

UNIVERSITY OF MISSOURI COLLEGE OF AGRICULTURE
AGRICULTURAL EXPERIMENT STATION

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Controlling American Foulbrood with Sulfa Drugs

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Beekeeping would be a most pleasant and profitable line of work if it were not for the fact that the bee, like every other animal, has its ailments and is subject to the whims of nature. Of these ailments, the so-called American foulbrood is the most important. It affects the larva during the first four or five days after the egg hatches and, like typhoid in man, is presumably acquired with the food given to the young by the nurse bees. It does not affect the mature bees, except to completely destroy morale in case of a badly affected colony. By robbing, strong colonies are able to pick it up with honey taken from weak affected colonies.

In the early days it was described as being so infectious that a beekeeper scarcely dared visit in an affected apiary lest he pick it up and carry it to his bees, and inspectors were required to thoroughly disinfect their equipment and person before going into another apiary. Also, it was claimed that one affected colony in an apiary was sure to give it to all the rest of the colonies and to all apiaries in the neighborhood. Bee inspection laws were passed by most of the honey-producing states in hopes that it could be exterminated by inspection and compulsory destruction of the affected colonies. Thousands of colonies, including bees, combs and equipment, have been and still are being burned in an effort to stamp it out. However, about 15 years ago, some effort was made to save and clean up infected combs. Formaldehyde and, later, chlorine solutions were found to be effective in destroying the resistant spores of American foulbrood in the dry scales in infected combs, but such treatments did not prove practical and were abandoned after a few years.

Some thirty years ago the senior author, cooperating with a local beekeeper, began a series of experiments designed to study the disease under controlled condition in the department apiary. Combs diagnosed by the federal specialists as showing infection with American foulbrood were given to colonies and the course of development followed by the disease was observed. In these early experiments we found that some colonies removed the dry scales and cleaned up the cells and reared normal brood in them throughout the season. This was contrary to all previous findings and when we reported these results at our annual winter beekeeping short course in 1916-17 it created considerable local discussion, which later spread to other states. As the disease studies progressed we repeatedly found in our experimental yard and in commercial

apiaries colonies which persisted in remaining free of the disease either when exposed to it in nature or when diseased combs were introduced.

Other workers began similar studies, with the result that, in time, many practical beekeepers questioned the earlier dread of the disease which at times bordered on hysteria. As a result, it was found that the disease did not necessarily spread like wildfire and that some colonies could both avoid it and clean it up if introduced. These findings were responsible for the more recent nationwide search for colonies which persisted in escaping American foulbrood and the present program of rearing so-called disease-resistant queens. A number of the men now interested in rearing and distributing resistant queens remember well those lively bee disease discussions held in connection with the annual Missouri beekeeping short courses.

Naturally, we have been interested in and have cooperated with the program of rearing and testing resistant strains of queens. We have urged our beekeepers who have had trouble with the disease to turn to the so-called resistant queens and many Missouri beekeepers have requeened their apiaries with these queens and some have found them of help in reducing disease losses. However, resistant queens are not solving the problem, and, even with more rigid selection, it will be a long time before all beekeeping regions can hope to rid the woods and neglected apiaries of susceptible colonies.

As a result, in 1942, we decided to include in our bee disease studies the use of sulfa drugs and possibly others of the newer bactericides as a possible means of cleaning up infection in diseased colonies and of preventing the infection getting started in healthy colonies. On the advice of members of the staff of the medical school at the University of Missouri, and of Dr. Perrin H. Long of John Hopkins School of Medicine, it was decided to begin by feeding the different sulfa drugs either in water or in sugar syrup, using a 10 milligrams percent dilution. This means 10 milligrams of the drug to each 100 grams or cubic centimeters of solution. However, in the early feeding experiments we gave some small feedings of ten times and, in one experiment, twenty times this concentration of sulfanilamide. However, these more concentrated solutions seemed to be somewhat toxic when considerable amounts were fed to weak colonies. For experimental feedings, 50 mgms. percent solutions have also been fed, but in most of our large scale feedings we have used approximately 15 mgms. percent solutions and these have shown no evidence of being toxic to either the adult bees or the brood.

