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Report for 1958

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Bee Department

C. G. Butler

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BEE DEPARTMENT

C. G. BUTLER

J. B. Free was awarded a Postdoctorate Fellowship by the National Research Council of Canada, and in September left to spend one year in the Apicultural Department, Ontario Agricultural College, Guelph. In March L. Bailey lectured in Oslo and Copenhagen at the invitation of the Beekeeping Associations of Norway and Denmark, and in August visited Prague as a guest of the Czechoslovak Academy and attended the 1st International Congress of Insect Pathology. In September C. G. Butler attended the 4th International Conference of Bee Research Workers at the University of Bologna and the 17th International Beekeeping Congress in Rome.

Members of the department have lectured at meetings of scientific societies, beekeepers' associations, etc., and have also continued to serve on various committees concerned with beekeeping, pollination, etc.

BEE BEHAVIOUR

Swarming

A method of determining, biologically, in the laboratory, the efficacy of any given substance to inhibit queenless worker honeybees from rearing queens (see *Rep. Rothamst. exp. Sta. for 1957*, p. 169) has been considerably improved, and a start has been made in determining the amount of queen substance that can be extracted from queens of various types. Preliminary results suggest that queens that have actually been superseded by their workers, and queens who have swarmed from uncrowded colonies, possess less of this substance than queens from colonies of similar size in which the workers have not reared any new queens. These results support the view that swarming is, like supersedure, initiated by a shortage of queen substance (Butler). It is, however, still uncertain why colonies sometimes replace their queens by the process of supersedure and at other times by swarming; nor do we know what causes swarming bees to leave the parental hive.

When a swarm is put into a hive a short distance away from the parental one, few, if any, of its worker bees return to the parent colony (Free), whereas if bees with a queen are taken experimentally from a colony and similarly treated, a large proportion of them return to the old position. Attempts are being made to discover whether bees in a swarm behave in this way because they have lost their memory of the old site, or know both sites and prefer the new, or have never learned the position of the old site. When individual bees were removed from a newly emerged swarm, marked and released about 40 yards away, those subsequently found were all in the colonies nearest to the point of release, and none was recovered either from the swarm or from the parent colony. In other experiments, series of marked bees of known age were built up in colonies and, subsequently, single marked bees and parts of the colonies were moved to new positions. Among the bees in the older age groups, the proportion failing to return to the old position was far less than the proportion which would have left the hives had the colonies swarmed (see Butler, *Bee World* 21 (1940), 9–10). Thus it appears that many of the bees in a swarm do not know, or have forgotten, the position of their old hive, but are not simply the bees which, in a normal colony, would have been too young to have flown out and become orientated to the site. (Simpson.)

When the queen is removed from a newly emerged swarm the swarm cluster breaks up and the bees fly back to the parent colony. Their behaviour while doing so is similar to that when a swarm is emerging. The swarm breaks up within 10–15 minutes of the removal of the queen and so cannot be due to exhaustion of the supply of queen substance obtained by licking the queen's body and transferring honeystomach contents from bee to bee. Bees in a swarm must therefore perceive the presence of a queen by some other means—perhaps by scent or sound.

The time taken for a non-swarming colony to show signs of queenlessness was investigated by the use of a pair of observation hives so constructed that a queen (or any other bee) can be removed without disturbing the colony. The queen of one colony was removed, kept temporarily in an incubator and presented in a wiregauze cage to both of the colonies at successive intervals of time. The bees of the queenright colony paid little attention to the queen in the cage at any time, in spite of the fact that she was a stranger. Those of the queenless colony paid little attention for several hours, but eventually became more attentive and, after 4 or 5 hours, had become decidedly hostile. When the queen was released in her own colony she was immediately attacked and " balled ", and her safe re-introduction was possible only after she had been caged in the colony for a lengthy period—in one case, 3 days. The basis of this behaviour is not clear, but it is important in relation to queen introduction, because it not only confirms previous conclusions that a queen to be replaced should not be removed until immediately before the new one is put in (see Butler & Simpson, Bee World 37 (1956), 105-114, 124), but also shows that introduction can sometimes only be achieved safely by periods of caging much longer than those obtainable with any existing system that allows the bees to release the queen. (Simpson.)

