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FEEDING OXYTETRACYCLINES AS TERRAMYCIN (R) DOES NOT AGGRAVATE CHALKBROOD INFECTIONS [1]

By DIANA M. MENAPACE [2] and WILLIAM T. WILSON [3]

Honey Bee Pesticides/Diseases Research, Agricultural Research Science and Education Administration, U. S. Dept. of Agriculture Laramie, Wyoming, U.S.A. 82071

SUMMARY

Oxytetracycline (Terramycin(R) or TM) formulated in extender patties and chalkbrood-contaminated pollen cakes were fed to colonies of honey bees *(Apis mellifera* L.); other colonies were fed chalkbrood-contaminated pollen cakes only, uncontaminated pollen cakes only, or TM extender patties only. The degree of chalkbrood infection that developed in colonies fed oxytetracycline plus chalkbrood was the same as the degree that developed in colonies fed only chalkbrood. The colonies fed only TM extender patties had less chalkbrood than the control colonies, which were fed uncontaminated pollen cakes with or without extender patties or were not treated.

INTRODUCTION

In the past few years, the incidence of chalkbrood (CB), a disease caused by the fungus *Ascosphaera apis* (Maassen ex Claussen) Olive and Spiltoir, has increased rapidly in North America (MENAPACE and WILSON, 1976, and NELSON *et al.* 1976). As a result, there has been concern that this increase may reflect the wide-spread use of oxytetracycline (Terramycin(R) or TM) in the control and prevention of American foulbrood (AFB) and European foulbrood (EFB). One reason is the fact that humans treated with tetracyclines often develop monilial superinfections caused by the yeast *Candida albicans* (SMITH, 1969 and KUCERS, 1972). Apparently, the antibiotic kills the normal bacteria of the gut and the yeast grows uninhibited and

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(2) Microbiologist.

(3) Research Entomologist.

without competition. Therefore, other animal species, i.e. honey bees, may likewise be subject to mycological superinfections (fungi, yeasts and molds) because of the use of the tetracyclines. In fact, adult honey bees normally have nonpathogenic fungi in their intestines (GILLIAM and PREST, 1972), and antibiotics (fumagillin and oxytetracycline) do depress the intestinal fungal flora (GILLIAM *et al.* 1974). Therefore, oxytetracycline might affect the growth of fungi such as *Ascosphaera apis*. The following experiment was designed to investigate the influence of oxytetracycline on chalkbrood infections.

MATERIALS AND METHODS

Eighteen colonies of honey bees installed in May 1977 from three-pound packages were standardized so they had equal amounts of brood and equal numbers of bees. All colonies consisted of two 10-frame-deep hive bodies and one 10-frame-shallow with a modified O.A.C. pollen trap under each colony. The 18 colonies were divided into four treatment groups : six controls (two received no treatment, two received uncontaminated pollen cakes, and two received uncontaminated pollen cakes plus TM extender patties), four colonies received TM extender patties only, four colonies received only pollen cakes contaminated with CB spores, and four colonies received both TM extender patties and contaminated pollen cakes. Prior to treatment, all colonies were thoroughly inspected and found to be free of all bee brood diseases.

The TM extender patties used in the test were similar to those developed by WILSON *et al.* (1970) except that they consisted of 1,362 grams granulated sugar, 454 grams solid vegetable shortening and 182 grams oxytetracycline (TM 25 formulation) per eight 250 gram patties. The pollen cakes used in the test contained 50 grams pollen, 50 grams granulated sugar, and 8-10 ml distilled water per cake. The uncontamined pollen cakes were made with pollen collected from disease free colonies. The contaminated pollen cakes each contained two pulverized black chalkbrood mummies.

The test began June 20, 1977 when uncontaminated pollen cakes were put in the broodnest of colonies designated to receive uncontaminated pollen cakes and contaminated pollen cakes were put in the broodnest of colonies designated to receive them. The colonies that were not to be treated and the colonies that were not to be fed only TM extender patties received nothing. The pollen cakes were replaced weekly through August 23 so the colonies received pollen cakes for a total of ten weeks. Beginning June 27, and once a week until August 23, the TM extender patties were put on the broodnest of the designated colonies (total of nine patties).