Our first few feeding tests were with sulfanilamide, but we also fed solutions of sulfathiazole and sulfadiazine. The early feedings

showed that sulfathiazole was most prompt and most effective in clearing up infection, so in all the later more extensive feeding experiments we used sulfathiazole. For convenience of preparing, we used a single 500 mgm. tablet to a gallon of solution in most cases. This is slightly more than a 15 mgms. percent solution and is convenient for practical use. To prepare the solution, break up the tablet into a fine powder and add to a gallon of hot water and give time for the white powder to disappear. Then add the sugar, using a pound to a pint or more of the hot water containing the sulfa drug, depending on the season of the year or the amount of sugar you have to spare. When the drug is given to the bees in the drinking water, simply leave out the sugar. However, in water the drug tends to settle out of the solution.

For late fall or winter and early spring feeding, we have also combined the sulfathiazole with a pollen substitute. The pollen substitute is prepared, as follows: Heat a gallon of water to about 180°F. and dissolve a one-half gram or 8 grain tablet of sulfathiazole in it, and add two gallons of sugar while water is hot. Then to a gallon of this thick syrup stir in one pound of dry skim milk powder until it is thoroughly wet and add four pounds of soybean flour, and allow to set overnight. In the morning, add at least a quart more of the solution and stir or knead in thoroughly. When ready to give to the bees it should be just thick enough that it will not run down between the top bars of the frames where it is placed as a flat cake.

In one single experiment, we also checked the possibility of sterilizing diseased combs with concentrated solutions of sulfa drugs. An abandoned comb from a diseased colony with most of the cells containing disease scales was used. On one side marked with a "D" each cell was filled by means of a medicine dropper with a 200 mgms. percent sulfadiazine solution and left at room temperature for 13 days. The comb was then turned over, marked with a "T", and each cell on this side was filled in the same way with a 100 mgms. percent sulfathiazole solution. Each cell was filled to overflowing with the solution and again left at room temperature about 90°F. for about a week. In each case, the sulfa solutions seemed to soften and loosen the scales and other cell contents and the mouth of each cell filled with tiny bubbles. Before giving the comb to a colony in the small apiary of the senior author, it was held under a water faucet to wash out the sulfa drug and other loose materials from the cells. This experiment was not repeated, but the bees, a 1943 swarm, cleaned up the comb and soon had both sides filled with healthy brood. No disease appeared in this or the other combs and one super of surplus honey was taken off in 1943. In 1944, this and two or three other colonies in the apiary had dysentery and two colonies had American foulbrood, but this colony still seems to be free of American foulbrood.

Experimental Results

Experiment 1. The first series of sulfa drug feeding experiments was begun May 22, 1942, in the Agricultural Experiment Station apiary in Columbia. For this experiment, three weak, diseased colonies, or they might better be termed nuclei, were selected.

Colony 1:—This was a three-banded Italian colony which was so far gone with disease that it had practically no cells containing healthy grubs and there was not more than one pound of bees. It was in a standard 10-frame hive with all the brood combs so reeking with disease that the queen had moved up into the super, and here too most of the brood was diseased.

On May 22 it was given 650 cc. of dilute sugar syrup containing 648 mgms. of sulfanilamide, which was approximately a 100 mgms. percent solution of the drug. The solution was sprinkled into the cells of the combs. On May 27, it was found that a large percent of the workers and some of the brood were dead. Later, it was found that the queen also was gone and a capped queen cell was given to it.

On June 19, the colony was still in bad shape, with no developing new brood noted but some capped brood and plenty of old dead brood. Then, on June 23, it was given a smaller feeding, 324 cc. of the 100 mgms. percent sulfanilamide in sugar solution. At this time the colony had considerable healthy young brood and showed signs of recovering from the toxic action of the first feeding. The smaller dose given at this time did not seem to cause any ill effects either on the adult bees or on the grubs, though some small grubs were floated up out of their cells when the medicated syrup was sprinkled over the combs.

On July 8, the colony was examined and the young brood all seemed healthy. though there was still some of the old dead larvae. The newly capped brood all seemed healthy.

Six days later, July 14, the colony was again fed 325 cc. of a 100 mgms. percent solution of sulfanilamide. At that time the young brood all seemed to be healthy but there were still plenty of disease scales and old dead larvae in the cells of the combs. The colony was definitely building up.