A peculiar type of dance, described as dorso-ventral abdominal vibration (D-VAV) (Milum, V. G., Amer. Bee J. 95 (1955), 97–104), is sometimes performed by worker bees with their front legs in contact with other worker bees, and during swarm preparations it is similarly performed on the queen. An attempt has been made to investigate the circumstances under which this dance is performed on workers, in the hope that it may subsequently be possible to make a similar study of its performance on the queen. The dance does not simply occur at random among the worker bees, because bees marked while doing it were more likely to be seen doing it afterwards than bees marked at random on the comb. Some bees 148

were seen to perform almost continuously for long periods, whereas others danced only occasionally. The minimum age at which individual bees (sixteen bees) began to perform the dance was 5 days and the mean about 10. Bees marked after being seen to dance were more likely to be seen foraging within the next 3 days than bees marked at random. Bees marked when returning from foraging for pollen were more likely to be seen dancing within the next 3 days than bees marked at random—in spite of the fact that they were out of the hive during much of the periods of observation. Ability to return to the hive when released a short distance from it (in an apiary in which other colonies were present) was at least as great among D-VAV bees as among foragers. It seems possible that the bees which perform this dance are either foragers or potential foragers, the latter probably being mostly young bees ready to forage but not yet doing so because nectar or pollen sources are few. (Simpson.)

Queen substance

The residue from an ethanol extract of queen honeybees (i.e., of queen substance), when fed in distilled water to queenless worker honeybees, can inhibit them from queen rearing (Butler & Gibbons, J. Insect Physiol. 2 (1958), 61-64). It has now been demonstrated that the inhibitory substance is produced in the mandibular glands of a queen and is probably distributed over her body surface, whence her workers can collect it when she grooms herself (Butler and Simpson). The contents of the mandibular glands of a queen can also inhibit ovary development in queenless worker honeybees when fed to them (Butler).

The improved biological method of determining the amount of queen substance in a given sample is being used, among other things, to determine the activity of certain fractions of the secretion of the mandibular glands of queen honeybees, being prepared by Dr. R. K. Callow and Miss N. Johnston, of the National Institute for Medical Research, with whom we are collaborating in an attempt to isolate and identify the active principle. We are greatly indebted to many beekeepers, both amateur and professional, in Britain and other countries, who have generously supplied us with hundreds of queens for this work.

Ethanol extracts of *Apis indica* queens (kindly supplied by Mr. L. A. S. Perera) have been shown to inhibit queen rearing by groups of queenless *A. mellifera* workers when fed to them.

GENERAL RESEARCH

Salivary glands

The mechanism by which the contents of a bee's mandibular glands are discharged has been elucidated. The duct of the gland is flattened and stiffened at the point of discharge at the base of the mandible. Tension on the integumentary membrane between the mandible and the hypopharyngeal plate separates the lips of the orifice and allows discharge of the secretion which is under pressure in the gland. This tension appears to be produced by lowering and retraction of the hypopharyngeal plate by the muscle which connects it with the tentorial bridge. (Simpson.)

Temperature regulation and food consumption

Groups containing various numbers of worker bees (10-200)were kept at given temperatures between 0° and 40° C. The temperature at which each group maintained itself and the amount of sucrose syrup of known concentration and water it consumed were measured at intervals. The percentage of bees in each group that were clustering together, and the mortality rate, were also determined.

With environmental temperatures between 25° and 35° C. the death-rate was low and similar in groups of different sizes. At 40° C. relatively fewer of the bees in groups of 200 survived than those in smaller groups. Below 25° C. the more bees in a group, the lower was the mortality rate. Groups of 50, 100 and 200 bees survived 24 hours or more at 10° C., and sometimes did so at 5° C.

At environmental temperatures up to 30° C. the bees forming a group maintained a temperature higher than that of the environment. The temperature of a group increased with that of the environment, and the lower the latter, the more it differed from that of the group. The larger a group, the higher its temperature was maintained above that of the environment. The difference between the temperatures of larger and smaller groups decreased with increase of environmental temperature until, at 35° and 40° C., all the groups were maintaining approximately the same temperature, which was either that of the environment or slightly less.

With environmental temperatures between 20° and 40° C. the percentage of bees clustering increased with the size of the group; the bees in groups of ten and twenty-five scarcely clustering at all. It is clear, therefore, that at this temperature range clustering does not take place in response to low temperature but results from mutual attraction. The more bees forming a cluster, the higher is the percentage of the available bees that joins it. However, at 15° C. 70 per cent or more of the bees clustered together irrespective of the size of the group, and at 10° C. nearly all the bees in each group were clustering.

