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SOME FACTORS AFFECTING THE SECRETION OF ABO MASAL JUICE IN YOUNG DAIRY CALVES

J. F. W. GROSSKOPF, Onderstepoort Laboratory

INTRODUCTION

While studying the secretion of gastric juice in calves some interesting observations were made on changes in the amount and composition of the abomasal secretion under different conditions.

Although it seems more natural that a calf should be fed from a nipple, various workers (Geddes, 1950; Alexander, 1954; Kesler, McCarthy & Knott, 1956), proved that there was no weight increase advantage in calves fed from nipples over those drinking from open buckets. Nipple feeding did, however, reduce the incidence of scours. The main object of the present study was to see how these two methods of feeding affected the secretion of abomasal juice.

Espe & Cannon (1937) found that teasing or sham-feeding had very little or no effect on the rate of abomasal secretion in calves, but that the actual drinking of milk had a stimulating effect on the secretory rate. Although not proved in these experiments, there were definite indications of psychic reflexes stimulating or inhibiting the rate of secretion.

TECHNIQUE

Five young grade Friesland bull calves were used. Heidenhain type Pavlov pouches, type E, as described by Hill & Gregory (1951), were produced in two calves (No. 1 and 2). Abomasal pouches, as described by Grosskopf (1954), were made in the other three (No. 3, 4 and 5). A tubular plastic fistula with a piece of rubber tubing leading to the outside was inserted into each pouch. The operations were carried out before the age of two weeks. The calves remained in good health but after three to five weeks the tissues around the fistula tended to become digested.

Abomasal juice was collected by connecting a light plastic test tube to the rubber tubing on the fistula. The juice dripped into this plastic tube while the calf was in the standing position (see Plate 1). Every ten minutes the plastic tube was replaced, so that the contents of each tube represented the secretion over that period. On most occasions collections took place over a three to five hour period. On one occasion the secretion over a 24 hour period was collected. Collection of juice was commenced as soon as it appeared to be free from blood i.e. about two to three days after the operation.

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The volume and pH of each sample were measured as soon as possible after collection, but the pepsin and rennin determinations were done after storage in a refrigerator, sometimes for as long as 24 hours. No change in pepsin or rennin value, however, could be demonstrated even after ten days such storage.

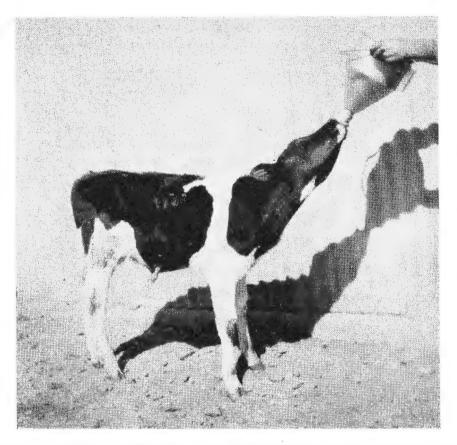


PLATE I.--Calf No. 3 drinking from a nipple while abomasal secretion collects in tube.

The pH was determined by a Beckman Model G pH meter. Total acid estimation was not done, as the amount collected during ten minutes was not sufficient for all determinations.

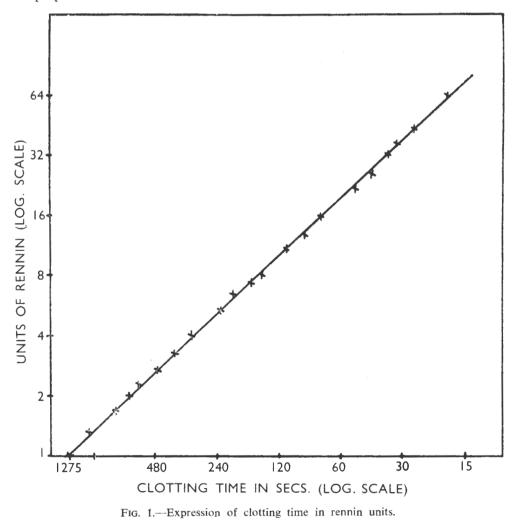
Rennin determination

This was done by adding one drop of the abomasal secretion to 5 ml. of fresh cow's milk which had been kept at 37° C for 15 minutes and shaking it well. The clotting time was then determined to the nearest second. To produce a standard amount, the drops were always added to the milk through a standard No. 20 Luer-Lok hypodermic needle held at approximately the same angle. One drop added in this way represented 0.033 ml. The clotting time of each sample was determined twice and the average taken.

At first it was thought that the varying acid values of the samples of abomasal juice might affect the clotting time. This was disproved by the following tests:—

- (a) Doubling the acid value of a sample of abomasal juice did not affect the clotting time of milk to which it was added.
- (b) A sample of abomasal juice was divided into two equal portions. One of these was then heated to 70° C for five minutes to inactivate the enzymes. The clotting time obtained from one drop of the inactivated juice plus one drop of the untreated juice was identical to that from one drop of the latter alone.

It was assumed, therefore, that the buffering action of the milk was sufficient to counteract the small variation in the pH of abomasal juice when added in the proportions used.