On July 27, the colony was again fed the same amount of syrup containing 100 mgms. percent sulfanilamide. The new brood all seemed to be healthy, but the colony was still weak and the combs still had old dead larvae.

Throughout August and September the colony received no further syrup containing the sulfanilamide and there was only a light fall nectar flow. As a result, on October 12, when the colony was examined it was found that some of the late brood was again diseased, and the colony had only about 10 pounds of stores, and it was no stronger than in late July when the feeding of the sulfa drug was discontinued. This seemed to indicate that so long as there

is evidence of the disease in the hive the sulfa drug must be fed continuously, or at least at short intervals, since in this case its effects were lost after two months. However, in this experiment the young brood was kept free of the disease while the sulfanilamide sugar solution was fed. The colony was too weak to survive the following winter.

Colony 2:—This three-banded Italian colony likewise was one that the disease had almost wiped out. It had recently reared a new queen, had perhaps one pound of bees and very little scattered healthy brood. All the diseased combs were left in the hive and the colony was given light feedings of 200 mgms. percent sulfanilamide-sugar solutions. The first feeding was made on May 27 and included only 325 cc. of the solution. With this smaller feeding of an even more concentrated dosage of sulfanilamide no ill effects on either workers, queen, or brood were noted. They immediately began to clean up and to produce healthy young brood.

A second feeding, including the same amount of the same solution, was made on June 23 by sprinkling over the combs. The young brood all appeared to be healthy, but the combs contained old dead diseased brood and scales. This colony was carefully gone over on July 8 and all new sealed and unsealed brood appeared to be healthy, but old diseased grubs and scales were still abundant in the combs.

The colony was fed on July 14, and again given the same amount of the same solution. At that time, healthy brood was present on six or seven of the combs, but the colony had but little honey.

On July 27, the colony was again fed and the same amount, 325 cc. of the same solution was sprinkled over the combs. At that time, the colony was building up rapidly and the morale of the workers was good. As in case of Colony 1, this was the last feeding this colony received.

When examined on October 12 the colony was only fairly strong. There was not much brood and some of this had evidently died of the disease since the last feeding in July. The colony was in better condition than Colony 1, for it had about forty pounds of honey and about enough bees to survive the winter. It had gone farther than Colony 1 in cleaning up and building up, but the long period in August and September, when no sulfanilamide was given, enabled the disease to build up again. The colony struggled along but did not survive the winter.

Colony 3:—This was a fairly strong colony of mixed Caucasians and Italians. Its new queen emerged as the feeding experiment began. The combs had an abundance of diseased brood and scales, but little healthy brood. This colony was given 650 cc. of a 50 mgms. percent sulfanilamide-sugar solution first on June 23. When examined on July 8 it had an abundance of diseased brood, but also much new healthy brood.

A second feeding of 650 cc. of a 50 mgms. percent sulfanilamide-sugar solution was given on July 14. At that time the young queen was laying rapidly and had eggs and healthy brood on most of the ten combs. However, there was plenty of old dead grubs and scales.

The next feeding was made on July 27, and the same amount of the same solution as before was given. On that date the colony was building up rapidly with all the combs about full of brood, most of which was healthy. Only an occasional dead larva was present, but there were still some capped cells with old dead larvae. This was the last feeding given this colony.

When examined on October 12 the combs were again overrun with the disease. There was little brood and it was scattered, but the colony had about 100 pounds of honey. The lighter dosage of sulfanilamide, combined with the larger amount of honey, probably so diluted the sulfa drug that in the two months when no sulfanilamide was given the disease was able to spread more rapidly than in either Colony 1 or 2. This colony survived the winter, but eventually succumbed to the disease.

Experiment 2. The second series of experiments on the control of American foulbrood with sulfa drugs was carried on in the commercial apiary of the junior author, beginning in July, 1942. It has been his habit to purchase a number of packages of bees from the South each spring, and in 1941 everyone of his new packages and all of his other colonies came down with the disease before fall. A diseased yard in the neighborhood was found to be the source of his yearly trouble. For three years the disease practically cleaned out his apiary.