The food consumption of the bees increased as the temperature of the environment fell, irrespective of the size of the group to which they belonged, and, consequently, their rate of metabolism and heat production rose. Little water was drunk at environmental temperatures of 30° C. and lower, but at 35° and 40° C. much was used. Such water consumption could have been due to the bees evaporating water by exposing it to the atmosphere, as the bees of colonies will do at high temperatures, or may, perhaps, have been associated with a high rate of water loss from their bodies at such high temperatures. (Free and Spencer-Booth.)

Longevity of workers

Groups of marked worker bees were introduced into each of four colonies at weekly intervals between March and October 1957 and their longevity determined. Although this varied considerably in consecutive weeks, the mean monthly value diminished steadily from March to June, and then increased slightly during July; the weekly death-rate of bees of all ages was greater in midsummer than at any other time. Bees reared in March had a mean life of just over 5 weeks, those reared in June lived only about 4 weeks. The maximum ages attained by bees reared in each month were: March, 67 days; April, 60 days; May, June, July, 53 days.

Some of the bees reared in August and September survived until late May of the following year, and a few that emerged in the first week of October survived until the following June. No relationship was found between the age of the bees going into winter and their chance of survival until the following spring; neither was the subsequent death-rate of bees during spring and early summer in accordance with their age.

There were marked variations in the longevity of the worker bees of the different colonies both in summer and winter. (Free and Spencer-Booth.)

Observations on Apis indica

Mr. L. A. S. Perera, who is visiting the department for a year in connection with the Colombo Plan, kindly brought two colonies of *Apis indica* with him from Ceylon. These colonies, carried in the luggage hold of the plane (i.e., without heating or special ventilation), arrived in very good condition.

An attempt was made to hybridize this species with A. mellifera by inseminating, instrumentally, six A. mellifera queens with semen from A. indica drones. Unfortunately, great difficulty was experienced in extracting sufficient semen from the drones to give large inseminations. Although on subsequent examination the spermatheca of one of these queens was found to contain a few living spermatozoa, none produced worker brood. It is, therefore, still uncertain whether hybrids between these species can be produced.

An attempt was also made to get an *A. mellifera* colony to rear *A. indica* queens in artificial queen-cell cups, but the larvae transferred were not accepted by the host colony.

Successful natural mating of an A. *indica* queen, presumably with one or more drones of the same species (the offspring showing no signs of being hybrids), took place when one of the A. *indica* colonies became queenless and reared a new queen, in spite of the fact that drones were few and all in the other colony.

An interesting difference in the behaviour of workers of A. indica and of A. mellifera was noted. When ventilation-fanning, the former do so with their heads facing out of the entrance, thus directing an air stream into the nest and not out of it as A. mellifera workers do. Communicative-fanning with the scent glands exposed does not differ greatly in the two species.

POLLINATION

Recent observations of honeybees working on red clover flowers showed that the great majority without pollen in their pollen-baskets were collecting nectar by "robbing" the flowers and thus not pollinating, whereas nearly all the bees with pollen-loads were entering the flowers in the orthodox manner and, presumably, pollinating them. Dissection showed that many of these latter bees had little nectar in their honeystomachs. It is clear, therefore, that honeybees will visit red clover flowers to collect pollen only, and that bees gathering nectar only from this crop are often useless as pollinators. Similarly, bees gathering pollen from apple flowers are more valuable pollinators than those collecting nectar, many of which fail to touch the anthers. Therefore, for pollination of red clover and apple flowers, pollen-gathering honeybees are more important than nectar-gatherers.

A number of workers claim to have significantly increased the number of honeybees foraging on selected crops, such as red clover, with a resultant increase in seed set, by feeding with colonies with sucrose syrup in which flowers of the crop were soaked for several hours and which contained fresh blossoms that the bees touched while drinking. The data published were not altogether satisfactory, and it seemed possible that the "directing" technique was merely increasing the number of bees seeking nectar, but not pollen, from the selected crop. As this would not increase pollination, further controlled experiments were made. The percentage of pollen of the crop concerned in the total amount of pollen of all kinds collected by bees of the "directed" colonies was compared with that collected by the foragers of control colonies fed with unscented syrup of the same concentration. The results do not indicate that "directed" colonies collected any more pollen from the experimental crops than the control colonies, so that in the present state of knowledge growers seem ill advised to try to increase the pollination of their crops by this "scent-directing" technique. Attempts are being made to discover a satisfactory method of

Attempts are being made to discover a satisfactory method of increasing the proportion of pollen-gathering foragers of colonies for purposes of pollination. (Free.)

