77% less at 100% R.H. than at 0% R.H. in one example. This contraction was there attributed to an elongation of the hatching strip drawing the pleats closer together. The progressive inward leaning of the hatching pleats provide another explanation, without postulating an active decrease in breadth of the hatching strip itself. It will be plain that the distance AB, measured from the outer margin of each hatching pleat will be reduced as they lean progressively inwards. In a wire model bent from shape I (fig. 13) to shape II to simulate these movements of the pleats, the distance AB was reduced from 55mm. in the former shape to 47 mm. in the latter, a reduction of about 14.5%. Allowing for the fact that this figure was obtained from a crude model this figure is of the same order as that obtained by measurements of an egg (8.77%).

This contraction due to the movements of the pleats provides a mechanism whereby the circumference of the egg may become reduced with increasing humidity, without necessitating an actual contration (or only a very small one) of the unspecialized chorion, since the movements of the hatching pleats 'take up the slack' so to speak. If the basic cause of the egg shape changes is due to the tendency of the chorion to increase its curvature round the egg, this can be achieved by a reduction in diameter of the egg and thus reduced circumference, which in turn can be achieved (by

the above movements of the hatching pleats) without a contraction of the chorion layers round the egg, or at most only a small one.

The movements of the pleats with humidity may also explain why the chorion usually ruptures longitudinally along the outer margin of one or other of the hatching pleats, without the necessity of postulating the existence of a line of weakness along the outer margin of each pleat, since the strain would be expected to be greatest at those lines with such movements of the pleats.

Observation 7. If a small perforation be made through the chorion and c.v.membrane to the yolk below, of an otherwise intact laid egg, a small blob of yolk appears covering the outer opening of the hole. This shows that the yolk contents of the egg are held under slight pressure by the egg membranes at room humidity (50 - 60% R.H.). If the egg is now exposed to increased humidity, causing it to change shape, this blob of egg yolk increased in size, and when the humidity dropped to its former level, the blob decreased to its former size, by flow of the liquid yolk back into the egg. This shows that the volume of the egg was slightly reduced as the humidity increased. In an intact egg therefore increased humidity causes a slight reduction in the volume of the egg leading to increased internal pressure, which is reduced again as the humidity falls and the egg regains its former shape.

Observation 8. If the hatching pleats and strip be removed entirely from an egg at room humidity the egg becomes slightly shorter than before, while the gap between the free edges of the chorion on either side of the original position of the hatching strip, becomes slightly bigger than the width of the hatching pleats and strip removed. This gape is of course covered by the c.v.membrane which under-

lies the chorion, and indicates that the chorion is under some transverse tension at room humidity. When an egg with hatching strip removed is exposed to high humidity (90 - 100% R.H.) the gape in the chorion becomes reduced, as shown in Fig. 14. This partial closing of the gape with

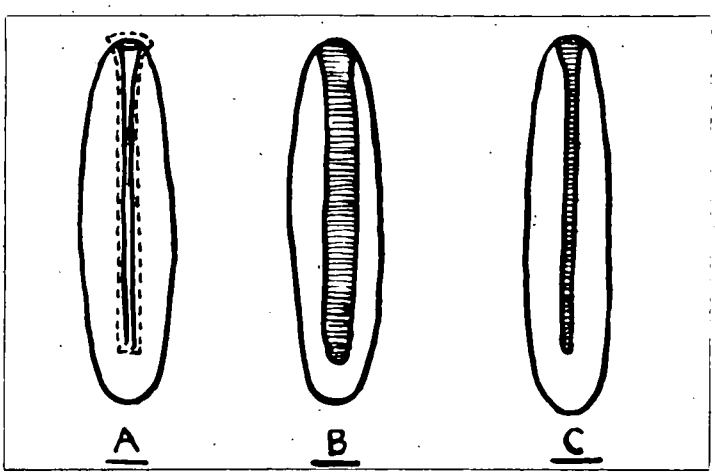

FIG. 14. A, area within broken line removed, to give appearance B in low R.H. and C in high R.H. Shaded areas represent c.v. membrane exposed.

rise in humidity suggests that the chorion is acting rather like a bimetallic strip does with rise in temperature. In Fig. 15 the cross-section of the egg with hatching strip removed, at high and low R.H. is compared with that of a bimetallic strip in the form of an incomplete circle at high and low temperature, and is drawn from the observations on the blowfly egg described above and on a bimetallic strip of the above form.

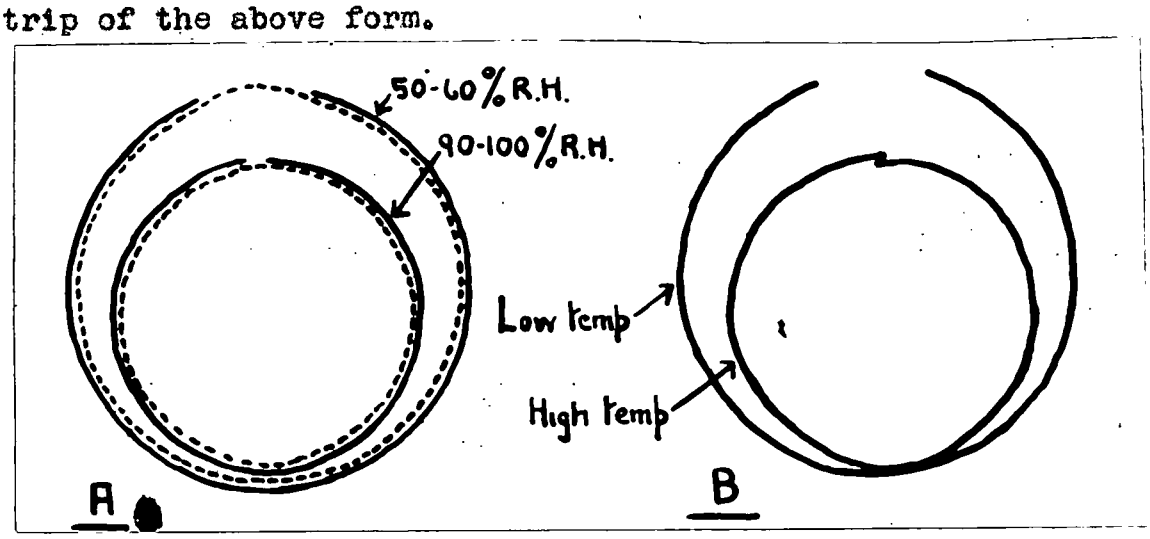

FIG. 15. A, T.S egg with hatching pleat area removed (as in fig. 14.). Unbroken line represents chorion, and broken line the c.v. membrane. B, diagram of behaviour of bimetallic strip with metal of high coefficient of expansion on outside.

Shape changes in eggs in relation to hatching. Since the above observations have shown that the chorion alone is responsible for the humidity dependent shape-changes, further analysis of the mechanism whereby they are accomplished depends on a knowledge of the structure of the chorion. It has already been shown that the chorion undergoes curling movements dependent on R.H. (page 12). This leads, with higher R.H., to increased curvature of the chorion. It has also been shown that the chorion is composed of at least two main layers of different composition (page9-12). It was concluded that the protein of the inner layer may be already tanned, or of such a nature that it is not suceptible to tanning by the usual agents. Embedded and partly separating the two layers from each other a row of dark bodies were observed. The curling movements of the chorion fragments suggests that the two layers are differentailly affected by absorbtion of water, leading to expansion of the outer layer with rising R.H., and contraction or non-change of the inner layer, both considered around the circumference and not along the length of the egg. The structure of the chorion leading to curling of chroion fragments with R.H. changes, leads to a change in shape of the intact chorion on the egg. That the curling movements and the shape change of the egg with rising R.H. are due to absorbtion of water by the chorion is supported by the fact that the elongation of blowfly eggs with increasing R.H. is affected very little by temperature; the increased length of eggs at 100% R.H. over their length at 0% R.H. was found to be the same at 37° as at 17°C. The process of absorbing water by organic materials from the atmosphere is well known to be affected very little by temperature, and to be largely dependent on R.H., as is shown by the curves of weight increase of washed wool at various

temperatures and humidities, used by the woollen industry. The almost instantaneous nature of the curling movements of chorion fragments, and of the egg shape changes means that the chorion must contain extremely hydrophilic protein, with a very large surface area in relation to volume. The latter condition is fulfilled by the outer chorion layer, whose thickness is but 2-3μ , and with a surface area of about 8600 sqμ, and since oily materials do not spread over it (page 132) it is also hydrophilic. The outer layer presumably expands by absorbtion of water, around the circumference of the egg, while the inner layer may contract in the same direction or maintain its initial length, thus producing curling of the chorion as in a bimetallic strip. The reduction in cross-sectional area of the egg (Observation 5) and the hatching pleat movements (Observation 6) shows that an increased curvature of the chorion is brought about by increasing R.H. with little or no absolute contraction of the chorion as a whole, so that the outer chorion layer is relatively longer compared with the length of the inner layer, around the circumference of the egg. Thus the tendency of the outer chorion layer to expand with increasing R.H. is achieved only relatively. The important factor in the curling movements is probably the relative stresses and strains put on each other by the two chorion layers. It is possible, although purely speculative, that the dark bodies embedded in the chorion are present in order to reduce the area of contact of the two contrasting chorion layers, so that length changes can occur without much shear strain between them.

The effects of humidity on the chorion can ultimately be fully worked out only after a knowledge of its molecular structure has been deduced. Such knowledge is at present completely lacking. Although highly hypothetical it is

interesting to speculate whether these effects are brought about by a molecular structure in some way comparable to that of mammalian hair and horn keratin. In the case of wool fibres, it has been shown by Speakman (1931) that the long keratin molecules are mainly arranged in a parallel manner to form micelles which are at least ten times as long as they are broad. The mechanism by which the great reversible extension of wool fibres (amounting to about 100% of their original length) is brough about under tension at high humidity, has been elucidated by Astbury and his co-workers. They have shown (Astbury and Woods, 1931, 1933) that the elongation is brought about by an intramolecular extension of the keratin involving, basically, the straightening out of what in the unstretched fibre is a folded chain molecule, so that two forms of keratin, (α and β), representing the stretched and unstretched form are identifiable, and which give X-ray diffraction patterns which are distinct from each other. A change in the length, on a small scale, of fairly long, parallel orientated protein molecules may occur in the blowfly egg chorion, caused by changes in the water content of protein micelles. A difference of only 4-5% in the length of the chorion molecules would be sufficient to account for the facts. The action of water in the chorion may be the same as in the wool fibre, where, according to Astbury and Woods (1933) the water molecules enter the keratin micelles and inter-micellar keratin, and become adsorbed to the chain molecules, causing swelling and a 'lubrication' of the chains, permitting the intramolecular extension.

Several different molecular arrangements on the above lines in the blowfly egg chorion can be visualized, which might be expected to cause effects which would fit in with observations. In one arrangement the outer chorion layer

may be though of as built of chains orientated around the egg. These chains would increase in length with rising R.H. by a partial straightening out of folded chains and produce an expansion of the layer. Co-existent with this the inner protein layer might be formed of chains orientated along the egg and which expand along the long axis of the egg on absorbtion of water, so that the layer tends to elongate but contracts in the transverse plane around the egg - that in which the outer layer tends to expand. A representation of this arrangement is given in Fig. 16.

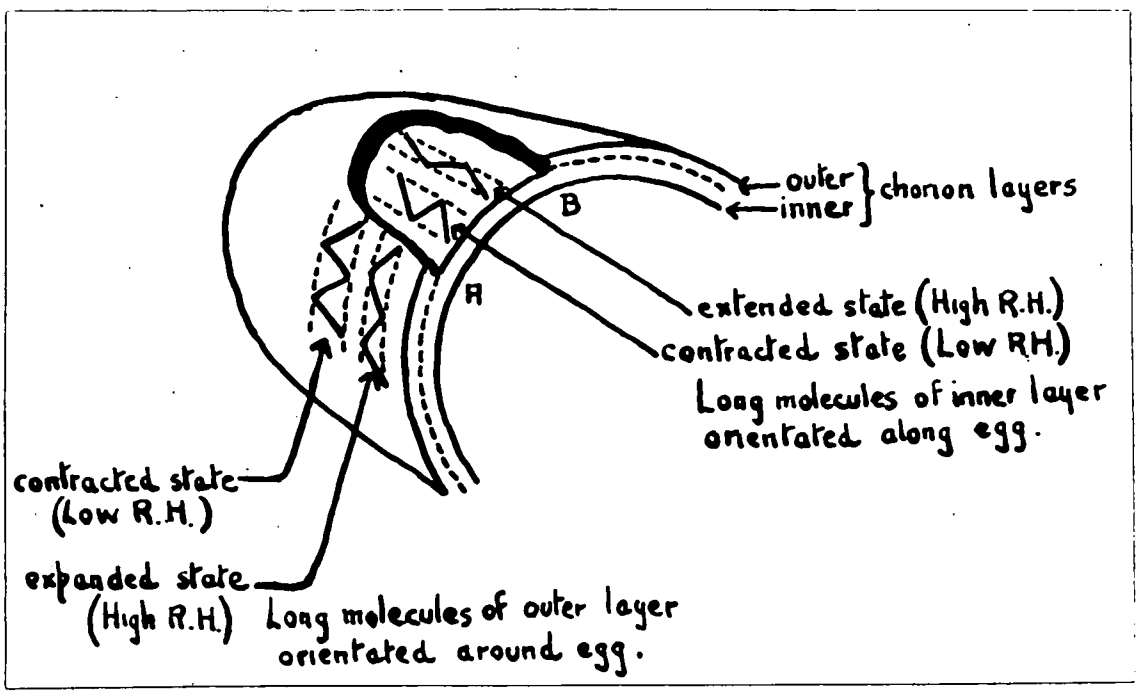

FIG. 16. Diagrammatic representation of possible molecular structure of blowfly egg chorion.

Other arrangements can be visualized, but no gain is made by considering them since no facts are available on the molecular structure of the chorion. It has been found that when the chorion is viewed in polarized light, it is anisotropic, which suggests that there is no marked parallel orientation of long molecules through most of its thickness. It may be that the chorion proteins are not built into long chains and that the expansion of the outer chorion layer in high R.H. is purely a swelling process due to water absorbtion, similar to that of gelatin and many other

proteins, and that the inner layer contracts because it 'gives' to the compression imposed on it by extension of the outer layer. There is some evidence that the inner chorion layer is of a spongy nature, the cavities of the sponge being filled with air. Where the inner chorion layer is thickened, at at the edge of the micropylar plate (see page 14 Fig.4), a distinct spongy structure could be made out with the highest power of the microscope. It is possible that the same is true, on a smaller scale, of the inner chorion layer over the general egg surface. The air content of the inner layer would thus explain the white colour of the chorion (see page 18). A parallel orientation of the protein molecules of the outer layer only, around the circumference of the egg might coexist with this arrangement of a spongy inner layer. The full explanation of the mechanical properties of the chorion, and thus of the humidity-dependent shape changes of blowfly eggs must await a study of its molecular structure, using specialized X-ray methods, similar to those used by Astbury for keratin structures, and by Fraenkel and Rudall (1940) for the blowfly puparium.

A more empirical conclusion to the observations on shape changes, and linking them with the hatching mechanism of blowfly eggs, may be set out as follows:-

With increasing R.H. from 0%, the cross-sectional area of the egg is reduced while the egg elongates. It may be that the primary shape change is the former, and that the latter is purely compensatory for it, since the enclosed egg resists the increased deformation imposed on it by the shape change of the chorion, and is forced to elongate. There may be a tendency in the chorion itself to elongate along the egg so that both elongation and reduction of the cross-sectional area of the egg proceed without the

latter causing the former and being resisted by it. If the primary shape change is one of forcing the enclosed egg to reduce its cross-sectional area, strains will be set up in the chorion liable to result in longitudinal rupture along the egg, and not transversely. Since the reduction in cross-sectional area increases cumulatively from dry air up to saturation, the strain on the chorion also increases with humidity. If there are two longitudinal lines of weakness in the chorion (on the outer side of each of the hatching pleats) rupture along these lines would become increasingly easy to accomplish by the larva inside the egg, with increasing humidity. Since the amount of elongation of the egg is almost certainly a measure of the reduction in cross-sectional area the amount is also a measure of the strain on the chorion. The curves of elongation of eggs of various species (Fig. 10 page 62) are thus indicative of the progressive increase in strain on the lines of weakness in the chorion, with increasing humidity. They show therefore an increasing probability of successful rupture of the chorion by the larva with increasing R.H. They can be related to the known effects of humidity on hatching in blowfly eggs - the minimum humidity for hatching, the final percentage hatch achieved by a group of eggs, and the increasing dispersion of hatching times within a group of eggs with decreasing R.H. From Fig.10, it is to be expected that at humidities of 100-90% R.H., the chorion is already under maximum strain due to the shape changes outlined above, and the blows of the larva on the inside of the micropylar plate (see page 9) are likely to cause rupture fairly quickly. With lower R.H's of 90-60% the chorion is under progressively less strain, and many more blows by the larva may be necessary to cause rupture of the chorion, and frequently exhaustion or desiccation of the larva supervenes before hatching is achieved. This

is shown by the increasing number of fully developed larvae found dead insede their shells with decreasing R.H. within the above range (page 38). Below 55-60% R.H., any number of blows cannot rupture the chorion, and no hatching occurs in the vast majority of cases. The decreasing strain on the chorion with decreasing R.H. means that the larva has to exert itself to a greater extent to achieve rupture of the chorion. Thus it was found that the time required for all the constituent eggs of a batch, to hatch, increased quickly with decreasing humidity from saturation (page 38).

In Observation 1 (page 62) it was shown that subjection of eggs to sudden humidity changes of 0 to 100% R.H. usually caused the chorion to split along the outer margins of one or other of the two hatching pleats. It was there pointed out that in eggs so treated the split varied in its site of origin and could occur anywhere along the length of the hatching pleats. When eggs hatch in the normal way, due to the activity of the enclosed larvae, the split in the chorion invariably originated at the extreme anterior end of the hatching pleats, where they diverge to partly surround the micropylar plate (page 10 , Fig. 1). This anterior localization of the split in hatching, compared with the greater variability in the site of its occurrence in eggs subjected to sudden humidity oscillations, may be explained in the follow way. When the larva has reached the hatching stage, it hammers on the inside of the micropylar plate with its mouth-hooks. When observed under a binocular, each blow of the larva was seen to cause the micropylar place to bulge out slightly in the middle, and also caused the front end of the egg to elongate slightly. At high humidities this elongation accentuates still further the elongation of the egg caused by high R.H. itself, and would be expected to reduce the cross-sectional

area of the anterior end, and so increase still further the strain on the anterior part of the hatching strip. This increase in strain on the anterior end would be expected to be localized there, since it is unlikely that strains set up in one part of the chorion would be transmitted freely to other parts of the shell. Thus in hatching the chorion split would be expected to occur near the anterior end of the egg.

It was observed that the chorion of the blowfly egg was considerably stiffer at all humidities below saturation that in saturated air itself. Vacated egg-shells, in saturated aie collapsed completely and became closely applied to the glass slide substratum. At 95% R.H. and down to the minimum humidity for hatching vacated shells did not become collapsed so that, if the rupture in the chorion was obscured for some reason it was found difficult at first glance to determine whether the larva had hatched or not. At 100% R.H., it was observed that the chorion was limp, like cardboard soaked in water, so that the two sides of the split in the chorion could readily be forced apart by the escaping larva. At humidities below saturated larvae seemed to find difficulty in forcing their way out by pushing apart the two sides of the split, rather like a man trying to force his way through a slit in a stiff cardboard sheet. At 17-18°C observation of times of hatching of groups of eggs, gave the following figures:- 100% R.H. - 1 larva escaped per minute; 90% R.H. - 1 per 3.63 min; 80% R.H. - 1 per 14.88 min. The longer time required at 90 and 80% R.H. is well marked. The activity of the larvae is low at 17-18°C, and the times required by them to escape from the shell following its rupture would be less at 30-37°C. After rupture has been accomplished at 37°C, larvae completely extricated themselves from the limp shells

within 10-30 secs. in saturated air. At the same temperature, after inital chorion rupture, at 65% R.H., it took from 15-35 mins for the same process to be accomplished. Thus humidity affects the rate of hatching in a group of eggs in two ways:- one, by making it more difficult for the larva to rupture the chorion with decreasing humidity from saturation, owing to the shape changes considered in the preceding pages, and two, by making it more difficult for the larve to escape from the shell after it has succeeded in initially rupturing the chorion, with decreasing R.H. from saturation.

(v) The effects of variation in humidity

It was found that if L.sericata eggs were placed in saturated air for a few minutes, at 37°C, they became more susceptible to desiccation if subsequently incubated at lower R.H's, than eggs which were placed directly after laying into the low humidity (page 41-2). Experiments on similar lines were carried out on the eggs of four of the five blowfly species considered in this section, to determine whether a similar effect could be detected with them. Eggs of these species were incubated at humidities known to be fairly near the minimum required by them to complete development, both directly after laying and after a preliminary period in saturated air. The two lots were in every experiment kept in the same vessel so that both were subjected to identical R.H's after one lot had been in saturated air. In each case a set of eggs from the same batch were kept in saturated air throughout their incubation period as controls. The experiments were carried out at temperatures slightly below the highest temperature at which the species was known to be able to complete development in the light of water bath experiments. The

TABLE 14

(a) P.TERRA-NOVAE EGGS

Expt.No.	Treatment of eggs	% eggs reaching various stages.* H	PH	MH	E	No. of eggs used.
1	CONTROL, 100% R.H. 37°C	88	0	0	12	25
	65% RH. 37°C for whole incub. period.	0	98	2	0	50
	100% RH 14°C for 1·5hr., then into 65%, 37°C.	0	84	0	16	50
	100% R.H. 37°C for 1·5hr. then into 65%, 37°C Y	0	0	0	100	100
	(b) L.ILLUSTRIS EGGS					
2	CONTROL, 100% R.H. 35°C.	35	0	25	40	20
	80% R.H. 35°C. for whole incub. period.	30	0	20	50	20
	100% RH. 35°C. for 1·2hr. then into 80%, 35°C	22	13	22	43	20
3	CONTROL, 100% R.H. 35°C.	52	2	11	15	44
	80% RH. 35°C. for whole incub. period.	81	2	0	17	47
	100% R.H. 35°C. for 1·2hr., then into 80%, 35°C	71	4	6	19	48
4	CONTROL, 100% R.H. 37°C.	98	0	0	2	64
	75% R.H. 37°C for whole incub. period.	78	5	14	3	58
	100% R.H. 37°C for 0·25hr. then into 75%, 37°C	71	18	3	8	63
5	CONTROL, 100% RH. 37°C.	64	3	6	17	36
	70% R.H. 37°C for whole incub. period.	0	18	12	70	34
	100% RH. 37°C for 0·25hr. then into 70%, 37°C.	0	0	3	97	36
	(c) C.ERYTHROCEPHALA EGGS					
6	CONTROL, 100% RH. 30°C.	100	0	0	0	36
	80% R.H. 30°C. for whole incub. period.	97	0	0	3	36
	100% R.H. 30°C for 0·5hr. then into 80%, 30°C	92	0	0	8	12
	100% R.H. 30°C for 1·0hr. then into 80% 30°C	92	0	8	0	12
	100% R.H. 30°C for 1·45hr. then into 80%, 30°C	100	0	0	0	12
	100% R.H. 30°C for 3·0hr. then into 80%, 30°C	100	0	0	0	12
	100% R.H. 30°C for 3·5hr. then into 80%, 30°C	100	0	0	0	12
7	CONTROL, 100% R.H. 33°C	71	9	5	15	21
	80% R.H. 33°C. for whole incub. period.	64	27	9	0	22
	100% R.H. 33°C. for 0·75hr. then into 80%, 33°C	75	12	8	5	24

* For details of classification of stages, see Appendix, page 161-2.

TABLE 14 (continued)

(d) C. VOMITORIA EGGS

Expt. No.	Treatment of eggs	% eggs reaching various stages * H	PH	MH	E	No. of eggs used.
8	CONTROL, 100% R.H. 30°C.	95	0	0	5	20
	70% R.H. 30°C. for whole incub. period.	0	55	30	15	20
	100% R.H. 30°C. for 1·0 hr. then into 70%, 30°C.	0	12	65	23	17
	100% R.H. 30°C for 2·0 hr. then into 70% 30°C.	0	15	65	20	20
9	CONTROL. 100% R.H. 30°C.	100	0	0	0	36
	80% R.H. 30°C. for whole incub. period.	92	0	3	5	36
	100% R.H. 30°C for 0·5 hr. then into 80%, 30°C	92	0	0	8	12
	100% R.H. 30°C for 1·0 hr. then into 80% 30°C	83	0	0	17	12
	100% R.H 30°C for 1·5 hr. then into 80%, 30°C	92	0	0	8	12
	100% R.H 30°C. for 3·0 hr. then into 80%, 30°C	92	8	0	0	12
	100% R.H. 30°C. for 3·5 hr. then into 80% 30°C	100	0	0	0	12
10	CONTROL, 100% R.H. 33°C.	71	12	4	13	24
	80% R.H. 33°C for whole incub. period.	21	0	0	79	24
	100% R.H. 33°C for 0·75 hr. then into 80%, 30°	62	8	17	13	24
11	CONTROL, 100% R.H. 35°C.	87	3	0	10	38
	80% R.H. 35°C. for whole incub. period.	36	36	14	14	36
	100% RH. 35°C for 0·75 hr. then into 80%, 30°C	14	60	19	7	42
12	CONTROL, 100% R.H. 35°C.	72	19	7	2	58
	80% R.H. 35°C. for whole incub. period.	38	55	5	2	120
	100% RH. 35°C. for 0·25 hr. then into 80%, 35°	28	55	11	6	282
13	CONTROL, 100% R.H. 35°C.	70	11	9	10	64
	75% RH. 35°C. for whole of incub. period.	2	3	3	92	65
	100% R.H. 35°C. for 0·45 hr. then into 75%, 35°C.	0	2	3	95	63
14	CONTROL 100% R.H. 34°C.	92	3	1	4	114
	75% RH. 34°C for whole incub. period.	12	35	12	41	208
	100% R.H. 34°C for 0·2 hr. then into 75%, 34°C.	8	43	27	22	226
15	CONTROL. 100% R.H. 34°C.	37	0	3	60	59
	75% R.H. 34°C for whole incub. period.	21	21	5	53	154
	100% RH 34°C for 0·25 hr. then into 75%, 34°C	27	29	4	40	140

* For details of classification of stages, see Appendix, page 161–162.

results of 15 experiments are given in Table 14. It will be seen from the results of Expt.1 that P.terra-novae eggs failed completely to survive at 65% R.H. 37°C after a 90 min. exposure to saturated air at the same temperature, while a high percentage (98%) completed development at the same humidity without prior exposure to saturated air, thus showing a 'conditioning' effect, as was noted with L.sericata eggs. From the same experiment it will be seen that no significant 'conditioning' took place if P.terra-novae eggs were exposed to saturated air at 14°C, another feature of similarity to L.sericata. L.illustris eggs showed a 'conditioning' effect in Expt. 5 only. In three other experiments subjection to saturated air did not reduce the number of eggs completing development at low humidities. Two experiments on C.erthrocephala eggs (Expts. 6 and 7) showed no 'conditioning' effect. Of eight experiments on C.vomitoria eggs (Expts. 8 - 15) only (Expt. 8) showed a clear cut 'conditioning' effect.

Although many experiments (Table 14, pp.80-81) showed no reduction in the number of eggs completing development in 'conditioned' compared with unconditioned eggs, there was some indication that in the former eggs fewer of those completing development succeeded in hatching. The experiments cannot be compared statistically because they are not homogenous, various combinations of temperature and humidity being employed. It is concluded from them that L.sericata and P.Terra-novae eggs showed a well marked 'conditioning' effect, while results on the other species were more erratic.

An alternative explanation for the 'conditioning' effect, other than that previously offered (page 42) may now be considered. It has been found that eggs undergo shape changes in relation to humidity (page 57-61) and that they are

due to the properties of the chorion, which forces the egg enclosed in it to deform. It is thus likely that there is some slight movement of the chorion in relation to the c.v.membrane when eggs are taken suddenly from say 70 to 100% R.H. Certainly an increased pressure of the chorion on the c.v.membrane must take place over some parts of the shell. Evidence has also already been given that there is a lipoid waterproofing layer in blowfly eggs situated between the chorion and the c.v.membrane (page 17-29). If this lipoid layer displays some molecular orientation, as is known to be the case in insect cuticle lipoid layers (Beament, 1945), it is possible that movement of the chorion or increased pressure of the chorion, in relation to the c.v.membrane, would cause some slight disorganization of the lipoid layer, possibly of a temporary nature. This would increase the permeability of the shell and cause increased water loss. Eggs exposed to saturated air for a few minutes and then transferred to a lower R.H. would thus be more susceptible to desiccation than eggs placed directly at this lower R.H. Greater water loss in 'conditioned' than in unconditioned eggs was shown to occur in L.sericata (page 43).

(vi) The size of the eggs of the various species.