For this experiment, six diseased colonies were selected, the first three of which, Colony 1, 2, and 3, were fed sulfathiazole, and Colony 4, 5, and 6 received sulfadiazine. Colony 1 was a strong one, Colony 2 was weak, and Colony 3 was medium strong. Colony 4 was weak, Colony 5 medium strong, and Colony 6 was strong.

For ease of mixing, two one-half gram tablets of the sulfa drugs were ground to a fine powder in a mortar and added to one-half gallon of hot water, to which a cupful of sugar was added as sweetening. Each colony was fed a third of a half-gallon of this treated syrup each week in a Boardman feeder. These solutions were close to 60 mgms. percent sulfathiazole and sulfadiazine solutions respectively. As the summer passed, it was observed that Colony 1, 2, and 3, which received sulfathiazole, were cleaning up the disease and their new brood was all healthy, while Colony 4, 5, and 6, which received the sulfadiazine, did not seem to be making much headway with the disease and eventually they all succumbed to the disease. It was noted that these drugs tended to settle out of the weak syrup solution, especially the sulfadiazine. Heavier sugar solutions seem to hold the drug better. A light nectar flow was available, but feeding of the drugs continued until October.

Colony 2, which started out weak, presented a convincing picture of what the sulfathiazole will do for American foulbrood. The combs were so full of dead grubs that the queen had moved over to one side of the hive. As the drug was used the new brood began to remain healthy and the colony started cleaning up the hive comb by comb to provide room for the queen. By October, or when brood-rearing began to let up, five combs were cleaned and in use and the colony had stored sufficient honey to carry it through the winter.

Besides building up and producing clean healthy brood after the sulfathiazole was fed to the three colonies, it was noted that the general temper and the morale of the colonies improved. Toward fall, no longer was one confronted with the spiteful zip up the sleeve and a sting. The colonies were completely transformed in disposition, health, and strength by the drug.

Experiment 3. The third series of experiments was carried out in the junior author's apiary in 1943 and, for a number of reasons, it did not turn out as well as we had reason to expect. The feeding experiments were begun too late in the season for one thing. Too many diseased colonies were included in the experiments for the amount of sulfa drug available. Only small quantities were given to each colony by means of a Boardman feeder. Small quantities of pollen substitute containing the sulfa drug were also fed to the bees up until corn began to tassel. In the flight range of the apiary in 1943 there was a fair nectar flow, which further diluted the inadequate sulfa drug given to the bees. As a result, while the use of the drug definitely inhibited the progress of the disease, when fall came there were a total of 31 colonies showing varying degrees of the disease.

Our experience in 1943 only convinced us that if the use of a sulfa drug to control American foulbrood is workable in a commercial apiary and not merely in a few experimental colonies we would have to lay out our plans on a much larger scale and make use of all known facts about bees and the way the disease operates. It is well known to every experienced beekeeper that immediately the main honey flow stops every field worker becomes an inveterate robber. Then, woe be to the weak colony not able to protect its stores. Also, woe be to the robber colony if its workers bring home honey from a weak diseased colony. Robbing is one of the most exasperating problems the beekeeper must contend with. Frequently, the strongest colony with the tallest stack of filled supers at the end of one season is the first colony to go down with disease the next year.

Experiment 4. The fourth series of experiments was laid out in the junior author's apiary in the fall of 1943. Of the 31 colonies showing disease, 16 were gassed and the other 15 were prepared for winter and spring treatment with sulfa drugs. From his later ex-

perience the junior author vows that he will never again gas a diseased colony that has even a few thousand bees. Most of the colonies were given a single brood chamber though a few were also given a partly filled super of honey. Each comb in the brood chamber was examined and those showing disease were removed and replaced with combs from the supers. A few of the colonies were given only six or seven combs for the winter, the others being too full of disease. This fourth experiment, therefore, began with 15 strong nuclei wintered on diseased honey but with no combs showing diseased grubs or scales. Each of them had enough honey to carry them through the winter, so no sugar syrup containing half a gram of sulfathiazole to the gallon was fed during the fall. However, later findings indicate that we should have given each colony at least some of the sulfa syrup for winter stores. The 15 colonies were not wrapped or packed for the winter, but they were sheltered in a wooded draw.