BEE DISEASES

Nosema disease

Of all colonies at Rothamsted, 45 per cent were found to be infected with Nosema apis in the spring of 1958; an apparent, but non-significant, increase since 1957. In 1957 all twelve colonies in one apiary, which were heavily infected, were transferred to combs that had been fumigated with the vapour of acetic acid, and their number was increased to twenty by subdivision. In 1958 infection was only found in five of the twenty colonies in this apiary. This was the only significant decrease of infection in any apiary since 1957. Three out of fourteen colonies in another apiary were found infected in 1957, and each colony was examined twice each month, for other experimental purposes, from September 1957 to April 1958, by which time four colonies had died and all of the remaining ten were infected. This was the only significant increase of infection in any one apiary since 1957, and was probably caused by the excessive manipulation of these colonies during the winter. However, there is also a tendency for infection to increase in colonies handled a lot during the summer. When colonies are handled, some infected bees are undoubtedly crushed, and healthy bees may become infected from licking the remains; furthermore, handling may cause infected bees to defaecate, and this can be a potent source of infection. Bees of colonies handled in winter and spring were often seen to void excrement on the combs. (Bailey.)

Acarine disease

All colonies were examined for infection with Acarapis woodi in autumn 1957. This completed a series of field observations that has given information for the first time of the relationship of the type of season to incidence of infection in colonies that are not treated for their infection. During 1955, a year of good nectarflows, infection fell abruptly; in 1956, a year of virtually no nectarflow, it rose again; and in 1957, an average year, infection remained steady. (Bailey.)

Mortality of colonies during the winter of 1957-58 showed virtually the same relation to degree of infection as that in previous winters: the minority of colonies with over 30 per cent of their bees infected were more likely to die than the remainder; all (eight) died with over 60 per cent infection. (Bailey.)

European Foul Brood disease

An improved method of isolating *Streptococcus pluton* has been developed. The contents of the mid-guts of diseased larvae are smeared on to microscope slides and allowed to dry. Most, usually all, of the bacteria associated with infection, except *S. pluton*, die in these smears after a few days; but *S. pluton* remains viable in such dry smears for at least 15 months. Cultures prepared from dried smears, made on the special medium developed for *S. pluton*, give many more colonies of this organism than do inocula of similar size from fresh material.

Growth of S. pluton is completely inhibited in vitro by penicillin G (concentration $10^{-7}-10^{-9}$); terramycin $(10^{-5}-10^{-7})$ and streptomycin $(10^{-4}-10^{-6})$. Small-scale field trials with terramycin have confirmed its ability to suppress visible signs of this disease, but the growth of S. pluton in heavily infected colonies was not entirely eliminated by treatment with this antibiotic. The walls of those cells which contained pupae that had developed from infected larvae became smeared with living S. pluton. This is a normal occurrence in untreated infected colonies, and seems likely to be the principal method by which S. pluton is transmitted; infected larvae which survive void the bacteria with their faeces at the time of pupation.

S. pluton, in dry smears, has been found to be extremely sensitive to fumigation by the vapour of formaldehyde or acetic acid at room temperature. No growth *in vitro* was obtained after exposure of smears to the fumes for between 5 and 15 minutes. Although comb fumigation is unlikely to provide a method of treating infected colonies, it may prove valuable for the treatment of empty combs and equipment suspected to be contaminated.

The distribution of *Bacterium eurydice*, which seems to cause visible signs of disease when superimposed on infection by *S. pluton*, is not confined to colonies with European Foul Brood disease. A survey, made with the assistance of members of the Bee Research Association, has shown it to occur in Britain and Northern Ireland in regions where European Foul Brood has rarely, if ever, been found.

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Three distinct types of *B. eurydice* have been identified. One grows very feebly *in vitro* as an anaerobe, and most adult and larval bees are infected with it at all times. The other two varieties are facultative anaerobes, grow vigorously *in vitro*, and occur in most adult and larval bees in summer, but they are difficult to find in winter.

larval bees in summer, but they are difficult to find in winter. B. eurydice grows well in a pollen extract prepared by soaking 1 part by weight of pollen, trapped from pollen-gathering bees on their return to the hive or taken from the comb, in 9 parts of distilled water. The suspension is sterilized by Seitz-filtration. Although it is an excellent medium for B. eurydice, it does not support or influence growth of S. pluton. (Bailey.)