Pollen traps and broodnest areas were examined for CB mummies weekly (weather permitting) for 10 weeks. Examining the broodnest meant checking three frames of capped brood in each of the two deep hive bodies of each colony. The CB infections were categorized as light (L) – an average of 1-5 mummies per each side of the frame, medium (M) – 6-10 mummies, and heavy (H) – 11 + mummies.

The data were evaluated by rating the level of CB in each colony each week (i.e. L = 5, M = 10, and H = 15) and calculating the average for each treatment group for the ten weeks. The average for the groups could then be compared. (If mummies were seen only in the pollen trap and not in the broodnest, the colony were considered to have a medium infection.) Also, each week, colonies without obvious CB infection was assigned « 0 », and colonies with obvious CB were assigned « 1 ». The CB infection in each treatment for the ten weeks was averaged, and treatment groups were compared. Any colonies with chronic « queen » problems (i.e. loss of a queen and difficulty in requeening) were eliminated from the computation.

RESULTS

At the initial examination, all colonies were free of visible signs of CB or any other bee disease. However, two colonies that were not in the test, but were in the same apiary were found to be infected with American foulbrood (AFB). Those colonies were removed to a hospital yard. The test colonies (except the two receiving no treatment and the two receiving only uncontaminated pollen cakes) were given TM extender patties for four days before the test to guard against the spread of AFB. (None of the treated colonies given the TM patties at this time ever showed signs of AFB, but one of the two control colonies did become infected and was replaced three weeks into the experiment.)

Within one week after the first CB-contaminated pollen cakes were put into the colonies, six of the eight colonies receiving this treatment had CB infections. Also, two control colonies (one receiving only uncontaminated pollen and the other receiving uncontaminated pollen plus TM patties) and one test colony (receiving only TM patties) developed CB infections. By the end of the ten-week test, all colonies except one of the control colonies (fed uncontaminated pollen) had shown signs of CB at least once. Those that were being fed CB had the most consistent infections, but one control colony that was receiving uncontaminated pollen plus TM patties was infected ten of the eleven times the colonies were examined (no CB was seen at the initial inspection).

One colony from each treatment group (except controls) was removed from the test because of chronic queen problems. This left three colonies in each treatment group and six control colonies.

By either method of computation, the colonies receiving only TM patties had the least amount of infection, even less than the six control colonies (Table 1 and Fig. 1). The infections in the control colonies were lighter than those in colonies fed CB but were definitely higher than those in colonies fed only TM patties (Table 1 and Fig. 1).

When the amount of CB in the colonies was rated L (5), M (10), or H (15) and the ratings for the ten-week period were averaged, it was found that colonies fed only CB had an average rating of 5.5; those fed CB plus TM patties had an average of 5.0 (Table 1).

Also, when one (1) was assigned for the presence of CB and zero (0) for the absence of CB and the average taken for each treatment group for the ten-week period, the results were essentially the same. As in the previous case, the colonies fed both CB and TM showed essentially the same amount of infection as the colonies fed CB only (0.73 compared with 0.70).

DISCUSSION

These results indicate that oxytetracycline does not enhance chalkbrood infections. Even looking at the results two different ways there was no great difference in the amount of infection in the colonies receiving only contaminated pollen and the colonies receiving contaminated pollen and extender patties. These two groups having basically the same amount of infection indicates two things. First, that oxytetracycline does not enhance CB infections. If the presence of TM in the hive enhanced CB infection, there would have been a noticeable difference in the amount of infection in the colonies fed only contaminated pollen and the colonies fed TM and contaminated pollen. The latter colonies would have had much more severe infections. Second, that oxytetracycline does not control or inhibit CB infections. If TM controlled or inhibited CB infections, the colonies fed contaminated pollen and TM would have had little or no infection compared to those colonies fed only contaminated pollen.

