

Absorption of ^{45}Ca and ^{89}Sr from the Abomasum of the Calf* (33767)

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(Introduced by F. R. Mraz)

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Previous studies of gastrointestinal absorption (1, 2) using unabsorbed marker ratio techniques, indicated that calcium is absorbed through the walls of the abomasum in the bovine and that this part of the ruminant stomach may be an important absorption site for calcium. The validity of the unabsorbed marker ratio technique depends upon the assumption that both the unabsorbed marker and the nutrient being measured move through the digestive tract in a uniform manner. Therefore, in the earlier investigations (1, 2), the evidence for absorption of calcium through the walls of the abomasum is indirect since the movement of $^{144}\text{Ce}^{144}\text{Pr}$ (or chromic oxide) and ingesta calcium were based on this assumption.

The additional studies reported in this paper were conducted primarily to obtain more evidence for abomasal absorption of calcium. However, since radiostrontium is an important fission product and also an alkaline earth metal, we have included ^{89}Sr in two of the three experiments reported.

Methods and Materials. Three experiments involving 4 holstein calves each were conducted. In Expt. 1, the calves were milk-fed. In Expts. 2 and 3 the calves were maintained on a hay and grain diet. The essential differences between the latter two experiments were in the volume of radioisotope dose solution given, this being 2 and 225 ml, respectively, and in Expt. 2, only ^{45}Ca was given. The dose volume in Expt. 1 was 2.5 ml.

The animals varied in weight from 54 to

165 kg. Each calf was anesthetized and prepared by surgically exposing the abomasum through a ventral incision. Equi-Thesin⁴ was used as the anesthetic agent in Expt. 1 and electroanesthesia (3) was used in Expts. 2 and 3. The contents of the abomasum of each calf were isolated from the remainder of the gut by ligatures at the omasal end and at the pyloric sphincter prior to dosing.

In Expt. 1, 0.25 mCi each of ^{45}Ca and ^{89}Sr were injected into the abomasum. The amounts of isotopes remaining in the abomasal contents and tissue were measured at the end of a 2-hr experimental period.

The calves studied in Expt. 2 had catheters placed in the facial branch of the left external maxillary artery and the left gastric epiploic vein of the abomasum. The latter catheter was inserted in the area of the omasum and directed retrograde toward the abomasum. A catheter was also placed into the abomasal cavity for a distance of 10 cm in the cardiac area. A dose of 2 mCi of ^{45}Ca was introduced into the abomasum through this catheter. Arterial and venous blood samples were taken at 0.25, 0.5, 1, 2, 4, 8, 16, and 24 min. following dosing. After 25 min, the animals were sacrificed and the digestive tracts were removed. Contents and tissue of the abomasum were taken for ^{45}Ca determinations.

Experiment 3 was similar to Expt. 2. The dose per animal was 0.28 mCi of ^{45}Ca and 0.34 mCi of ^{89}Sr given in balanced solution⁵ at pH 2.8 and in a solution volume of 225 ml.

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⁵ This solution contained the following ingredients (mg/liter): P, 480; Mg, 239; Ca, 720; Na, 1310; K, 630; the pH was adjusted using HCl; Compounds used were KH_2PO_4 , $\text{Mg SO}_4 \cdot 7\text{H}_2\text{O}$, CaCl_2 , NaCl , NaHCO_3 and KCl . The composition was designed to be comparable to abosomal contents (7).

Analytical procedures were similar for all experiments. Blood plasma samples were placed into crucibles, dried and ashed at 650° . The ash was dissolved in acid solution and the ^{45}Ca and ^{89}Sr were precipitated as oxalates (4). The ^{45}Ca and ^{89}Sr were counted with an end-window Geiger-Mueller tube using an aluminum absorber to distinguish between the beta energies when both isotopes were present. Ingesta and abomasal tissue samples were treated in a manner similar to blood plasma. Standards were counted along with the samples and all results are expressed as a percentage of the administered dose.

Results and Discussion. In Expt. 1, two calves exhibited absorptions of ^{45}Ca and ^{89}Sr and two other calves exhibited no detectable absorption. An average of $21.4 \pm 16.0\%$ of ^{45}Ca and $24.6 \pm 15.8\%$ of ^{89}Sr were absorbed from the abomasum of 2 calves over a 2-hr period. No discrimination was shown against ^{89}Sr and the observed ratio for the average absorption was 1.15 for these animals, indicating that moderately more ^{89}Sr than ^{45}Ca was lost from the abomasum. These animals were milk-fed and it is possible that the small volume of administered dose was trapped in the milk curd in the two animals that exhibited no absorption. Since both isotopes were administered in a single dose solution, their relative absorptions within a given animal should not be subject to the absolute magnitude of absorption. However, in view of the difficulties encountered with the milk-fed calves, a hay and grain feeding system was used in the additional two experiments.