The junior author keeps systematic field notes on each of his colonies and the following records on five colonies taken at random from the lot of 15 colonies used in this experiment will show what treatment was given and what effect it had on the disease.

On February 21, we lifted the hive covers and spread on a large thin layer of pollen substitute cake. Thereafter the weather turned cold and the hives could not be opened until March 14, at which time every hive in the yard was carefully inspected and more pollen cake given. It was observed that the pollen cake must be spread immediately over the brood nest, for in cold weather the bees will not forage for it and no benefit will accrue. By turning the inner cover over, space is provided for the thin layer of pollen cake.

Colony 20

- 11-24-43. Foulbrood bad, 5 frames badly diseased. Removed them and gave five shallow frames of honey.
- 3-14-44. Bad start of disease in one frame. Abundance of brood and bees. Short of honey. Gave treated syrup and pollen substitute.
- 4- 1-44. Two cells with dead brood, many scales, are cleaning up. Eating pollen cake, queen clipped, feeder can nearly empty.
- 4-19-44. Saw no indication whatever of foulbrood. Gave frame of honey, new cake of pollen, and can of syrup. Have 5 frames of brood and eggs.
- 4-30-44. No disease—pollen cake gone.
- 7-10-44. No disease, lots of brood, two supers nearly full of honey.

Colony 17

- 11-24-43. Foulbrood bad, four frames taken out. Have food chamber and left that on.
- 3-14-44. Disease in the old brood nest. Were eating pollen cake. No disease in new brood. A good hive.
- 4- 1-44. Some disease in old nest, none in the new brood. Gave new supply of pollen cake and syrup.
- 4-19-44. No disease, cake and syrup gone. Gave more. Brood expanding rapidly.

- 4-30-44. Saw two dead grubs. No diseased scales. Six full frames of brood, three of honey.
 7-10-44. No disease. Two brood chambers well-filled with brood, and one large and one small super of honey ready to extract.

Colony 15

- 11-24-43. Foulbrood bad. Removed 5 frames, none given back.
 3-14-44. Foulbrood in old brood nest, lot of it. Had eaten all of pollen cake and there was no disease in the circle of new brood.
 4-19-44. Saw no evidence of disease. The queen had laid through the old brood nest and young bees were emerging from cells which still had in them the scales of old diseased brood.
 4-30-44. Could see no disease at all.
 7-10-44. No evidence whatever of disease, hive full of bees and 40 pounds of honey ready to take.

Colony 24

- 11-24-43. No foulbrood. This hive went into winter apparently clear of disease. It had two brood chambers.
 3-14-44. Lot of foulbrood. Three frames bad, took away one brood chamber and gave medicated pollen substitute and syrup.
 4- 1-44. Most disease of any of the hives, all located in old brood nest. New brood appears clear of it.
 4-22-44. Could see no disease. Had eaten all pollen cake and syrup, had five full frames of brood. Gave pollen cake and syrup.
 4-30-44. No disease, 7 full frames of brood, now ready for second brood chamber.
 7-10-44. No disease, two supers of honey.

Colony 16a

- 11-24-43. No foulbrood. Hive clear. Two brood chambers of honey. Leave as is.
 3-14-44. Bees all in top brood chamber. Four frames two-thirds full of brood and fully half of it dead with disease. Took away bottom brood chamber, removed four frames of honey, extracted them, put back the empty combs and fed syrup and pollen cake. This was a large hive and it took five or six pails of the syrup as food. We poured it in.
 4-30-44. There was but little evidence of disease left.
 7-10-44. It is clear of disease and has two supers of honey.

In 1942, the junior author purchased fifty-one two-pound packages headed with resistant queens. By November, thirty of these had come down with the disease, and by November of the next year all but seven were gone. One of these is still in the yard and has never had disease. The other six had disease this spring, but cleaned up readily with the treatment. In all fairness, it must be said that this heavy mortality of resistant bees was due to the presence of seventy-five neighboring colonies in old-fashioned "gums" that were keeping his bees supplied with disease. His bees are still combating that menace, and if the bees in this yard come through another winter without infection that will be an added score for sulfa treatment. It might be added that in this state there is no bee inspection law.