Berridge (1952 *a*, *b*) used skim milk powder in a 0.01 M calcium chloride solution to determine the rennin activity but in this study a bulk sample of fresh cow's milk proved to be quite satisfactory.

For the purpose of these trials the rennin activity was expressed in arbitrary units. A sample of abomasal secretion with a high rennin content was collected from calf No. 5 after the subcutaneous injection of carbamylcholine. This sample was diluted in two-fold dilutions with hydrochloric acid having the same pH as the abomasal secretion. The clotting time of milk was then determined with each dilution. When clotting time was plotted against dilution (using log. scale) a straight line was obtained. The highest dilution used (1 in 64) was found to give a clotting time of 21 minutes 15 seconds and this was taken as one unit (see Fig. 1).

Pepsin determination

This was done according to the method described by Anson (1938). A 2 per cent solution of haemoglobin in 0.06 N hydrochloric acid (pH 1.6) was digested by a known quantity of abomasal juice at a fixed temperature for a fixed time. The amount of tyrosin so formed was then determined colorimetrically with a Leitz photo-electric colorimeter. The proteolytic activity was found to be in direct proportion to the amount of tyrosin formed. By slightly modifying the technique it was found that the digestion could be done at 37° C for five minutes instead of at 25° C for 10 minutes. To obtain suitable colorimeter readings the gastric juice was diluted to twice its volume with distilled water before it was added to the haemoglobin substrate.

For the purpose of these experiments the proteolytic activity was also expressed in arbitrary units. One unit of proteolytic activity was taken as that which would produce 0.00015 milli-equivalents of tyrosin per 5 ml. from 100 mgm. of haemoglobin in an acid medium during five minutes at 37° C.

EXPERIMENTAL PROCEDURE

All five calves used for these experiments were subjected to the following procedures: ----

Three to four 10-minute samples of abomasal secretion were collected in the morning before feeding. The calves were then fed 800 ml. of fresh cow's milk at body temperature from either a nipple feeder or open bucket. All calves were accustomed to both types of feeding throughout. The collection of the 10minute secretion samples continued during and after feeding and even until after a second, third or fourth four-hourly feeding. The time it took to drink the 800 ml. of milk was recorded in each case. All samples were numbered and examined as soon as possible for volume, pH, proteolytic and milk-clotting activity.

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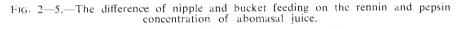
A total number of 654 10-minute samples were collected before, during and after 50 feeding times. Most of the samples were collected from calves No. 2 and 3, while the least were taken from calf No. 1.

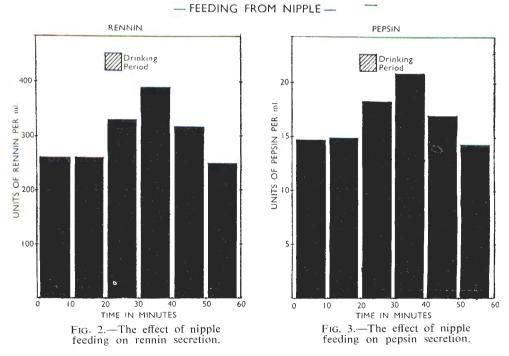
On other occasions the effect of carbamylcholine and atropine on the amount and composition of the gastric secretion was determined. Due to the quick action of these drugs it was decided to collect 5-minute samples instead of the usual 10-minute samples.

After the collection of five such samples, calf No. 5 (48 Kg.) was given, on two occasions, 1 mgm. of carbamylcholine subcutaneously, care being taken to disturb the animal as little as possible. Five subsequent specimens were then collected on both occasions. The same procedure was followed with the subcutaneous injection of 24 mgm. of atropine.

RESULTS

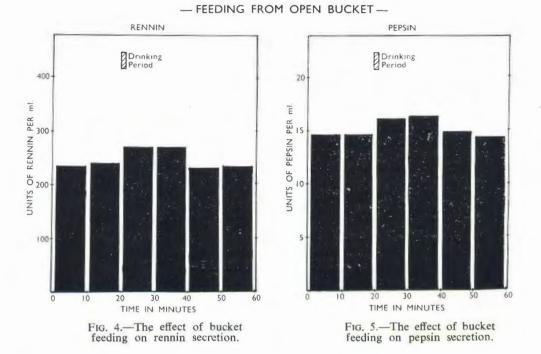
As can be seen from the accompanying graphs (Fig. 2 to 5) the drinking of milk had a stimulating effect on the concentration of rennin and pepsin in the abomasal secretion. The maximal effect was reached during the second 10minute period after starting to drink. It will be noticed further that the average time taken to drink from the nipple was five minutes, while it took the calf only 75 seconds to finish the same amount of milk from the bucket.





Drinking from the nipple caused an average increase of 50.3 per cent in the concentration of rennin and an increase of 42.2 per cent in the proteolytic activity of the abomasal secretion. Bucket feeding, on the other hand, only caused an increase of 14.7 per cent and 10.7 per cent in the rennin and pepsin concentrations respectively.

