THE PROBLEM OF ANTIBIOTICS IN MILK*

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The antibiotics used in the treatment of mastitis are very effective inhibitors of starter culture organisms. When very small amounts of these antibiotics get into the milk supply, the milk is unfit for use. There is no practical method by which the dairy plant operator can overcome the presence of these "wonder drugs." A comprehensive education program must be instituted to inform the dairy farmer of the dangers of allowing these antibiotics to get into the milk supply. He must be taught methods of avoiding this hazard. This program must be accompanied by legal action when necessary.

DURING the past years, much emphasis has been placed on increasing the production of the dairy cow. The breeding and selection of our herds have reflected this philosophy. As a result, we have today an animal that is capable of producing a much greater amount of milk than her ancestors. No doubt this places a strain on the milk producing organs, leaving them more susceptible to infection and disease.

The removal of this milk from our animals requires considerable labor. To solve this problem we have developed milking machines and have concentrated our producing animals into larger herds. These advancements reduced the amount of labor necessary to produce a given amount of milk. However, progress in greater milk production created new problems. One of these is the prevalence of mastitis in milking herds.

ANTIBIOTIC TREATMENT

Some progress is being made in combating mastitis. The main weapon used in this fight is antibiotics. These substances are sometimes referred to as the new "wonder drugs." Antibiotics have the ability to halt the growth or in some cases to kill some of the organisms that cause mastitis. The use of these antibiotics has enabled many dairy farmers to halt the ravages of mastitis in their herds.

The more common antibiotic substances used in the treatment of mastitis are penicillin, streptomycin, and aureomycin. Some of the other antibiotics that have been used to a lesser extent for this purpose are chloromycetin, subtilin, and bacitracin. Sometimes a sulfonamide drug, i.e., sulfamethazine, is used in conjunction with penicillin in the treatment.

There are various ways that these antibiotics may be administered to the animal. Some are given orally. Others are injected as saline or water solutions. A very common method of treatment is by the use of small tubes called bougies. These bougies contain the antibiotic suspended in an ointment or oil base. They are inserted into the teat canal and thus the antibiotic is infused into the udder. These bougies are readily available to the dairy farmer. A prescription is not required for their purchase. Seldom are they administered under the guidance of a qualified veterinarian.

The dosage of antibiotic used in the treatment of mastitis varies. When penicillin is used, the dosage may be anywhere from 25,000 to over 100,000 units. Roughly defined, the unit of penicillin represents approximately 0.6 microgram of the substance. A microgram is one millionth of a gram. Therefore the normal dosage represents about 0.02 to 0.06 gram of penicillin. These extremely small amounts of penicillin are most helpful in treating mastitis and reducing infection.

ANTIBIOTICS DECREASE BACTERIA COUNT IN MILK

The dangers caused by the increased use of these "wonder drugs" often confronts the dairy processor as a bolt from the blue. Many dairy technologists have been introduced to the problem by a hurried phone call from a worried dairy processor. The stories may vary. As a rule they may be summarized by any of these statements of exasperation. "My starter will not develop acidity," "I'm having trouble, I can't get any acid development in my cultured buttermilk." The problems all have this in common. They are associated with dairy processors that are using starter cultures in the preparation of their dairy products.

Troubles of this sort have been reported by processors of cottage cheese, cultured buttermilk, and other fermented dairy beverages. This unwanted experience is also being shared by cheesemakers of most all types of cheese. The magnitude and extent of this trouble is increasing daily.

Those doing laboratory control work on raw milk supplies have had their attention called to this problem by sudden drops in plate counts or by undue lengthening of the inactivating organisms that cause trouble. The presence of 100,000 units of penicillin per ml of milk is no longer a concern. The problem still lies in the fact that the cultured dairy products can be traced to the presence of these so-called "wonder drugs."