TABL. 1. — *Chalkbrood infections (a) in colonies fed oxytetracycline (TM) and/or chalkbrood-contaminated pollen for a ten-week period.*

Treatment	Colony No.	Date Examined										Total	Average
		6/27	7/5	7/11	7/18	7/26	8/1	8/8	8/16	8/23	8/30		
None	1 (21) *(b)*	0	0	0	0	0	0	0	5	5	0	10	
	12	0	0	0	0	5	15	15	15	5	10	65	3.75
Plain *(c)* pollen + TM	2	0	0	0	0	0	0	0	0	5	0	5	
	6	10	10	5	10	5	5	15	15	5	10	90	4.75
Plain *(c)* pollen	29	0	0	0	0	0	0	0	0	0	0	0	
	30	0	10	5	5	5	10	5	0	0	0	40	2.0
TM	11	0	0	0	0	0	5	0	0	0	5	10	
	16	0	0	0	0	0	0	5	0	0	0	5	
	18	10	0	0	5	0	0	5	0	0	0	20	1.17
	14 *(d)*												
CB	4	10	10	5	0	5	0	5	5	0	5	45	
	8	10	0	15	0	5	0	0	0	0	5	35	
	15	10	10	15	5	5	10	15	10	5	0	85	5.5
	17 *(d)*												
CB + TM	3	10	10	5	0	5	0	0	5	5	0	40	
	5	10	0	10	5	5	5	5	5	10	0	55	
	13	10	0	0	5	10	10	5	10	5	0	55	5.0
	7 *(d)*												

(a) 5 indicates light infection 1-5 mummies/each side 6 frames of capped brood.
10 indicates medium infection 6-10 mummies/each side 6 frames of capped brood.
15 indicates heavy infection 11+ mummies/each side 6 frames of capped brood.
(b) Colony No. 1 replaced with colony No. 21 on 7-11-1977.
(c) Plain pollen = pollen uncontaminated with CB.
(d) Queenless colony removed from test.

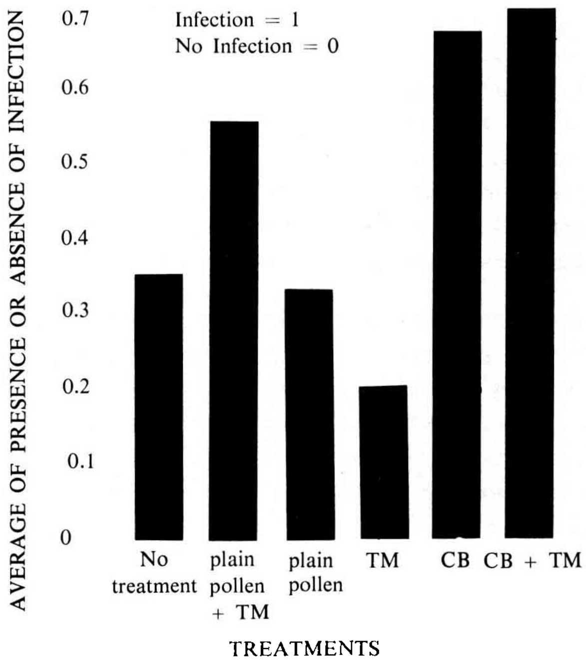

FIG. 1. — *Ten-week average of ratings for the presence or absence of CB infection in colonies receiving CB spores and/or oxytetracycline (TM), Laramie, Wyoming, 1977.*

The CB infections in the six control colonies and the colonies fed TM only are of interest because all were « natural » in that they were introduced from sources other than contaminated pollen cakes. These infections may have been the result of : 1) natural infections, undetected previously, 2) spores and/or hyphae carried on hive tools and gloves, though precautions were taken, 3) spores and/or hyphae carried by drifting bees and by bees that were robbing due to a dearth of nectar, or 4) CB spores present in the pollen used for the pollen cakes, though the colony it was collected from appeared disease free.

To establish more definitely the lack of obvious relationship between the feeding of oxytetracycline (TM) and the occurence of CB, the history of both chalkbrood and the use of oxytetracycline was reviewed. Oxytetracycline has been used extensively in North America since the early 1950's to clean up AFB and EFB infections (KATZNELSON *et al.* 1952, KATZNELSON and JAMIESON, 1953; STUDIER, 1953, and GOCHNAUER, 1953) with no confirmed cases of CB for about 15 years. However, once established and positively identified in 1968 (THOMAS and LUCE, 1972), CB spread rapidly across the U.S. (HITCHCOCK, 1972, MENAPACE and WILSON, 1976). Also chalkbrood has been a problem in Europe for 50 years (HITCHCOCK, 1972), long before antibiotics came into use.