Experiment 5. A fifth experiment, begun the first of April, 1944 in the junior author's apiary, included the continuous feeding of small quantities of a dilute sugar solution containing sulfathiazole to fifty packages of bees received from the South. The solution contained a one-half gram tablet and one pound of sugar to a gallon of water, which is approximately a 15 mgms. percent sulfathiazole-sugar solution. This experiment had two objectives, first, to determine whether or not colonies developed from packages in the presence of American foulbrood can be kept free of the disease, and second, to determine what possible effect the sulfathiazole might have on the *Nosema* organism.

These fifty packages were shipped on April 4 and arrived about noon on the 6th. Half of them were installed that afternoon and the remaining half the next afternoon. The bees came through in splendid shape and they were a superior lot. There was scarcely any dead bees in the bottom of the cages. The hives had been previously made up and arranged on their stands, 10 feet one way and 20 feet the other. They were numbered from 1 to 50 and each contained five empty, dry, drawn combs. Treated syrup, containing half a gram of sulfathiazole to a gallon, and treated pollen substitute had been prepared and was waiting. The method of installing used was not very different from the standard, except that after wetting the bees the wooden block was pried off the cluster jarred down and, before they could get reorganized, the cage was inverted and everything was shaken into the hive. With a half-gallon fruit jar filled with sweetened water and prepared pepper box fashion, the mass was sprinkled while the feeder can and queen cage were being retrieved. Then the combs were slid over the struggling bees. The queen was then wetted, the screen pried off, and care taken to see that she fell down among her bees. The feeder can was then put in place, the hive closed and not opened again for about five days to let the queen get adjusted, and at which time pollen substitute was spread over the cluster.

We were particular to see that the feeder cans always had syrup in them and it is surprising the amount of pollen substitute package bees will eat. Special pains were taken to keep that before them. Too much emphasis cannot be placed on this item for package bees. Most of them are young nurse bees and should be husbanded to hover the brood. Package bees never contain enough old bees to gather adequate pollen for rapid development. The progress of these bees was rapid though we had four snows in April and it was the second coldest April in the history of our weather records.

The bees were fed treated sugar syrup, as described, until the first of June and a few hives until the 17th of June. During the last days of May the rationing board advised that the sugar allotment for feeding these bees was running low and that no more could be had. That threw a scare into the enterprise. There were available

several supers of unextracted honey stored the year before, partly by diseased colonies and partly by disease-free colonies. No effort had been made to keep it separate. But this threat looked serious and in the quandary we went carefully over the results of our feeding trials on the fifteen diseased colonies in Experiment 4. We had reason to believe our treatment would control any disease that might develop, so this honey was divided so as to give each package two frames. Care was taken that no frame of honey was used in which a queen had ever laid. Part of this honey undoubtedly contained American foulbrood spores and those colonies receiving the infected honey were certainly exposed to the disease. However, to this day, September 10, there has never been a single cell of American foulbrood in any of these colonies that we could find. They have all done well, stored surplus, and are busy with the fall nectar flow. The prophylactic action of sulfathiazole in protecting these fifty colonies developed from packages is particularly noteworthy, in view of the fact that each season for the past four years over fifty percent of the new packages in this apiary developed American foulbrood by the first of September. These bees had access to American foulbrood infection in a neighboring apiary and they were also given combs of honey stored by diseased colonies, so it seems evident that the sulfathiazole fed to them prevented them from coming down with the disease.

The second objective of Experiment 5, namely, that dealing with a study of the possible effect of sulfathiazole on Nosema was inspired by the articles by Farrar, Rahmlow and Gardner in the bee journals. Their findings reminded the writers that in the early feeding experiments sulfathiazole seemed to give to the worker bees themselves a virile look and to restore morale which all diseased colonies, sooner or later, lose completely. In spite of the general claim that American foulbrood has no ill effects on the adult workers, it is perfectly evident to those who have worked extensively with this disease that the individual workers in a diseased colony are below par and that they are ailing from some cause. While we were engrossed in following through the study of sulfa drugs as a protection to the brood, we felt that we should also study their possible beneficial effects on the adult bees affected with Nosema. In the early feeding experiments, we did not check the adults specifically for Nosema, but the fifty packages of bees used in this experiment gave us an excellent opportunity to study the action of sulfathiazole on Nosema.