The eggs of the species were measured in the course of the work. They were selected at random from egg batches, and the length and greatest width measured. The apparatus used for measuring was that described on page 57 . In view of the fact that the lengths of eggs varied with R.H. precautions were taken against breathing on them while they were being measured. The means of the measurements and the standard deviations, for the eggs of the various species are given in Table 15, page 84 . From this table it will be seen that there was considerable overlap between the extremes

TABLE 15

Dimensions of blowfly eggs (μ)

25 eggs from each batch

Species	Mean length and S.D.	Extremes of length variation	Mean width and S.D.	Extremes of width variation
L.sericata (batch a)	1261 ∓ 29	1219 - 1291	315 ∓ 8	301 - 333
" (batch b)	1142 ∓ 21	1097 - 1170	305 ∓ 6	292 - 317
L.caesar	1174 ∓ 28	1121 - 1243	325 ∓ 8	309 - 340
L.illustris	1371 ∓ 37	1284 - 1414	313 ∓ 14	292 - 340
L.ampullacea	1283 ∓ 55	1202 - 1373	359 ∓ 12	333 - 374
P.terra-novae (batch a)	1097 ∓ 51	942 - 1154	339 ∓ 6	325 - 349
" " (batch b)	1273 ∓ 25	1219 - 1309	304 ∓ 8	288 - 313
C.erythrocephala	1404 ∓ 55	1284 - 1487	382 ∓ 12	350 - 406
C.vomitoria	1225 ∓ 17	1178 - 1267	339 ∓ 7	325 - 355

of variation in each species. The mean length of L.illustris was greater than of the other Lucilia species, but the smallest L.illustris eggs fell into the range of the largest L.sericata eggs. If greater numbers of eggs had been measured it is likely that the overlap would have been greater. That the mean egg length varied from batch to batch in one species is also shown by the figures . It will be seen that the C.erythrocephala eggs measured were bigger (mean length 1404 ∓ 55μ) than those of C.vomitoria (mean length 1225 ∓ 17μ) although the variation means that the smallest eggs of C.vomitoria are within the range of the larger eggs of P.terra-novae and the Lucilia spp.

In all the species studied, one or two abnormally short eggs were fairly frequently found in batches of otherwise normal eggs. These short eggs had the same width as full-length eggs, so that they were relatively fatter. Other types of abnormal eggs were sometimes encountered, and occurred among all the species. These abnormalities

concerned the hatching strip and hatching pleats (Fig. 17). In L.caesar eggs particularly the hatching strip and pleats were frequently extremely short (Fig. 17).

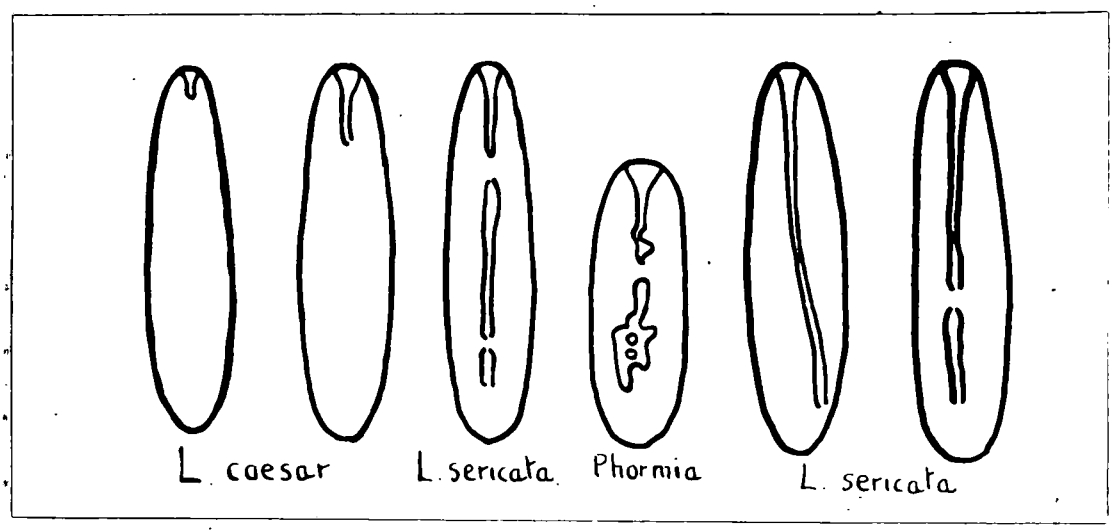

FIG. 17. Examples of eggs encountered with abnormal hatching pleats.

(vii) Size of L.sericata eggs in relation to size of parent female

A large batch of eggs was obtained from normal sized L.sericata stock-culture flies, and divided into two approximately equal parts. Both lots were bred out to flies using the normal technique. One lot of larvae was allowed to feed to full size (i.e. for four days at 25-26°C) and produced normal full sized flies - the females weighing 50 - 70 mg. when gravid. The other lot of larvae were allowed to feed for only 1.5 days and produced extremely small flies, the gravid females weighing only 15-20 mg. Both lots of flies were kept in separate cages and fed with meat in the usual way until egg laying commenced. Eggs from both lots were selected at random from batches and their lengths measured. The mean length, standard deviation, and extremes of variation of eggs from three batches laid by small and three by large flies were as follows:-

(μ)

Eggs laid by full sized flies

Batch 1 1196 ∓ 52 (1071 - 1285)

Batch 2 1225 ∓ 30 (1145 - 1285)

Batch 3 1150 ∓ 37 (1097 - 1259)

Eggs laid by small flies

Batch 1 1077 ∓ 69 (988 - 1202)

Batch 2 1264 ∓ 23 (1211 - 1309)

Batch 3 1186 ∓ 10 (1161 - 1202)

The means of the three batches pooled in each case were as follows:-

Eggs laid by full sized flies

1194 ∓ 43μ

Eggs laid by small flies

1174 ∓ 87μ

It will be seen that there were no marked differences in egg size correlated with size of fly; the differences between the mean lengths are smaller than the standard deviations. The small flies laid both the smallest (988μ) and the largest (1309μ) eggs. The greater variation found in the size of eggs laid by small flies is probably purely accidental, and if greater numbers of batches had been examined the variation ~~between~~ in the mean length would probably have been more similar in both groups. It is therefore concluded that the eggs laid by small females are not smaller than those laid by normal full sized females.

Observations on the numbers of eggs laid by the two groups of flies were carried out. The small flies were found to lay very much fewer eggs than did the large flies. Although both lots of flies contained approximately the same number of females, two days after commencing egg laying, the small flies had laid only 58 eggs, while by the same time the large flies had laid nearly 400. Before they died the small flies only laid about 150 more eggs while the

full sized flies laid several hundreds. Counts of ovarioles in flies of both sizes yielded the following results (Table 16).

TABLE 16

No of ovarioles in L.sericata

4 Small Flies	4 Large flies
58, 74, 60, 69, (Average 67)	278, 184, 227, 280 (Average 242)

It appears that the undersized female L.sericata does not lay eggs of reduced size, but maintains the egg size at about the normal level, while accommodating them in the abdomen of greatly reduced volume by having a reduced number of functional ovarioles, so that fewer eggs mature together. Mackerras (1936) states that small L.cuprina females produce fewer eggs than large ones.

(viii) The waterproofing mechanism of blowfly eggs

In view of the evidence obtained of the existence of a lipoid waterproofing layer between the chorion and the c.v.membrane of the L.sericata egg (page 27-8), experiments on similar lines were carried out on the eggs of the five other blowfly spp. studied.

The effect of abrasion with fine alumina of the intact c.v.membrane after removal of the chorion from the egg, and of dusting the c.v.membrane with alumina which was kept static and not allowed to abrade the membrane, was studied by means of experiments similar to those carried out on the L.sericata egg, and identical results were obtained. In all five spp. slight abrasion of the c.v.membrane led to such rapid water loss from eggs that they became completely collapsed within 5 min. in dry air at 19°C, by which time untreated controls with chorions removed kept under the same conditions had not yet lost their original sausage shape.

Measurements of the rates of water loss from eggs in dry air at various temperatures were carried out. The technique was similar to that described for the L.sericata egg (page 22-3). Surface area measurements of the egg batches were however not made owing to the variation in the sizes of egg within a single batch. Instead the eggs in each batch were counted and the water loss expressed in terms of mg. water loss per hr. per 100 eggs. The curves obtained and expressed in this way are given in Fig.18. From Fig.18

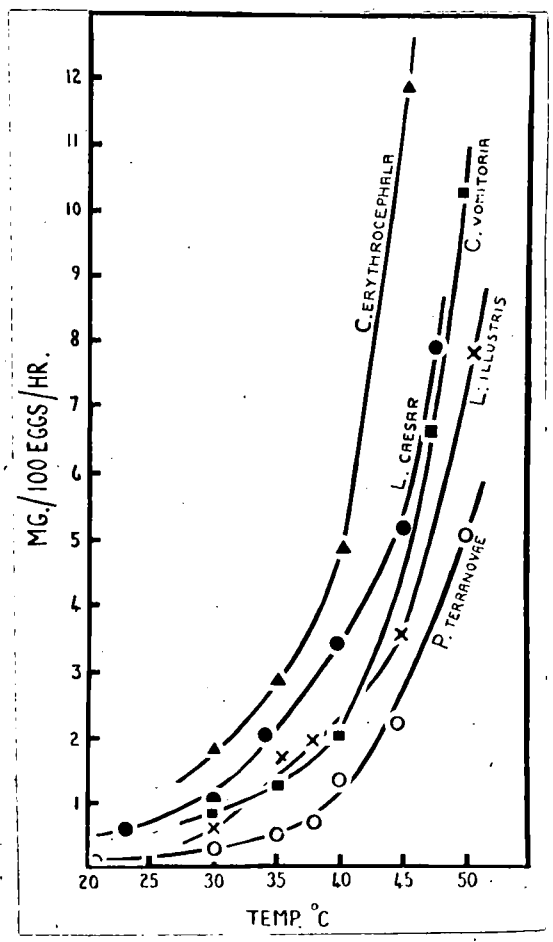

FIG.18. Temp./water-loss curves of eggs of species studied other than L.sericata.

it will be seen that the water loss/temperature curve of P.terra-novae eggs showed a 'critical' temperature in the region of 37-39°C, but was not so well defined and abrupt as in the L.sericata egg. The curves for the eggs of L.caesar, L.illustris, C.erythrocephala and C.vomitoria showed no abrupt 'critical' temperature. The waterproofing lipoid of these eggs would therefore appear to be different to that of L.sericata. The abrasion experiments however indicate its presence. It is unlikely that the eggs of such closely related species to L.sericata as

blowflies, would be waterproofed in a fundamentally different manner.

(ix) Ovoviviparity in the blowflies studied.

The occurrence of ovoviviparity in C.erythrocephala had long been known. Lowne (1890-92) states that two or even three fertilized eggs may be retained in the uterus of this species. In the present work ovoviviparity in this species was frequently found to occur in laboratory cultures, but in every case examined only one fertilized egg was retained in the uterus, never more.

Ovoviviparity was also found commonly among culture flies of C.vomitoria, and also in wild caught flies of both Calliphora spp. It appears therefore that ovoviviparity in these two species occurs under natural conditions and is not merely a result of artificial laboratory treatment.

In both Calliphora spp. it was observed that the retained fertilized egg could be one of the first eggs of the batch matured by the fly since they were found in flies which had had no opportunity for laying eggs since emmergence. It was also observed that the retained egg might be the last egg of a batch, since flies were found containing a fully developed egg in the uterus, while the eggs in the ovarioles were in an early stage of growth. These flies had already matured and laid one or more batches of eggs.

The length of time that a fully developed larva imprisoned within the egg in the uterus of the adult, can remain alive was estimated in C.vomitoria in the following way. The complete reproductive organs, including the ovipositor, were dissected out from gravid females. Those cases where a retained fertilized egg was present in the uterus were placed on glass slides and kept moist with

Ringer at 16-18°C. In many of these cases the retained eggs had not yet completed development when removed from the parent flies. They were observed at 2 hr. intervals until the fully developed larva could be observed moving in the eggs. These observations were continued until the movements of the larva ceased permanently, when it was then judged to be dead. In this way it was found that the first instar larvae could survive imprisonment in the uterus for up to about 45 hr. at 16-18°C. This figure represents the length of time that the first instar larvae can withstand starvation at that temperature.

Whether ovoviviparity also occurred in Lucilia spp. and P.terra-novae was studied by dissecting stock culture females, wild caught females and specially bred and treated flies. The latter involved the breeding of flies, feeding them on meat until a high proportion of the females were gravid, and then removing the meat, and subsequently dissecting a number of them each day for a fortnight. Neither in flies so treated, nor in culture or wild flies was a single case of the retention of fertilized eggs found in the above species.

DISCUSSION

The water-bath and incubator experiments showed that the eggs of L.caesar, L.illustris, C.erthrocephala, C.vomitoria and P.terra-novae were more susceptible to dessication than the eggs of L.sericata. It was noted in the introduction (page 47) that L.sericata is the main cause of sheep myiasis in Britain, the other species being of less importance. Since microclimatic studies have shown (Macleod 1940, Davies & Hobson, 1935) and Part III of this thesis) that conditions in sheep fleeces are frequently very dry it appears that the eggs of L.sericata are better adapted to withstand the rigorous conditions than those of the other

blowfly species studied. The eggs of L.sericata species require a shorter incubation period than those of other species. This may be favourable to it in an environment such as the sheep's fleece where rapid fluctuations in R.H. may occur, (page 107-110). The humidity and temperature relations of the eggs of the individual species are discussed in relation to information on conditions in sheep fleeces on page 139-140.

The details of the hatching mechanism of blowfly eggs has already been discussed (page 71-79).

The ovoviviparity which was examined in C.erythrocephala and C.vomitoria cannot be of great importance in sheep myiasis, because of the fact that only single eggs are retained at the time. Since fairly large numbers of larvae are necessary before myiasis is successfully established, a single retained egg laid with each batch of Calliphora eggs, is of no consequence.

The failure to observe or induce ovoviviparity in the other blowfly species indicates that it must at least be so rare as to be of little importance.

SUMMARY

1) The minimum humidities for completion of development at various temperatures, for the eggs of L.caesar, L.illustris, C.erythrocephala, C.vomitoria and P.terranovae were determined. It was found that they all required higher R.H's for some of the eggs to complete development, than those of L.sericata at the same temperatures.

2) The eggs of the above five species required high humidities for hatching as with those of L.sericata.

3) It was found that blowfly eggs changed shape in relation to R.H. changes. These shape changes were due to the mechanical properties of the chorion, which forced the enclosed egg to deform. Evidence that they form a hatching mechanism was produced, and correlated with the known effects of R.H. on the percentage hatch, and the speed of hatching in groups of eggs.

4) The 'conditioning' effect noted in L.sericata eggs was also found with P.terra-novae egg, but seemed less well marked and more variable in the other species studied.

5) Determinations of the incubation periods of eggs of the six species studied, at various temperatures, showed that those eggs of L.sericata and L.illustris completed development in a shorter time than those of the four other species studied.

6) Small L.sericata flies, produced from underfed larvae, were found to lay fewer eggs than normal sized females. Eggs laid by flies of both sizes were found to be of the same length.

7) Evidence is produced that the eggs of the five species studied are waterproofed by a lipoid layer as was shown for the L.sericata egg.

8) Ovoviviparity to the extent of retaining a single egg at a time was found and studied in C.erythrocephala and C.vomitoria. No retention of fertilized eggs was observed in the Lucilia and Phormia species.

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PART III

FIELD OBSERVATIONS ON FLEECE-ATMOSPHERE HUMIDITY AND ON THE SURVIVAL OF BLOWFLY EGGS IN SHEEP FLEECES

Introduction

Davies and Hobson (1935), making use of the natural hygroscopicity of wool, measured the relative humidity of fleece atmosphere. They studied a variety of fleece types under various weather conditions, but confined attention to Welsh Mountain sheep. They found that the R.H. in the fleece, (next to skin) except on the rump and anal regions, rarely exceeded 70% even during wet weather, when the fleece was moist on the outside. They concluded that the humidity in the fleece was the deciding factor in determining susceptibility to blowfly strike, and that the humid conditions in soiled wool around the breech, accounted for the frequency of strike in that region. This conclusion was supported by the laboratory work of Evans (1934) on the humidity relations of L.sericata eggs. Also Macleod (1940) showed that the fleece R.H. near the skin under summer conditions rarely exceeded 50%, and, in the sheep he used, rarely became saturated even after heavy rain.

Recent laboratory work (Part I, page 41) has shown that L.sericata eggs can survive fairly long periods under rather dry conditions at the temperatures expected near the sheep's skin, and thus would be expected to complete their development in fleeces if rapid increases in R.H. occurred during their incubation period. The question of fleece humidity was therefore reinvestigated in order to determine whether such fluctuations in R.H. did occur during summer weather. Humidity readings were made, in the first place, by means of cobalt chloride papers as modified by Solomon (1945). This method was particularly suitable for the purpose since consecutive readings at particular sites in the fleece could

be carried out, and a record of day to day changes in fleece R.H. obtained with the minimum of disturbance of fleece conditions. Later in the work, a modification of the cobalt chloride paper technique, and also cobalt thiocyanate paper were employed in addition.

In conjunction with readings of fleece atmosphere R.H., a study was made of the fate of laboratory laid egg batches placed in sheep fleeces, in order to see how far laboratory results paralleled the behaviour of eggs under field conditions. This study also formed a basis for checking the accuracy of the fleece R.H. readings.

I WORK IN 1946

(i) A survey of methods of measuring R.H.

The problem of R.H. measurement is one presenting considerable practical difficulties in spite of the fairly wide range of methods available and theoretically utilizable. In entomological work perhaps the main difficulty is that the information on R.H. is required for the air actually surrounding the insect, i.e. the microclimatic R.H. needs to be measured, often with very small air samples available. Methods used by meteorologists are mainly quite unsuited on this account. The small air sample accentuates the difficulty that some methods actively change the humidity of the air while the measurement is being carried out. Also the relative amount of disturbance of humidity conditions brought about by the actual process of making the determination, is usually greater with a small volume of air in the environment.

In the present work R.H. measurements were required for the atmosphere of the sheep's fleece - i.e. of the air entangled between the wool and other fibres of sheep. Information on the humidity at various known distances from the sheep's skin was aimed at, since blowfly eggs may be laid

at various distances from the skin within the thickness of the fleece. In the fleece atmsophere a thermal stratification occurs, with higher temperature close to the skin with a falling temperature gradient away from the skin. It is to be expected that the temperature falls to a level very near that of the outside air, at or near the tips of the wool fibres. This thermal stratification, if it were the only factor affecting fleece R.H. would produce a gradient of decreasing R.H. from the outer part of the fleece towards the skin. The temperatures at various distances from the skin of the sheep was thus of importance as well as theR.H.

Some methods of measuring R.H. are briefly considered below:-

1) Movements in hygroscopic materials. Those most commonly used are hair, paper, a paper + silver foil strip and wood. Usually the movements of the material are magnified by a lever system. The bending movement of a wood shaving with R.H. change was examined in the laboratory. It was found that the method could give accurate results, and could be made on a small enough scale to insert into a sheep's fleece without causing excessive disturbance. Also it could give a continuous record of R.H. changes over a period of a few hours at least. The disadvantages of the method were too great, since the apparatus needed frequent recalibration, and was too fragile for daily use among live sheep.

2) Determination of moisture content of a hygroscopic material. The materials usually employed are paper, hair or wool. The material to be used, e.g. a piece of paper of a particular quality and size is weighed at various R.H's and a curve of weight change against R.H. compiled. Slade (1933) employed paper hygrometers in this way, and the use of weighing hair-hygrometers was popularized by Buxton (1931). Paper hygrometers were also used by Macleod (1940) for fleece

readings. The method has several advantages among which are the accuracy possible if used carefully, and the fact that any change in weight of the paper used due to picking up foreign material while being exposed to the unknown R.H., can be measured by weighing the paper both before and after the exposure in a known fixed R.H. For the present work the method was found to be too laborious, and unsuited to obtaining large numbers of readings on several sheep during one day. Also the slight hysteresis effect obtained with paper and other organic materials may introduce some error. An important objection to the method is the relatively large size (see Macleod 1940) of the papers placed in the fleeces, so that some disturbance of conditions is likely. In addition, the method does not permit measurements of humidity gradients.

3) Wet and dry bulb-thermometers. A current of air is required past the bulbs so that disturbance of conditions is implied, and evaporation for the wet bulb itself alters the R.H. in a closed space. The method can only be used for macroclimatic R.H. measurements.

4) Dew-point determinations. The possibilities of this method for measuring fleece R.H. were investigated with an apparatus using a 5 cc air sample. Since the writer was mainly interested in the R.H. of the air near the skin surface and at various known distances from the skin, the method was abandoned since it was impossible to be certain whether all the air in one sample came from one particular layer of the fleece atmosphere.

5) Electrical conductivity of salt impregnated fabric. This method involves the exposing of a small 'element', essentially composed of a salt impregnated fabric, to the unknown R.H. The fabric is wired to a sensitive galvanometer, and the conductivity of the element measured. The conductivity of the element varies with the degree of hydration of the

salt on the fabric which will in turn vary with R.H. The method is potentially very accurate. The disadvantages are that the apparatus is expensive and delicate, and needs a mains electric supply for its operation. For the purposes of the present work, the method was precluded, since the 'element' would be very liable to contamination with dirt in fleeces. In addition, such factors as the CO_2 concentration in the fleece atmosphere might affect the conductivity of the fabric, apart from R.H.

6) Chemical hygrometers. These involve the absorbtion of the water-vapour from the air sample, usually by means of pure sulphuric acid, and measuring the contraction in volume of the sample (Rideal and Hanna, 1915). The apparatus was considered too delicate for field use, and unsuited for large numbers of rapid readings.

7) Colour changes of cobalt salts. This method has been modified for microclimatic humidity determinations by Solomon (1945). The salt most commonly used is cobalt chloride, and was alone employed in the work carried out in 1946. The procedure adopted in making and employing cobalt chloride papers is described on page 98-101. Here, the main attributes of the method will be considered. Paper impregnated with cobalt chloride gives a range of colours, from intense blues at 0-30% R.H, through various shades of blue-lilac, and pink-lilac at 30-65% R.H., and pink shades from 65-95%. The method depends on the matching of the colours of the exposed papers with those of standard papers previously exposed to a series of known humidities and maintained at the colour they had attained, by sealing them in a cell filled with liquid paraffin. Owing to the nature of the colour range, it is only between 40 and 70% R.H. that an accurate matching of the papers can be made. Less accurate matching can be made between 30 and 40% R.H.

and 70 and 80% R.H. For the present work the range 40-70% R.H. happens to be the most important part of the R.H. scale, since these humidities predominate in sheep fleeces. The advantages of the method are that it is easy to employ in the field, and can be used to obtain large numbers of readings simultaneously by placing papers at several points in the fleeces of several sheep. It can be used to obtain R.H. readings at various fixed points in fleeces for several consecutive days. The papers are small and can be inserted into the fleece with the minimum of disturbance of fleece conditions. The main disadvantages are that the papers have to be left in the fleece for 30 mins. without any movement, and the accuracy of the readings within the 40-70% R.H. range is to the nearest 2%. The fact that free moisture washes the cobalt salt off the paper is not a serious disadvantage.

(ii) Method used in present investigation

The cobalt chloride paper method was that chosen for measuring fleece atmosphere humidity, in the field work carried out in the summer of 1946. Papers for use in the field and standards for comparison were made according to the methods described by Solomon (1945). Standard papers and the papers placed in fleeces were of Whatman No. 1. filter paper, impregnated in a 25% $^{w}/_{v}$ solution of Analar cobalt chloride ($CoCl_2.6H_2O$). The circular filter paper sheets were immersed for 1 min. at room temperature in the above solution, dried between two changes of desiccator-dried filter paper with a squeegee. They were then hung up and allowed to dry at room humidity and stored in desiccators over phosphorus pentoxide, in the dark. Standards were prepared by exposing pieces of cobalt chloride paper 30 x 10 mm. in size to constant humidities of from 30 to 90% R.H. at intervals of 5%, at 30°C in an electric incubator for 30 min. The constant humidities

were maintained by sulphuric acid/water mixtures (Wilson 1921) placed in honey jars, while the papers were hung at a standard distance from the surface of the acid by means of a hook attached to the underside of the tight fitting metal lids. Since the standards were prepared by exposure to constant humidity for 30 min. papers used in field determinations of fleece humidity were left in position for the same period. The temperature of 30°C for the exposure of the standards was chosen as being about half way between the temperature at the skin surface (about 37°C) and at 2-3 cm. off the skin (probably about 25°C but variable). The comparison of the colours of the field papers with the standards was always carried out at room temperature, so that both standards and field papers were at the same temperature (16-18°C). Thus the papers were being compared at a temperature considerably below the temperature of the air to which they had been exposed and with which they had gained moisture equilibrium. No correction for the slight effect of temperature on the colours of cobalt chloride papers was necessary, since standards and field papers were compared at the same temperature.

Papers used in the field were of various sizes from 1 x 1 - 1 x 4 cm. They were numbered before use so that the precise position on a particular sheep where the paper had been placed was known. The papers were carried to the sheep in dry stoppered tubes, so that they were always a brilliant blue colour when they went into the fleeces. In the case of the strips (1 x 2 - 1 x 4 cm) they were carefully inserted end ways into the fleece, so that one end of the paper rested against the skin, and the paper lay along the wool fibres. The position was marked by loosely tying the tips of the wool staples with string, and care was taken to cause as little disturbance as possible to fleece structure. Papers were

always handled with an entomological pinning forceps. On removal from the sheep they were quickly transferred to a bottle of pure medicinal paraffin, which preserved the colour they had attained in the fleece. Comparison with standards was made in the laboratory under conditions of fairly constant illumination, usually within 6 hr. of removing the papers from the fleeces. It was found that papers could be kept in paraffin in 3 x 1" tubes for about a week without appreciable change in colour.

The range of accurate readings has already been mentioned (page 97). At R.H's of 30-40% no estimate of the precise humidity was made, and papers giving such readings were classed as 'below 40% R.H.', and are represented on subsequent figures as 35%. When papers were exposed to air well below 40% (eg. 15%) their intense blue colour could be distinguished from that of the 30-40% range. Papers giving this intense blue were thus classified as 'below 30'. Such readings were encountered under winter conditions only (Table 18). At humidities of over 70% owing to the small differences between the pink colours, readings could be made only to the nearest 5% R.H. In the tabulated results of fleece humidity readings (Appendix page 183-227), readings from 80-95% are classified as 'over 80'. A humidity of 100% could be recognized as such by the pronounced leaching of salt from the paper, giving it a much paler colour.

At any early stage in the field work it was found that cobalt strips frequently apparently registered very steep humidity gradients between the skin surface and points 2 - 4 cm. off the skin. In order to test whether the strips were reliable for registering such humidity gradients, laboratory experiments were carried out as follows:- Conditions on sheep wet with rain were simulated by sticking a large piece of fleece, 5 cm. thick, on to a cardboard sheet. The base of the fleece

next to the cardboard was maintained dry by packing it with silica gel, while the free wool tips were maintained moist by spraying with water. In this way a steep R.H. gradient in the fleece atmosphere, of below 40% at the base to almost 100% R.H. at the wool tips, was maintained. Cobalt chloride strips measuring 1 x 3 cm. were placed in this gradient, and the readings at the ends of each paper recorded after various intervals. It was found that very little 'creep' of water along the papers occurred with 30 min. - the time employed in the field readings. After 80 min. appreciable 'creep' had occurred, but it was insufficient to affect the colour at the extreme 'dry' end of the paper strips.

The positions at which papers were placed in fleeces were standardized as much as possible. Details of these standard positions are given in the Appendix (page 179). The main positions employed may be listed here:- Withers; midback; tailhead; right and left crutch; right and left flank; right and left belly. Papers were placed at other positions when such readings were likely to be of interest. In practice it was found that the papers could be placed in very nearly the same position on each sheep on successive days, so that standardization of positions was achieved to a considerable degree.

In the course of the 1946 work, readings by cobalt-chloride papers were obtained for twenty eight sheep. Daily readings over periods of several days were obtained for 9 sheep - 5 at Houghall Farm, Durham, and 4 at Crag Farm, Ravenglass, Cumberland. Details of all sheep employed will be found in the Appendix (page 180-1).

(iii) Comparison of the method with those previously used.

A series of parallel fleece humidity readings using cobalt-chloride papers and the wool weighing technique of Davies & Hobson (1935) were made on three sheep under summer conditions.

A further series of triple comparative readings using the present method, the wool method and the paper hygrometer technique as used by Macleod (1940), were made on three sheep under winter conditions in December.

The Davies & Hobson (1935) method involved the removal of wool samples from the basal 2 cm. next to the skin, conveying them in stoppered tubes for weighing, and by successive weighings after exposure to various humidities the R.H. at which the samples were in equilibrium before removal from fleeces were determined. This figure depended on the assumption that the relationship between weight and humidity is linear. The authors found that this was so over a certain R.H. range and gave the curve of weight of a wool sample at different R.H's (Davies and Hobson, 1935, page 284, Fig.1). This curve showed a slight hysteresis effect, in that the weights of the sample with rising and falling R.H. did not exactly coincide.

Macleod's paper hygrometer method. Macleod states that he used a single batch of good quality writing paper. After many trials with different types of paper the writer selected a writing paper with high hygroscopicity. But even in this paper the weight differences were considered to be smaller than would be desired. Pieces of this paper weighing about 400 mg. were stored at 30% R.H. 30°C. and always brought to the unknown humidity from this fixed starting point. Cylinders of wire gauge about 4 x ½in. closed at one end were employed for keeping the papers in the fleece. The papers were transferred from the standard 30% R.H. 30°C., system to tightly corked test tubes, in scrolls of about 1½ turns. In the field they were quickly transferred to the gauze cylinders, which were placed against the skin in a natural 'shed' of the wool. After a fixed period of 1-2 hr the end of each gauze cylinder was exposed and the scroll

transferred by forceps quickly into a tightly corked test-tube and brought to the laboratory. Weighings were made on a torsion balance (500 mg./0.5 mg.). After weighing each paper was exposed for 24 hr. at 30% R.H. and again weighed as a check for alteration of weight by soiling or other cause.

In carrying out the comparative readings with cobalt chloride papers, the wool weighing method of Davies & Hobson and with paper hygrometers as used by Macleod, care was taken to adhere to the technical details of the methods as given by these authors in their papers.

