

Sulphonamides and American Foul Brood Disease of Bees

AN editorial note in a recent issue of *Gleanings in Bee Culture* (72, 493; 1944) reports on the use of sulphathiazole in the treatment of American foul brood disease of bees by Prof. Haseman, University of Missouri, Columbia. Sugar syrup containing sulphathiazole fed to the bees enabled them to raise healthy brood in combs containing the scales of larvæ which had died of the disease.

In 1943, Mr. C. A. Ekins, of Brookwood Hospital, Surrey, sent to this Department an account of some experiments he had been carrying out since May of that year, in which sulphapyridine was being used in the treatment of five colonies of bees affected with American foul brood. He continued his experiments until July 1943 (with further treatment as a preventative measure in the following spring), since which date, he reports, there has been no recurrence of the disease. No evidence of disease was found when the colonies were examined in May 1944 by the local officer appointed for the inspection of apiaries under the Foul Brood Disease of Bees Order.

Following upon Mr. Ekins' claims of complete success with his treatment, and after consultation with him, it was decided to carry out a trial of sulphapyridine in the experimental disease apiary at Rothamsted. Accordingly, in June 1944, two colonies of bees of approximately equal strength, and headed by hybrid queens of the same age, were infected with American foul brood by feeding them a suspension of the spores of *Bacillus larvæ* in sugar syrup. The disease was allowed to run its course in both colonies until August 3, when one of them was fed 600 ml. of sugar syrup containing 3 gm. of soluble sulphapyridine. This treatment was repeated at weekly intervals, four doses being given in all. The other colony was fed plain syrup in equal quantities on the same dates. On September 8 the bees in both colonies were killed and the combs then examined. No major honey flow occurred during the period of treatment.

There was a marked difference between the two sets of combs. In those from the treated colony all the recent brood appeared healthy, though somewhat scattered and irregular in arrangement. No larvæ in the early or rosy stage of the disease were found, with the exception of two individuals situated on a comb remote from the three combs forming the actual brood-nest. Scales, the formation of which normally takes about three weeks from the death of the larva, were, however, present in eight out of the eleven combs in the hive. In the combs from the untreated colony many rosy larvæ were present, along with others showing the progressive stages leading to the formation of the scale.

It would appear, therefore, that during the course of the treatment the progress of the disease within the colony had been arrested, and that only healthy brood was being reared in combs where the disease had previously been established. The test was a severe one owing to the length of time which elapsed between infection and the beginning of the treatment; it was purposely not carried to its ultimate conclusion because of the lateness of the season and because of the danger of robbing by bees from other hives nearby.

It should be noted that the conditions of the experiment were not identical with those of the ex-

periments carried out by Mr. Ekins, who combined manipulative methods with the sulphapyridine treatment, and a complete elimination of the disease, as claimed by Mr. Ekins with his methods, was not obtained. The result does, however, justify the planning of future tests on a larger scale.

P. S. MILNE.

Bee Department,
Rothamsted Experimental Station,
Harpenden, Herts.

Phagocytosis and Storage of Trypan Blue in the Appendix of the Rabbit

THE observations of Baker and Enticknap¹ substantiate the claim of Bizzozero and Ruffer (cited by McEwan²) that an active phagocytosis occurs in the appendix of the rabbit, the agents concerned being large mononuclear cells present in the lymph follicles and lymph papillæ of the submucosa. The following observations made on the appendices of vitally stained adult rabbits throw additional light on this process.

The dye, trypan blue, was administered subcutaneously in a 2 per cent solution in distilled water and the animals were killed at various intervals after stopping the injection. The amounts of the dye used, the frequency of the injections and the times when the animals were killed are given in the accompanying table.

Rabbit	Daily dose of trypan blue (c.c.)	No. of injections	Time between last injection and killing
1	2.5	1	12 hr.
2	2.5	2	2 days
3	2.5	2	10 days
4	5.0	2	2 days
5	5.0	2	10 days
6	7.0	2	2 days
7	7.0	2	5 days
8	10.0	2	2 days
9	10.0	2	10 days

Following the administration of trypan blue in moderate concentration (R 1, R 2 and R 3) particulate segregation of dye particles occurred in the cytoplasm of cells in the serous coat, in reticulum cells surrounding the follicles and in certain cells of the mucosa. The dye-containing cells in the serosal coat were frankly macrophagic. Those in the peri-follicular tissue were rather smaller, but the majority of them exhibited the staining characters of macrophages. A few showed an early rosette arrangement of the dye particles often seen in monocytes. These cells were abundant in the region of the peripheral lymph sinus of the follicles, and at many sites appeared to project into its lumen. Cells of similar size and with identical staining characters occurred near the lymph channels in the mucosa. At other situations in this zone, notably around encysted sporozoa (abundant in larger or smaller groups), macrophages with a more bulky cytoplasm were electively stained, and the appearances suggested that many of them play an important part in the defence reaction of the mucosal tissues towards these parasites. The follicular lymphocytes were not affected by the dye; but numerous large moribund cells and disintegrating cell debris in the centre, and to a less extent at the periphery of the follicles, were diffusely stained (see photo.).

The administration of the dye in larger amounts (R 4, R 5, R 6 and R 7) produced a marked increase in the number of dye-containing cells in the peri-follicular zone and around the mucosal lymph