In Expt. 2 an average of $31.4 \pm 16.1\%$ of the administered ^{45}Ca dose (radiostrontium was not given in this experiment) disappeared from the abomasum during the 25-min absorption period. Calcium-45 was detected in venous blood of three animals taken 30 sec after dosing, while the earliest detection in arterial blood was in samples taken at 2 min for one of these animals and at 8 min for the other two. The fourth animal had detectable ^{45}Ca in venous blood taken at 4 min and was not detected in arterial blood. After equilibrium was established (at

8 min for 3 animals) the venous plasma/arterial plasma ^{45}Ca concentration ratio was about 10. It is obvious from the large standard deviation for absorption in this experiment that considerable variation between calves occurred.

It was concluded from Expt. 2 that the large variability associated with ^{45}Ca absorption could be explained by an unequal distribution of ^{45}Ca in the abomasal contents. The ^{45}Ca was administered in 2.0 ml of saline and it is possible that the dose could have been released near the walls of the abomasum resulting in a very high absorption, or on the other extreme it could have been released within a mass of ingesta further from absorption sites, resulting in a very low absorption of ^{45}Ca during the 25-min period. It was anticipated that moderate manipulation of the abomasum after infusion of the dose would have caused adequate mixing but apparently it did not.

Experiment 3 was designed to overcome this difficulty. The larger volume of dosing solution used in this experiment (225 ml) resulted in considerably less variability in abomasal absorption (Table I). An average of $20.8 \pm 3.1\%$ of the ^{45}Ca and $3.2 \pm 3.0\%$ of the ^{89}Sr administered to these animals was absorbed over the 25-min period. The ^{45}Ca and ^{89}Sr were associated with abomasal tissue in similar amounts (2.6% of the administered dose). This suggests that these amounts were adsorbed to the abomasal surface rather than being taken into the abomasal tissue.

The attempt to isolate the venous drainage from the abomasum in Expt. 3 was not satisfactory. It was hoped that this would give a more direct measure of the extent of

TABLE I. ^{45}Ca and ^{89}Sr in Abomasum Ingesta and Tissue; Expt. 3 (four calves).

Item	% of administered dose* (mean $\pm$ SD)	
	^{45}Ca	^{89}Sr
Abomasum	76.6 ± 4.0	94.2 ± 3.1
Abomasum tissue	2.6 ± 1.1	2.6 ± 1.1
Absorbed	20.8 ± 3.1	3.2 ± 3.0

* 25 min following dosing.

abomasal absorption. Since ^{45}Ca and ^{89}Sr appeared in arterial blood in Expt. 3, it was concluded that either the venous drainage was not completely isolated or that another system of transport was operating. Recent observations in this laboratory have shown that the lymphatic system contributes significantly to the transport of absorbed cations (5). As a check against leakage of radioisotopes from the abomasum into the upper small intestine, samples of ingesta from the small intestine were analyzed. Negligible amounts of ^{45}Ca and ^{89}Sr were found in these samples.

Earlier studies, using unabsorbed marker ratio techniques, gave indications that a net calcium absorption of approximately 60% occurred from the abomasum of 12–20-week-old calves (1, 2). In Expts. 2 and 3 at least 20% of the administered ^{45}Ca was absorbed within 25 min. Storry (6) in 1961 reported that practically all of the calcium in the abomasal contents is ultrafilterable and thus in the best possible state for absorption. Therefore, the observed absorption should be a measure of the efficiency of the abomasal tissue transport mechanism.

Essentially no discrimination was observed between calcium and strontium absorption in Expt. 1 (however, only 2 of the 4 milk-fed calves showed absorption). Conversely, ^{45}Ca was absorbed in a sixfold greater quantity than ^{89}Sr in Expt. 3 when hay and grain-fed calves were studied. The change in diet is probably the explanation for the change in Sr/Ca ratio. It has been demonstrated in calves averaging 5 weeks of age that the calcium–strontium discrimination factor is not a constant throughout the gut but varies

with the site of absorption (7). Calves on a milk diet would have a higher digestibility and a different total nutrient absorption pattern than calves on the hay and grain diet.

Summary. *In vivo* experiments, using 12 anesthetized calves, were conducted to determine the extent of abomasal absorption of ^{45}Ca and ^{89}Sr . In two experiments, one with milk-fed calves and another with hay and grain-fed calves, a large amount of variation in absorption was noted. This was attributed to a lack of mixing of the isotope with abomasal ingesta. Variability was reduced in a third experiment by increasing the dose volume to 225 ml. An average of $20.8 \pm 3.1\%$ of an administered dose of ^{45}Ca and $3.2 \pm 3.0\%$ of ^{89}Sr were absorbed from the abomasum within 25 min. These estimates were based on the amounts of isotope recovered from ingesta and abomasal tissue of four animals and is substantiated by the rapid appearance of ^{45}Ca in venous blood drained from the abomasum.

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