The comparative readings with cobalt chloride papers and the wool weighing method obtained under summer conditions, are given in Table 17. This table shows several points of interest. The humidity readings obtained by basal wool

Table 17. *Comparative humidity readings (summer conditions)*

Wool weighing method (samples of basal 2 cm.) (% R.H.)	Readings by cobalt-chloride papers (% R.H.)				Remarks
	At time of sampling		6–12 hr. before sampling		
	Skin surface	2 cm. off skin	Skin surface	2 cm. off skin	
Series 1: 100*	100	100	90+	100	Wool samples taken during long rainy period, when sheep were continuously wet
86	50	70	50	70	
91	—	—	100	100	
74	—	—	51	51	
Series 2: 100*	100	100	100	100	Samples taken 1 day after preceding set. Sheep still wet
78	67	75	70	72	
97*	100	100	100	100	
74	56	67	60	72	
Series 3: 75	56	63	50	65	Samples taken 5 hr. after preceding set. Rain ceased and sheep almost dry
79	65	68	75	75	
86	55	75	58	75	
76	55	65	55	67	
Series 4: 65	40	45	56	63	Samples taken 24 hr. after preceding set. Dry weather
64	42	42	—	—	
70	43	43	55	65	
Series 5: 62	46	46	—	—	Samples taken 3 days after preceding set. Sheep dry. Weather in intervening period dry
50	Below 40	40	—	—	
50	42	45	—	—	

* Samples contained free water. Rain had penetrated to basal 2 cm. of the fleece.

sample weighings in Table 17 (col. 1) show that their water content was consistently higher than would be expected on the basis of the fleece readings by cobalt-chloride papers. For example, in the second reading of Series 1 (Table 17) the wool weighing technique gave a reading of 86% R.H. Cobalt-

chloride paper readings taken at this site both immediately before sampling and 6-12 hr. before sampling showed a humidity of 50% R.H. close to the skin with an ascending gradient to 70% R.H. at 2 cm. off the skin at both times. The water content of the wool samples would appear to have been higher than would be expected on the basis of fleece R.H. readings by the cobalt chloride method. The basal wool, it seems, was not in equilibrium with the basal fleece atmosphere. Further readings by the wool method (Table 20) showed that this condition apparently existed in winter too. This phenomenon was also found by Macleod when he compared the results of wool calibrations with those of paper hygrometers. He suggested that the positive discrepancy in results of the former technique was probably due to continuous skin gland secretion (possibly yolk secretion). However this may be, it seems that the wool weighing technique is subject to such positive errors that it cannot legitimately be used to assess the suitability of fleece humidity for blowfly strike near the skin.

Wool weighings (Table 17, col.1) gave higher readings when the sheep were wet than when they were dry. The readings are grouped in series each of which was taken 5 hr. to 3 days after the preceding series, from the same individual sheep (nos. 1, 2, 26). It will be seen from the table that the readings in series 1 and 2 taken when the sheep had been continuously wet during a rainy period lasting several days, are much higher than those in series 4 and 5 taken when the sheep were dry, after the end of the rainy spell. The lower readings obtained in series 4 and 5 however still higher than the corresponding cobalt chloride paper readings. In series 1 and 2, free rain water had penetrated to the basal 2 cm. of the fleeces in only 3 of the 8 samples, so that this factor alone could not account for the higher readings in these series. It appears therefore that rain on the

outsides of the fleeces although not penetrating to the basal 2 cm. did increase the moisture content of the basal fleece during the wet period. This phenomenon was also described by Macleod in his paper, and he suggested the possibility of some vapour diffusion process at work along the lengths of the wool fibres.

In the present work it will be shown that considerable humidity changes can occur in the basal fleece atmosphere within periods as short as 1 - 3 hr; therefore the time taken by pieces of fleece to regain equilibrium with the fleece atmosphere after a sudden humidity change is of practical importance if the wool weighing method is to be used. In the laboratory it was found that unwashed naked fleece samples, about 400 mg. in weight, transferred from 0 to 95% R.H. at 37°C required well over 3 hr. to regain complete equilibrium in the higher humidity, while cobalt-chloride strips showed that the air entangled in the fleece samples reached the new humidity (95% R.H.) within 1 hr. This considerable lag would invalidate results by the wool calibration method during showery weather when the basal fleece humidities may fluctuate rapidly.

The triple comparative humidity readings obtained under winter conditions are given in Table 18. The higher humidity readings obtained by the wool method than with the

Table 18 (a) *Comparative humidity readings (winter conditions)* (% R.H.)

Sheep no.	Cobalt-chloride paper method	Paper hygrometer method (Macleod, 1940)	Wool sample method (Davies & Hobson, 1935)
1	Below 30 (30)	19	50
1	Below 30 (40)	26	48
2	40 (58)	50	67
2	Below 30 (42)	21	55
2	Below 30 (40)	22	—
3	42 (42)	40	50
1	Below 30 (40)	16	—

The numbers in brackets refer to the readings 3 cm. off the skin.

other two is again apparent. Although cobalt-chloride papers cannot be used with accuracy below 40% R.H, Table 18 shows fairly close agreement between R.H. measurements by this method and those by paper hygrometers, at identical sites in the fleeces. Cobalt-chloride papers can register

humidity gradients while paper hygrometers cannot. For example, in Table 18, it will be seen in the third set of readings (sheep 2) that cobalt-chloride papers indicated an R.H. gradient in the fleece atmosphere, rising sharply from 40% near the skin to 58% R.H. at 3 cm. off the skin. A paper hygrometer placed as close as possible to the skin at this point in a natural shed of the fleece, failed to give any indication of this gradient but showed an intermediate R.H. - namely 50%.

(iv) Fleece humidity fluctuations as measured by cobalt-chloride papers

In the following pages, unless otherwise stated, the terms 'fleece R.H.' and 'basal fleece R.H.' refer to the R.H. of the fleece atmosphere within 1 cm. of the skin of the sheep. The following figures in this section refer to the R.H. at this position unless otherwise indicated. Only specimen results of the cobalt-chloride fleece readings are discussed to illustrate the main features of fleece R.H. fluctuations found. The full results are tabulated in the Appendix (page 180 et seq.).

Sheep nos. 1-3 and 7, the fleece R.H. readings of which are graphed in Figs. 21-3, were at Houghall Farm, Durham, and should be considered in the light of the meteorological data taken at this farm and summarised in Fig. 19. The conditions under which the sheep were kept at Houghall were typical for grass sheep on a lowland farm in northern England. Sheep nos. 4 and 5 were at Crag Farm, Cumberland, and were hill sheep kept under conditions typical for the lower fields of upland farms, where the pasture is of a poor type, poorly drained, and with considerable bracken growth, and with heavier rainfall than at Houghall.

The time of day at which particular fleece readings were made is given as G.M.T.

Sheep 4 and 5 (Fig.20). Observations on these sheep were

made during cool summer weather. It will be seen that the fleece R.H. near the skin at the midback of sheep 4 was below 40% during most of the period; this is shown in Fig.20 as 35%. It will be seen that when these sheep were wet after rain (e.g. 6, 8 Aug.) the fleece R.H. was appreciably raised, especially in sheep 5, in which after heavy rain it frequently reached 60-80%. The fleece of this sheep (5)- a Herdwick wether - was short and coarse (see Appendix page 180) and penetration of rain drops to the basal cm. of the fleece along

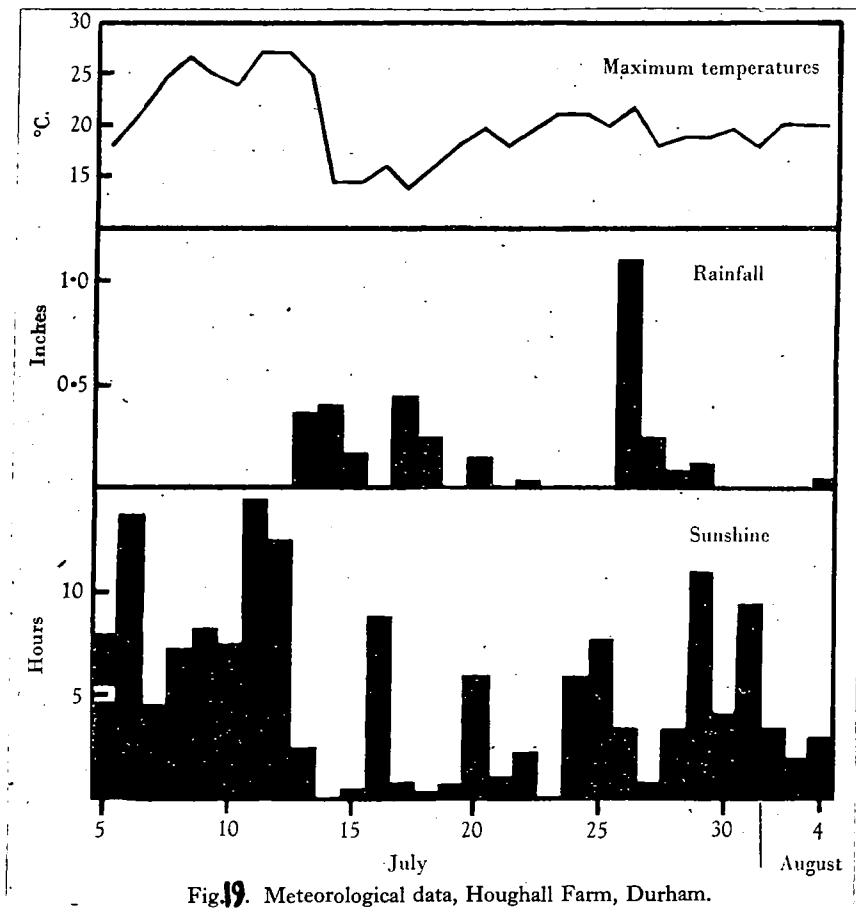

Fig.19. Meteorological data, Houghall Farm, Durham.

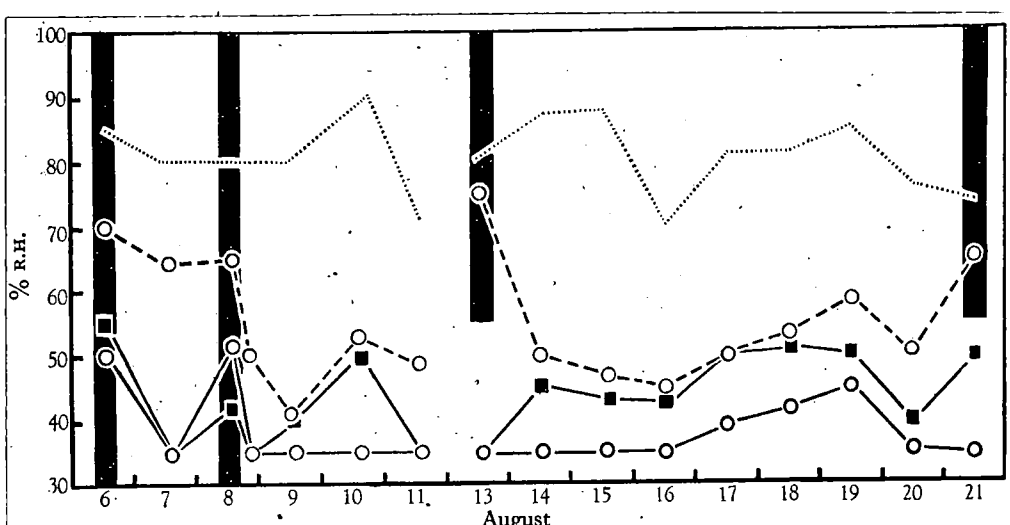

Fig.20 Fleece atmosphere R.H. variations in sheep 4 and 5. Black columns indicate when the particular sheep was wet with rain, in this and subsequent figures. Sheep 4 ———; sheep 5 - - - - - -; macroclimatic R.H. (read by a sling psychrometer at the same times as the fleece readings were taken) ·····; ○, midback position; ■, right crutch position.

the back and flanks was the rule rather than the exception. That the effect of rain on the basal fleece R.H. was of short duration is shown on 8 Aug. where two sets of readings were made - one at 09.00 hr. when the sheep were wet after night rain, and another at 14.30 hr. when the sheep had dried. The readings showed that after the rain dried from the outsides of the fleeces, the R.H. near the skin had fallen rapidly in all cases. The general picture of fleece humidities in sheep 4 (Fig.20) is one of low R.H. raised for short periods by the effect of rain and dew, and agrees with the accounts given by Macleod (1940), who used mainly Down Cross lambs. The same figure, however, shows other features. In sheep 4 the R.H. near the skin at the right crutch was, during most of the period, significantly higher than over the rest of the body, although the fleece was not contaminated with any dung or urine. By estimating the percentage development of freshly laid L.sericata egg batches, placed in various parts of the fleece of this sheep, it was shown that only in the crutch was the humidity suitable for blowfly egg development; other parts were too dry (see page 116). This higher R.H. in the crutch region seems to have been due to more rapid skin secretory activity in that region compared with the rest of the body surface, since cobalt-chloride strips in the former position frequently showed a higher R.H. at the skin surface that at 2 cm. up the wool staple. Other experiments on sheep 4 also pointed to the same conclusion (page 144-5).

Comparing fleece R.H. readings for sheep 4 with those for sheep 5 (Fig.20) it will be seen that the fleece of the latter sheep was consistently more humid. In sheep 5, only 37% of the ninety-three readings taken were at 50% R.H. or below that level, compared with 79% in sheep 4 (ninety-four readings). These differences may be due to greater skin secretory activity in sheep 5. This view was confirmed by the fact that in this

sheep, when the fleece was not wet with rain, the R.H. was frequently higher near the skin surface than at points farther out in the fleece; this occurred less frequently in sheep 4.

Sheep 2 and 3 (Fig.21). These were two short-fleeced Swaledale x Border Leicester ('Mule') lambs. Humidities are again seen to be below 40% R.H. when the sheep were dry, but rain on the outside of their fleeces raised the basal humidities considerably, e.g. 22 July. Since their fleeces were short, rain was frequently found to penetrate to the basal cm. of the fleece, and sometimes to the skin along their backs. On

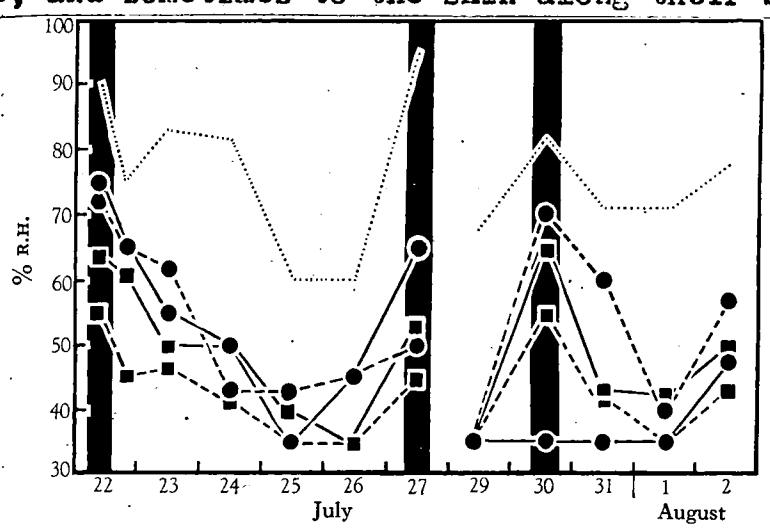

Fig. 21. Fleece atmosphere R.H. variations in sheep 2 and 3. Sheep 2 ———; sheep 3 - - - - -; ●, withers; other symbols as in Fig. 2.

their flanks, the wool locks formed an efficient 'roof-tile' system whereby the rain was turned off their bodies. These two lambs, however, did show idiosyncrasies in fleece R.H., possibly correlated with differences in their fleeces (see Appendix page 180). After rain, the fleece R.H. tended to be higher in the close-fleeced sheep 2 than in the slack-fleeced sheep 3 (Fig.21: 22, 29, 30 July). Macleod (1940) claims to have shown that rain penetrates farther into a close than into a slack fleece. It was noticed in the field that the slack fleece of sheep 3, after rain had ceased, always became dry to the touch in a shorter time than did that of sheep 2. These observations tend to support the contention of Macleod. It is also to be expected that drying processes within the thickness of the fleece would be more rapid in a slack fleece.

Fig.21 indicates that the fleece humidities of these

short-fleeced lambs tended to follow fluctuations of macroclimatic R.H., apart from the direct effect of rain drops in their fleeces.

Suggestion of a diurnal rhythum of fleece R.H., which would be well developed in hot weather with heavy dew or rain during the night, is afforded by the data in Fig.21 (22 July) when humidity readings were taken in the morning, after night rain, and again in late afternoon (cf. Fig.20, 8 Aug., when a similar result was obtained).

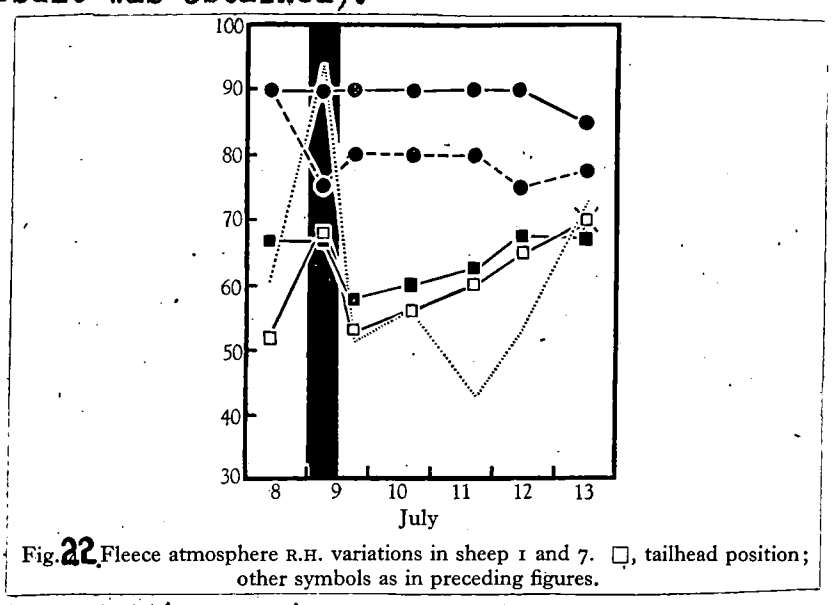

Fig.22. Fleece atmosphere R.H. variations in sheep 1 and 7. □, tailhead position; other symbols as in preceding figures.

Sheep 1 and 7 (Fig.22). The picture of fleece R.H. in these two Down Cross lambs differs markedly from that of the sheep hitherto considered. Humidities in the fleeces of these sheep from 8 to 16 July were above 60% and frequently above 85% R.H. For the most part high R.H's were confined to the basal 1 cm. or so next to the skin, and cobalt-chloride paper strips indicated very steep humidity gradients, from 60 to 90% R.H. near the skin, to below 40% R.H. 3 cm. up the wool staples. To confirm the existence of a high humidity near the skin, on 13 July a batch of laboratory laid L.sericata eggs, which had been incubated to within half an hour of hatching, was placed in the fleece, at the withers of sheep 1. Half of the batch was placed at the base of the fleece against the skin, and the other 3 cm. away from the skin. The eggs placed near the skin hatched immediately, and a strike was established at that spot without any addition of moisture;

those eggs 3 cm. from the skin did not hatch. This shows that the humidity near the skin was at least 80% R.H., and nearer the outer surface of the fleece very much lower. These two lambs (sheep 1 and 7) were healthy and undipped. Drops of yellow-stained watery material were observed on wool fibres near the skin, and were wet to the touch. When cobalt-chloride strips come into contact with them, they produced a characteristic decolorization in which the cobalt salt was washed up the paper, leaving a colourless edge. This phenomenon was similar to that produced by contact with water droplets. Contact with greasy material, causing a translucent stain on the papers, was much rarer.

Since sheep 1 and 7 were sweating to an extent sufficient to maintain suitable humidities for the development and survival of blowfly eggs and young larvae during the period 8-16 July (Fig.22), the limiting factor for strike on these

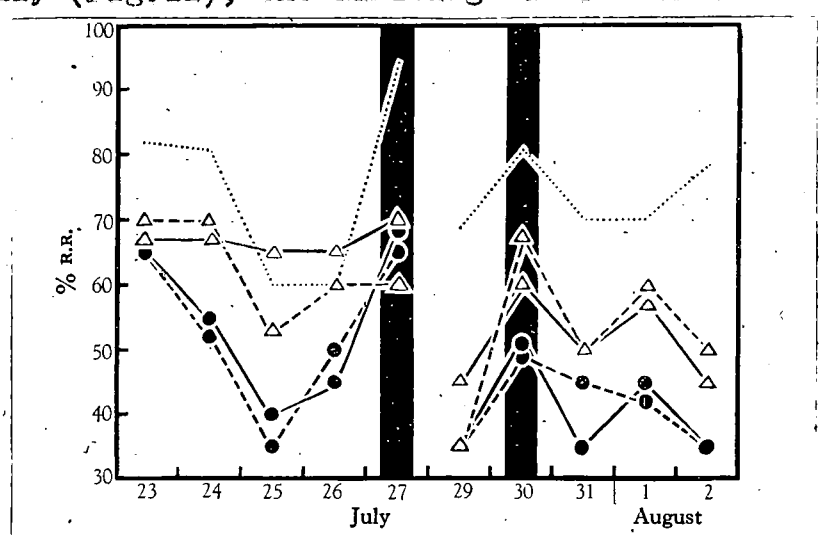

Fig.23. Fleece atmosphere variations at a later period in sheep 1 and 7. △, right flank position; other symbols as in preceding figures.

sheep must have been that they did not attract blowflies to oviposit on them. The weather during that week was exceptionally hot and dry (Fig. 19). Sweating, as measured by a high humidity close to the skin, was particularly rapid in the withers region in both sheep (Fig.22), and it appeared to spread to other parts of the body as the hot weather continued.

From Fig.19 it will be seen that the weather after 13 July became cooler and wetter. Sheep 1 and 7 were dipped

in a D.D.T. emulsion on the morning of 18 July and daily fleece readings recommenced on 23 July; some of the results are graphed in Fig. 23. Comparing Figs. 22 and 23 it will be seen that in the second period their fleeces were considerably drier, and quite in keeping with Figs. 20 and 21. In both sheep, however, during the period 23-27 July (Fig.23), humidities were still high in the flank position. It is concluded that sweating had diminished over most of the body surface, but was still sufficiently rapid on the flanks to maintain high R.H's close to the skin. Additional daily humidity readings taken in dry weather showed that rapid sweating frequently occurred over large areas of the flanks of both sheep at this time. Fig. 24 shows that these flank sweat-areas were not constant, but varied from day to day.

Some indication of the relationship of fleece atomsphere R.H. to macroclimatic R.H. is given in Fig. 25, where humidity readings at the skin surface are compared with those at 3 cm. off the skin, and with the macroclimatic R.H., read by a sling psychrometer at the time the fleece readings were taken. The fleece readings refer to the right flank position of sheep 1 during a period when sweating was occurring in that region. From Fig. 25 the following will be noted:

(1) Quite apart from the direct effect of rain, the fleece R.H. at 3 cm. off the skin tended to follow variations in macroclimatic R.H. (e.g. 31 July-2 Aug.).

(2) The skin surface R.H. bore no relation to macroclimatic R.H.

(3) During rapid sweating the fleece R.H. was found to be highest close to the skin: this represents a reversal of the gradient to be expected on grounds of thermal stratification of the fleece atmosphere (e.g. 24-

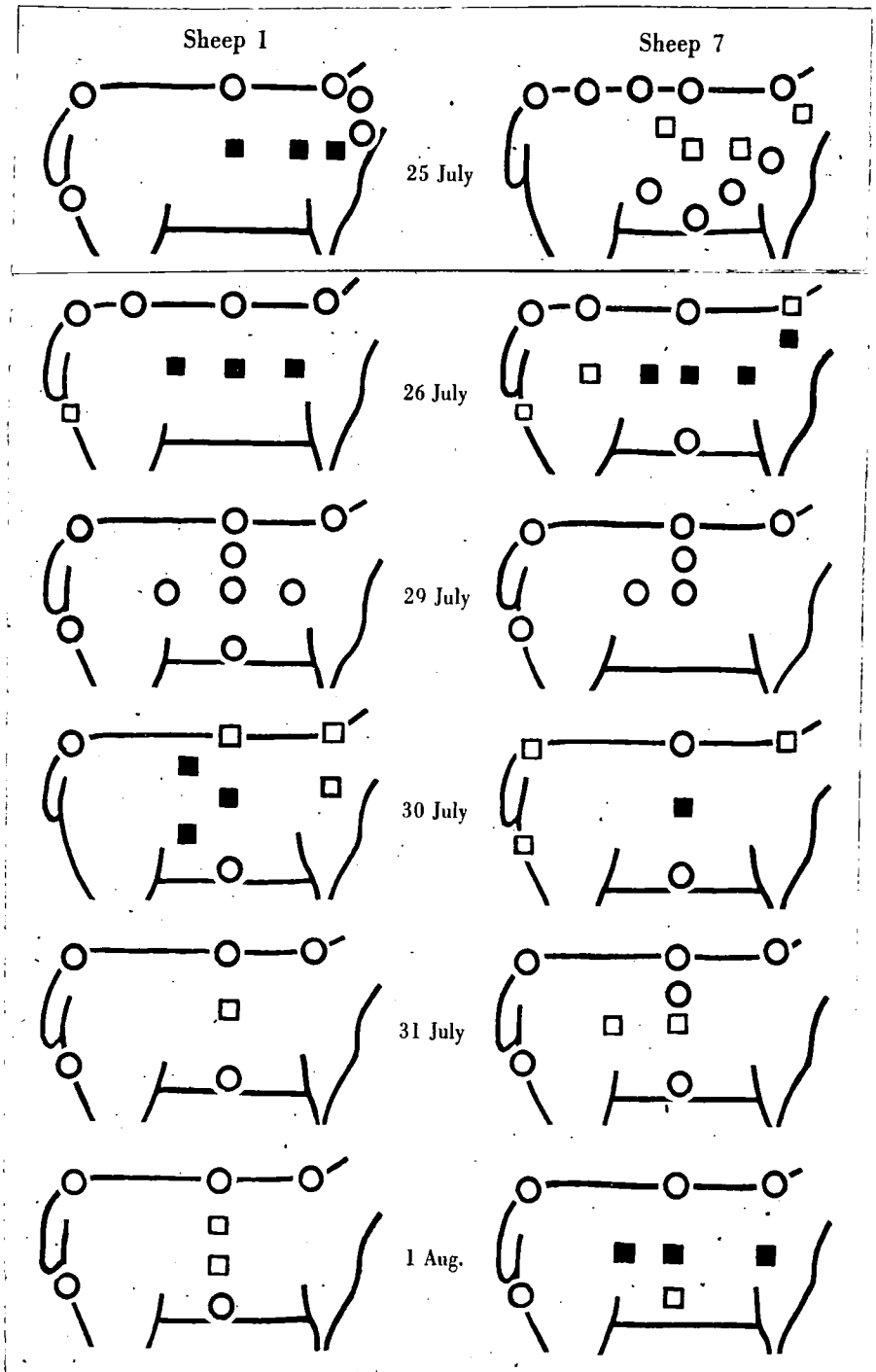

Fig. 24. Fleece humidity readings, sheep 1 and 7. ○, below 50% R.H.; □, 50–59% R.H.; ■, over 60% R.H.

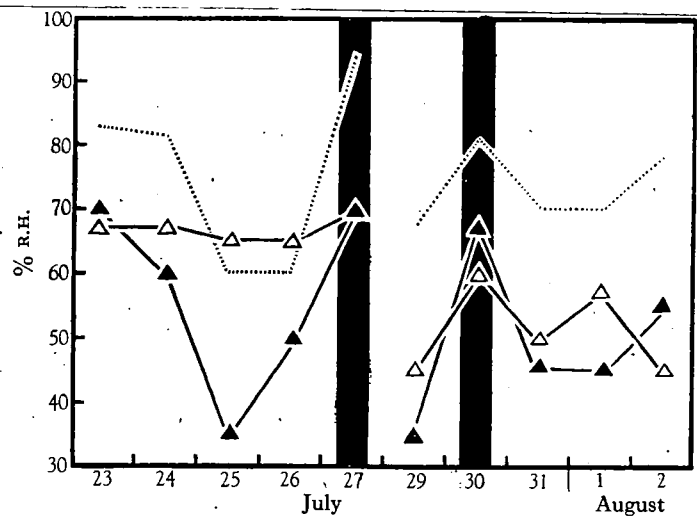

Fig. 25. Effect of sweating and macroclimatic R.H. on humidity within the fleece. △, right flank position, at skin surface; ▲, right flank position, 3 cm. off the skin; ..... macroclimatic R.H.

26 July, 31 July- 1 Aug.).

(4) The R.H. gradient, due to sweating, may be abolished (27 July) or its direction may be reversed (30 July) by heavy rain.

It will be shown (Part IV) that increased sweating can be produced, in some sheep, by causing them to run. In four such running experiments on one sheep, sweating was detected at the withers in each case, but never at the midback. Since the withers and midback positions were only about 30 cm. apart, sweating would thus appear to be sometimes sharply restricted to particular parts of the body surface. This fact may account for some strikes on clean wool, otherwise difficult to explain. Examples of such strikes, occurring at the same spot on one sheep in successive years, are quoted by Macleod (1943). The following observations show that factors other than muscular exertion also influence the rate of sweating. Fleece humidity readings were taken on sheep 10, a Swaledale Ram lamb, at 09.30 hr. on 20 Aug.; these readings are given in Table 19. The higher humidities next the skin indicate that rapid sweating was occurring, although the sheep had not been running nor was it excited abnormally during the period of the observations. Readings at the same positions on this sheep were carried out 3 hr. later under similar weather conditions. They showed that the fleece atmosphere throughout the thickness of the fleece was below 40% R.H., indicating that sweating had ceased, or at least was proceeding at a slower rate. Simultaneous observations on sheep 4 showed no such variations, although both sheep were kept together and treated identically. It is evident that skin secretory activity in sheep may be governed by factors, other than muscular activity, and which are probably not directly dependent on external conditions such as atmospheric humidity or air temperature.

Table 19 *Sheep* 10. % R.H. *at* 09.30 hr., 20 August

	Position in fleece				
	Withers	Midback	Tailhead	Right crutch	Right belly
Skin surface 3 cm. off the skin	62 Below 40	60 Below 40	51 Below 40	50 50	Below 40 45

The above observations also show that moisture at the base of even a thick, close fleece, is very rapidly evaporated, and that to maintain high humidities at its base, suitable for blowfly eggs to develop, a continuous supply of moisture is needed.