The amount of CB in the hive seems to be related to the ratio of adults to brood (DEJONG, 1977), so a weakened colony would have fewer adults available for removal of mummies than a healthy colony. Colonies infected with AFB or EFB generally become weak and may die; and CB apparently is seen more often in such weakened colonies. They are not necessarily more susceptible to CB infection, but the bees cannot or do not keep the mummies cleaned out of the cells (DEANS, 1940). Consequently, CB may show up in some colonies, not because of feeding oxytetracycline, but because weak colonies cannot effectively control the CB infection.

Commercial beekeepers and inspectors have reported instances in which large bee outfits on heavy « drug » programs have CB infections. However, beekeepers that feed their colonies no drugs have also reported CB (WILSON, 1977). Therefore, there appears to be no correlation between feeding TM and the incidence of CB. It can be seen from these facts, general observations, and the data obtained that there is not an obvious relationship between CB infections and the feeding of oxytetracycline (TM), and that TM does not enhance CB infections.

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RÉSUMÉ

LE NOURRISSEMENT AUX OXYTETRACYCLINES (TERRAMYCINE) N'AGGRAVE PAS LES INFECTIONS DE COUVAIN CALCIFIÉ

Afin de tester l'influence de l'oxytetracycline sur les infections de couvain calcifié dans les colonies d'abeilles *(Apis mellifera* L.), nous avons nourri celles-ci avec des galettes contenant de l'oxytetracycline et avec des pâtés de pollen contaminé par le couvain calcifié. Au cours de l'expérience on a également utilisé des colonies qui n'avaient pas été nourries et des colonies qui n'avaient reçu que des galettes à l'oxytetracycline, ou que des pâtés de pollen contaminé par le couvain calcifié, ou que des pâtés de pollen non contaminé, ou bien à la fois des galettes à l'oxytetracycline et des pâtés de pollen non contaminé. On a examiné les colonies chaque semaine pendant onze semaines et on leur a fourni au total dix pains de pollen et neuf galettes à l'oxytetracycline.

Les colonies nourries à la fois avec les galettes à l'oxytetracycline et avec le pollen contaminé, ainsi que les colonies nourries seulement avec le pollen contaminé, avaient presque le même degré d'infection de couvain calcifié. Il est montré en conséquence que l'oxytetracycline n'intensifie ni ne freine les infections de couvain calcifié.

ZUSAMMENFASSUNG

KEINE VERSCHLIMMERUNG VON KALKBRUT-INFEKTIONEN DURCH VERFÜTTERUNG VON OXYTETRACYCLIN (= TERRAMYCIN(R))

Um den Einfluss von Oxytetracyclin (TM) auf das Auftreten von Kalkbrutinfektionen (CB) bei Völkern der Honigbiene *(Apis mellifera* L.) zu überprüfen, wurde TM in Form von Dauertabletten (eine Präparation, welche die Lebensdauer des Antibiotikums verlängert) und mit CB infizierte Pollenkuchen an die Völker verfüttert. Ferner wurden Völker in die Versuchsreine miteinbezogen, die kein Futter erhielten, und solche, die nur TM-Tabletten, nur CB-infizierte Pollenkuchen, nur nichtinfizierte Pollenkuchen, oder TM-Tabletten und nichtinfizierte Pollenkuchen bekamen. Die Völker wurden 11 Wochen lang einmal wöchentlich untersucht und erhielten insgesamt 10 Pollenkuchen und 9 TM-Tabletten.

Die Völker, die sowohl TM-Tabletten wie auch CB-infizierten Pollen bekamen und jeune, die nur CB-infizierten Pollen erhielten, zeigten fast denselben Grad der Kalkbrutinfektionen. Somit wurde festgestellt, dass Oxytetracyclin Kalkbrutinfektionen weder fördert noch verhindert.

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