STARTER DIFFICULTIES

Many of the bacteria used in starter cultures possess some characteristics similar to those organisms that cause mastitis. Therefore, the antibiotics that have been so effective in combating the organisms that cause udder infections have also been quite effective in inactivating the desirable types of bacteria that are responsible for the action of our cultures. Recently much time and effort have been spent by several

groups of investigators to determine the critical amount of an antibiotic that must be present in the milk to inhibit the action of starter organisms. The results of these investigations indicate that in numerous instances of some of the bacteria commonly used in starter cultures are more sensitive to such antibiotics, as penicillin, than are the organisms that cause mastitis. It has been revealed that extremely small amounts of penicillin in the milk or starter cultures will inhibit the growth and activity of the acid-producing organisms.

Krienke has reported that as little as 0.1 unit of penicillin per ml of milk can retard acid production to a great extent. He found that a sample of milk containing this small amount of penicillin would develop only 0.45 percent acidity when inoculated with a good starter culture and then incubated at the proper temperature for 16 hours. A control sample of good milk under the same conditions would develop 0.75 percent acidity. When the penicillin content of the milk was increased to just 0.25 unit per ml, the acid production under the same conditions was reduced to only 0.27 percent. Similar results to those of Krienke have been obtained by Katznelson and Hood, Hunter, and others. It is true that different bacteria vary in their sensitivity to the antibiotics. In fact there is probably a difference in sensitivity of the various strains of the same organisms. Yet, all of the more common types of starter organisms are inhibited by very small amounts of these antibiotics.

To illustrate this difference in sensitivity, Hunter has reported that to obtain the same degree of inhibition produced by 0.1 unit of penicillin per ml of milk on Streptococcus cremoris requires 0.25-0.30 unit on Streptococcus lactis. Hargrove and co-workers have shown that marked inhibition on the growth of Streptococcus thermophilus is obtained by as little as 0.01 unit of penicillin per ml of milk. To obtain the same results with Lactobacillus bulgaricus and Propionibacterium shermanii required 0.1 unit. Of the antibiotics previously mentioned, penicillin seems to be the most effective in inhibiting starter bacteria. Katznelson and Hood indicated that chloramphenicol was the least potent. The critical amounts of sulfamethazine and aureomycin that inhibit acid production by lactic starters has been discussed in a recent publication by Krienke. Though there is a difference in the potency of these antibiotics, remember, the amounts are all relative and even with the least potent, an extremely small amount in the milk can ruin the best starter.

**Antibiotics Affect Quality Control**

As further evidence that antibiotics will inhibit the growth of bacteria in milk, several studies have been made concerning the feasibility of their use as milk preservatives. The advisability of this practice is quite doubtful. In a recent publication, Foley and Byrne suggested the use of penicillin for this purpose. Curran and Evans concluded that penicillin has no application in the preservation of food even though it might be used in certain non-food materials. In discussing the possibility of the preservation of foods with antibiotics, Anderson and Michener stated that the possible physiological effects of continued use of foods containing antibiotics has not been determined. They believed that much more information is needed on the subject.

If antibiotics are present in a raw milk supply, they may influence the results of some of the milk quality tests. Johns and co-workers have reported a concentration of penicillin as low as 1 part in 167,000,000 parts of milk may retard the dye reduction. It is their opinion that this may not be as serious in the resazurin test as in the methylene blue test. According to Ruehe, the presence of penicillin in milk prolonged the methylene blue time to some extent but in most cases not enough to change the general quality classification of the milk.

**Duration of Bactericidal Effect**

When an antibiotic has been infused into an udder it would seem natural that the milk drawn from this udder would contain a certain amount of antibiotic. With this thought in mind, various research workers have sought to determine the amounts of antibiotics that will be present in a milk after treatment. Also they have tried to determine the time it will require after treatment until the milk will be free of the antibiotic or when its content in the milk will be of no significance. It is difficult to summarize the findings of the research workers on this subject. None has arrived at an exact conclusion as to how long after treatment the milk will contain critical amounts of the antibiotic. Some antibiotics longer than others. This might be caused by the differences in the udder structure of cows. It is quite evident that when the antibiotic is administered in an ointment or oil base, it is retained for a longer period of time. Krienke has calculated that it is possible for the milk from one treated quarter to render unfit the mixed milk of 20 cows. This would be the case if one quarter is treated with 75,000 units of penicillin and the milk drawn at the first milking after treatment is mixed with the herd milk. Even with this dilution the milk would not be suitable for use in cultured dairy products. Thus a cow treated in all four quarters would render unfit the milk from 80 normal cows. Further calculation that after two milkings (36 hours after treatment) the milk from one quarter would ruin the milk from 5 cows.