(v) The fate of freshly laid L.sericata egg batches placed in sheep fleeces

Egg batches, freshly laid by laboratory bred flies, were placed in various positions on the sheep mentioned in the preceding section. A small number of eggs of each batch were set aside in saturated air as controls. Batches were removed from the sheep after 12-16 hr., the constituent eggs teased out after wetting, and the proportion of eggs containing fully developed larvae estimated (Table 20).

The humidity readings in Table 20(b), col.4, were taken 6-12 hr. after the end of the incubation period of the eggs in question. Thus, they do not necessarily represent the humidity conditions to which the eggs were subjected during development.

Table 20 *Development of eggs in fleeces*

Eggs were placed on the skin of the sheep unless otherwise stated.

(*a*) *Sheep at Houghall Farm, Durham*

Sheep no.	Position of eggs	Cobalt-chloride paper readings at site of egg placement (% R.H.)		Estimated percentage of eggs completing development
		Readings taken 1–5 hr. before eggs were placed in the fleece	Readings taken at time eggs were removed from the fleece	
2	Withers	Below 40	48	>50
	Withers	Below 40	47	0
3	Withers	60	55	>50
	Tailhead	Below 40	Below 40	<50
	Withers	45	56	100
	Tailhead	Below 40	Below 40	<50
1	Rump	Below 40	—	0
6	Right thigh	Below 40	—	0
	Rump	Below 40	—	0
7	Right flank	51	60	<50
	Right flank	62	50	<50
	Right flank	Below 40	55	<50

Table 20. (*b*) *Sheep at Crag Farm, Ravenglass*

Sheep no.	Position of eggs	Cobalt-chloride paper readings at site of egg placement (% R.H.)		Estimated percentage of eggs completing development
		Readings taken at time eggs were placed in the fleece	Readings taken 6–12 hr. after end of incubation period	
4	Withers	Below 40	Below 40	0
	Withers	Below 40	Below 40	<50
	Midback	45	Below 40	0
	Midback	Below 40	Below 40	0
	Midback	70	43	<50
	Right crutch	50	—	<50
	Right crutch (4 cm. off skin)	66	—	>50
	Right crutch	50	—	100
	Right crutch	50	50	>50
5	Right flank	57	63	<50
	Right crutch	45	57	0
	Withers	75	75	100
	Midback	46	67	0
	Right belly	60	69	100
	Withers	63	—	0
	Withers (3 cm. from skin)	Below 40	—	0
	Withers	66	70	<50
	Midback	55	68	>50
8	Left flank	74	75	100
	Right flank	68	62	100
	Right flank	55	—	100
	Withers	70	100	0
	Left flank	73	Below 40	>50
9	Withers	Below 40	60	0
10	Withers	56	61	<50
	Withers (3 cm. from skin)	Below 40	70	<50
	Midback (3 cm. from skin)	—	64	100
	Right flank	57	50	100
	Right crutch	50	—	100
	Withers	61	Below 40	0

In general, the results in Table 20 confirmed laboratory experiments on the L.sericata egg, and emphasized the reliability of readings of fleece humidity by cobalt-chloride papers. Using the latter technique, it was shown for sheep 4, that humidities in the crutch region were higher than in most other parts of the fleece (Fig.20, Appendix page 203-219). The humidity in the crutch often approached 50% R.H. - the minimum humidity for the successful development of eggs at 37°C. These results were confirmed by placing eggs in various parts of the fleece of this sheep (Table 20(b)). Of five batches placed in the midback and withers regions, in three, all eggs failed to complete development, whilst in the other two only 10% reached the prehatching stage. Of four batches placed in the right crutch, three showed practically 100% completion of development, whilst 20% of the remaining 4th batch reached the same stage. Thus only the crutch region of the sheep was sufficiently humid for the

development of L.sericata eggs.

Altogether 42 egg batches were placed in the fleeces of 9 sheep; in 16 batches more than 50% of the eggs completed development; in 12 between 1 and 50% of the eggs reached the same stage; whilst in the remaining 14 batches all eggs died through desiccation at an early stage in development. In a few batches, several eggs hatched whilst in the fleece. Of the batches in which more than 50% of the eggs completed development, some were taken out of the fleece after 10-12 hr. and placed in saturated air; from these many larvae hatched within a few minutes.

It is clearly evident from the above experiments, that humidity conditions in sheep fleeces were sometimes maintained at a level sufficient for L.sericata eggs to complete development. Humidities high enough for hatching, however occurred less frequently.

II. WORK IN 1947

Field work in the summer of 1947 was confined to obtaining readings of fleece R.H. for sheep nos.29-42. Methods were exactly similar to those in 1946 (page 99-100), and the full results will be found in the Appendix page 225-227. The readings were carried out in June, during moderately warm, sunny weather using Border Leicester x Half bred cross lambs, about 6 months old (Appendix page 225-227).

The readings obtained throw some light on the comparative frequency of sweating at the various standard positions in the fleeces where readings were taken. As before, the incidence of a higher humidity at the 'skin' end of the cobalt-chloride paper strip, than at the other end 2-3cm. off the skin, was adopted as the criterion of sweating at the time the readings were taken. An analysis of the

incidence of such 'sweat' readings is given in Table 21.

TABLE 21

Frequency of 'sweat' readings at various points in fleeces, Sheep nos. 29-42. June 1947.

Position	Total No. of readings.	No. of readings showing sweating	% readings showing sweating
Withers	39	15	38.4
Midback	19	5	26.3
Tailhead	39	6	15.3
R. flank	16	2	12.5
L. flank	14	3	21.4
R. crutch	8	0	0.0
Totals	135	31	22.9

The figure of 38.4% of readings showing sweating at the withers is the highest of the positions studied. This re-emphasizes the conclusion reached during the 1946 work (page 111) that sweating was often most marked at the withers, which is a position known to be the most susceptible to blowfly strike, considering its small area, outside the breech region (Macleod, 1943). In Table 21, the next most frequent site of sweating was the midback position, again a region where 'body strikes' are comparatively frequent. The number of readings for this site (19) is however too small to base a definite conclusion on the results.

III WORK IN 1948

From July to Sept. 1948, laboratory laid egg batches of the 5 blowfly species other than L. sericata were placed in sheep fleeces in conjunction with fleece R.H. readings.

(i) Methods

Solomon (1945) described, in addition to cobalt-chloride

paper, other papers impregnated with cobalt salts for the measurement of R.H. by colour-matching methods. Since paper impregnated with cobalt-chloride alone gives fairly accurate humidity readings only within the range 40-70% R.H., it was decided to adopt other methods in conjunction with cobalt-chloride papers, to give accurate readings over a wider range of humidities. For readings within the 40-70% R.H. range, cobalt-chloride papers were retained. For the 30-40% range, a modification of cobalt-chloride paper, mentioned by Solomon (1945) and used by Darrow (1943) was employed, involving the addition of glycerol to the cobalt-chloride solution in which papers were impregnated. For readings within the 70-95% R.H. range, cobalt thiocyanate papers were employed. In the case of all 3 methods, sufficient papers were impregnated in the salt solutions at the beginning of the season's work to last through the season, and to make the standards which remained effective for at least 6 months (Solomon, 1945).

Darrow (1943) mentions that if glycerol be added to the impregnating solution the sensitive R.H. range of the paper, where various distinctive lilac shades occur, is moved further down the range than the 40-70% R.H. of normal cobalt-chloride paper. The ammount of this shift is directly proportional to the ammount of glycerol present in the solution. Solutions containing 25% $^{W}/_{V}$ of cobalt-chloride were made up in mixtures of water and 5, 10 and 20% 'Analar' glycerol. Pieces of dessiccator-dried No.1 filter paper were impregnated in each of these solutions and dried at room humidity. These 'glycerol-cobalt-chloride' papers were exposed at 30°C to several humidities. The colours they attained were compared with the colours of normal cobalt-chloride paper standards of the same humidities. It was decided from these trials that paper impregnated in

20% glycerol-cobalt-chloride solution (hereinafter referred to as glycerolpapers) gave the most suitable sensitive range for the purpose in view. This range was 25-55% R.H. An unexpected result of the addition of glycerol was that the colours were appreciably more brilliant than the corresponding colours with cobalt-chloride alone. 'Glycerol' papers and standards were then prepared as described for straight cobalt-chloride paper (page 98-99).

The cobalt thiocyanate papers were prepared as described by Solomon. Crystals of pure cobalt thiocyanate ($Co(CNS)_2.\frac{1}{2}H_2O$) were obtained and dried over phosphorus pentoxide for several days. A solution containing 23.0 gr. of this substance per 100 ml. distilled water was then made, and papers were impregnated in it. A special thin tissue paper (Electrolytic 'B' Condenser Tissue) was used for this purpose, as recommended by Solomon, since the brilliance of the colours imparted to paper with this solution rendered the use of thicker paper unnecessary. The best range for accurate colour matching and thus for accuracy in humidity readings is 70-95% R.H. with cobalt thiocyanate papers. Standards were prepared at 30°C as described for cobalt-chloride paper (page 98-99) and the same length of exposure of standard papers and of papers in fleeces was used.

In the fleece R.H. determinations, the glycerol and cobalt-chloride papers were always used as strips 20 x 7.5 mm. in size, and were stuck together in parallel pairs by a thin transverse strip of adhesive paper, each strip being numbered at the top left hand corner with a pencil before use, and stored in a dry air chamber. These 'double stripe' were inserted by forceps, endways into the fleece so that the bottom end (without the number) lay against the skin. In preliminary trials using cobalt-chloride, glycerol and cobalt thiocyanate papers, strips of the 3 types of paper were

joined together with adhesive paper to form 'triple strips.' They were discarded in favour of the 'double strips' because it was found that the thin tissue thiocyanate papers usually curled up by the time they were removed from fleeces, so that the exact distance of the ends of the strip from the sheep's skin was variable. In subsequent field work the thiocyanate papers were inserted into the fleeces separately in the form of small pieces about 1 cm. square, which were pushed into fleeces until they lay on the skin or approximately 2 cm. off the skin. The preliminary field trial of the three types of paper was carried out on sheep in July. Where the sensitive range of the cobalt-chloride and 'glycerol' papers tended to overlap (45-55% R.H.) the R.H. reading given by the two papers in a double strip usually coincided to within 2%. In this field trial, the humidities in the sheep fleeces happened to be too low for accurate readings by the thiocyanate papers.

Readings given by cobalt-chloride papers were classified as already described (page 100). When readings by glycerol papers were above the upper limit of their sensitive range (55% R.H.) they were classified as 'over 55% R.H.' and are given as '55+' in Tables .

(ii) Survival of egg batches of species other than L.sericata in sheep fleeces

Between 8 July and 3 Sept. 1948, 143 egg batches laid by laboratory cultures of the undermentioned species were placed in the fleeces of 17 lambs (sheep nos. 43-56/60-62) at Houghall Farm, Durham. They were made up as follows:-

25 L.caesar egg batches
26 L.illustris egg batches
43 Phormia egg batches
47 C.vomitoria egg batches
2 C.erythrocephala egg batches

The batches were usually placed in the fleeces some 30-45 min after laying, during which period they were kept

as cool as possible, at 12-17°C. At these temperatures blowfly eggs develop very slowly (Wardle 1931) so that the amount of development that the eggs had undergone was negligible by the time they were placed at higher temperatures within fleeces. A number of eggs were set aside as controls in saturated air in the laboratory. The batches were transported to the sheep in separate 3 x 1⅝ tubes, within which was a small piece of sheep wool on which the eggs lay. When putting eggs into the fleeces, the fleece at the site chosen was slightly parted and the eggs shaken off the piece of wool and pushed with the finger until they lay at the distance chosen from the skin. A 'double strip' was placed beside the batch, with the end of the strips against the skin, at the same time, and the fleece closed over them, and tied at the wool tips with string. The double strips were removed after 30 min. quickly transferred by forceps to a bottle of liquid paraffin and taken to the laboratory for comparison with standards. After removal of the double strip from the site of the batch, the fleece tips were tied up again. The batches were removed after 19-26 hrs in the fleeces, placed in tubes and taken to the laboratory for examination. Second humidity readings at the egg sites were made at the time of their removal.

The sheep were kept in a series of paddocks about 20x10 yards in size. A thermohygrograph was kept at about 3' above the ground in a screen, in one of the paddocks, throughout the period of the experiments. The thermohygrograph records were used to obtain some idea of the weather conditions prevailing during the period that each particular batch was in a fleece, while rainfall and hours of sunshine were recorded at a stationabout a mile from the paddocks. The amount of rainfall during the period that

a batch remained in a fleece might give a guide to the fleece humidities and their fluctuations during that period, since it has been shown that rainfall causes fluctuations in fleece R.H. (Part III page 107). It has been noted previously (page 37) and by Macleod (1940) that fleece temperatures can be considerably higher when the sheep are exposed to sunshine, owing to absorbtion of radiant energy by their fleeces so that sunshine affects the precise temperatures to which eggs may be subjected during development.

The results obtained are given in Tables 22 - 25 (page 124-6, 128), and are discussed below under the various species to which they appertain.

L.caesar eggs (Table 22 page 124). Of 25 batches placed in fleeces, in 12 all the eggs failed to complete development. In the remaining 13 batches, in 3 only did more than 50% of the eggs complete development. In 5 of the 13 batches some eggs succeeded in hatching but as seen in the table the percentage batch in each case was low. It has been shown (Part II page 51, Table 8) that the minimum humidity for successful development of L.caesar eggs lies between 60 and 65% at 37°C. From the results in Table 22 it is not possible to correlate the humidities found at the beginning and end of the incubation period of the various batches with the percentage development of the eggs. This is very likely due to the fact that nothing is known about the R.H. to which the eggs were subjected between the humidity readings taken at the beginning and end of the period in the fleece. In addition, the absence of temperature records at the egg sites constitutes a serious gap in the work. Another difficulty is that the number of egg batches employed was small, and this applies to all the experiments. In consequence, no general conclusions about the suitability or otherwise of different parts of sheep fleeces under various weather conditions,

TABLE 22 L.CAESAR EGGS.

Period in fleece (G.M.T.)	Sheep No.	Position †	Distance of eggs from skin (cm.)	%R.H. at egg site: Time eggs IN ς	IN N*	Time eggs OUT ς	OUT N	Weather during expt.: Rainfall, inches	Sunshine, hours.	% development in CONTROL eggs	Fate of eggs in FLEECE: % eggs HATCHING	% eggs PREHATCHING
13.30,18 July- 14.00,19 July	49	W	0	55+	58	55+	62	0.0	8.0	90	0	5
	49	W	2	55	50	52	50			70	20	10
	54	M	0	55+	68	55+	67			70	0	40
	43	RC	0	48	49	41	40			90	0	0
13.30,19 July- 14.40,20 July	44	RC	0	41	40	38	40	0.0	9.7	100	0	0
	44	M	2	45	45	50	49			90	0	60
10.10,24 July- 10.30,25 July	56	RC	0	55+	65	55+	66	0.0	9.0	20	0	0
	56	RC	2	55+	65	55+	57			30	0	5
	52	RC	0	55+	68	55+	57			20	0	0
	51	RC	0	55+	63	55	55			10	0	0
19.30,26July- 16.30,27 July	56	M	0	55+	62	55	55	0.0	9.0	80	0	0
13.30,28 July- 14.15,29 July	45	W	0	55+	75	55+	66	0.0	10.0	90	0	0
14.15,29 July- 13.45,30 July	54	M	0	55+	59	55+	61	0.0	11.0	50	0	0
15.15, 3 Aug.- 17.50, 4 Aug.	54	W	0	55+	58	55+	70	0.0	0.3	20	20	10
14.10,23 Aug.- 14.50,24 Aug.	46	W	0	50	50	55+	70	0.0	9.0	10	0	0
09.50,25 Aug.- 14.15,26 Aug.	47	W	0	55+	80+	55	53	0.37	9.0	25	0	15
	47	W	2	100	100	55+	61			40	0	25
14.20,26Aug.- 10.50,27 Aug.	47	M	0	50	50	55+	69	0.08	2.0	40	0	30
	47	RF	2	55+	67	55+	80+			20	10	15
	47	RC	0	42	41	55	55			50	0	0
13.10,31 Aug.- 10.05, 1 Sept.	56	M	0	50	50	55	55	0.15	3.0	30	0	0
	56	M	2	55+	60	55+	61			50	10	15
	60	RC	0	54	52	50	50			60	0	0
13.55, 3 Sept.- 09.30, 4 Sept.	55	R	0	-	80	-	-	2.1	0.0	70	0	70
	55	M	0	-	80+	-	-			65	10	85

†, for key to position symbols see Appendix, page 179. * ς = reading by 'glycerol' papers. N = reading by cobalt-chloride papers.

for blowfly egg survival, can be drawn. The results are intended only as a record of the fate of specific blowfly eggs in particular fleeces at the time that the experiments were carried out.

L.illustris eggs (Table 23 page 125). As with the L.caesar eggs employed, the fertility of some of the

TABLE 2 L.ILLUSTRIS EGGS

Period in fleece (G.M.T.)	Sheep No.	Position	Distance of eggs from skin (cm.)	% R.H at egg site: Time eggs IN G	N	Time eggs OUT G	N	Weather during expt.: Rainfall, inches.	Sunshine, hours	% development in CONTROL eggs	Fate of eggs in FLEECE: % eggs HATCHING	% eggs PREMATCHING
10.30,12 July-10.00,13 July.	54	W	2	55+	67	-	-			25	0	20
	54	RC	2	54	54	-	-			20	0	60
	53	M	0	55+	60	-	-			50	0	30
	53	M	2	55+	60	-	-			10	0	0
	53	RC	0	55	55	-	-			40	0	50
	53	LC	0	55+	63	-	-	0.14	0.0	40	0	10
	47	T	0	55+	66	-	-			40	0	90
	47	W	0	55+	68	-	-			70	0	80
	56	RC	0	55+	63	-	-			40	0	0
	56	LC⅜	0	55+	59	-	-			40	0	0
	54	W	0	55+	67	-	-			60	0	20
13.30,19 July-14.30,20 July	44	T	0	42	40	40	40	0.0	9.0	80	0	0
19.20,26 July-16.20,27 July	51	W	0	55+	80+	55+	66			20	0	20
	51	T	0	49	48	45	43			60	0	0
	47	RC	0	55+	56	55	53			70	0	0
	62	RC	0	53	52	48	48	0.0	8.5	40	0	0
	53	RC	0	55+	68	55+	66			30	0	40
	56	M	0	55+	61	55	55			10	0	0
10.20,22 Aug.-14.15,23 Aug.	49	W	0	53	51	53	50			90	0	0
	49	M	0	45	45	55+	61	0.18	10.5	65	0	0
	49	RC	0	45	45	45	45			90	0	0
14.40,30 Aug.-14.15,31 Aug.	52	M	0	-	40	-	46	0.0	1.0	60	0	0
	60	T	0	-	46	-	60			20	0	0
14.00, 3 Sept.-09.30, 4 Sept.	55	R	0	-	80	-	-			20	10	80
	55	M	0	-	80+	-	-	2.1	0.0	55	20	5
	55	W	0	-	65	-	-			40	5	80

L.illustris batches was low so that some of the resulte are valueless. Of 26 L.illustris egg batches placed in fleeces, in 13 of them all eggs failed to complete development, while in the other 13 some eggs did survive, in 6 of which batches, 50% or more of the eggs completed development. In 3 batches only did a small percentage of the eggs succeed in hatching (those in the fleece of sheep 55 during the period 14.00, 3 Sept. - 09.30 4 Sept.). This period was one of continuous heavy rain - about 2.0 inches of rain fell (Table 23). The eggs were therefore in the fleeces at a time of high humidity,

TABLE 24 PHORMIA TERRANOVAE EGGS

Period in fleece (G.M.T.)	Sheep No.	Position	Distance of eggs from skin (cm.)	% R.H. at egg site: Time eggs IN G	IN N	Time eggs OUT G	OUT N	Weather during expt.: Rainfall, inches	Sunshine, hours	% development in CONTROL eggs	Fate of eggs in FLEECE: % eggs HATCHING	% eggs PREHATCHING
14.15, 8 July–14.30, 9 July	51	M	0	55+	67	–	–			70	5	70
	51	W	0	55+	67	–	–			90	0	40
	53	W	0	45	46	–	–	0.01	0.8	40	0	40
	53	W	2	50	52	–	–			90	10	80
	53	M	0	47	50	–	–			50	0	0
14.00,12 July–10.30,13 July	43	W	0	–	80+	–	–			70	0	90
	43	RC	0	47	47	–	–			80	0	0
	48	RC	0	57	56	–	–			60	0	0
	48	LC	0	55	55	–	–			90	0	0
	48	T	0	50	50	–	–	0.14	0.0	60	0	0
	49	T	0	55+	61	–	–			70	0	0
	49	T	2	55+	65	–	–			100	10	50
	49	RC	0	55+	73	–	–			60	0	0
	49	RC	2	55+	73	–	–			60	30	30
13.30,18 July–14.00,19 July	43	M	0	55+	63	42	40			90	0	50
	43	M	2	50	50	40	40			90	15	60
	49	M	0	55	55	47	45	0.0	5.0	90	0	20
	49	M	2	49	48	42	40			70	2	80
	54	M	0	55+	68	55+	67			80	0	30
14.50,20 July–13.40,21 July	44	RC	0	38	40	40	40	0.05	9.0	60	0	0
19.15,26 July–17.15,27 July	51	W	0	55+	80+	55+	66			80	10	70
	51	T	0	49	48	45	43			80	0	0
	47	RC	0	55+	56	55	53			65	0	0
	62	RC	0	53	55	48	48	0.0	9.0	45	0	0
	53	RC	0	55+	68	55+	66			20	0	5
	56	M	0	55+	62	55	55			80	0	5
15.30,27 July–14.00,28 July	54	W	0	55+	68	55+	75			70	0	0
	54	W	2	55+	59	57	58	0.0	11.0	90	0	0
	50	RC	0	42	43	49	48			100	0	0
	50	RC	2	42	43	49	48			60	0	0
14.15,29 July–13.45,30 July	54	M	0	55+	59	55+	61	0.0	3.5	70	0	0
10.30,31 July–12.00, 1 Aug.	53	W	0	55+	70	–	–			90	0	0
	53	T	0	55+	80+	–	–			90	0	0
	53	RC	0	55+	80+	–	–			50	0	0
	55	M	0	55+	67	–	–	0.79	4.5	30	0	0
	55	RC	0	55+	68	–	–			30	0	0
	56	W	0	55+	80+	–	–			95	0	0
	56	RC	0	55+	68	–	–			50	0	0
14.10,23 Aug.–14.00,24 Aug.	46	M	0	41	40	–	–			80	0	0
	46	M	2	41	40	–	–			90	0	0
	45	M	0	42	40	–	–	0.0	9.0	80	0	0
	45	M	0	42	40	–	–		0	95	0	0
	45	RC	0	50	50	–	–			70	0	0

when it was unlikely that naturally laid eggs would be present in fleeces, since wild flies are probably inactive during continuous rain. It may thus be concluded that in the fleeces of the sheep used humidities high enough for hatching of L.illustris eggs were not encountered, although humidities suitable for completion of development were fairly frequently found.

P.terranovae eggs (Table 24 page 126). Of 43 egg batches placed in 28 all the eggs died through desiccation at an early stage. Of the remaining 15 batches, in 9 more than half the eggs completed development. Hatching occurred in only 7 of these batches, and the percentage hatch was always low.

C.vomitoria eggs (Table 25 page 128). Of 47 batches used, in only 9 did some eggs complete development. Conditions suitable for C.vomitoria eggs were thus rarely found at the time of the experiments, in the fleeces of the sheep used. It is noteworthy that of the 9 batches which were partially successful, 7 had been placed 2 cm. off the skin of the sheep, where temperatures are lower than at the skin surface. It has been shown in the present work that maximum temperature for the development of C.vomitoria eggs is about 36°C (page 52). The average temperature at the skin surface of sheep was assumed to be 37°C by Davies & Hobson (1935), and this temperature has been adopted as a guide in the present work. It is therefore probable that the very low number of batches successfully completing development at the skin surface of the above sheep was due to the fact that temperatures prevailing there were usually too high. (A total of 28 C.vomitoria egg batches were placed near the skin). In this case humidity is not itself the limiting factor for blowfly egg survival. The two batches which completed development at the skin surface were on sheep 44 (Table 25, page 128) during a cool period (14.20, 26 Aug. - 11.00, 27 Aug.) with some rain

TABLE 25 C.VOMITORIA EGGS

Period in fleece, (G.M.T.)	Sheep No.	Position.	Distance of eggs from skin. (cm.)	% R.H. at egg site: Time eggs IN G	N	Time eggs OUT G	N	Weather during expt.: Rainfall, inches	Sunshine hours.	% development in CONTROL eggs	Fate of eggs in FLEECE: % eggs HATCHING	% eggs PREHATCHING
15.15, 3 Aug.-	54	M	0	55	55	55+	61			90	0	0
17.50, 4 Aug.	54	M	2	57	57	55+	65	0.0	0.3	90	0	5
	47	M	0	50	50	55+	69			45	0	0
	47	RF	0	55+	67	55+	80+			65	0	0
	47	RF	2	55+	67	55+	80+			20	15	25
14.20,26 Aug.-	47	RC	0	42	41	55	55			10	0	0
11.00,27 Aug.	44	T	0	50	50	55	54	0.08	2.0	65	0	0
	44	M	0	55+	58	55+	58			90	0	50
	44	M	2	55+	66	55+	58			80	10	90
	44	W	0	55+	75	55+	75			65	0	60
	47	T	0	55+	55+	-	54			75	0	0
11.00,27 Aug-	47	T	2	44	40	-	47	0.0	11.0	60	0	0
11.15,28 Aug.	49	RF	0	55+	80+	55+	70			70	0	0
	49	RF	0	-	70	-	62			60	0	0
11.20,28 Aug.-	49	RF	2	-	65	-	62	0.09	5.0	100	0	0
11.00,29 Aug.	47	RF	0	-	69	-	-			100	0	0
	47	RF	2	-	60	-	-			100	0	0
	48	T	0	-	60	38	40			100	0	0
	48	T	2	-	80	40	40			60	0	0
	48	M	0	-	61	40	40			50	0	0
	48	M	2	-	53	40	40			75	0	0
	48	W	0	-	65	-	56			80	0	0
	48	W	2	-	75	-	54			60	0	0
	48	RC	0	-	53	-	40			90	0	0
	48	RC	2	-	66	-	45			60	0	10
	53	W	0	-	60	-	50			70	0	0
	53	W	2	-	67	-	50			95	0	0
	53	RF	0	-	65	-	59			90	0	0
10.30,29 Aug.-	53	RF	2	-	65	-	62	0.0	3.8	95	0	0
15.40,30 Aug.	53	RC	0	-	60	-	-			100	0	0
	53	RC	2	-	60	-	-			70	5	50
	53	LC	0	-	60	-	47			70	0	0
	53	LC	2	-	66	-	47			90	0	0
	43	T	0	-	56	-	47			70	0	0
	43	T	2	-	80	-	50			50	0	0
	47	T	0	-	55	-	40			50	0	0
	47	T	2	-	62	-	43			65	0	0
	49	M	0	55+	66	-	47			100	0	0
	49	R	0	55+	68	-	53			50	0	0
	49	R	2	55+	68	-	53			60	0	0
14.40,30 Aug.-	52	M	0	41	40	46	46	0.0	1.0	20	0	0
14.15,31 Aug.												
	56	M	2	-	60	55+	61			60	0	10
	56	M	0	-	50	55	55			70	0	0
13.20,31 Aug.-	60	RC	0	50	52	51	50	0.15	3.0	60	0	0
10.15, 1 Sept.	61	W	0	55+	66	55	55			90	0	0
	61	W	2	55+	80+	55	55			95	10	80
	61	RF	0	55+	80	55+	58			95	0	0

(0.08 inches) and only 2 hr. sun, so that fleece temperatures would tend to be low. The thermograph record showed that the period was a cool one, being 10-15°C during the day and 5-10°C during the night. Further experiments on the development of C.vomitoria eggs in fleeces are described later (page 129-130).

C.erythrocephala eggs. Owing to various difficulties, only 2 batches of this species were placed in fleeces. Since the maximum temperature for the development of C.erythrocephala eggs is lower (35°C) that that of C.vomitoria eggs (36°C) (page 52) it is to be expected that temperatures low enough for the eggs of the former species to complete development will rarely be obtained in view of the results with C.vomitoria eggs (page 129-30).

Attempts were made to estimate the average temperatures in the fleeces of the sheep used, utilizing information previously obtained on the maximum constant temperatures at which the eggs of various species could complete development, and the duration of incubation at constant temperatures (page 55-6). The eggs of C.vomitoria (max. const. temp. for development - 36°C), and of L.sericata, which hatch in 7.6 - 7.9 hr. at 37°C, were used.

Thirteen batches of C.vomitoria eggs were placed under small cotton wool pads moist with water, at various positions in the fleeces of 6 sheep (nos. 52, 57-61) during the period 30 Aug. - 2 Sept.,1948. The eggs were thus subjected to saturated air to eliminate the effects of low R.H., so that the temperatures in the fleeces were the important factors. Of 11 batches placed directly on the skin of the sheep under the wet pads, in 7, all the eggs failed to complete development. Control eggs showed high fertility. In the other 4, many eggs hatched, but in these cases the wet pad and the eggs had worked themselves loose so that the egg lay about 1-2 cm. off the skin by the time they were

removed for examination. They had not completed development at the skin surface. The failure of the other 7 batches at the skin surface can be accounted for only as due to the fact that temperatures there were lethal to them. No wool grease had spread over them (see page 130-5). A further 2 batches placed in contact with cotton wool pads in the fleeces of sheep, so that they lay 2 cm. off the skin, successfully developed and hatched. The average temperatures near the skin at the positions used on these sheep at the time of the experiments must therefore have been above 36°C, and so lethal to C.vomitoria eggs, while temperatures at 1-2 cm. off the skin were low enough to permit their development. The weather during these experiments was rather cool and cloudy, shade temperatures during the day time being 15-20°C, and 5-12° during the night.