After the fifty packages of bees were all settled in the hives, but before they received the sulfa drug, we collected samples of dead bees from each hive, numbered them from 1 to 50, and on April 26 sent them to the North Central States Bee Culture Laboratory at Madison, Wisconsin, to be checked for Nosema. Twenty-nine of the

samples were infected with *Nosema*, nine of them (Nos. 11, 17, 21, 22, 26, 31, 41, 46 and 48) heavily infected.

After feeding the colonies treated syrup and pollen substitute, as previously described, we sent a second sample of bees from the nine colonies previously found heavily affected with *Nosema* on July 22. The diagnosis showed only one sample, #26, infected with *Nosema* at that time. A third series of ten samples, including one from Colony #26, which still showed *Nosema* in July, were submitted on September 2 and the microscopic examination showed none of the samples infected with *Nosema*. Further tests are to be made to determine if all of the colonies which showed *Nosema* infection last spring, before they began receiving the sulfa treatment, are actually free of *Nosema*. If they prove to be free of the disease we shall be inclined to give the sulfa drug credit for the cure as well as for the numerous other striking cases of improvement in vigor and morale of the adult bees in the various colonies treated for American foulbrood. Also, our results and observations lead us to raise the question whether *Nosema* and American foulbrood may not work in teams with perhaps a much closer relation existing between these two ailments than past findings might indicate.

Experiment 6. A sixth experiment was begun in the junior author's apiary in May, 1944. Two early May swarms were selected for the trial. Both were given young queens from the same parent colony. These swarms were hived on old drawn combs and they both started off with brood disease. Each was fed treated pollen substitute when the disease was discovered and also two one-half gallon jars of sugar syrup containing a half gram of sulfathiazole to a gallon. That was all the feeding they had at the time. Both colonies cleaned up all signs of American foulbrood. Then on July 10 two combs with American foulbrood showing dead brood not yet dried to scales (see Figure 1) were put down in the middle of each hive and two more one-half gallon jars of the prepared feed were given along with treated pollen substitute. Both colonies cleaned out the diseased debris and burnished the cells, but the queens did not accept the combs until the odor was gone. Hive No. 2 accepted her comb first but laid around sporadically. While we did not see any signs of disease, we thought best to treat them again and, therefore, gave this colony a jar of water containing one-half gram of sulfathiazole to a gallon. No signs of disease appeared later. The queen in Hive No. 1 did not accept her worst comb for nearly three weeks, but thereafter she never faltered. Figure 2 shows the second cycle of her brood and four healthy queen cells which were drawn out after the comb with adhering bees was set over in a hive as a nucleus for further study. Both colonies stored considerable pollen around the brood and they are going into the winter free of any signs of foulbrood.

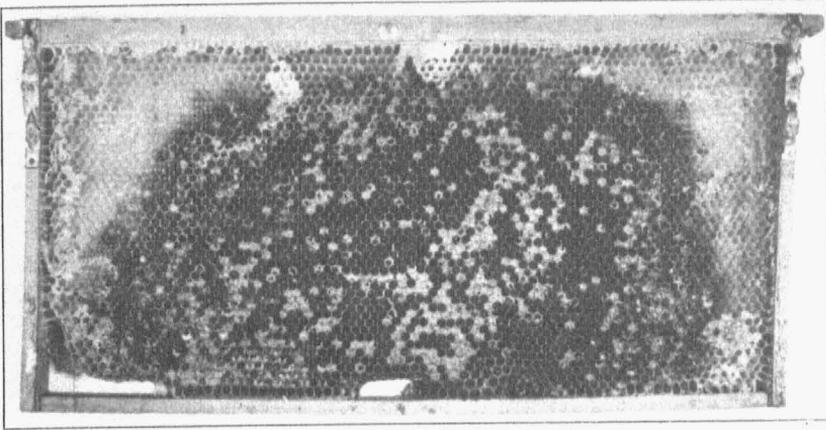


Fig. 1.—One of two combs badly infected with American foulbrood given to Colony 1 on July 10, 1944, as described in Experiment 6.