Ruehe has reported an experiment in which 10 ml of an aqueous solution of 200,000 units of penicillin was injected into each quarter of the udders of two cows. His results indicated that under these conditions the milk produced three hours after treatment contained considerable amounts of penicillin. However, there was not enough penicillin retained in the milk after the third milking (27 hours after treatment) to inhibit his starter.

Hansen, Wiggins, and Boyd have shown that there was no diffusion of antibiotics from the treated quarters to the untreated quarters. Therefore, they recommend that the milk from the treated quarters be discarded for three days or six milkings after treatment.

Fuller has studied the treatment of mastitis by the combined use of sulfamethazine and penicillin. He reports that the milk was not available for use until 3-5 days after treatment. From the above reports it is obvious that the milk from a treated cow should not be used for at least several days after treatment. Yet, many conscientious farmers discard
only the first milking after treatment. Needless to say what happens in the case of those who are not so conscientious.

When a cow is treated with an antibiotic and her milk included in the milk supply too soon after treatment, then the milk will be contaminated with the antibiotic. Doan® has listed the following items as being responsible for the amount of penicillin in herd milk: number of quarters treated, number of cows in herd, vehicle carrying the antibiotic, time elapsing after treatment, and level of production.

**Effect on Other Dairy Products**

The effects of antibiotics in dairy products other than fresh raw milk have been studied by Krienke and Fouts. They added penicillin at the rate of 1 unit per ml to whole milk. This milk was then made into evaporated and condensed milk. Portions of these products were inoculated with 3 percent starter culture and incubated at 95°F for 7–8 hours. The acid production was compared to that of a control or penicillin-free sample. They found that when the evaporated milk containing the penicillin was held for 9 days at room temperature, then subjected to this treatment, its acid production was about normal. Acid production could not be obtained in the condensed milk even after it was held for 13 days before being subjected to the above treatment. In the same series of experiments, 1 unit of penicillin per ml was added to skim milk. The skim was then dried. Acid development could not be obtained in this reconstituted product even after 10 weeks of storage in the dry form. This was still true when the non-fat, dry milk solids were diluted 50 percent with a normal product. All of these experiments point to the conclusion that the presence of antibiotics in milk is a definite hazard to several phases of the dairy industry.

** Attempted Neutralization of Antibiotic Effects**

Several attempts have been made to overcome the presence of antibiotics in milk. The great difficulty to this problem is to detect their presence. As a rule, we do not know that a milk supply contains these "wonder drugs" until it is too late. The cottage cheese already has been set or the culture added in the preparation of buttermilk. Then we wait for the acid development that never comes. Tests have been proposed to detect the presence of these antibiotics. Many of the tests that have been advanced are too time-consuming. Their results are not available soon enough to be of practical help to the dairy processor. Recently a test based upon the principle that the activity of the phosphatase enzyme in raw milk is retarded in the presence of antibiotics has been proposed by Stolz and Hanlind. If further work confirms the preliminary reports, then this test will be quite helpful. Its main advantage is that its results can be obtained in less than an hour.

Heat treatments of milk containing antibiotics have little effect in reducing their inhibitory action on starter cultures. Heat treatments that are common in the dairy industry such as pasteurization, autoclaving, and steaming have little effect on the antibiotics. The addition of various oxidizing or reducing compounds or the use of surface active agents in milk containing the drugs are of little help.