On 1 Sept. 3 L.sericata egg batches were treated in the same way as those of C.vomitoria above. One was placed under a small wet pad directly on the skin, of each of sheep 57-59, at 07.30 hrs. These batches were removed from the fleeces at 15.30 - 15.40 hr. on the same day and were found to be in process of hatching at that time i.e. after 8.0-8.2 hr. in the fleeces. Since the incubation period of L.sericata eggs at 37°C (constant) is 7.6-7.9 hr., the above duration of 8.0-8.2 hr. in the field shows that the average temperatures near the skin of the 3 sheep used must have been very near to 37°- probably between 36 and 38°C.

(iii) Effect of wool 'yolk' on blowfly egg survival in sheep fleeces

It will be noticed in Tables 22 -25 (pages 124-8) that several egg batches of all four species failed to complete development in fleeces when humdility measurements at the start and finish of the experiment indicated humidities high enough for egg development, at the sites used. The

explanation for some of these cases of unexplained failure may well have been that the R.H. at the sites fell to a low level between the taking of the two readings. It was however noticed in several such cases of failure that the eggs on removal from fleeces had aquired the golden yellow colour of wool 'yolk'. Examination of such eggs in the laboratory showed that they were covered with a thin film of wool yolk, rendering the chorion transparent. Yolk occurred in the fleeces in the form of irregularly shaped, greasy yellow blobs, varying from about a half to 3 mm. in diameter, and which adhered to groups of wool fibres, or were strung at intervals along individual fibres. In some sheep these blobs seemed more numerous than in others, but were observed on sheep used in all 3 of the seasons over which work extended (1946-48). Wool yolk or grease is well known to be composed of a mixture of the oily secretion of the sebaceous glands, and of varying amounts of 'suint' which has been considered to be derived from the 'sudoriferous' sweat glands (Freney 1940, Bonsma & Starke 1924). The suint fraction is known to act as a wetting agent (Hobson 1941) owing to its content of soaps, and to be hygroscopic (Freney 1940; Hambrock et al, 1934).

Laboratory experiments were carried out on the spreading powers of raw wool yolk over blowfly eggs, and its effects on their survival observed. Blobs of yolk as described above were obtained from a sheared fleece, by picking them with needles off locks of wool partly separated under a binocular microscope. The yolk so obtained was unchanged from its natural mixed condition. Small pieces of yolk were separated from these blobs and used to treat blowfly eggs in various ways.

It was found that raw wool yolk so obtained had very little spreading power over the outer surface of the blowfly

egg chorion, which is known to be hydrophilic (see page 72). A piece of yolk placed half way along an egg on the outside of the intact chorion usually failed to spread at 37°C, and eggs so treated completed development at appropriate humidities. On the other hand, if the yolk was placed at the anterior end of the egg so that the micropyle was covered, it penetrated the chorionic micropyle and spread over the egg as a thin film between the inner surface of the chorion and the outer surface of the c.v. membrane (see page 14-15). A high proportion of eggs so treated failed to complete development, being almost certainly asphyxiated by the covering film of yolk. This film appeared to fill the air spaces in the chorion (page 75) as shown by the fact that wherever spreading had occurred via the chorionic micropyle, the area of spread was indicated by the transparency of the chorion. The amount of spread was therefore judged by the area of the transparency. It was plain in all the experiments that there existed a high correlation between the amount of yolk spread over the egg, and its survival or non survival. Table 26 illustrates the above features. In this table, of

TABLE 26

The effect of wool yolk on L.sericata egg survival, 70% R.H., 37°C.

Position of wool yolk	No. of eggs used	% reaching various stages: Hatching	Prehatching	Early Stage
Half way along eggs	38	65	0	35
On micropyle	39	15	0	80
Untreated controls	35	100	0	0

the eggs which were treated with yolk on the micropyle, those that were able to hatch (15%) were those in which for some reason the yolk had failed to spread over a large area. In the 80% of the eggs which had died at an early stage yolk

had spread over most of their surface area in each case.

Experiments on the spreading powers of raw wool yolk over eggs at various humidities at 37°C showed that spreading readily occurred at 70% R.H, less readily at 80%, and hardly at all at 90%. This differential spread at different humidities is reflected in the percentages of treated eggs completing development at three humidities (Table 27). It will be seen that few treated eggs survived at 70% R.H.,

TABLE 27

Effect of wool yolk on L.sericata egg survival at 70, 80 and 90% R.H. 37°C.

(Wool yolk placed on micropyle of each egg)

% R.H.	No.of eggs used	% reaching various stages		
		Hatching	Prehatching	Early stage
70	36	11	5	84
80	49	43	4	53
90	17	94	0	6

while nearly half (43%) did survive at 80% R.H., and 94% at 90% R.H. It was very clear in the experiments that at 80% R.H. those eggs that did survive were those over which hardly any spreading of yolk had occurred. The explanation of the failure of yolk to spread between the chorion and c.v.membrane of blowfly eggs at 90% R.H. appears to be that at this humidity, hygroscopic substances (presumably part of the 'suint' fraction) in the yolk absorbed sufficient water to make an emulsion, so that it was unable to spread. Such yolk blobs at 90% R.H. assumed a slightly white opaque appearance, rather like vaseline after prolonged contact with water, while yolk at 70% R.H. had a translucent greasy appearance, presumably because it contained less absorbed water. At 80% R.H. the yolk blobs had an intermediate appearance, and had intermediate spreading

properties, (Table 27). Very high humidities in sheep fleeces therefore not only provide suitable conditions for eggs to complete development, but would also emancipate them from the risk of asphyxiation by spreading of wool yolk.

It was noticed that more egg batches became covered with yolk when placed on the skin of the sheep than when placed 2 cm. off the skin. This may have been due in some cases to the presence of relatively more yolk in the basal part of the fleece than further out. It may also have been due to the higher temperatures near the skin causing more rapid spread of yolk over eggs. Burtt (1945) states that wool grease extracted from fleece became liquid at about 35°C. The raw unchanged wool yolk used in the present work was observed at various temperatures, through a horizontal microscope, in a melting point apparatus. Wool yolk that had been dried for several days over sulphuric acid was used. No sharply defined melting point was observed, as would be expected from its mixed character, but a gradual melting was observed which was not complete until about 55°C was reached. It appears therefore that the raw yolk was more liquid at 37°C, than at 25°C, an average temperature for the outer parts of the fleece. Thus more rapid spreading would be expected to occur near the skin (about 37°C) than at points further out in the fleece. It was found in experiments at 70% R.H. 24°C, that spreading of the yolk over blowfly eggs did occur at that temperature, but it was too slow to prevent them from hatching, since it took about 30-50 hr. for extensive spreading to occur, while at that temperature development was completed in 16-18 hr.

It appears therefore that at least some of the cases of unexpected failure of eggs to complete development, and of low percentage development in batches, mentioned on

page 130 were due to the asphyxiation of eggs through spreading of wool yolk over them. In blowfly eggs naturally laid on sheep the spread of grease may kill them and prevent myiasis being subsequently set up. No information is available as to whether this factor is of any practical importance, and the present work merely points to its existence as a feature in the ecology of blowfly eggs laid in sheep fleeces.

DISCUSSION

Since a proportion of L.sericata eggs can complete development at 50% R.H. 37°C (page 33) the present observations on fleece atmosphere humidity show that suitable humidities for the development of the eggs of this species occurred comparatively frequently in the fleeces of some of the sheep used. The existence of sheep with more humid fleeces than others (page 108) may throw light on the high susceptibility of some sheep to blowfly myiasis, while other sheep of the same breed and kept in the same flock remain free from maggots. Such differences in susceptibility between sheep are familiar to every shepherd. In sheep with humid fleeces, suitable for L.sericata egg development the crucial factor would appear to be the extent and rapidity of rises in fleece R.H. which would enable hatching to occur. The rise in basal fleece humidity, caused in some way by the presence of free water on the outside of the fleece, described by Macleod (1940) was also found in the present work. When the outside of the fleece was wet, basal fleece humidities frequently rose to 60-80% R.H. when some hatching would occur. The fleece atmosphere near the skin did not however become saturated, again agreeing with Macleod's findings. Rain drops were observed to penetrate within 1 cm. of the skin along the back, in short fleeced Swaledale x Border Leicester lambs, and also in short coarse

fleeced Herdwick and Swaledale wethers. It can be visualized that sudden heavy rain, or sunny showery weather, in addition to promoting fly activity could provide sudden rises in fleece R.H. and enable eggs to hatch. Under such conditions, the ability of the larvae to remain imprisoned within the shell for 3 hr. at 37°C (page 40) is of potential importance, since, especially in hill districts, heavy rain may occur within short periods during weather which is mainly sunny and suitable for fly activity. Blowflies are particularly active during the early forenoon, and eggs laid at such a time would be reaching the hatching condition during a period when dew was falling, or when the sheep might be lying in dew-laden vegetation. The sudden rise in basal fleece R.H. when the outside of the fleece is wet, might lead to hatching of the eggs, especially if they were not laid near the skin. For example, in sheep 1, the basal humidities at the tailhead and right crutch positions at 04.00 hr. on 9 July, 1946, a cool morning with very heavy dew, were 68 and 67% R.H. respectively. At these positions at 14.00 hrs. on the same day when the sheep were dry and the afternoon hot, the humidities had fallen to 53 and 57%, respectively.

Laboratory experiments (page 41) have shown that about 50% of L.sericata eggs survived in saturated air after exposure for 7 - 8 hr. to air of 40% R.H. 37°C, provided the eggs had not previously been exposed to high humidities. Eggs laid under dry fleece conditions in the field would therefore be expected to survive if the humidity rose sharply during the incubation period. The present work showed that such rapid rises in humidity near the skin of the sheep, do occur. It seems, however, that the transitory effect of rain in raising the basal fleece R.H. would rarely cause strike to develop, since first instar larvae of L.sericata survive for only about 1 hr. at 50% R.H. 37°C (Davies & Hobson, 1935) - the larvae would be killed as the R.H. fell again as the rain

drops evaporated from the outsides of the fleeces. The possibility that numerous newly hatched first instar larvae closely congregated on a small patch of skin are able to cause a rise in R.H. in their immediate vicinity by causing serous exudate to be poured out due to skin irritation, cannot however be neglected.

It was also shown (page 41-3) that L.sericata eggs, if incubated in saturated air at 37°C for a few minutes, could withstand very little desiccation when subsequently incubated at a low R.H. This property of the eggs may be of importance in preventing myiasis development in the field, particularly when eggs are laid immediately after rain, since water present on the outside of the fleece raises the R.H. near the skin for so short a time.

A shade air temperature of 16°C and a humidity of 70% R.H. may be considered to be fairly average conditions in British summer weather. This represents a vapour pressure of 9.5 mm. Hg. A basal fleece R.H. of 40% and above may be considered to be common in the sheep used in the present work. Assuming that the temperature of the air at the base of a fleece is 36°C a humidity of 40% R.H. there represents a vapour pressure of 17.8 mm. Hg, which is considerably in excess of that typically found in the external air. Both Davies & Hobson (1935) and Macleod (1940) also noted the higher vapour pressure near the skin of sheep, even in dry summer weather. They considered the excess to be due to evaporation from the skin. It appears from the fleece R.H. readings given by Macleod (1940) that in the sheep he used, evaporation of water from the skin as measured by the positive balance of vapour pressure near the skin was much less rapid than was found in some of the sheep used in the present investigation. Among the latter the high R.H. of the basal fleece, due presumably to active sweating, showed marked

variations in intensity and in distribution over the body. Of the 28 sheep of various breeds investigated in detail in 1946, 5 were Down cross lambs of which 2, studied in detail, showed continuous sweating in warm weather to such an extent that their fleeces were humid enough near the skin for strike to develop. In these sheep during one period, the usual descending R.H. gradient towards the skin (owing to the higher temperatures near the latter) was not present. Instead, a gradient in the reverse direction occurred with humidities of 70 -90% R.H. near the skin with 50 - 70% R.H. in the outer fleece. In none of the other sheep used (various Herdwicks and Swaledales) was such continuous general sweating found over large areas of the body. In the latter sheep sweating over more restricted areas and for shorter periods sometimes occurred.

Many farmers consider Down breeds and their crosses to sweat more especially when fat than other classes of British breeds and in addition they are considered to be more susceptible to blowfly myiasis. In 2 Down cross lambs (page 110-1) sweating was found for considerable periods to be more rapid at the withers, a region where strikes occur comparatively frequently (Macleod 1943).

It appears probable that some part of the suint fraction (Freney 1940) of the living fleece represents the dried residue of the watery skin secretions which, in the present work was found at times to raise the R.H. near the skin of some sheep. That rapid changes may occur in the rate of this secretory activity is suggested by R.H. changes, in the basal fleece atmosphere which can occur (page 114). In view of these observations the work of Hobson (1936) appears in a new light. He carried out suint analyses on wool samples from struck and unstruck sheep and concluded that strike could occur when the suint content was low. This result does

not preclude the possibility that susceptibility to strike may sometimes be linked with rapid sweating, leading to high basal fleece humidities. Such activity might render a region suitable for strike development, before any appreciable local rise in the suint content of that particular part of the fleece could occur. Hobson (1936) also found that in Welsh Mountain sheep, flank and belly wool contained more suint than wool from other parts of the body. In sheep studied in the present work, 2 Down cross lambs (page 111-2) showed sweating restricted to the flanks for a considerable period, and one Swaledale ram (sheep 10) not included in the text figures, was also found to sweat continuously on the flanks alone for a period of 3 weeks. These observations tend to confirm the belief that the suint fraction does have some connexion with sweating. Freney (1940) has already suggested that suint may be a constituent of true sweat, utilized to regulate body temperature.

The subject of sweating in sheep is considered further in Part IV (page 143-154).

Although the study of the fate of laboratory laid blowfly egg batches, placed in sheep fleeces in conjunction with R.H. readings at the sites at which they were placed, was not carried out on as large a scale as would be desired, it confirmed in general the evidence obtained in the detailed study of fleece R.H. and the laboratory experiments on the humidities required by the eggs of the various species to complete development. A total of 185 egg batches of the 6 blowfly species studied were placed at various positions in the fleeces of 27 sheep. The results showed that humidity conditions in 1946 were frequently maintained at levels sufficient for L.sericata eggs to complete development. The results obtained with the eggs of the various species in fleeces, are not comparable with each other, for various

reasons. It was found that conditions for the successful development of L.caesar, L.illustris, P.terranovae and C.vomitoria eggs were more rarely found. Experiments showed that temperatures near the skin of 7 of the sheep used, were too high for C.vomitoria eggs to develop, and must therefore have been too high for C.erythrocephala eggs as well (page 52). Measurements of the length of incubation of L.sericata eggs under wet pads on 3 of the above 7 sheep at the same time, showed that the temperatures near their skin surface must have averaged about 37°C.

Field work and laboratory experiments suggested that blowfly eggs in sheep fleeces may sometimes be asphyxiated by becoming covered with a film of wool grease. This spreading of grease was found to be more rapid and common at temperatures of about 37°C and humidities of below 70% R.H.

In conclusion, it may be stated that the present work shows that blowfly eggs laid in sheep fleeces face 3 main hazards. These are - too low a humidity leading to death by desiccation; too high a temperature leading to heat death; and the danger of wool grease spread causing death by asphyxiation. A consideration of these sources of mortality plainly indicate that the outer parts of sheep fleeces are far more suitable for blowfly egg survival than the inner parts close to the skin. In the outer parts the temperatures are lower, and thus humidities tend to be higher, and the risk of wool grease spread is reduced. Near the skin of sheep, the temperatures are higher and humidities lower, and the risk of wool grease causing death of eggs is increased. The occurrence of rapid sweating, already discussed tends to reverse the above relationships.

SUMMARY

1) Comparative determinations of fleece atmosphere R.H., using cobalt chloride papers and paper hygrometers showed that the former were reliable, although their use was subject to a limited humidity range (40-70% R.H.). Cobalt thiocyanate and cobalt chloride papers containing glycerol, extending the range of reliable readings to 25-95% R.H. were also used. Determinations made by means of wool samples were found to be subject to large positive errors, since the wool near the skin of sheep did not appear to be in with the fleece atmosphere.

2) Measurements of fleece R.H. by cobalt-chloride papers on 28 sheep of various breeds showed that the R.H. varied considerably from day to day in individual sheep, and in different sheep at the same time. The considerably more humid conditions found in the fleeces of some sheep is suggested as one of the contributory factors to the differential susceptibility to blowfly myiasis of the individual sheep in a flock. The common condition in most of the sheep was one of low R.H. unsuitable for blowfly eggs to develop.

3) R.H. measurements and a study of the fate of L.sericata egg batches placed in fleeces showed, however, that conditions suitable for development were relatively frequently experienced in the sheep used. Humidities high enough for hatching were more rarely found.

4) Observations on L.caesar, L.illustris and P.terranovae eggs placed in fleeces showed that conditions suitable for their development were sometimes found.

5) Evidence is produced that C.erythrocephala and C.vomitoria eggs are unable to develop near the skin in fleeces because the high temperatures occurring there are lethal for them.

6) It was found that blowfly eggs may sometimes be killed in

fleeces by the spreading of wool grease over them. Laboratory experiments showed that when small quantities of raw wool yolk were placed on the micropyles of eggs, spreading of grease between the chorion and the underlying c.v. membrane, via the micropyle, readily occurred under certain conditions causing asphyxiation of the embryos.

7) Rapid secretion of watery material by the skin (i.e. sweating) leading to suitable humidity conditions for blowfly eggs, was detected. The various sheep used showed great differences in sweating rates, the most rapid sweating being found in 2 Down cross lambs, where at one period, the limiting factor for the development of myiasis on them must have been that blowflies were not attracted to them.

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PART IV

SWEATING IN SHEEP IN RELATION TO BLOWFLY EGG SURVIVAL

Introduction

It has been generally held that sheep do not produce watery sweat over the general body surface in order to regulate body temperature, and that the coil glands that are numerous and well distributed over the body (Carter, 1939) were mainly sudoriferous glands of the apocrine type, producing mainly solid or semi solid secretions with negligible amounts of water from the point of view of temperature regulation. Such sudoriferous glands are well developed on certain parts of the body, such as the axillae and groins. Evaporation of moisture from the skin was found by Davies & Hobson (1935) and Macleod (1940), since the vapour pressure of the air next to the skin was usually considerably in excess of that of the external air. The moisture responsible for this excess when the basal fleece R.H. is 30-40% R.H. can be accounted for as 'insensible perspiration'. (Kuno, 1934, page 46) rather than to evaporation of rapidly produced watery secretions. Readings of fleece R.H. in the present work (page 110- et seq) showed that in some sheep at certain times evaporation of moisture from the skin proceeded on a sufficient scale to maintain humidities of 70-90% R.H. near the skin for considerable periods. This suggested that 'sensible perspiration' or sweating was occurring. The work of Lee and Robinson (1941) on the effects of hot atmospheres on Merino sheep produced inconclusive evidence that they sweated considerably. They found that, in neither a hot dry nor in a hot wet atmosphere could respiratory evaporation account for more than one third of the observed

weight loss of experimental sheep, and suggested that the skin sweating may help to account for the difference. Lee and his co-workers in addition experimented with the rabbit, pig, cat and dog (Lee, Robinson & Hines, 1941) Robinson & Lee, 1941a, b, c), and found that the sheep showed the greatest tolerance to hot atmospheres of all species studied.

Freney(1940) attempted to detect sweating in sheep after muscular exertion. He made moisture content determinations on basal fleece wool samples from Merino sheep, before and after they had been made to run for several minutes, pursued by men on horseback, on a hot sunny day. Freney failed to find any significant increase in the moisture content of the samples after the sheep had been so exerted, and concluded that sheep probably did not sweat. He id not however claim to have settled the problem conclusively.

It was decided to reinvestigate the problem of sweating in sheep, since humidity readings taken indicated that in some sheep, it might produce conditions suitable for blowfly eggs to develop and hatch. A case has already been cited (page 110) of one sheep on which a strike was induced merely by placing eggs near the skin, without any artificial addition of water. The humid conditions on this sheep and on others used during the work appeared to be due to rapid sweating.

More sensitive methods were employed in the detecting of sweating in sheep, than the wool sample moisture determinations of Freney (1940). The use of cobalt-chloride paper strips enabled determinations of the humidity gradients in sheep fleeces, from the skin surface outwards, to be carried out. The use of cobalt-chloride paper is basically similar to the use of the colour changes of cobalt salts in the detection of sweat areas, employed in medical research (for references see Darrow, 1943).

I THE EFFECT OF MUSCULAR EXERTION ON SWEATING

Experiments on the effects of running on fleece R.H. were

carried out on 2 sheep (nos. 4 and 10) at Crag Farm, Ravenglass, Cumberland during cool weather in August, 1946.

Method. Cobalt-chloride strips were placed at standard positions in the fleece of the sheep to be run, removed after 30 min. and stored immediately in liquid paraffin as in routine fleece readings (page 100). A second set of strips was placed in the same positions and tied securely in position, and the sheep, accompanied by others were then caused to run rapidly around a field for some 10-15 minutes by means of dogs. At the end of this period open-mouthed panting generally occurred in the sheep which were then penned and about 15 mins. allowed to pass before the second set of papers was removed. A third set of papers was sometimes placed in the fleece to measure possible changes in fleece R.H. taking place some time after cessation of running. As in the case of routine readings, care was taken not to disturb the fleece structure.

Results. The information gained in the 4 experiments carried out is given in Table 28 (page 146-7). Readings were taken at the two ends of each 2 cm. strip, so that readings given in Table 28 refer to the skin surface and to a point 2 cm. off the skin in each case. The position symbols are the same as those previously used (see Appendix page 179). It will be seen from the table, by comparing the readings obtained at the various positions 5 min before the sheep was run, with the corresponding readings 20-30 min. after running, that increases in fleece R.H., especially near the skin, occurred at some positions in all four experiments. In some cases these were small, but sometimes the increases were big, for example in Expt. 4 (Table 28(d)) the R.H. at the skin at the withers position of Sheep 4 increased from below 40% R.H. 5 min. before running to 60% R.H. 20 min. after running. These increases in fleece R.H. produced by exertion of the

TABLE 28

Effect of muscular exertion on fleece R.H.

Position	% R.H. in fleece: 5 min. before running, skin surface	5 min. before running, 2 cm.	30 min. after running, skin surface	30 min. after running, 2 cm	1 hr. after running, skin surface	1 hr. after running, 2 cm
(a) Expt.1. 11 Aug. 13.15-15.00 hr. Air temp. 16°C. Sunny with west breeze. Sheep 4.						
W	below 40	below 40	43	43	44	48
M	below 40	" 40	below 40	below 40	below 40	below 40
T	" 40	" 40	44	44	-	-
RF	" 40	" 40	42	45	44	below 40
RB	" 40	" 40	-	-	-	-
RC	" 40	" 40	52	-	-	-
(b) Expt.2. 13 Aug. 13.30-15.30 hr. Air temp. 16°C. Heavy rain previous night. Sheep dry by 13.00. Sheep 4.	5 min. before running		20 min. after running		2 hr. after running	
W	below 40	below 40	55	48	below 40	below 40
M*	57	100	70	-	" 40	75
T	below 40	40	57	-	" 40	-
RF	" 40	below 40	-	-	" 40	41
RB	" 40	-	-	-	40	-
RC	" 40	50	62	62	45	-
(c) Expt.3. 15 Aug. 13.00-14.30 hr. Air temp. 13°C. Sunny. Sheep kept in doors overnight. Sheep 4.	5 min. before running		20 min. after running		50 min. after running	
W	below 40	below 40	46	below 40	below 40	40
M	" 40	" 40	below 40	below 40	" 40	below 40
T	" 40	" 40	50	44	" 40	45
RF	40	" 40	44	44	" 40	45
RB	40	49	46	-	" 40	45
RC	43	59	45	48	" 40	45

* Rain head penetrated to the skin at this point during previous night, and had not yet completely evaporated from within the fleece by 13.30 hr.

TABLE 28 (continued)

Position	% R.H. in Fleece: 5 min. before running — skin surface	5 min. before running — 2 cm.	20 min. after running — skin surface	20 min. after running — 2 cm
(d) Expt. 4. 20 Aug. 12.30–13.30 hr. Air temp. 16°C. Sunny, west breeze. Sheep 4 and 10.				
(1) Sheep 4.				
W	below 40	below 40	60	below 40
M	below 40	below 40	below 40	below 40
T	below 40	below 40	45	below 40
RF	below 40	below 40	41	41
RB	below 40	40	55	50
RC	below 40	44	55	52
(2) Sheep 10.				
W	below 40	below 40	below 40	below 40
M	below 40	below 40	below 40	below 40
T	below 40	below 40	43	below 40
RF	below 40	below 40	below 40	below 40
RB	below 40	45	below 40	45
RC	below 40	45	50	54

sheep suggest strongly that sweating was caused by the exertion. Details of the two sheep used will be found in the Appendix (page 180), and both throughout the experiments were normal healthy animals as far as could be judged by fleece condition, fatness, colour of the inner membrance of the eyelids, and general appearance. Thus the results were not obtained on sick animals, and the sweating cannot have been a pathological phenomenon. Sheep 4 was first selected for the running experiments because routine fleece readings had shown that its fleece atmosphere was usually considerable drier than many of the other sheep used (see Fig. 20 , page 107).

Comparison of readings taken 20 min. after running, with those taken 1-2 hr. after running (Table 28 , Expt.1,2, & 3) shows that by the latter time the R.H. near the skin at many positions had fallen again to their original low level. In Expt.3 (Table 28) carried out on a cool day (air temp.13°C), readings taken only 50 min. after the end of running showed that the fleece R.H's of sheep 4 had already fallen to their original values. The effect of a running period of 10-15 mins. on the basal fleece R.H's was thus very transitory.

A constant feature of Expt. 1, 3 and 4 (Table 28) is the complete absence of any detectable rise in R.H. at the midback position of sheep 4 after running (Expt. 2 is invalidated in this respect, owing to the chance penetration of rain to the skin at the midback position, during heavy rain on the previous night). This constant absence of sweating at the midback of sheep 4, while positions quite close to it (e.g. withers and tailhead) showed sweating in all 4 experiments, is interesting. The distance between the midback and the withers position in sheep 4 was only about 30 cm. This shows that sweating rates in sheep can vary considerably between points fairly close together on the body. Macleod (1943) has drawn attention to the existence of highly susceptible small areas on some sheep, where myiasis may become established in successive seasons, to the exclusion of others. These susceptible areas may be due to the occurrence of rapid sweating on them, rendering them in some way suitable for myiasis development.

The running experiments described above can be considered only as preliminary experiments on the subject of sweating in exerted sheep. Since only 2 sheep were used information on the possible variation in sweating between individual sheep when similarly exerted is not available from these results.

II THE EFFECT OF HIGH EXTERNAL TEMPERATURES ON SWEATING

Method. In August, 1948, experiments on the effects of high temperatures on fleece R.H. were carried out on 2 sheep (nos. 4 and 53). High temperatures were obtained in an insectary fitted with electric heaters. A wooden crate (7'0" long x 3'6" wide) was built into the insectary to hold the sheep. With 2 sheep within it, some movement of the sheep was possible - they could walk about twice their own length before having to turn around; complete inactivity was not imposed on them. Soiling of fleeces was prevented by covering the floor with straw. They were fed on freshly cut grass of the species palatable to sheep, and were given in addition about 2 lb. crushed oats daily to reduce the effects of the close confinement on their condition. After completion of the series of experiments it was estimated that the sheep used were in better condition than before, so that the information on the effects of high temperatures was not obtained on sheep in declining condition. Since the dimensions of the insectary were small, the great heat output of the heaters enabled temperatures of 35-38°C to be attained within it, in as short a time as 90 min. The effects of sudden large temperature increases on fleece humidity could then be studied. ~~Thermostat~~ Temperature records in the insectary were kept by means of a 24 hr. thermohygrograph and a mercury thermometer. The thermohygrograph was situated close to one side of the crate, at 2' above the floor, i.e. about the same height as the back of the sheep. The temperature readings it gave during an experiment were checked against the mercury thermometer. It was found that when the temperature within the insectary was changing, the thermograph showed a time-lag of 15-30 min. In the temperature curves for experiments in figs. 26-28 the time-lag in the

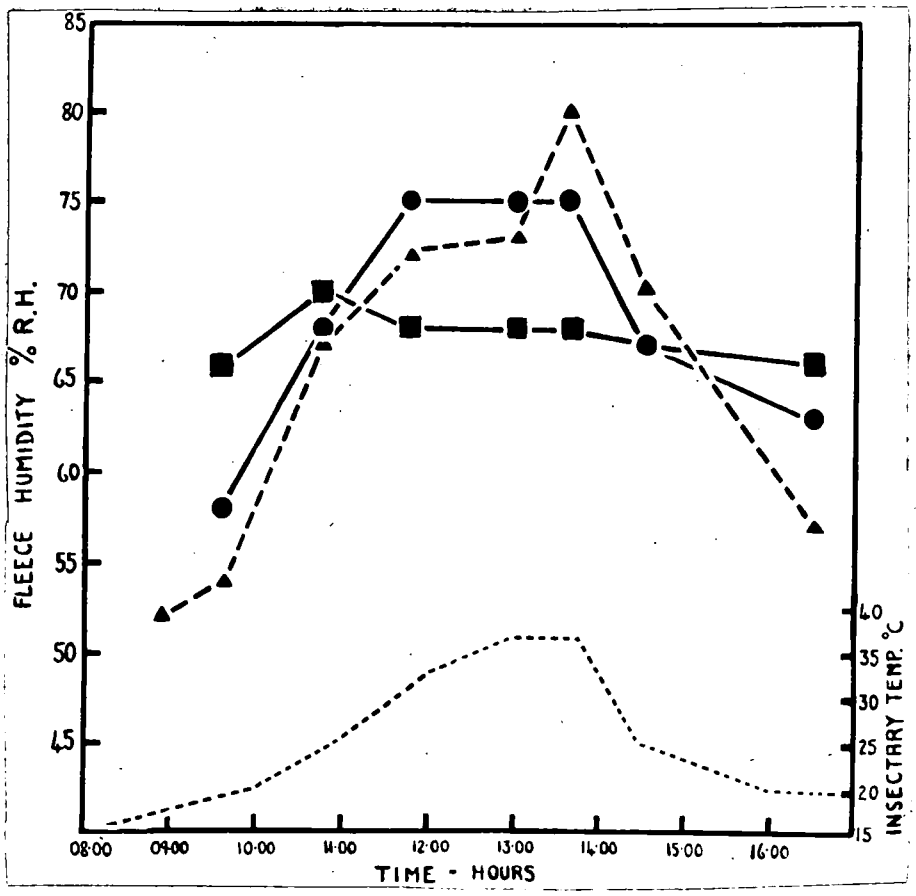

FIG. 26. Fleece R.H. fluctuations in sheep 43 (— — —) and 53 (——) ; ▲, R. flank position; ●, withers position, skin surface; ■, withers position, 2 cm. off skin; -----, insectary temperature. (Expt. 2. Appendix. 229-231).

