

A SURVEY OF PENICILLIN CONTAMINATION OF MARKET MILK SUPPLIES

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Although attention was previously directed to the subject of antibiotic contamination of the milk supply as a veterinary and public health problem^{1,2} information is still lacking in regard to South African milk supplies. An antibiotic survey of milk supplies has therefore been carried out as an initial step towards deciding whether the problem is worthy of further attention. The survey has been undertaken in respect of herd milk supplies received in Johannesburg from the Transvaal highveld area, with particular reference to penicillin contamination of the milk. The area of milk supply extends in a vast semi-circle stretching from Ermelo - Volksrust in the east, of Potchefstroom - Klerksdorp in the west, and as far south as Frankfort - Koppies in the Orange Free State.

The reason for concentrating on penicillin to the exclusion of other antibiotic preparations is threefold:

1. Penicillin is still extensively used for the intramammary treatment of bovine mastitis, in spite of the fact that other antibiotic preparations such as dihydrostreptomycin, aureomycin, terramycin, etc., are tending to supplant penicillin.

2. Many medical authorities in the field of antibiotic therapy, allergy and paediatrics believe that, of the various forms of antibiotic therapy employed for bovine mastitis, it is penicillin that is most likely to make the milk harmful to consumers.³

3. The presence of penicillin in milk is established relatively easily by an identification test using the enzyme penicillinase, which specifically inactivates penicillin.

Material and Method

Herd milk samples collected at random from Johannesburg dairies during the period November 1958 to June 1959 were subjected to a disc assay procedure based on the $\frac{1}{2}$ " double disc method of Cerny and Morris.⁴ The test organism employed was a strain of *Sarcina lutea* obtained from the Department of Biological Standards, National Institute for Medical Research, London. The organism is sensitive to a low concentration of penicillin, of at least 0.01 units of penicillin per ml. of milk, by the procedure employed for this $\frac{1}{2}$ " double disc assay method.

The possibility that natural inhibitors in milk from untreated cows might produce false reactions (by bacteriophage and naturally occurring heat-labile inhibitory substances) was eliminated by preliminary heating of the milk to a temperature of 180°F for 5 minutes. When the milk is heated to this extent antibiotics which are present may be partially inactivated, and recent workers are content to employ a temperature of 180°F for only 1 minute.⁵ The results obtained after heating the milk in this way do emphasize the fact, not generally appreciated, that the process of pasteurization normally carried out at 161°F for 10 seconds does not destroy antibiotic substances in the milk.

Whenever a zone of inhibition was noted the test was repeated for confirmation of the result, and an additional duplicate milk specimen treated with penicillinase was simultaneously tested in identical manner. The absence of inhibition in the penicillinase-treated sample was accepted as positive indication that the inhibitory effect of the normal sample was due to penicillin contamination.

Although the magnitude of the zone of inhibition was always recorded these comparative measurements are not employed here as a criterion of the concentration of penicillin in the milk, largely on account of the many variations in the *in vitro* tests that are difficult to control on a day-to-day basis.⁶

Results of Survey

The results are summarized in Table I. Of the 1,229 milk samples that were tested for penicillin contamination, 66 showed inhibitory characteristics (5.37% of samples tested). The cause of the inhibition was undecided in respect of 2.28% of the samples, and in the remaining 3.09% the inhibition was due to penicillin.

TABLE I. PENICILLIN CONTAMINATION OF HERD MILK SUPPLIES

No. of herd milk samples	1,229
No. of inhibitory milk samples	66 (5.37%)
No. of samples containing penicillin	38 (3.09%)

It was not possible to complete the penicillin identification procedure in respect of certain inhibitory milk samples. Especially during the December - February midsummer period a number of samples soured to such a degree as to invalidate the test by the time the penicillinase test was applied the following day. In yet other instances the inhibitory effect could not be reproduced, presumably owing to a dissipation of the inhibitory substances as a result of natural changes in the milk, perhaps assisted also by the preliminary heating process. There is however no doubt that a minimal proportion of at least 3.09% of milk samples considered typical of Transvaal market milk supplies were contaminated with traces of penicillin.

Comment

This limited survey shows that, as in Europe, the USA and Great Britain, penicillin traces are commonly present in a small proportion of South African herd milk supplies. It could hardly be otherwise in a country where the farmers can freely obtain antibiotic remedies for treating udder infections from chemists, agricultural cooperatives, veterinary firms and other agencies.

After intramammary therapy the antibiotic substance is not completely absorbed in the cow's udder and a quantity becomes excreted in the milk, commencing from the first milking after treatment. The amount of antibiotic is usually greatest at the first milking and then decreases progressively,

TABLE II. EXCRETION OF PENICILLIN IN MILK UP TO 3 DAYS AFTER INTRAMAMMARY THERAPY

Penicillin Dosage (units per quarter)	Number of Samples	Units of Penicillin per ml. of Milk, after:		
		24 hours	48 hours	72 hours
25,000	4	5.7	0.2	—
50,000	22	9.0	0.9	0.2
100,000	6	8.1	1.3	0.5

but effective quantities may remain for 3-5 days according to the preparation used and the dosage administered. The excretion of penicillin in milk up to 3 days after treatment with various rates of dosage is exemplified by the results obtained by Danish workers, as shown in Table II (Voordeckers⁷).

Dosage of 300,000 units is commonplace and a dosage of less than 100,000 units per quarter is seldom employed. These results afford some indication of the quantity of penicillin which may be present in the milk obtained from an infected cow shortly after treatment. For instance, after 24 hours a two-gallon cow treated in each of the 4 quarters with 100,000 units excretes about 74,000 units of penicillin, or approximately 2,000 units of penicillin in a glassful of milk.

The degree of contamination of the bulked herd milk supply ultimately depends on a number of factors such as the dosage administered, the number of quarters infused, the amount of milk produced by the infected cows, the number of infected animals in a herd, and the volume of milk from treated cows in relation to the total herd milk supply. In general terms, however, the interaction of all these variable factors, as well as the possibility of allergic or sensitizing ill-effects to persons consuming the milk, can be easily eliminated if the milk from each treated quarter is discarded or used for purposes other than human consumption for a period of 72 hours after treatment.

Control

The fundamental requirement is that the milk from an infected cow shall be withheld from the herd milk supply for 3 days after antibiotic treatment. Since the milk often *appears* to be normal shortly after treatment the farmer may believe that the milk is suitable for use, but apart from its antibiotic content such milk is also adulterated by pus and is generally unsuitable for consumption.⁸

Obviously there is need for considerable educational and propaganda work to overcome ignorance of this type. Another possibility, which may be of eventual assistance in ensuring the discarding of milk, is the incorporation of a

suitable dye-stuff in the antibiotic preparation, in order to discolour the milk for a considerable period after treatment. Workers in the USA and Scandinavia have already achieved a limited degree of success in this direction.

Unfortunately there is at the moment no rapid platform test which can be used on the incoming milk to detect antibiotic contamination of milk supplies and to prevent their being used for consumption. Thus the control procedure would seem to depend essentially on the routine antibiotic testing of herd milk supplies, and subsequently either (a) possibly prosecution for supplying contaminated milk, or (b) suspension of the milk supply until the milk has been demonstrated to be free from contamination. As procedure (b) is likely to require several days to complete, the penalty is sufficiently severe to deter the dairy farmer from taking the risk of including doubtful milk in his herd milk supply.

SUMMARY

Samples of market milk supplies collected from dairies in Johannesburg were examined for penicillin contamination.

Penicillin in concentrations exceeding 0.01 units per ml. of milk was found in 3.09% of the 1,229 samples tested.

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