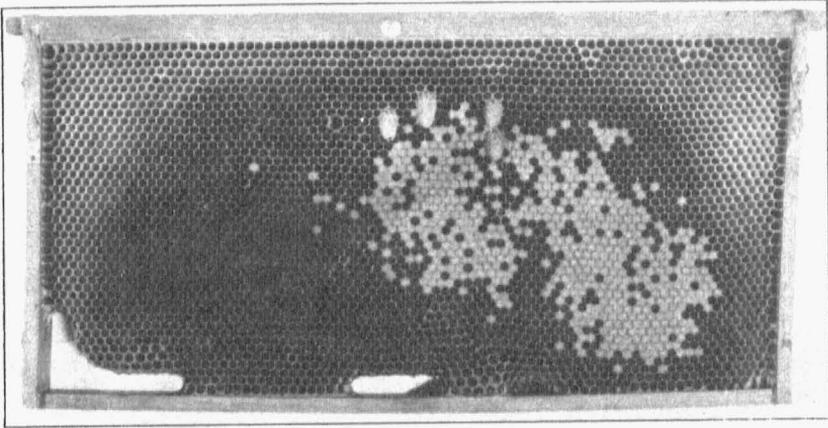


Fig. 2.—The same comb shown in Figure 1 as it appeared September 8, 1944, after being cleaned out by colony of bees fed sulfathiazole, as described in Experiment 6.

This experiment alone should be sufficient to convince the most skeptical of the possibilities of sulfathiazole in actually cleaning up American foulbrood. It should be observed, however, that these infected combs were given to colonies which, six weeks before, had been cleared of American foulbrood and indications of *Nosema*, and the bees were, therefore, strong and vigorous and the queens were young and virile. It would probably have taken them longer to clean up, had the colonies been headed with old queens and rundown workers.

These experiments are being continued at the Experiment Station. It is felt that if one of these new drugs can be effectively and universally used to prevent and control American foulbrood and possibly *Nosema*, it will prove the greatest boon to beekeeping since

the discovery of the movable comb hive. It is believed that if all beekeepers would include the feeding of sulfathiazole in sugar syrup and pollen substitute as a regular part of the routine of fall and spring bee management, we might safely expect to control American foulbrood and possibly *Nosema* fully as well as we now control hog cholera.

The results of these experiments to date indicate, first, that sulfathiazole is harmless to the brood and adult bees when fed continuously for 60 days, at the rate of one-half gram tablet in a gallon of water or sugar syrup; second, that in the presence of sulfathiazole the American foulbrood bacillus will not develop; and third, that it has a very definite beneficial action on bees infected with the *Nosema* parasite. These investigations are not yet completed, and this therefore is in the nature of a progress report. However, to those wishing to test out the use of sulfathiazole for protecting bees from disease or in helping rid colonies of the disease we would recommend the following procedure:

(1) In the fall, when bees are prepared for the winter, check each colony and where American foulbrood appears remove combs showing disease scales and dead grubs and replace them with combs of honey from the super.

(2) To each colony, feed at least one gallon of sugar syrup containing a one-half gram tablet of sulfathiazole to the gallon of syrup, prepared as previously described. Give more of the treated syrup to those showing considerable disease, even replacing some of their winter stores with treated syrup.

(3) Prepare and give to each colony late in the fall and again beginning at least three weeks before natural pollen is available in the spring, a few pounds of the pollen substitute containing sulfathiazole, and prepared as previously described.

(4) Give all colonies winter protection to help conserve their strength.

(5) In the spring, keep a close watch on every colony and feed more treated syrup and pollen substitute when spring brood rearing begins.

(6) Continue to give small feedings of the syrup at least until the regular honey flow opens.

(7) To package bees, feed both treated syrup and pollen substitute until they have developed into strong nectar-gathering colonies.

(8) Requeen as necessary to make sure each colony keeps up to full strength, and be careful to prevent robbing.

(9) Cooperate with neighbor beekeepers and promptly dispose of any infection by rendering diseased combs.

(10) Arrange to secure sulfathiazole through your local medical officials, veterinarians, or others authorized to dispense this drug.