Note. In figs. 26-28, fleece R.H. readings refer to skin surface unless otherwise stated.

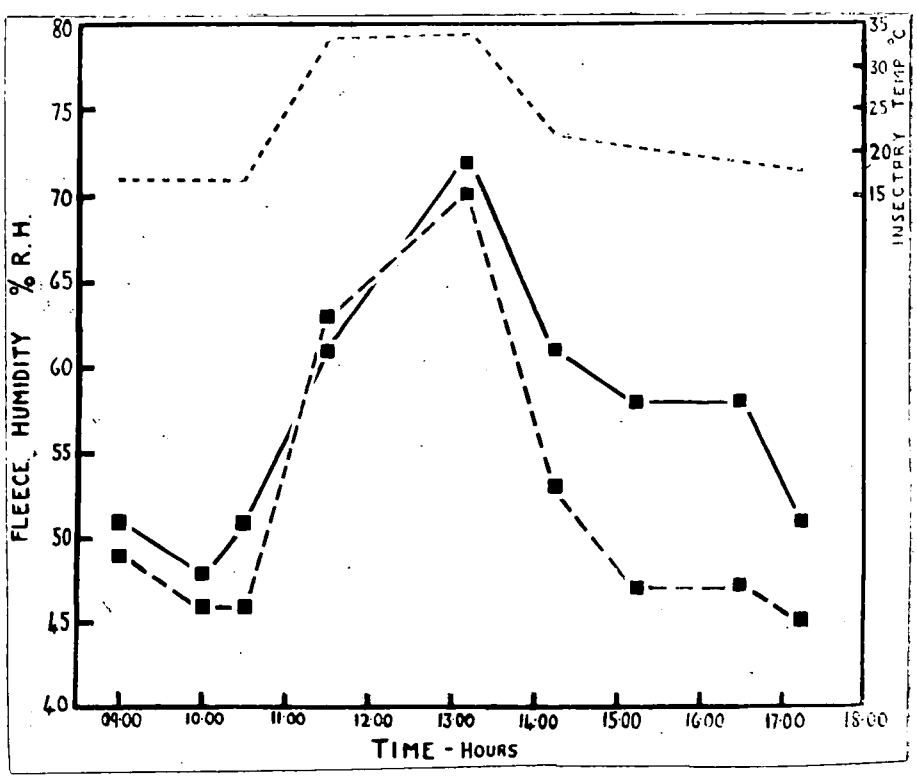

FIG. 27. Fleece R.H. fluctuations in sheep 43 (— — —) and 53 (——) ; ■, midback position; --------, insectary temperature. (Expt. 5, Appendix, page 233-234.)

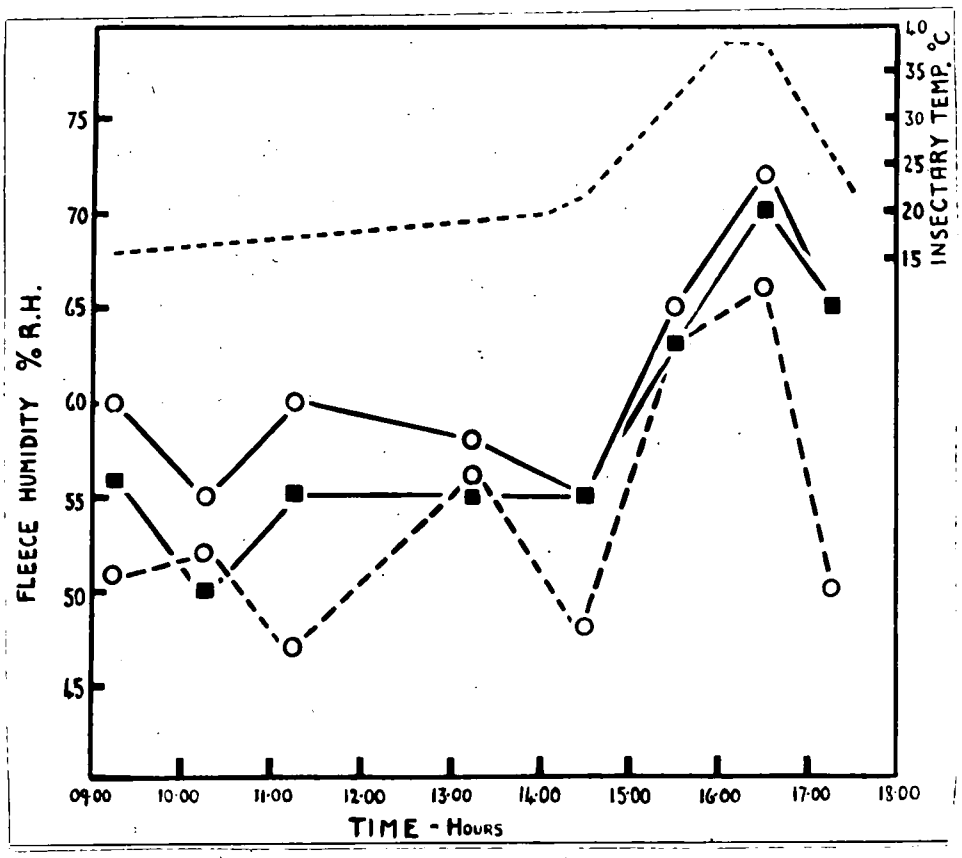

FIG. 28. Fleece R.H. fluctuations in sheep 43 (– – –) and 53 (——); O, midback position; ■, R. crutch position; ------, insectary temperature. (Expt. 3. Appendix, page 231-232)

thermograph records has been corrected. It was found that the maximum temperature recorded on the thermograph record coincided to within ± 1°C of the temperature recorded by the mercury thermometer.

Readings of fleece R.H. were carried out on the sheep by means of 2 cm. double strips (page 120) in a manner already described (page 99-100).

Results. The results of two preliminary experiments were unsatisfactory because the fleece R.H's of the 2 sheep were found to be already high (mainly over 60%) without their having been subjected to hot atmospheres. (These initially high fleece humidities may have been due to 'psycholgical' sweating on the part of the sheep, owing to disturbance and strange surroundings). The sheep were allowed 2 days to become accustomed to the insectary conditions and frequent handling.

Five separate experiments were carried out. The procedure in each was the same. Successive fleece R.H. readings were taken at standard points on the sheep commencing

before the insectary temperature was raised and continuing at intervals of 0.5 or 1.0 hr. during the period of high temperature and ending after the temperature had been allowed to drop again. Insectary R.H. (measured by thermohygrograph) fluctuated in a similar manner during all experiments, being 70-80% during low temperatures and falling to 45-55% by the time the temperature rose to 33-37°C. The humidity record during high temperatures is probably not accurate owing to the long temperature range.

Some records of fleece R.H. from selected positions during expts. 2, 3 and 5 are given in figs. 26-28 (page 150-1) together with the corrected thermograph records in each case. The full results will be found in the Appendix (page 229-234).

Fig. 26 shows pronounced changes in skin surface R.H. at one position in each of sheep 43 and 53. Great increases in R.H. near the skin occurred as the insectary temperature rose. These high humidities were maintained at 70-80% R.H. from 11.45 until about 13.30 hr., while the temperature remained at 33-37°C. From 13.30 to 16.30 hr. the insectary temperature fell from 37° to 20°C, and during the same period fleece R.H. at the 2 positions fell to the region of 55-65%. Changes of the same type occurred during the experiment in R.H. at 7 other positions on these 2 sheep.

The rapid rise and fall in R.H. near the skin correlated with rise and fall in external temperature, suggest strongly that rapid sweating was induced in the sheep during the period of high temperature. In Fig. 26 is included the graph of R.H. at 2 cm. off the skin at the withers position of sheep 53. It indicates that R.H. changes at this point were much less than at the skin surface. This shows that during the period of high temperature the main R.H. changes occurred in the basal position of the fleece next to the skin, and confirms the hypothesis that the changes were due to sweating, and

TABLE 29

Proportion of cobalt-chloride papers showing sweating in hot atmosphere experiments

	When heaters were OFF		When heaters were ON	
Expt.No.	No sweating	Sweating	No sweating	Sweating
1	12	0	7	11
2	30	1	16	20
3	25	0	10	5
4	30	0	5	2
5	23	0	4	8
Totals	120	1	42	46

not primarily to changes which occurred in the R.H. of the insectary. The latter factor may have had some effect on fleece R.H's, particularly at 2 cm. off the skin.

In Expt.3 (Fig. 28) during the period 09.15 - 14.30 hr. when the insectary temperature rose very slowly from 16° to about 22°C, fleece R.H's fluctuated in an indeterminate manner. From 14.35 to 16.30 hr. the temperature rose from 22 to 38°C and simultaneously the fleece R.H. at all 5 of the positions used (3 only are graphed in Fig. 28) rose sharply to the region 65-75% and fell later as the temperature dropped to 22°C. The R.H's in Expt. 5 (Fig. 27) and in Expts. 1 and 4 underwent comparable changes, and bear out the suggestion that sweating occurred in the sheep during high temperatures.

An analysis was made of the proportion of cobalt-chloride paper strips used which showed rapid sweating. The criterion adopted was the occurrence of a higher R.H. next to the skin than at 2 cm. off it, as was done with papers used in field work (page 117). The results are given in Table 29 (page 153). It will be seen from this table that while the insectary heaters were switched on i.e. when the temperature was high,

or rising, 46 readings showed sweating by the above criterion, while 42 did not. When the heaters were off, i.e. when the temperature was low or rapidly falling, only 1 reading showed sweating while 120 did not. This shows fairly conclusively that sweating was induced by the high temperatures, and disproves the possible objection to the work that the sheep would have sweated in any case without their subjection to high temperatures. Table 29 and the fleece R.H. curves given in figs. 26-28 also show that sweating commenced in the sheep almost simultaneously on raising the temperature, and ceased promptly as soon as the temperatures fell. It should of course be pointed out that the temperature fluctuations to which the sheep were subjected were very large compared with those encountered in the field in this country.

DISCUSSION

The results of running and hot atmosphere experiments on sheep described above leave little doubt that some sheep at least do sweat to some extent in order to regulate body temperature. Since the experiments were carried out on such small numbers of sheep detailed discussion of the possible role of sweating in the development of sheep blowfly myiasis is not at present warranted. The detection of rapid and continuous sweating on sheep earlier in the work (page 111) and the demonstration that humidities high enough for blowfly egg survival and myiasis development were maintained in their fleeces, indicates the possible importance of sweating in this respect.

The hot atmosphere experiments fit in with those of Lee and Robinson (1941) on weight losses from Merino sheep at high temperatures.

SUMMARY

1) Four experiments on 2 dry-fleeced sheep in cool weather showed that muscular exertion caused their fleece R.H's to rise for a period, and then to drop to their former low levels. This is interpreted as evidence for sweating in these sheep, caused by muscular exertion.

2) In 1 sheep, sweating at the withers position but not at the midback was detected in 3 experiments. This suggests that sweating may vary markedly between various points fairly close together.

3) Increases in fleece R.H. near the skin were also detected in sheep when subjected to hot atmospheres (25-37°C). The synchronization of fleece R.H. changes with those of temperature indicate that sheep sweat in hot atmospheres in order to regulate body temperature.

4) The results are regarded as being purely preliminary in nature.

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GENERAL SUMMARY OF WORK

Of the blowflies whose eggs were studied, those of L.sericata were found to be the most desiccation resistant. Those of L.caesar, L.illustris and P.terranovae were found to need rather higher humidities, while those of C.erythrocephala and C.vomitoria needed both higher humidities and lower temperatures to survive.

Experiments indicated that blowfly eggs are waterproofed by a lipoid layer, situated between the chorion and the chorionic vitelline membrane, in a manner fundamentally similar to the egg of Rhodnius and to insect cuticle as elucidated by other workers.

The sensitivity of the hatching of blowfly eggs to R.H., so well demonstrated by Davies & Hobson by transferring eggs from saturated air shortly before hatching into 50% R.H. and thus preventing hatching, is amplified by the observation of humidity-dependent shape shanges in the egg shell in the course of the present work.

Fleece R.H. observations bore out the frequent occurrence of low R.H.'s there, stressed by previous investigators. The fluctuations of fleece R.H. due to rain or dew were prominent features of the present study, and their possible role in genesis of strike has been pointed out.

The desirability of a biological study of the fleece microclimate, by studying the fate of blowfly eggs in fleeces is indicated by the results of preliminary experiments on these lines described in this thesis.

The occurrence of high R.H's close to the skin in sheep under certain conditions led to the detection of sweating, which was subsequently demonstrated by means of muscular exertion of the animals and subjecting them to hot atmospheres. Experiments showed that sweating might provide suitable conditions in fleeces for blowfly eggs, a state of affairs not recognized hitherto.

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APPENDIX

LABORATORY RESULTS

WATER-BATH EXPERIMENTS

EXPLANATORY NOTE ON CLASSIFICATION OF THE VARIOUS STAGES REACHED BY EGGS IN WATER-BATH HUMIDITY EXPERIMENTS

In the following tables giving the results of the above experiments, and in various Tables in the text giving the proportion of the eggs used reaching various stages of development, the columns are headed as follows:-

H = Hatched; PH = Prehatching stage; LMH = Late Mouth-Hook stage; EMH = Early Mouth-Hook stage; and E or Early = Early stage.

This classification is to a large extent arbitrary. In tables where only an 'EMH' or 'MH' column is given and 'LMH' is omitted, the eggs in that column represent those reaching the 'EMH' stage plus those reaching the 'LMH' stage. The distinctions between eggs reaching 'LMH' stage and 'EMH' stage were not well defined and their separation is perhaps too arbitrary to warrant classification into these two categories.

A description of the appearance of the various stages is now given:-

HATCHED - The larva had vacated the shell. The few cases where the larva had begun to hatch but had succumbed half way out of the shell were classified as having hatched.

PREHATCHING STAGE - A fully developed larva was present in a contracted state in the shell, indicating that it had moved within it before succumbing. The tracheal system was full of gas.

LATE MOUTH-HOOK STAGE - The bucco-pharyngeal armature was visible together with the dark circles of minute cuticular spines near the junctions of the body segments. The cuticle was distinct but had not adopted its final semi-opaque white character of the fully developed larva. The 'larva' at this stage died without visible evidence of any of the prehatching movements.

EARLY MOUTH-HOOK STAGE - The only larval organs visible were the dark sclerites of the bucco-pharyngeal apparatus. No larval cuticle could be detected with cursory examination under a binocular microscope.

EARLY STAGE - No larval organs were distinguishable under a binocular microscope (30 x)

An examination of L.sericata eggs at 100% R.H. 35°C, showed that the above stages were reached in the following times after incubation immediately on laying:- Early Mouth-hook stage in 7.0-7.5 hr; Late Mouth-hook stage in 7.5-8.0 hr; Prehatching stage in 8.0-9.0 hr; Hatching in 9.0-9.5 hr.

LUCILIA SERICATA

Expt. No.	Temp. °C.	%RH	% Stages reached by eggs H	PH	LMH	EMH	Early	No. of eggs	Remarks
22	30	100	87.5	0.0	0.0	17.9	1.6	56	
	37	65	13.0	38.9	1.9	0.0	46.2	54	
	37	60	14.0	28.1	0.0	3.5	54.4	57	
	37	55	0.0	48.0	6.0	12.0	34.0	50	
	37	50	0.0	5.6	0.0	27.8	66.6	36	
	37	45	0.0	0.0	0.0	12.4	87.6	41	
23	30	100	92.5	0.0	0.0	2.8	5.7	144	
	37	65	38.8	36.9	2.9	8.7	12.7	103	
	37	60	12.6	57.3	1.8	0.9	23.4	111	
	37	55	1.5	52.7	14.8	11.7	2.3	95	
	37	50	0.0	6.9	6.9	37.9	48.3	58	
	37	45	0.0	6.3	10.1	29.2	35.4	79	
30	30	100	79.5	0.0	0.0	0.0	20.5	39	
	37	70	84.8	9.1	0.0	0.0	6.1	33	
	37	65	48.3	29.6	0.0	0.0	22.1	27	
	37	60	35.8	35.8	0.0	3.6	25.8	28	
	37	55	3.7	81.5	0.0	0.0	14.8	27	
	37	50	0.0	69.7	0.0	3.3	27.0	33	
9	30	100	100.0	0.0	0.0	0.0	0.0	57	
	38	100	18.2	5.5	0.0	32.7	43.6	55	
	38	95	78.4	5.4	2.7	5.4	8.1	37	
	38	90	21.4	14.3	0.0	5.3	59.0	56	
	38	85	94.0	0.0	2.0	2.0	2.0	51	

LUCILIA SERICATA (continued)

Expt. No.	Temp. °C.	%RH.	% Stages reached by eggs H	PH	LMH	EMH	Early	No. of eggs.	Remarks
	30	100	59.2	0.0	0.0	4.5	36.3	22	N.B. Low fertility of controls
	38	100	77.8	0.0	0.0	5.6	16.6	18	
10	38	90	73.4	0.0	0.0	0.0	26.6	15	
	38	85	75.0	4.2	4.2	4.2	12.4	24	
	38	80	58.3	16.6	8.3	16.8	0.0	12	
	30	100	87.5	0.0	0.0	4.2	8.3	48	
	38.8	100	22.0	2.0	6.0	8.0	62.0	50	
	38.8	95	11.0	1.4	13.7	6.8	67.1	73	
16	38.8.	90	62.5	6.2	0.0	9.4	21.9	64	
	38.8	85	43.3	14.9	1.4	6.7	33.7	74	
	38.8	80	7.8	6.5	2.6	11.7	71.4	77	
	30	100	94.6	0.0	0.0	1.8	3.6	57	
	39	100	7.4	3.7	5.6	3.7	79.6	54	
	39	95	5.7	22.7	11.6	32.0	32.0	53	
14	39	90	15.2	2.2	0.0	17.4	65.2	46	
	39	85	24.1	20.4	3.7	3.7	48.1	54	
	39	80	24.0	18.0	4.0	10.0	44.0	50	
	30	100	81.6	0.6	1.3	3.8	12.7	157	
	40	100	9.9	1.0	10.8	13.7	64.6	102	
	40	95	0.0	0.0	0.0	0.0	100.0	160	
33	40	90	0.0	4.5	0.0	10.2	85.3	157	
	40	85	0.0	5.5	3.2	10.3	81.0	126	
	40	80	0.0	0.9	0.0	7.5	91.6	106	

LUCILIA SERICATA (continued)

Expt. No.	Temp. °C.	%RH	% Stages reached by eggs					No. of eggs.	Remarks
			H	PH	LMH	EMH	Early		
34	30	100	73.2	4.6	0.0	3.5	18.7	131	
	40	100	0.0	0.0	0.0	0.0	100.0	132	
	40	95	1.2	0.6	1.2	4.0	93.0	171	
	40	90	0.0	0.0	0.0	1.2	98.8	97	
	40	85	0.0	0.0	0.0	1.0	99.0	106	
	40	80	0.0	0.0	0.0	0.0	100.0	138	

LUCILIA CAESAR

Expt. No.	Temp. °C.	%RH	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
	30	100	76.7	0.0	0.0	3.5	19.8	150	
	37	70	4.0	17.5	0.8	10.3	67.4	126	
36	37	65	0.0	2.9	0.0	20.7	76.4	140	
	37	60	0.0	1.8	0.0	7.1	92.1	112	
	37	55	0.0	0.0	0.0	1.0	99.0	140	
	30	100	69.2	0.0	0.0	1.7	29.1	120	
	37	65	0.0	1.2	0.0	5.1	93.7	98	
37	37	60	0.0	0.0	0.0	0.0	100.0	100	
	37	55	0.0	0.0	0.0	0.0	100.0	110	
	30	100	33.3	0.0	0.0	0.0	66.7	33	Low fertility of controls
	38	80	0.0	0.0	0.0	0.0	100.0	35	
	38	75	0.0	0.0	0.0	0.0	100.0	39	
38	38	70	0.0	0.0	0.0	0.0	100.0	46	
	38	65	0.0	0.0	0.0	0.0	100.0	40	
	38	60	0.0	0.0	0.0	0.0	100.0	25	
	30	100	88.8	0.0	0.0	5.6	5.6	18	
	38	100	0.0	9.5	33.3	19.1	38.1	21	
	38	95	59.3	0.0	3.7	0.0	37.0	27	
40	38	90	58.7	0.0	3.5	0.0	37.8	29	
	38	85	57.7	11.5	3.8	3.8	23.2	26	
	38	80	40.6	15.7	3.1	0.0	40.6	32	

LUCILIA CAESAR (continued)

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
	30	100	65.2	0.0	0.0	0.0	34.8	46	
	39	100	0.0	0.0	0.0	0.0	100.0	46	
41	39	95	0.0	0.0	0.0	0.0	100.0	60	
	39	85	0.0	0.0	0.0	0.0	100.0	40	
	39	80	0.0	0.0	0.0	0.0	100.0	59	
	30	100	86.3	1.3	1.2	0.0	11.2	80	
	40-40.5	100	0.0	0.0	0.0	0.0	100.0	69	
	40-40.5	95	0.0	0.0	0.0	0.0	100.0	65	
42	40-40.5	90	0.0	0.0	0.0	0.0	100.0	48	
	40-40.5	85	0.0	0.0	0.0	0.0	100.0	63	

LUCILIA ILLUSTRIS

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs H	PH	LMH	EMH	Early	No. of eggs	Remarks
	30	100	22.9	0.9	1.2	6.1	69.9	212	Low fertility in control eggs. Temp. rose to 38.5°C by the end of the experiment.
	37	70	1.6	9.8	0.0	7.1	81.5	184	
	37	65	0.0	5.5	0.0	9.8	84.7	163	
28	37	60	0.0	0.0	0.0	2.5	97.5	162	
	37	55	0.0	0.0	0.0	0.0	100.0	178	
	37	50	0.0	0.0	0.0	0.0	100.0	93	
	30	100	44.5	0.0	0.0	7.0	48.5	128	Low fertility of controls. Temp. rose to 37.5°C by the end of the experiment.
	37	70	28.2	14.5	0.0	8.1	49.2	124	
	37	65	3.8	14.4	0.0	3.8	78.0	104	
29	37	60	0.0	9.4	0.9	5.7	84.0	106	
	37	55	0.0	0.0	0.0	0.0	100.0	110	
	37	50	0.0	0.0	0.0	0.0	100.0	97	
	30	100	38.4	1.4	0.0	4.1	56.1	73	NB. Low fertility of controls.
	37	70	3.5	27.9	1.2	5.8	61.6	86	
	37	65	0.0	4.6	0.0	5.7	89.7	87	
30	37	60	0.0	2.0	0.0	9.0	89.0	100	
	37	55	0.0	0.0	0.0	2.4	97.6	84	
	37	50	0.0	0.0	0.0	0.0	100.0	90	
	30	100	39.1	0.0	0.7	0.7	59.5	138	NB. Low fertility of controls.
	37	70	3.1	8.1	0.0	1.9	86.9	160	
35	37	65	0.9	25.0	0.0	16.6	57.5	108	
	37	60	0.0	11.5	0.0	11.5	77.0	78	
	37	55	0.0	0.0	0.0	2.8	97.2	68	

LUCILIA ILLUSTRIS (continued)

Expt. No.	Temp. °C.	%RH.	% Stages reached by eggs H	PH	LMH	EMH	Early	No. of eggs.	Remarks
37	30	100	68.0	1.2	1.2	9.0	20.6	120	
	37	65	0.0	26.5	1.7	12.8	59.0	117	
	37	60	0.0	0.0	0.0	4.2	95.8	144	
	37	55	0.0	0.0	0.0	1.0	99.0	108	
9	30	100	9.8	0.0	0.0	3.3	86.9	61	Low fertility of control eggs.
	38	100	5.0	0.0	0.0	3.3	91.7	60	
	38	95	5.6	2.8	1.4	12.4	77.8	72	
	38	90	2.9	5.9	0.0	7.4	83.8	68	
	38	85	10.1	8.7	0.0	13.0	68.2	69	
31	30	100	65.3	0.8	0.0	0.0	33.9	121	
	38	80	0.0	5.5	2.8	11.7	80.0	128	
	38	75	0.0	0.0	0.0	8.5	91.5	118	
	38	70	0.0	0.0	0.0	4.3	95.7	141	
	38	65	0.0	0.0	0.0	0.0	100.0	136	
	38	60	0.0	0.0	0.0	0.0	100.0	134	
32	30	100	32.0	4.0	0.0	8.0	56.0	25	N.B. Low fertility of controls
	39	100	0.0	0.0	22.2	7.4	70.4	27	
32	39	90	0.0	10.5	10.5	31.6	47.4	19	
	39	85	0.0	9.5	0.0	33.3	57.2	21	
	39	80	0.0	0.0	0.0	0.0	100.0	23	
39	30	100	32.6	0.0	0.0	11.2	56.2	89	
	40	100	0.0	0.0	0.0	0.0	100.0	89	

LUCILIA ILLUSTRIS (continued)

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
39 (continued)	40	95	0.0	0.0	0.0	0.0	100.0	112	
	40	90	0.0	0.0	0.0	0.0	100.0	109	
	40	85	0.0	0.0	0.0	0.0	100.0	73	
	40	80	0.0	0.0	0.0	0.0	100.0	83	

PHORMIA TERRA-NOVAE

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
22	30	100	86.9	0.0	2.1	1.3	9.7	145	
	37	65	0.0	1.5	1.4	18.7	78.4	139	
	37	60	0.0	0.0	0.0	5.9	94.1	118	
	37	55	0.0	0.0	0.0	0.0	100.0	112	
	37	50	0.0	0.0	0.0	0.0	100.0	62	
	37	45	0.0	0.0	0.0	0.0	100.0	98	
23	30	100	78.5	0.0	5.0	0.0	17.5	40	
	37	65	6.5	6.5	3.2	32.3	52.5	31	
	37	60	2.9	2.9	2.8	20.0	71.4	35	
	37	55	0.0	0.0	0.0	0.0	100.0	50	
	37	50	0.0	0.0	0.0	0.0	100.0	45	
	37	45	0.0	0.0	0.0	0.0	100.0	33	
17	30	100	100.0	0.0	0.0	0.0	0.0	72	
	40	100	0.0	7.2	18.9	5.8	69.1	69	
	40	95	4.1	8.2	8.2	16.5	63.0	73	
	40	90	0.0	0.0	5.0	12.5	82.5	80	
	40	85	0.0	0.0	1.8	18.2	80.0	55	
	40	80	0.0	0.0	0.0	6.0	94.0	50	
16	30	100	85.0	1.2	0.0	1.1	12.7	86	
	38.8	100	13.8	6.4	13.8	13.8	52.2	109	
	38.8	95	43.2	311	12.1	6.2	35.4	65	
	38.8	90	47.3	22.8	2.6	0.8	26.5	114	
	38.8	85	27.6	9.2	0.0	1.2	62.0	87	
	38.8	80	22.3	39.5	2.9	5.7	29.6	139	

CALLIPHORA ERYTHROCEPHALA

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
21	30	100	41.0	12.4	8.6	22.2	16.8	81	
	30	65	49.4	21.6	0.0	2.5	26.5	79	
	30	60	15.6	26.6	9.4	12.5	15.9	64	
	30	55	4.5	23.9	16.4	11.9	43.3	67	
	30	50	0.0	5.6	19.6	37.4	37.4	72	
	30	45	0.0	2.8	1.4	26.4	69.4	72	
25	30	100	80.0	10.0	0.0	5.0	5.0	20	Temperature rose to 31.5°C for short time during expt.
	30	45	0.0	0.0	0.0	0.0	100.0	30	
26	30	100	59.2	4.5	0.0	4.6	31.7	22	
	30	55	0.0	71.5	0.0	0.0	28.5	22	
	30	50	0.0	30.0	0.0	5.0	65.0	20	
	30	45	0.0	13.6	0.0	22.8	63.6	14	
27	30	65	13.3	13.4	0.0	5.3	68.0	75	No controls in this expt. Eggs accidentally killed.
	30	60	8.0	3.4	1.1	6.8	80.7	88	
	30	55	0.8	13.9	0.8	6.5	77.7	122	
	30	50	0.0	8.5	3.2	6.4	81.9	94	
	30	45	0.0	1.3	1.3	18.4	79.0	76	
43	30	100	72.3	2.8	5.6	0.0	19.3	36	
	30	60	6.8	31.8	15.9	2.3	43.2	44	
	30	55	4.2	22.9	8.3	14.6	50.0	48	
	30	50	0.0	0.0	0.0	0.0	100.0	54	
	30	45	0.0	0.0	0.0	0.0	100.0	13	

CALLIPHORA ERYTHROCEPHALA (continued)