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On a few occasions there was an increase in the volume of secretion after both forms of feeding. After calculation of the averages, however, it was found that there was no variation in the rate of secretion either with bucket or nipple feeding. The average volume secreted during a 10-minute period was 4 to 5 ml.

Carbamylcholine had a marked stimulating effect on the rate of abomasal secretion as well as on the pepsin and rennin concentration. (Fig. 6, 8 and 10.) The effect on the secretion rate was only maintained during the three 5-minute periods after administration of the drug, whereas the stimulating action on the enzyme concentration was still in evidence after 30 minutes.

The injection of atropine caused a marked decrease in the rate of secretion and also in both the rennin and pepsin concentrations (Fig. 7, 9 and 11). Moreover, it blocked the effect of a subsequent nipple feeding given 10 minutes after the administration of the drug.

Although it seemed that there was a slight decrease in pH of the abomasal juice after most feeding times, the results were inconclusive. Larger quantities would be required for the determination of the actual acid value. The pH of the samples usually varied between 1.3 and 1.7.

Fig. 12 represents the changes in average daily pH and rennin concentration of the abomasal juice of calf No. 2 over a period of seven weeks. It will be seen that both the pH and rennin concentration showed a tendency to fall as the calf grew older.

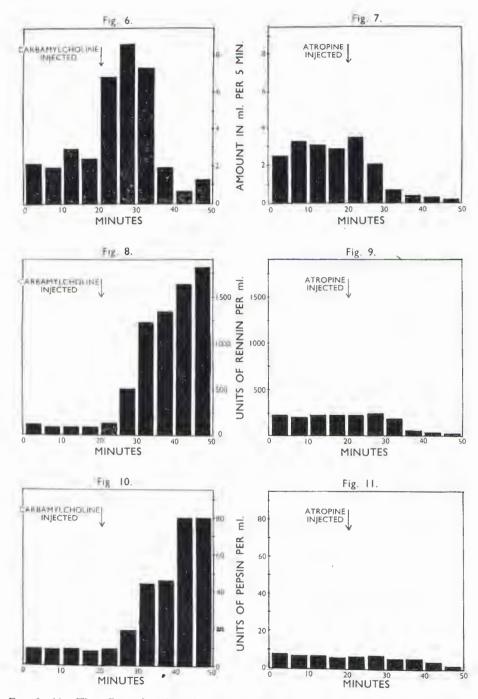


FIG. 6-11 -The effect of carbamylcholine and atropine on the volume and rennin and pepsin concentration of abomasal secretion.

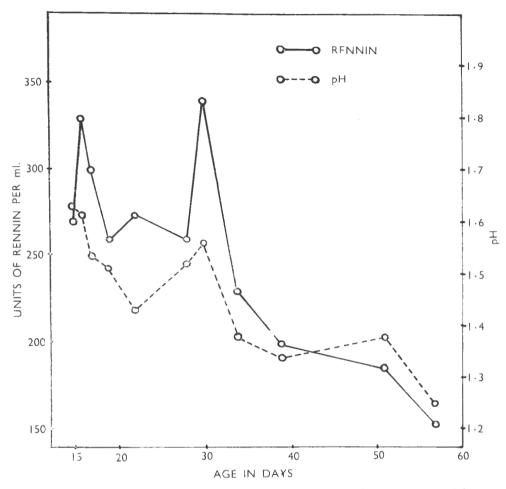


FIG. 12.--Changes in average daily pH and rennin concentration of abomasal juice (Calf No. 2).

DISCUSSION AND CONCLUSIONS

From these experiments it is quite clear that the drinking of milk caused an increase in the concentration of both rennin and proteolytic activity of the abomasal juice in calves. The increase was more marked in nipple-fed calves than in those that drank from the open bucket. This difference is probably due to the longer time required when sucking from the nipple. On one occasion the valve was removed from the nipple so that the calf finished the milk in less than half the usual time. The result then appeared to be similar to that obtained after bucket feeding. It was, therefore, concluded that the time taken to drink the milk played an important rôle in the composition and rate of abomasal secretion.

As carbamylcholine had a stimulating and atropine a depressing effect on the amount and concentration of the abomasal secretion it could be concluded that secretion is influenced by parasympathetic stimulation through the vagus nerve. Hill (1952), however, showed that there was no difference in the secretion of goats with severed or intact vagus nerves.

On comparing the graphs representing the pepsin and rennin secretions one is immediately impressed by the striking similarity in the secretion of these two components. There is still some doubt as to whether they are in fact two different enzymes or whether a single enzyme performs two separate functions.

SUMMARY

Methods are described for the collection of abomasal secretion samples from calves and for the examination of the juice for proteolytic and rennin activity. The effect of the rennin and pepsin concentration of the secretion was studied during two different methods of feeding milk viz. nipple feeding and drinking from the open bucket. It was found that both ways of feeding caused an increase in the rennin and pepsin concentration of the juice but that the effect of nipple feeding was greater.

The administration of carbamylcholine stimulated the abomasal secretion rate and also caused an increase in the rennin and pepsin content of the juice. The injection of atropine, on the other hand, inhibited these functions.

It was also found that during a period of seven weeks the rennin concentration as well as the pH of the abomasal secretion of one calf fell steadily with increasing age.

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