The use of the enzyme penicillinase has been suggested. This would be a means of destroying the penicillin that might be present in a milk supply. However, it would be difficult to calculate the amount to use unless the penicillin content of the milk was known. In many cases, the cost of the penicillinase needed to treat a milk would be greater than the value of the milk.

The use of larger amounts of starter may overcome the inhibitory effects of penicillin. Krienke suggested this remedy but points out that this will work only when the amount of penicillin in the milk is relatively small. Furthermore, he questioned the quality of the finished dairy product. The possibility of developing starter organisms that are resistant to the action of antibiotics has not been overlooked. Katznelson and Hood developed a penicillin-resistant starter culture which coagulated milk in the presence of 3 units of penicillin per ml of milk. This starter retained its resistance after 20 passages in absence of penicillin. While this approach to the problem of antibiotics in milk may have possibilities, actually it has undergone little practical progress.

** Educational Program**

One solution to the problem would be to establish a long-range education program. This has been done to solve other quality problems. Educational programs on such subjects as sediment control are now bearing fruit. Why would it not work on this problem?

Any educational program on the control of antibiotics in milk must involve the help of all veterinarians, sanitarians, dairy processors, leading dairy farmers, dairy schools, and appropriate governmental agencies. Each of these groups could assist by giving proper instructions and demonstrations of methods for preventing mastitis. The elimination of mastitis would do wonders to improve our milk supply. If we can reduce or do away with mastitis we will also have eliminated the need for antibiotics. This approach to the problem may be idealistic, but it is logical and wholly in accord with existing legal definitions of milk.

Another and more immediate approach would be to tell the story of the effects of antibiotics in milk where it would do the most good. Most of our dairy farmers would cooperate if they had the facts. Teach them the necessity of discarding milk that may contain antibiotics. Make the dairy farmer aware of the fact that the loss of a small amount of milk from a few cows may save the milk from many cows.

** Legal Action**

Some consideration should be given to the desirability of invoking existing legal regulations to meet this problem. The definition of milk points out that the milk must be obtained from a healthy cow. A cow with mastitis is not healthy. Therefore, her lacteal secretion does not fulfill the definition of milk. It cannot legally be placed in the commercial milk supply. Legal action should be pressed to keep mastitic milk out of the public milk supply.
Also, we must keep in mind that when milk contains antibiotics such as penicillin, streptomycin, aureomycin, etc., it is adulterated. The legal machinery for action against producers of adulterated milk has been functioning for some time. Steps should be taken to control the indiscriminate use of antibiotics. They should be administered only upon the advice and under the direction of a qualified veterinarian. Antibiotics should be made unavailable to the layman except by prescription.

EDUCATIONAL PROGRAM VS.
LEGAL ACTION

It would seem most desirable that, if possible, the problem should be solved by an education and information program rather than by one involving legal action. A good educational program backed by legal action, when necessary, would probably solve the problem. Whatever approach is taken must be started soon. We who are interested in the progress of the dairy industry must do all within our power to help keep antibiotics out of the milk supplies.

REFERENCES


Comparative Study of Stains (Continued from page 51)

CONCLUSIONS

This study was extremely valuable in emphasizing the availability of improved milk stains, the variation which can occur in their use, and the need for extensive ground work prior to conducting a comparative study in order to assure uniformity in methods of procedure.

The results reported in this preliminary trial by nine laboratories show that improved stains have been developed, that they can be expected to yield higher clump counts than the present Standard Methods stain, and may possess other desirable characteristics resulting in less fatigue to the laboratory workers and in more accurate results.

The results show that the Acid-and-Water-Free Stain (Stain III) yielded the highest average count, ranked highest when scored according to the frequency of yielding the highest count; second highest, etc.; and showed least variation in score among eight laboratories. Until such time as further comparative studies conclusively demonstrate the relative merits of stains proposed for the direct microscopic examination of milk, the results presented suggest that the Acid-and-Water-Free stain should replace the Alcohol-Containing Methylene Blue Stain (Stain I) now recommended in Standard Methods, which showed the lowest average count.

REFERENCES