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
12	30	100	61.1	2.8	22.2	0.0	13.9	36	
	34	100	6.1	3.0	33.1	33.1	24.7	66	
	34	95	2.0	5.6	15.1	54.7	22.4	53	
	34	90	0.0	18.6	18.5	44.5	18.4	54	
	34	85	0.0	7.8	9.4	56.3	26.5	64	
	34	80	0.0	3.3	0.0	13.3	83.4	60	
13	30	100	73.8	2.3	11.9	4.8	7.2	42	
	34	100	3.1	0.0	49.0	47.0	1.9	47	
	34	90	1.9	11.5	17.3	34.6	34.7	52	
	34	85	0.0	11.5	15.4	50.0	23.1	52	
	34	80	0.0	3.7	5.6	46.3	44.4	54	
18	30	100	66.6	7.9	9.5	9.5	6.5	63	
	34	80	0.0	0.0	0.0	25.0	75.0	48	
	34	75	0.0	0.0	0.0	0.0	100.0	53	
	34	70	0.0	0.0	0.0	0.0	100.0	54	
	34	65	0.0	0.0	0.0	0.0	100.0	54	
	34	60	0.0	0.0	0.0	0.0	100.0	34	
1	30	100	68.6	5.6	3.7	0.0	22.1	54	
	35	100	1.0	1.0	13.0	15.0	70.0	100	
	35	95	0.0	1.0	8.6	22.0	68.4	104	
	35	90	0.0	0.0	1.1	9.2	89.7	87	
2	30	100	77.3	5.7	0.0	0.0	17.0	35	
	35	100	0.0	14.7	5.9	11.8	67.6	34	
	35	95	0.0	12.9	19.5	9.6	58.0	31	

CALLIPHORA ERYTHROCEPHALA (continued)

Expt. No.	Temp. C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
2 (continued)	35	90	0.0	0.0	5.0	5.0	90.0	40	
3	30	100	74.3	13.6	3.1	0.0	9.0	66	
	35	85	0.0	0.0	0.0	0.0	100.0	246	
4	30	100	63.2	0.0	5.3	5.3	26.2	19	
	35	100	0.0	4.5	18.1	9.1	68.3	22	
	35	95	0.0	0.0	31.8	18.2	50.0	22	
	35	90	0.0	0.0	0.0	4.5	95.5	22	
	35	85	0.0	0.0	0.0	0.0	100.0	20	
5	30	100	91.0	3.7	0.0	3.7	1.6	54	
	35	100	0.0	0.0	9.4	24.1	66.5	54	
	35	95	0.0	0.0	0.0	5.7	94.3	70	
	35	90	0.0	0.0	0.0	2.9	97.1	68	
	35	85	0.0	0.0	0.0	0.0	100.0	80	
11	30	100	68.7	2.0	9.8	13.6	5.9	51	
	35	100	0.0	0.0	0.0	21.0	79.0	100	
6	30	100	95.0	0.0	0.0	2.0	0.0	42	
	36	100	0.0	0.0	0.0	0.0	100.0	48	
	36	95	0.0	0.0	0.0	0.0	100.0	42	
	36	90	0.0	0.0	0.0	0.0	100.0	42	
	36	85	0.0	0.0	0.0	0.0	100.0	46	

CALLIPHORA ERYTHROCEPHALA (continued)

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
	30	100	38.1	1.2	4.8	3.6	52.3	84	N.B. Low fertility of controls
	36	100	0.0	0.0	0.0	0.0	100.0	88	
7	36	95	0.0	0.0	0.0	0.0	100.0	70	
	36	90	0.0	0.0	0.0	0.0	100.0	80	
	36	85	0.0	0.0	0.0	0.0	100.0	74	

CALLIPHORA VOMITORIA

Expt. No.	Temp. °C.	%RH.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
21	30	100	86.4	1.1	3.4	4.6	4.5	88	
	30	65	11.0	61.0	9.4	6.2	12.4	64	
	30	60	1.5	73.6	8.8	1.5	14.6	68	
	30	55	0.0	21.3	21.3	30.8	26.6	65	
	30	50	0.0	11.2	11.2	38.8	38.8	72	
	30	45	0.0	1.4	1.4	34.3	62.9	70	
12	30	100	58.6	6.9	12.2	0.0	24.3	58	
	34	100	78.9	5.8	5.8	1.9	7.6	52	
	34	95	90.0	2.9	4.3	0.0	2.8	70	
	34	90	53.8	35.4	3.8	1.5	6.5	65	
	34	85	23.2	43.5	2.9	8.7	21.7	69	
	34	80	7.2	85.5	0.0	2.9	4.4	69	
18	31	100	94.3	2.9	0.0	0.0	2.8	68	
	34	80	42.5	57.5	0.0	0.0	0.0	66	
	34	75	0.0	61.2	1.9	12.9	24.0	54	
	34	70	0.0	11.8	7.9	36.8	43.5	76	
	34	65	0.0	1.3	2.7	44.0	52.0	75	
	34	60	0.0	0.0	0.0	0.0	100.0	73	
5	30	100	68.0	5.7	0.0	1.9	24.4	53	
	35	100	39.2	7.8	9.8	5.9	37.3	51	
	35	95	30.4	12.2	9.1	13.6	34.7	66	
	35	90	1.4	14.8	8.2	36.5	39.1	74	
	35	85	0.0	0.0	8.2	9.8	82.0	61	

CALLIPHORA VOMITORIA (continued)

Expt. No.	Temp. °C.	%R.H.	% Stages reached by eggs					No. of eggs	Remarks
			H	PH	LMH	EMH	Early		
	30	100	100.0	0.0	0.0	0.0	0.0	13	
8	35	100	90.0	6.0	4.0	0.0	0.0	50	
	35	90	65.5	20.7	13.8	0.0	0.0	29	
	30	100	35.8	14.3	4.8	0.0	45.1	42	
	35	100	84.0	2.3	2.3	4.5	6.9	44	
11	35	90	47.5	11.9	15.2	3.4	22.0	59	
	35	85	23.0	44.3	9.6	9.6	13.5	52	
	35	80	31.4	1.5	7.5	8.9	50.7	67	
	30	100	89.3	3.8	0.9	0.0	6.0	104	
	35	80	0.0	67.0	7.2	21.9	3.9	128	
	35	75	0.0	4.2	10.0	63.3	22.5	120	
20	35	70	0.0	0.0	2.0	44.0	54.0	50	
	35	65	0.0	0.0	0.0	0.0	100.0	76	
	35	60	0.0	0.0	0.0	0.0	100.0	68	
	30	100	100.0	0.0	0.0	0.0	0.0	54	
	36	100	6.0	40.0	42.0	12.0	0.0	50	
6	36	95	0.0	45.3	21.4	33.3	0.0	42	
	36	90	0.0	0.0	0.0	50.0	50.0	48	
	36	85	0.0	0.0	0.0	18.7	81.3	48	
	30	100	71.2	9.6	5.8	3.8	9.6	52	
	36.8	100	0.0	0.0	0.0	0.0	100.0	64	
15	36.8	95	0.0	0.0	0.0	0.0	100.0	66	
	36.8	90	0.0	0.0	0.0	0.0	100.0	63	

DETAILS OF POSITIONS OF FLEECE R.H. READINGS

AND OF THE SHEEP USED IN FIELD WORK 1946 - 48

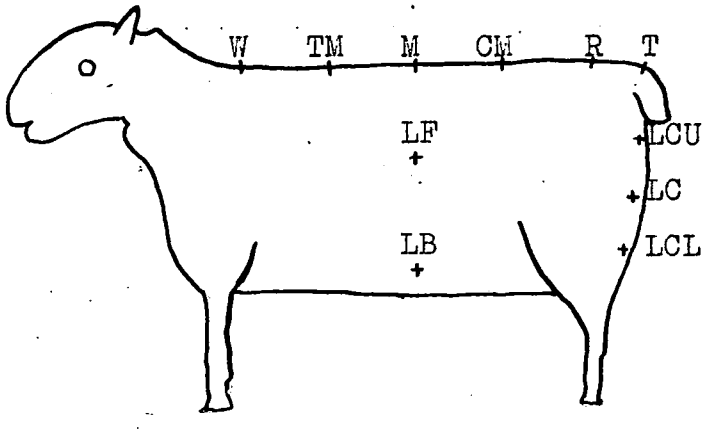
APPENDIX

KEY TO POSITION SYMBOLS IN TABLES OF FLEECE R.H. READINGS.

W	=	Withers	LCU	=	Upper left crutch
M	=	Midback	LCL	=	Lower left crutch
R	=	Rump	RB	=	Right belly
T	=	Tailhead	LB	=	Left belly
RC	=	Right crutch	RF	=	Right flank
RCU	=	Upper Right crutch	LF	=	Left flank
RCL	=	Lower Right crutch	CM	=	Quarter back
LC	=	Left crutch	TM	=	Threequarter back.

These position symbols are used in all tables in the text, relating to fleece humidities, in the tabulated results of fleece R.H's and in the results of hot atmosphere experiments given in the Appendix.

Diagram of Location of above positions. Sheep viewed from left side only. 'Right' positions correspond to 'left', but are on the other side of the body.



Notes on Sheep used for fleece humidity readings. 1946.

Sheep 1. 'Mule' x Suffolk Lamb. Female. Age 6 - 7 months. Close compact fleece of Suffolk Type, with fine wiry wool. Staple length about 8 cm.

Sheep 2. Swaledale x Border Leicester Lamb. Female. Age 3 - 4 months. Short fleece, close in comparison with sheep 3, but very open compared with sheep 1. No 'shed' in the wool along the back. Wool falls into small crimped locks. Staple length about 5 - 6 cm.

Sheep 3. Swaledale x Border Leicester Lamb. Female. Age 3 - 4 months. Very slack fleece, long course fibres, with a marked 'shed' in the fleece along the back. Staple length about 5 - 6 cm.

Sheep 4. Swaledale x Herdwick wether. Age 18 months. Fleece entirely brown, short, and fairly close. Staple length about 5 - 6 cm.

Sheep 5. Herdwick (some Swaledale blood) wether. Age 18 months. Fleece shorter than sheep 4. Staple length only 4 - 5 cm. Fleece fairly close. Wool fibres coarse and stiff.

Sheep 6. Swaledale x Border Leicester Lamb. Male. Age 3 - 4 months. Fleece similar to sheep 2.

Sheep 7. 'Half-Bred'x Oxford Lamb. Male. Age 6 - 7 months. Close fine fleece. Wool less wiry than in sheep 1, fleece of 'Oxford' type. Length of staple about 8 cm.

Sheep 8. Swaledale x Herdwick wether. Age 18 - 19 months. Coarse fleece rather open in character.

Sheep 9. Swaledale x Herdwick wether. Age 18 months. Finer fleece than sheep 8 but even more open.

Sheep 10. Pure Swaledale Ram (possible some Herdwick blood). Age 18 months. Good thick close fleece, characteristic of best Swaledale sheep. No sheds in the fleece. Staple length about 8 cm.

Sheep 11. Herdwick wether (some Swaledale blood). Age 18 - 19 months. Fleece similar to sheep 8.

Sheep 12. Suffolk x ? Lamb. Female. Age 11 months. Fleece similar to sheep 1, but by now longer in staple.

Sheep 13. Half-Bred x Oxford. Male. Age 11 months. Fleece similar to sheep 7, but by now longer in staple.

Sheep 15. Half-Bred x Oxford Lamb. Sex not recorded. Age 6 - 7 months. Fleece similar to sheep 7.

Sheep 16. Half-Bred x Suffolk Lamb. Sex not recorded. Age 6 - 7 months. Fleece similar to sheep 1.

Sheep 17. Half-Bred x Suffolk Lamb. Sex not recorded. Age 6 - 7 months. Fleece similar to sheep 1.

Sheep 18. Half-Bred x Oxford Lamb. Female. Age 6 - 7 months. Fleece similar to sheep 7.

Sheep 19. Half-Bred x Oxford Lamb. Male. Age 6 - 7 months. Fleece similar to sheep 7.

Sheep 20. Swaledale x Border Leicester Lamb. Age 3 - 4 months. Sex not recorded. Moderately dense fleece, with fairly fine wool but with some sign of a 'shed' in the fleece along the midback. Fleece therefore intermediate between sheep 2 and 3. Staple length 5 - 6 cm.

Sheep 21. Herdwick Ram. Age 2 or 3 years. Fleece coarse but fairly close. Staple length 5 - 6 cm.

Sheep 22. Herdwick wether. Age 2 or 3 years. Entirely unshorn. Very large thick fleece, and densely matted giving a thickness of fleece of about 13 - 16 cm.

Sheep 23. Swaledale Lamb. Female. Age 4 - 5 months. Fleece similar to sheep 10, but staple length shorter - about 6 cm.

Sheep 24. Herdwick Ram. Similar to sheep 21.

Sheep 25. Swaledale lamb. Details as for sheep 23.

Sheep 26. Swaledale x Border Leicester Lamb. Male. Age 5 - 6 months. Fleece similar to sheep 2.

Sheep 27 & 28. Herdwick Lambs. Sex not recorded. Age 4 - 5 months old. Good close fleeces.

Note on sheep used for fleece humidity readings. 1947

Sheep 29 - 42 These were a mixed batch of cross bred lambs, all either Border Leicester x 'Mule' or Oxford x 'Mule', and were 5-6 months old when used in experiments.

Note on sheep used for experiments in 1948

All were lambs born in the period Dec. 1947 - Jan. 1948.

Sheep 43 & 51 These were Suffolk x 'Half-Bred' crosses.

Sheep 47-49 & 53,56,61 These were Oxford x 'Half-Bred' crosses.

Sheep 44-46 & 50,52,54, 55,57-60, 62 These were Border Leicester x 'Half-Bred' crosses.

FLEECE R.H. READINGS, 1946

SHEEP AT HOUGHALL FARM, DURHAM

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface.	2cm. off skin.
3 July. 11.45 hrs. 22°C. 60% R.H. Bright Sun. Light westerly breeze.	1	RC	45	-
	15	T	51	-
	15	RF	65	-
	16	W	47	-
	16	M	below 40	-
	17	RC	below 40	-
4 July. 10.15 hrs. 20°C, rapidly rising. 85% R.H. Stiff westerly breeze.	7	LC	65	-
	7	T	65	-
	16	R	45	70
	16	TM	63	63
5 July. 10.15 hrs. 15°C. 75% R.H. Cloudy, still.	1	T	47	-
	1	LCU	45	-
	1	LCL	48	-
	1	RC	46	-
	1	TM	67	-
	1	W	65	-
	7	LF	67	-
	7	RC	50	-
	7	LCU	50	-
	7	LCL	53	-
	7	T	50	62
	7	M	55	60
	7	TM	55	-
	7	RF	60	-
6 July. 15.30 hrs. 19.8°C. 58% R.H. Sunny Periods. Fresh Westerly Breeze.	1	RC	below 40	-
	1	LCU	below 40	-
	1	LCL	below 40	-
	1	T	-	below 40

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface.	2cm. off skin.
6 July. 15.30 hrs. (continued)	1	M	65	44
	1	W	63	45
	1	TM	70	45
	1	RF	48	-
	1	LF	50	-
	19	T	45	-
	19	LC	below 40	-
	19	RC	below 40	-
	19	W	51	below 40
	19	LF	62	50
	19	M	65	-
8 July. 09.45 hrs. 22.5°C. 62% R.H. Bright sunny morning. No dew left @ 9.0 a.m.	1	LF	above 80	-
	1	M	75	-
	1	T	52	-
	1	W	above 80	-
	1	LC	63	-
	1	RC	67	-
	1	RF	above 80	-
	7	T	67	-
	7	M	above 80	-
	7	W	above 80	above 80
	7	LF	above 80	-
	7	RF	above 80	-
	16	T	67	48
	16	LC	63	-
	16	RC	above 80	-
	16	M	72	63
	16	W	77	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
9 July. 06.30 hrs. 12.5°C. 95% R.H. Cool misty morning, heavy dew. Sheep wet.	1	T	70	-
	1	M	76	-
	1	W	above 80	-
	1	RF	80	-
	1	LC	65	-
	1	RC	67	-
	7	T	65	above 80
	7	M	80	-
	7	RF	above 80	-
	7	LF	68	above 80
	7	W	75	100
	7	LC	68	75
	7	RC	75	-
	15	T	above 80	-
	18	RC	68	-
	18	T	77	-
9 July. 16.40 hrs. 21.8°C. 52% R.H. Warm, dull. Slight breeze. Sheep dry.	1	T	53	-
	1	LC	58	-
	1	RC	58	-
	1	M	100	70
	1	W	100	75
	1	RF	62	-
	1	LF	64	-
	7	T	63	-
	7	RC	62	-
	7	LC	57	-
	7	M	70	-
	7	W	above 80	-
	7	RF	67	-
	7	LF	70	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin
9 July. 16.40 hrs. (continued)	15	W	95-100	75
	16	W	75	66
10 July. 14.45 hr. 24.5°C. 56% R.H. Close and sunny. Slight westerly breeze.	1	T	56	45
	1	M	70	-
	1	W	100	68
	1	RC	60	-
	1	LC	62	-
	1	LF	58	-
	1	RF	56	-
	7	T	70	-
	7	M	above 80	below 40
	7	W	80	below 40
	7	LC	68	-
	7	RC	68	-
	7	RF	73	-
	7	LF	68	53
11 July. 14.30 hrs. 26.5°C. 43% R.H. Very bright sun, no cloud, no breeze.	1	T	60	40
	1	M	above 80	below 40
	1	RF	72	-
	1	LF	70	63
	1	W	100	75
	1	LC	63	-
	1	RC	63	-
	1	RB	63	-
	7	T	50	below 40
	7	M	73	45
	7	W	80	40
	7	LF	70	64
	7	RF	72	65

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface.	2cm. off skin.
11 July. 14.30 hrs. 26.5°C. (continued)	7	RC	68	-
	7	LC	66	-
	16	RB	55	-
12 July. 10.30 hrs. 24°C. 53% R.H. Clear, bright sun. Dew in early morning.	1	T	65	46
	1	M	above 80	above 80
	1	RF	77	-
	1	W	above 80	75
	1	RC	66	-
	1	LC	67	-
	1	LF	77	-
	7	T	63	-
	7	M	above 80	below 40
	7	W	73	43
	7	LC	63	-
	7	RC	65	-
	7	RF	70	-
	7	LF	70	-
13 July. 11.00 hrs. 23°C. 72% R.H. Very close, thick haze, thunker expected.	1	T	70	70
	1	M	above 80	-
	1	W	80	-
	1	RF	75	-
	1	RC	67	-
	7	T	above 80	-
	7	M	100	70
	7	W	75	-
	7	LC	above 80	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm. off skin.
15 July. 10.30 hr. 14°C. 84% R.H. Showery. Sheep wet on outsides of fleeces.	1	T	68	100
	1	M	80	100
	1	W	70	100
	1	LF	above 80	-
	1	RC	62	-
	1	LC	65	100
	7	T	66	above 80
	7	M	68	100
	7	W	67	100
	7	LC	55	63
	7	RC	55	65
	7	RF	67	80
	7	LF	67	-
16 July. 11.00 hr. 13.5°C. 74% R.H. Cool breeze, cloudy but bright periods. Sheep very wet.	1	T	67	-
	1	M	above 80	-
	1	W	66*	-
	1	RF	73	-
	1	LF	78	-
	1	RC	57	-
	1	LC	64	-
	7	T	65	-
	7	M	68	-
	7	W	76	-
	7	LC	62	-
	7	RC	56	-
	7	LF	78	-
	7	RF	78	-
	16	T	43	-
	16	M	68	-

* This position dusted with Derris at 10.45 a.m.13 July.

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2 cm. off skin.
16 July. 11.00 hr (continued).	16	W	65	-
	16	LF	77	-
19 July. 11.00 hr. 14°C. 89% R.H. Cloudy. Heavy rain during last 5 nights. Sheet wet.	2	T	47	65
	2	R	55	60
	2	M	64	75
	2	W	75	80
	2	RB	50	58
	2	RC	42	-
	3	T	50	72
	3	R	52	68
	3	M	58	70
	3	W	68	73
	3	RC	47	65
	3	RF	66	76
	20	T	-	63
	20	R	67	-
	20	M	63	-
	20	W	67	-
	20	RC	50	-
	20	RF	68	-
20 July. 09.50 hr. 16°C. 81% R.H. Cloudy but some sun. Sheep dry. No rain during night.	2	T	45	60
	2	M	65	65
	2	W	55	65
	2	RC	47	-
	2	LF	55	-
	2	LC	60	-
	3	T	47	50
	3	M	42	47
	3	W	45	-
	3	RC	43	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm. off Skin.
20 July. 09.50 hr. (continued)	3	RF	50	-
	3	RB	45	54
	5	W	50	60
	5	M	45	50
	5	RC	47	67
	20	T	45	52
	20	M	45	-
	20	W	47	-
	20	RC	45	-
	20	RF	-	57
	20	RB	45	-
22 July. 11.30 hr. 16°C. 90% R.H. Heavy rain during night - sheep very wet.	2	T	55	62
	2	M	above 80	-
	2	W	77	-
	2	RF	69	above 80
	2	RC	64	-
	3	T	65	-
	3	M	75	-
	3	W	70	80
	3	RF	69	-
	3	RC	55	62
	20	T	55	62
	20	M	68	-
	20	W	67	-
	20	RF	55	65
	20	RC	50	56
	20	RB	45	50
22 July. 20.00 hrs. 70°C. 90% R.H. All day good drying breeze. Sheep dry.	2	T	47	60
	2	M	-	67
	2	W	-	68

Date etc.	Sheep No.	Position	Relative Humidity % Skin Surface	Relative Humidity % 2cm off Skin.
22 July. 20.00 hrs. (continued)	2	RC	61	-
	2	RF	65	72
	2	RB	55	60
	3	T	50	56
	3	M	67	67
	3	W	68	68
	3	RC	46	58
	20	T	46	50
	20	M	56	-
	20	W	55	60
	20	RC	52	-
	20	RF	50	56
	20	RB	45	50
23 July. 11.10 hr. 20°C. 83% R.H. Still drying weather. Sheep dry.	2	T	44	-
	2	M	64	-
	2	W	55	65
	2	RC	50	-
	2	RF	65	-
	2	RB	45	-
	3	T	55	-
	3	M	65	-
	3	W	67	-
	3	RC	47	-
	3	RF	65	-
	3	RB	45	-
	20	T	45	-
	20	M	51	-
	20	W	58	-
	20	RC	48	-
	20	RF	59	-
	20	RB	66	-

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm off skin.
23 July. 11.10 hr. (continued)	1	T	50	55
	1	M	66	-
	1	W	66	66
	1	LF	72	-
	1	RC	50	-
	1	RB	55	67
23 July. 12.45 hr. Conditions as before.	1	T	45	60
	1	M	65	69
	1	W	65	67
	1	RF	67	70
	1	RC	49	64
	7	T	67	67
	7	M	56	61
	7	W	66	67
	7	RC	56	-
	7	LF	70	-
	7	LB	50	-
24 July. 11.30 hr. 19 C. 82% R.H. Some dew during night. Dull.	1	T	51	-
	1	M	49	-
	1	W	55	50
	1	RC	48	-
	1	RF	67	-
	1	RB	45	-
	2	T	50	50
	2	M	61	55
	2	W	50	-
	2	RC	50	55
	2	RF	55	-
	2	RB	45	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2 cm. off skin.
24 July. 11.30 hr. (continued)	3	T	42	46
	3	M	below 40	-
	3	W	43	-
	3	RC	42	-
	3	RF	54	-
	3	RB	below 40	-
	7	T	47	56
	7	M	47	-
	7	W	52	52
	7	RC	55	-
	7	RF	70	-
	7	RB	40	-
	20	T	45	50
	20	M	44	50
	20	W	49	-
	20	RC	55	-
	20	RF	53	-
	20	RB	below 40	-
25 July. 11.30 hrs. 20°C. 60% R.H. Fresh breeze. Sheep dry.	1	T	42	42
	1	M	40	below 40
	1	W	40	below 40
	1	RC	40	44
	1	RF	65	below 40
	2	T	below 40	below 40
	2	M	below 40	below 40
	2	W	below 40	below 40
	2	RC	40	-
	2	RF	below 40	below 40
	2	RB	below 40	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
25 July. 11.30 hrs. (continued)	3	T	below 40	-
	3	M	below 40	-
	3	W	43	below 40
	3	RC	below 40	-
	7	T	below 40	below 40
	7	M	40	below 40
	7	W	below 40	below 40
	7	R	40	below 40
	7	RC	40	45
	7	RF	53	45
	20	T	45	below 40
	20	M	45	below 40
	20	W	55	45
	20	RF	46	46
	20	RC	45	-
	20	RB	below 40	-
26 July. 12.00 hr. 21°C. 60% R.H. Cloudy and close. Slight breeze only.	1	T	45	below 40
	1	M	45	below 40
	1	W	44	below 40
	1	RC	52	-
	1	CM	44	below 40
	1	RF	65	50
	2	T	below 40	below 40
	2	M	45	below 40
	2	W	44	below 40
	2	RC	below 40	-
	2	RB	below 40	-
	2	RF	55	below 40
	3	T	below 40	below 40

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin.
26 July. 12.00 hr. (continued)	3	M	below 40	below 40
	3	W	45	below 40
	3	R^C	below 40	-
	3	RF	46	46
	3	RB	below 40	-
	7	T	42	below 40
	7	M	below 40	below 40
	7	W	50	below 40
	7	RC	55	55
	7	RF	60	47
	7	RB	46	-
	20	T	below 40	below 40
	20	M	below 40	below 40
	20	W	42	below 40
	20	RC	below 40	-
	20	RF	41	-
	20	RB	below 40	-
27 July. 11.30 hrs. 15.5°C. 94% R.H. Cloudy. Rain before the readings were taken. Sheep wet.	1	T	64	70
	1	M	69	100
	1	W	72	100
	1	RC	60	60
	1	RF	72	75
	1	RB	56	75
	2	T	52	-
	2	M	above 80	-
	2	W	65	70
	2	RC	53	68
	2	RF	75	above 80
	2	RB	45	63
	3	T	47	65

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	2cm. off skin.
27 July. 11.30 hr. (continued)	3	M	45	-
	3	W	50	66
	3	RC	45	60
	3	RF	55	67
	3	RB	42	-
	7	T	55	70
	7	M	70	100
	7	RF	60	75
	7	RB	46	-
	7	W	65	100
	20	T	51	67
	20	M	55	75
	20	W	70	100
	20	RC	50	63
	20	RF	50	60
29 July. 09.50 hrs. 17°C. 68% R.H. Cool and windy. Sheep all dry to touch.	1	T	below 40	-
	1	M	below 40	-
	1	W	below 40	-
	1	RF	45	below 40
	1	RB	below 40	-
	1	RC	below 40	-
	2	T	below 40	-
	2	M	below 40	-
	2	W	below 40	-
	2	RC	below 40	-
	2	RF	43	below 40
	2	RB	below 40	-
	3	T	below 40	below 40
	3	M	below 40	below 40
	3	W	below 40	below 40
	3	RC	below 40	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2 cm. off skin.
29 July. 09.50 hrs. (continued)	3	RF	below 40	-
	3	RB	below 40	-
	7	T	below 40	42
	7	M	below 40	below 40
	7	W	below 40	below 40
	7	RC	48	48
	7	RF	below 40	below 40
	20	T	42	42
	20	M	below 40	-
	20	W	below 40	-
	20	RF	below 40	below 40
	20	RB	below 40	-
30 July. 12.45 hr. 18°C. 82% R.H. Rain during night, and a shower 3 hrs. before readings.	1	T	46	42
	1	M	55	48
	1	W	50	44
	1	RF	60	68
	1	RB	46	-
	2	T	below 40	47
	2	M	45	50
	2	W	below 40	50
	2	RB	below 40	-
	2	RF	62	-
	2	RC	65	48
	3	T	55	-
	3	M	67	67
	3	W	70	-
	3	RF	66	-
	3	RB	45	-
	3	RC	55	-
	7	T	50	-
	7	M	47	42

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
30 July. 12.45 hr. (continued)	7	W	49	45
	7	RC	50	-
	7	RF	67	67
	7	RB	44	50
	20	T	46	52
	20	M	46	50
	20	W	46	-
	20	RC	53	-
	20	RF	45	50
	20	RB	42	-
31 July. 10.30 am. 15.3°C. 71% R.H. Cool, strong breeze. Sheep dry.	x1	T	below 40	below 40
	1	M	below 40	below 40
	1	W	below 40	below 40
	1	RC	below 40	-
	1	RF	50	40
	1	RB	below 40	-
	2	T	below 40	-
	2	M	below 40	below 40
	2	W	below 40	below 40
	2	RC	43	-
	2	RF	45	below 40
	2	RB	below 40	-
	3	T	below 40	-
	3	M	48	below 40
	3	W	60	40
	3	CM	55	40
	3	RF	46	46
	3	RC	42	-
	7	T	42	42
	7	M	45	below 40
	7	W	45	below 40

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
31 July. 10.30 a.m. (continued)	7	RF	50	45
	7	RC	below 40	-
	7	RB	44	-
	20	T	below 40	-
	20	M	40	below 40
	20	W	44	below 40
	20	RF	44	42
	20	RB	below 40	-
	20	RC	41	-
1 Aug. 10.30 hr. 15.9°C. 71% R.H. Cloudy. Light breeze. Sheep dry to touch.	1	T	below 40	-
	1	M	45	below 40
	1	W	45	below 40
	1	RF	58	45
	1	RB	below 40	-
	1	RC	42	-
	2	T	below 40	below 40
	2	M	45	-
	2	W	below 40	-
	2	RF	48	-
	2	RC	42	-
	2	RB	below 40	-
	3	T	below 40	-
	3	M	below 40	-
	3	W	45	below 40
	3	RF	40	-
	3	RC	below 40	-
	3	RB	below 40	-
	7	T	40	-
	7	M	42	-
	7	W	42	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
1 Aug. 10.30 hr. (continued)	7	RF	60	50
	7	RC	45	-
	20	T	below 40	-
	20	M	below 40	-
	20	W	below 40	-
	20	RC	below 40	-
	20	RF	below 40	-
	20	RB	below 40	-
2 Aug. 11.45 hrs. 18°C. 78% R.H. Strong breeze. No rain. Sheep dry.	1	T	below 40	-
	1	M	below 40	below 40
	1	RB	42	-
	1	RC	45	55
	1	RF	50	-
	2	T	45	55
	2	M	60	55
	2	W	48	-
	2	RF	57	-
	2	RC	50	-
	2	RB	46	-
	3	T	45	50
	3	W	57	57
	3	RF	54	-
	3	RC	44	-
	3	RB	41	-
	7	T	below 40	-
	7	M	below 40	-
	7	W	below 40	45
	7	RF	50	60
	7	RC	50	-
	7	RB	49	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm off skin.
2 Aug. 11. 45 hrs. (continued)	20	T	45	50
	20	M	below 40	50
	20	W	below 40	-
	20	RF	below 40	55
	20	RB	below 40	-

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SHEEP AT CRAG FARM, RAVENGLASS, CUMBERLAND

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin.
6 Aug. 14.15 hrs. 15°C. 90% R.H. Heavy rain overnight. Cloudy. Sheep dry on outside, feel moist near skin.	8	W	over 80	-
	8	M	75	-
	8	T	75	-
	8	RF	over 80	-
	9	W	over 80	-
	9	M	60	67
	9	T	51	65
	9	RF	over 80	-
	9	RB	53	-
	11	W	100	-
	11	M	100	100
	11	T	65	75
	11	RF	over 80	-
	11	RB	67	-
6 Aug. 16.15 hrs. 16°C. 85% R.H. Cloudy. No rain during day. Sheep feel dry to touch.	4	W	64	-
	4	M	50	63
	4	T	60	-
	4	RC	55	65
	4	RF	60	-
	5	W	73	-
	5	M	73	-
	5	T	70	-
	5	RC	100	-
	5	RF	100	-
	5	RB	over 80	-
	8	W	73	-
	8	M	74	-
	8	T	61	-
	8	RF	over 80	-
	8	RC	60	68
	8	RB	68	-

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	2cm. off skin.
6 Aug. 16.15 hrs. (continued)	9	W	47	-
	9	M	44	-
	9	T	55	-
	9	RF	76	-
	11	W	100	100
	11	M	over 80	-
	11	T	100	100
	11	RC	60	-
	11	RF	73	-
	11	RB	78	-
	21	W	below 40	51
	21	M	below 50	-
	21	T	47	-
	21	RF	52	-
	21	50	73	
7 Aug. 12.20 hrs. 15°C. 80% R.H. No rain during night. Sheep dry.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	below 40
	4	RC	below 40	-
	4	RF	below 40	-
	4	RB	below 40	-
	5	W	50	below 40
	5	M	64	44
	5	T	50	below 40
	5	RC	64	-
	5	RB	68	-
	8	W	47	44
	8	M	65	45
	8	T	below 40	-
	8	RC	50	50

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm. off skin.
7 Aug. 14.20 hrs. (continued)	8	RF	56	56
	9	W	below 40	below 40
	9	M	below 40	below 40
	9	T	below 40	below 40
	9	RC	42	46
	9	RF	59	-
	9	RB	55	55
	10	W	below 40	below 40
	10	M	below 40	below 40
	10	T	below 40	45
	10	RC	below 40	-
	10	RF	60	-
	10	RB	42	-
	22	M	below 40	-
	22	LF	63	-
	22	LC	68	-
	22	RB	73	-
	23	W	61	61
	23	T	45	45
	23	RC	45	-
	23	RF	67	65
	23	RB	50	62
8 Aug. 12.30 hrs. 17°C. Heavy rain during night. Sheep wet.	4	W	65	75
	4	M	52	65
	4	T	60	66
	4	RF	66	73
	4	RB	64	-
	5	W	68	75
	5	M	65	78
	5	T	68	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin
8 Aug. 12.30 hrs. (continued)	5	RC	70	over 80
	5	RB	over 80	-
	8	W	-	70
	8	M	73	73
	8	T	65	69
	8	RC	68	68
	8	RF	over 80	over 80
	8	RB	-	73
	9	W	below 40	68
	9	M	64	75
	9	T	60	66
	9	RC	63	-
	9	RF	over 80	over 80
	9	RB	45	-
	10	W	67	72
	10	M	50	60
	10	T	50	67
	10	RC	60	-
	10	RF	55	63
	10	RB	60	-
	22	M	56	60
	22	LC	-	73
8 Aug. 18.00 hrs. 15°C. 80% R.H. Sunny. Strong Breeze. Sheep by now dry to touch.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	below 40
	4	RC	below 40	-
	4	RF	below 40	below 40
	4	RB	below 40	below 40
	5	W	51	45
	5	M	50	50

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	2cm. off skin.
8 Aug. 18.00 hrs. (continued)	5	T	47	55
	5	RC	45	50
	5	RF	56	50
	5	RB	63	-
	8	W	45	below 40
	8	M	54	54
	8	T	40	44
	8	RC	44	-
	8	RF	66	66
	8	RB	below 40	-
	9	M	below 40	43
	9	T	below 40	-
	9	RC	below 40	-
	9	RB	below 40	-
	10	W	below 40	44
	10	M	below 40	42
	10	T	below 40	43
	10	RC	below 40	50
	10	RF	below 40	45
	10	RB	below 40	-
9 Aug. 11.40 hrs. 14.5°C. 80% R.H. No rain during night. Sheep dry.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	below 40
	4	RC	40	-
	4	RF	below 40	below 40
	4	RB	below 40	-
	5	W	60	66
	5	M	40	45
	5	T	51	45
	5	RF	51	60
	5	RC	46	60

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm. off skin.
9 Aug. 11.40 hrs. (continued)	5	RB	56	68
	8	W	63	63
	8	M	70	61
	8	T	48	45
	8	RC	45	55
	8	RF	76	80
	9	W	below 40	43
	9	T	44	50
	9	RC	below 40	55
	9	RF	60	69
	9	RB	41	-
	10	W	44	below 40
	10	M	below 40	below 40
	10	T	below 40	below 40
	10	RF	45	45
	10	RB	below 40	-
	22	LC	49	60
	22	RB	43	55
10 Aug. 11.00 hrs. 14.9°C. 90% R.H. No wind. Heavy dew during night. Sheep wet.	8	W	75	over 80
	8	M	78	over 80
	8	T	65	60
	8	RC	76	-
	8	RF	73	80
	8	RB	62	-
10 Aug. 15.35 hrs. 15.2°C. 66% R.H. Sheep dry.	4	W	below 40	below 40
	4	M	below 40	-
	4	T	below 40	45
	4	RF	45	50
	4	RC	52	-
	4	RB	40	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin Surface	2cm. off Skin
10 Aug. 15.35 hrs. (continued)	5	W	55	55
	5	M	53	56
	5	T	55	60
	8	W	68	75
	8	M	over 80	over 80
	8	T	56	66
	8	RC	48	-
	9	W	below 40	50
	9	M	below 40	43
	9	T	below 40	48
	9	RC	below 40	-
	9	LC	43	-
	9	RF	-	57
	9	RB	42	-
	10	W	58	58
	10	M	50	-
	10	T	45	50
	10	RF	51	57
	10	RC	55	-
	10	RB	45	55
11 Aug. 16.00 hrs. 16°C. 71% R.H. No rain during night. Sheep dry.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	below 40
	4	RB	below 40	-
	4	RF	below 40	below 40
	4	RC	below 40	below 40
	5	W	50	50
	5	M	49	49
	5	T	47	-
	5	RC	55	57
	5	RF	60	60

Date etc.	Sheep No.	Position	Relative Humidity % Skin Surface	2cm. off skin
11.Aug. 16.00 hrs. (continued)	5	RB	58	-
	8	W	55	60
	8	M	63	63
	8	T	52	52
	8	RC	50	-
	8	RF	76	over 80
	8	RB	below 40	-
	9	W	below 40	-
	9	M	below 40	-
	9	T	below 40	below 40
	9	RC	below 40	-
	9	RF	41	49
	9	RB	below 40	-
	10	W	below 40	45
	10	M	41	-
	10	T	below 40	below 40
	10	RC	47	55
	10	RF	47	47
	10	RB	below 40	-
12 Aug. Sheep 27 & 28 when fleece readings taken were on the fell @ ft.	27	W	50	69
	27	M	60	73
	27	T	50	70
	27	RF	-	over 80
	27	RC	67	70
	27	RB	67	-
	28	W	67	100
	28	M	68	over 80
	28	T	65	72
	28	RF	-	75
	28	RC	67	73

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm. off skin.
12 Aug. (continued)	28	RB	60	75
13 Aug. 15.30 hrs. 16°C. 80% R.H. Sheep wet in morning but dry to touch by now.	4	W	below 40	below 40
	4	M	77	100
	4	T	below 40	40
	4	RF	below 40	below 40
	4	RB	below 40	-
	4	RC	below 40	50
	5	M	over 80	-
	5	T	63	63
	5	RF	64	70
	5	RC	65	over 80
	8	W	63	63
	8	M	over 80	-
	8	T	68	68
	8	RC	60	69
	8	RB	66	-
	8	RF	over 80	-
	8	LF	over 80	-
	9	W	below 40	below 40
	9	M	below 40	below 40
	9	T	below 40	below 40
	9	RF	41	55
	9	RC	45	63
	9	RB	45	-
14 Aug. 15.40 hrs. 14.5°C. 89% R.H. Heavy rain overnight. Sheep feel dry by now.	4	W	below 40	45
	4	M	below 40	42
	4	T	40	45
	4	RF	below 40	45
	4	RB	42	50
	4	RC	45	66

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin
14 Aug. 15.40 hrs. (continued)	5	W	47	47
	5	M	50	65
	5	T	49	55
	5	RF	57	66
	5	RB	60	70
	5	RC	46	66
	8	W	46	56
	8	T	45	48
	8	LF	over 80	100
	8	RF	65	65
	8	RB	55	68
	8	RC	44	55
	9	W	below 40	below 40
	9	M	40	45
	9	T	42	50
	9	RF	45	52
	9	RB	below 40	45
	9	RC	42	57
15 Aug. 10.00 hrs. 12.5°C. 89% R.H. Sheep slightly wet.	4	W	40	55
	8	LF	74	over 80
	8	RF	64	70
15 Aug. 15.40 hrs. Sheep dry.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	40
	4	RF	40	below 40
	4	RC	43	59
	4	RB	40	49
	5	W	45	45
	5	M	47	47
	5	T	50	53

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin
15 Aug. 15.40 hrs. (continued)	5	RF	57	57
	5	RC	45	60
	5	RB	60	64
	8	W	45	55
	8	M	58	58
	8	T	below 40	43
	8	RC	45	50
	8	LF	70	over 80
	8	RF	76	76
	9	W	below 40	below 40
	9	M	below 40	below 40
	9	T	below 40	42
	9	RF	below 40	42
	9	RC	40	50
	9	RB	44	50
16 Aug. 15.30 hrs. 15.4°C. 75% R.H. Sheep dry. Sunny.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	43
	4	RF	below 40	42
	4	RC	43	50
	4	RB	below 40	50
	5	W	44	48
	5	M	45	45
	5	T	50	50
	5	RF	50	56
	5	RC	42	55
	5	RB	53	66
	8	W	43	50
	8	M	55	55
	8	T	below 40	below 40
	8	RF	58	65

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin
16 Aug. 15.30 hrs. (continued)	8	LF	65	69
	8	RC	45	55
	8	RB	45	62
	9	W	below 40	below 40
	9	M	below 40	below 40
	9	T	below 40	41
	9	RF	below 40	40
	9	RC	42	57
	9	RB	below 40	below 40
17 Aug. 10.30 hrs. 15.6°C. 81% R.H. Some sheep feel slightly moist to touch.	4	W	40	below 40
	4	M	40	below 40
	4	T	below 40	below 40
	4	RF	-	63
	4	RC	50	66
	4	RB	50	68
	5	W	43	below 40
	5	M	50	50
	5	T	47	42
	5	RF	60	73
	5	RC	50	65
	5	RB	58	72
	8	W	69	69
	8	M	74	76
	8	T	50	50
	8	LF	69	77
	8	RF	70	78
	8	RB	55	70
	8	RC	55	63
	9	W	below 40	below 40
	9	M	below 40	below 40

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin.
17 Aug. 10.30 hrs. (continued)	9	T	below 40	below 40
	v9	RF	52	62
	9	RC	48	63
	9	RB	65	72
	10	W	56	below 40
	10	T	55	48
	10	RF	50	57
	10	RC	63	63
18 Aug. 15.00 hrs. Sheep dry.	4	W	below 40	55
	10	W	60	72
	10	RC	50	55
	10	M	45	64
	10	RF	49	49
19 Aug. 11.30 hrs. 16.3°C. 85% R.H. Sheep dry except for Sheep 5.	4	W	45	45
	4	M	45	45
	4	T	40	40
	4	RF	42	42
	4	RC	50	60
	4	RB	47	58
	5	W	46	42
	5	M	58	64
	5	T	60	60
	5	RF	64	70
	5	RC	46	64
	5	RB	54	65
	8	W	72	72
	8	M	72	72
	8	T	58	68
	8	RC	60	66
	8	RF	68	72

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2 cm. off skin
19 Aug. 11.30 hrs. (continued)	8	LF	67	72
	8	RB	65	68
	24	W	64	64
	24	M	53	44
	24	RF	65	65
	24	RC	65	65
20 Aug. 11.00 hrs. 15°C. 76% R.H. Heavy dew. Sheep moist to touch.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	below 40
	4	RF	below 40	45
	4	RB	40	65
	4	RC	40	40
	5	W	50	45
	5	M	42	below 40
	5	T	50	50
	5	RF	58	65
	5	RC	40	68
	5	RB	57	68
	8	W	68	68
	8	M	62	40
	8	T	41	45
	8	RF	57	67
	8	RC	below 40	45
	8	RB	below 40	55
	8	LF	46	46
	10	W	60	below 40
	10	M	60	below 40
	10	T	50	below 40
	10	RC	50	50
	10	RB	below 40	46

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	2cm. off skin
20 Aug. 14.30 hrs. Sheep dry.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	below 40	below 40
	4	RF	below 40	below 40
	4	RC	below 40	44
	4	RB	below 40	40
	8	RF	55	55
	8	LF	68	68
	10	W	below 40	below 40
	10	M	below 40	below 40
	10	T	below 40	below 40
	10	RF	below 40	below 40
	10	RC	below 40	45
	10	RB	below 40	45
21 Aug. 12.15 hrs. 15.3°C. 74% R.H. Heavy dew in early morning, but sheep dry by now except sheep 8 which was moist to the touch.	4	W	below 40	below 40
	4	M	below 40	below 40
	4	T	40	40
	4	RF	below 40	below 40
	4	RC	50	61
	4	RB	50	66
	5	W	63	below 40
	5	M	65	43
	5	T	69	69
	5	RF	68	56
	5	RC	52	67
	5	RB	60	67
	8	W	70	68
	8	M	77	68
	8	T	63	66
	8	RF	72	72
	8	LF	70	77

Date etc.	Sheep No.	Position	Relative Humidity % skin surface	Relative Humidity % 2cm. off skin.
21st Aug. 12.15 hrs. (continued)	8	RC	68	66
	8	RB	67	67
21 Aug. 15.00 hrs. Sheep dry.	8	RF	68	68
	8	LF	57	62
	25	W	below 40	below 40
	25	M	below 40	below 40
	25	T	45	47
	25	RF	40	40
	25	RC	43	49
	25	RB	50	58
22 Aug. 11.15 hrs. 15°C. 95% R.H. Heavy rain overnight. Sheep wet.	4	W	100	100
	4	M	100	100
	4	RB	65	over 80
	4	T	55	69
	4	RC	64	-
	5	W	100	100
	5	M	100	100
	5	T	70	over 80
	5	RF	65	70
	5	RC	65	69
	5	RB	69	75
	8	W	73	over 80
	8	M	over 80	100
	8	T	68	70
	8	RB	55	66
	8	RF	over 80	-
	8	LF	72	75
	8	RC	65	-

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
22 Aug. 18.30 hrs. 15.0°C. 76% R.H. Sunny. Sheep 8 still moist at crutch.	8	T	45	60
	8	M	over 80	over 80
	8	RB	42	76
	8	LF	50	57
	8	RF	65	65
	8	W	46	52
	8	RC	66	70
23 Aug. 11.30 hrs. 16°C. 72% R.H. Heavy dew overnight. Some sheep moist to touch.	4	W	46	below 40
	4	M	below 40	below 40
	4	T	43	43
	4	RF	below 40	below 40
	4	RC	62	65
	4	RB	50	68
	5	W	67	below 40
	5	M	55	55
	5	T	59	43
	5	RF	55	68
	5	RC	45	66
	5	RB	44	60
	8	W	75	over 80
	8	M	76	over 80
	8	T	65	65
	8	RF	75	78
	8	LF	68	68
	8	RC	59	66
	8	RB	55	66
	9	W	49	below 40
	9	M	40	below 40
	9	T	54	below 40
	9	RF	below 40	47
	9	RC	55	55
	9	RB	-	60

FLEECE R.H. READING, 1946

MADE IN CONJUNCTION WITH R.H. READING BY

WOOL SAMPLE METHOD

SHEEP AT SCIENCE LABORATORIES, DURHAM

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	Relative Humidity % 2cm. off skin.
2 Sept. 10.30 hrs. 16°C. 81% R.H. Heavy dew. Sheep moist to the touch.	1	W	65	-
	1	M	64	-
	1	T	60	-
	1	RF	68	-
	26	W	60	67
	26	M	50	62
	26	T	55	-
	26	RF	55	-
	26	RC	55	62
2 Sept. 17.30 hrs. 15°C. After a heavy shower. Sheep very wet.	1	W	55	-
	1	M	43	47
	1	T	60	-
	1	RF	100	100
	6	W	65	-
	6	RF	67	-
	26	W	68	-
	26	M	50	55
	26	T	46	-
	26	RF	54	-
	26	RC	40	48
3 Sept. 12.30 hrs. Sheep wet.	1	W	62	67
	1	M	55	-
	1	T	60	68
	1	RF	48	62
	6	W	58	64
	6	M	45	55
	6	RF	45	-
	26	W	62	66
	26	M	45	63

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
3 Sept. 12.30 hrs. (continued)	26	T	50	-
	26	RF	45	52
	26	RC	44	-
3 Sept. 17.40 hrs. After a moderate shower. Sheep wet.	1	W	50	-
	1	M	65	-
	1	T	65	65
	1	RF	60	-
	6	W	67	-
	6	M	62	-
	6	RF	62	68
	26	W	100	-
	26	M	61	-
	26	RC	43	-
	26	RF	50	-
4 Sept. 11.30 hrs. Heavy rain all night. Sheep wet.	1	W	over 80	100
	1	M	over 80	100
	1	T	65	68
	1	RF	67	70
	6	W	100	100
	6	M	68	68
	6	RF	59	59
	26	W	100	100
	26	M	50	72
	26	T	55	68
	26	RF	47	60
	26	RC	51	53
4 Sept. 19.45 hrs. Heavy rain all day. Sheep wet.	1	M	100	100
	1	T	70	70
	1	RF	72	72

Date etc.	Sheep No.	Position	Relative Humidity % Skin surface	2cm. off skin.
4 Sept. 19.45 hrs. (continued)	6	W	100	100
	6	M	70	70
	6	RF	62	73
	26	RF	55	75
5 Sept. 12.00 hrs. Rain up till 09.00 hrs. Sheep wet.	1	M	100	100
	1	T	67	73
	1	RF	67	75
	6	W	100	100
	6	M	58	74
	6	T	50	65
	6	RF	55	67
	26	RF	55	67
5 Sept. 16.45 hrs. Sheep almost dry by now.	1	RF	65	68
	6	M	55	-
	6	T	56	63
	26	RF	55	65
6 Sept. 17.30 hrs. No rain since 6 Sept.09.00 hrs. Sheep dry.	6	T	40	45
	26	T	42	42
	26	RF	43	43
9 Sept. 10.15 hrs. No rain. Sheep dry.	1	RF	42	52
	1	TM	40	45
	6	TM	below 40	40
	26	TM	below 40	below 40
	26	LC	46	48

FLEECE R.H. READINGS. 1947

SHEEP AT HOUGHALL FARM, DURHAM

Sheep No.	Position	% Fleece R.H. skin Surface	% Fleece R.H. 2cm. off skin	Sheep No.	Position	% Fleece R.H. skin Surface	% Fleece R.H. 2cm. off skin
			19 June				
36	W	58	58	38	W	50	48
36	M	60	60	38	T	50	-
36	T	60	60	38	RC	48	52
36	RF	58	60	38	RF	52	22
37	W	70	63	39	M	64	61
37	T	65	65	39	T	56	51
37	LC	63	70	39	LF	53	58
37	LF	67	64				
			20 June				
29	T	50	50	35	W	48	54
30	M	60	62	35	T	50	55
30	T	60	66	35	LF	58	56
30	RC	below 40	60	37	M	77	77
32	T	53	60	37	T	61	61
33	M	60	64	37	LF	67	-
33	RC	50	54	38	M	45	45
31	M	52	60	38	T	below 40	45
31	RF	50	55	39	W	54	58
34	W	75	75	39	T	55	57
34	M	74	74	39	LF	60	64
34	RC	58	64				
			23 June				
29	W	52	52	31	W	53	55
29	M	56	56	31	M	52	52
29	T	50	56	31	T	52	52
30	W	55	52	31	RC	45	45
30	T	52	52	32	W	57	59
30	RC	53	53	32	M	57	60

Sheep No.	Position	% Fleece R.H. skin surface	% Fleece R.H. 2cm off skin	Sheep No.	Position	% Fleece R.H. skin surface	% Fleece R.H. 2cm. off skin
			23 June (continued)				
32	T	50	50	37	M	75	55
33	W	55	61	37	T	64	64
33	T	48	55	37	RF	66	66
33	RC	53	62	38	W	45	45
33	RF	-	55	38	M	42	42
34	W	80	over 80	38	RF	43	43
34	T	68	68	39	W	60	52
34	RC	60	60	39	M	60	56
35	W	below 40	45	40	W	48	45
35	M	50	52	40	M	57	54
35	T	45	56	40	RF	45	45
37	W	75	64	40	RC	47	55
			25 June. Sheep moist				
31	W	50	48	38	W	45	45
31	T	65	75	39	W	55	60
32	W	75	75	39	M	60	49
32	T	52	below 40	39	T	52	45
33	W	70	51	39	LF	50	50
33	LF	70	-	40	W	55	58
34	W	73	55	40	M	57	61
34	T	68	65	40	T	50	50
34	LF	64	80	41	W	60	60
37	M	over 80	over 80	41	T	66	66
37	T	69	69	41	LF	51	48
			26 June. Sheep dry				
29	W	45	42	31	W	63	57
29	T	below 40	below 40	31	T	55	51
29	RF	54	54	31	RF	67	63

Sheep No	Position	% Fleece R.H. skin surface	% Fleece R.H. 2cm. off skin	Sheep No.	Position	% Fleece R.H. skin surface	% Fleece R.H. 2cm off skin
			26 June (continued)				
32	W	80	80	37	RF	80	80
32	T	75	75	38	T	below 40	below 40
32	RF	80	80	38	W	43	43
33	W	55	48	39	W	53	50
33	T	52	52	39	T	47	47
33	LF	59	59	39	LF	60	60
34	W	71	60	40	W	45	45
34	T	71	63	40	T	45	50
34	RF	80	75	40	RF	53	53
35	W	51	51	41	W	63	63
35	M	60	60	41	T	65	65
35	T	60	60	41	LF	68	68
35	RF	59	62	42	W	53	55
37	W	80	80	42	T	50	55
37	T	68	68	42	LF	35	62

FLEECE R.H. READINGS

HOT ATMOSPHERE EXPERIMENTS

EXPERIMENT I

11.Aug. Sheep 53 used.

Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.	Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.
	W	62	64		W	68	65
	M	54	57		M	68	65
13.45	T	61	61	16.30	T	68	65
(20°C)	RF	66	68	(38°C)	RF	75	70
	RC	57	60		RC	70	65
	RB	64	70		RB	70	68
	W	66	66		W	70	70
	M	62	62		M	65	65
14.40	T	63	63	17.20	T	67	67
(34°C)	RF	70	70	(22°C)	RF	70	above 80
	RC	62	60		RC	60	62
	RB	66	66		RB	56	70
	W	66	64				
	M	64	60				
15.30	T	75	67				
(36°C)	RF	77	77				
	RC	67	60				
	RB	68	70				

EXPERIMENT 2

13.Aug. Sheep 43 and 43 used.

(1) SHEEP 43

09.00	T	54	66
(17°C)	RF	52	60

EXPERIMENT 2 (continued)

Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.	Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.
	W	57	60		W	76	76
	M	55	61		M	68	62
09.30	T	55	66	10.55	T	60	67
(18°C)	RF	54	60	(25°C)	RF	67	67
	RC	50	54		RC	55	57
	RB	56	64		RB	68	70
	W	68	66		W	65	60
	M	61	50		M	64	55
11.45	T	72	72	13.00	T	73	73
(29°C)	RF	72	72	(37°C)	RF	73	73
	RC	53	55		RC	58	58
	RB	60	67		RB	75	75
	W	66	63		W	58	58
	M	65	59		M	60	60
13.30	T	70	70	14.30	T	65	68
(38°C)	RF	80	80	(23°C)	RF	70	70
	RC	60	58		RC	51	55
	RB	70	70		RB	60	66
	W	55	60				
	M	51	56				
16.30	T	56	63				
(20°C)	RF	57	63				
	RC	46	50				
	RB	50	55				
(2) SHEEP 53					W	75	68
09.00	M	58	75	13.00	M	72	65
(Temps. as above)	RF	67	75		T	70	66

EXPERIMENT 2 (continued)

Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.	Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.
	W	58	66		W	75	68
09.30	M	60	66	13.30	M	75	75
	T	52	60		T	70	65
	W	68	70		W	67	67
10.45	M	75	65	14.30	M	66	65
	T	75	65		T	60	60
	W	75	68		W	63	66
11.45	M	68	52	16.30	M	63	66
	T	66	60		T	58	65

EXPERIMENT 3

16.Aug. Sheep 43 and 53 used.

(1) SHEEP 43

09.15	W	55	64	14.30	W	63	68
(16°C)	M	51	62	(21°C)	M	48	56
10.15	W	55	65	15.30	W	66	66
(17.0°C)	M	52	61	(36°C)	M	63	60
11.15	W	50	61	16.30	W	75	75
(17.0°C)	M	47	62	(38°C)	M	66	64
13.15	W	60	64	17.15	W	61	61
(20°C)	M	56	62	(23°C)	M	50	57

(2) SHEEP 53

09.15	RC	56	66		RC	55	62
(Temps.	LC	53	62	14.30	LC	52	58
as above)	M	60	66		M	55	58

EXPERIMENT 3 (continued)

Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.	Time (G.M.T.) and Temp.	Position	% R.H. skin surface	% R.H. 2 cm.
	RC	50	64		RC	63	63
10.15	LC	51	64	15.30	LC	66	62
	M	55	63		M	65	60
	RC	55	62		RC	70	70
11.15	LC	55	64	16.30	LC	68	68
	M	60	63		M	72	68
	RC	55	66		RC	65	65
13.15	LC	55	64	17.15	LC	64	64
	M	58	64		M	65	65

EXPERIMENT 4

17 Aug. Sheep 43 and 53 used.

(1) SHEEP 43

09.15	T	52	65	11.15	T	50	54
(16°C)	RF	48	61	(19°C)	RF	50	53
10.15	T	44	53	13.45	T	50	62
(18°C)	RF	41	52	(22°C)	RF	60	60
14.45	T	68	68	16.45	T	42	53
(27°C)	RF	62	62	(18°C)	RF	61	61
15.45	T	47	60				
(19°C)	RF	51	60				

(2) SHEEP 53

	W	51	66		W	70	65
09.15	RF	60	67	14.45	RF	75	80
(Temps. as above)	RC	42	62		RC	61	60

EXPERIMENT 4 (continued)

Time (G.M.T.) and temp.	Position	% R.H. skin	% R.H. 2 cm.	Time (G.M.T.) and temp.	Position	% R.H. skin	% R.H. 2 cm.
	W	60	70		W	47	55
10.15	RF	60	66	15.45	RF	74	74
	RC	48	61		RC	55	57
	W	60	62		W	60	60
11.15	RF	65	65	16.45	RF	67	70
	RC	55	56		RC	51	58
	W	62	62				
13.45	RF	68	68				
	RC	54	61				

EXPERIMENT 5

20 Aug. Sheep 43 and 53 used.

(1) SHEEP 43

09.00	M	49	60	11.30	M	63	55
(17°C)	T	49	65	(32°C)	T	42	50
	RC	45	48		RC	50	50
10.00	M	46	53	13.15	M	70	63
(17°C)	T	55	68	(33°C)	T	80	80
	RC	42	45		RC	65	60
10.30	M	46	50	14.15	M	53	55
(18°C)	T	50	65	(21°C)	T	64	69
	RC	41	45		RC	53	55
15.15	M	47	55	17.15	M	45	56
(19°C)	T	55	68	(16°C)	T	60	68
	RC	42	50		RC	43	45
16.30	M	47	54				
(18°C)	T	65	70				
	RC	45	49				

EXPERIMENT 5 (continued)

(2) SHEEP 53

Time (G.M.T.) and Temp.	Position	% R.H. skin	% R.H. surface	% R.H. 2 cm.
09.00	M	51	60	
(Temp. as above)	T	50	70	
	RC	68	68	
	M	48	54	
10.00	T	55	68	
	RC	56	75	
	M	51	55	
10.30	T	-	-	
	RC	56	70	
	M	61	56	
11.30	T	70	65	
	RC	65	65	
	M	72	64	
13.15	T	75	68	
	RC	68	65	
	M	61	61	
14.15	T	66	66	
	RC	61	63	
	M	58	62	
15.15	T	65	68	
	RC	59	59	
	M	58	62	
16.30	T	68	68	
	RC	61	61	
	M	51	62	
17.15	T	65	70	
	RC	55	65	