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### I. THE FORESTOMACH GROWTH IN THE YOUNG DAIRY CALVES FED ON HAY OR DRIED NATIVE GRASS AS ROUGHAGE, AND ON STARTER.

By

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#### Introduction

The recent studies on the developing forestomach of young dairy calves have showed that the very early weaning system based on the early development of this organ is possible practically as well as theoretically (1-8). Castle and Watson (9) and Harrison et al. (10) reported that the calves were successfully weaned at 35 days of age. The latter authors claimed that nearly one-third of the whole milk required in the conventional weaning system could be saved in their system without any loss of weight gains. The weaning as early as 21 days of age was found successful by Preston (11) and Quayle (12). Similar results were obtained by Noller et al. quite recently (13). They reported that Holstein calves gained approximately 1 lb daily, whereas weaning at 28 days of age did not particularly increase the weight gain.

A high roughage system of feeding calves has been strongly recommended by the Ohio State workers (14). In their system, the early rumen function was encouraged by feeding rations high in roughage with limited grain, provided that the roughage consists of a good quality hay. No efforts were paid, however, to the determination of the morphological development of the rumen.

In connection with these early weaning or high roughage systems for raising calves, pasturing of young calves before and after the weaning has been

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investigated with increasing interest (15-16). Recently Kesler (17) reported that the growth of the pasture-fed and barn-fed calves was not different, though affectd by the season and by forage quality. In general, most authors agree that the young calves can make good use of pasture even before weaning.

It is clear that in these systems, either early weaning or early pasturing, the cost of feed can be reduced remarkably due to the decrease in the amounts of milk and concentrates fed to the calves. It remains doubtful, however, whether such systems may be recommended to the Japanese dairymen, since they have been facing difficulties in securing ample supply of tender forage or good quality hay. They were often enforced to utilize native grass as roughage. The recent research in our faculty suggested that dried native grass can be utilized as roughage for dairy cows within certain limitations (18). Thus the object of the present study is to obtain informations on the utility of dried native grass as roughage for rasing young calves and to answer the question whether such early weaning sytems are of practical values for Japanese dairy farmers.

The accumulated data have indicated that the development of the forestomach in the calf is determined by the quality as well as the quantity of feed given, while the efficiencies of feed digestion is mutually dependent on the stomach development itself (1-8). Accordingly the development was observed with eight dairy calves allotted to three dietary systems, using a few criteria previously proved most reliable (6-8).

#### Materials and Methods

Nine male Holstein calves purchased in Miyagi Prefecture within three days after birth were fed whole milk during the first two weeks of age. Then the whole milk was gradually replaced by a commercial calf starter ("Calf Meal", Nippon Formula Feed Mfd. Co., Ltd.), and the calves were weaned at the end of seventh week of age (Table 1). After the feeding of a commercial feed mix (Nippon Feed Mix Mfd. Co., Ltd.) for the following two weeks, the calves were allotted to three dietary groups, which will be termed as Groups HC, NC, and C, respectively. Group C received an experimental concentrate mixture, 60 per cent feed mix and 40 per cent ground corn, at the level of 2.5 per cent of body weight daily, to meet with the requirement of the Morrison Standard (Table 1). The amount of the experimental mixture was reduced to 1.75 per cent of body weight daily in Groups HC and NC, whereas hay in Group HC or dried native grass in Group NC was given ad libitum. The consumption of hay and dried native grass was relatively low. The composition of the concentrate mixture and those of the hay and dried native grass in this trial are shown in Table 2. Each dietary group consisted of three calves. In

Group NC, one calf (No. 2) suffered from serious disorder of the digestive tract and gained little. The animal was then excluded from the present trial.

Age (week)	Milk (kg/week)	Calf starter (g/week)	
1 – 2	4—5	0	
3	5	250	
4	5—4	500	
. 5	4—3	1000	
6	3—2	1200	
7	2—1	1500	
8 – 9	0	2000	

Table 1. Feeding schedules in the experiment (per one calf).

Table 2. Composition of feeds employed (%).

Feed	Moisture	Crude protein	Crude fat	Nitrogen free extract	Crude fiber	Ash
Experimental concentrate mixture	14.35	16.54	3.23	54.17	6.93	4.79
Hay*	12.70	9.62	3.01	37.53	30.27	6.87
Dried native grass*	12.97	7.71	2.59	39.62	29.62	7.49

<sup>\*</sup> Harvested in the Summer of 1960 at the Koiwai Farm in Iwate Prefecture.

All of the remaining eight calves grew normally.

All calves were slaughtered at 13 weeks of age, and development of their forestomachs was observed following the techniques previously employed by the senior author (6-7). The observation conducted were, (1) stomach capacities, (2) stomach tissue weight, (3) papillary growth in the rumen, and (4) histology of the stomach epithelia. The stomach capacities were measured by the volume-displacement method of Flatt et al. (3). The stomach tissues were fixed in Helly's fluid, embedded in paraffin, and cut into  $6\mu$  paraffin sections. The stainings employed were, hematoxylin-eosin, Crossman's modification of trichrome stain, and periodic acid-Schiff method for polysaccharides (PAS). Further details of the above methods are available in the previous reports (6-7).

#### Results

#### General growth

As shown in Table 3, the average weight gain of the three calves in Group C was 0.265 kg daily. The value was significantly lower than those in Group HC and NC. The gains of the latter two groups, 0.350-0.352 kg daily, were slightly lower than the level of Ragsdale and Beltsville standard for young calves (0.412 kg daily). The calves in Group C appeared less fleshing and

thrifty than those in Group HC and NC. No serious disorder of the digestive tract was experienced in the calves, except Calf No. 2 mentioned in th previous section.

#### Stomach capacities

The capacities of the reticulo-rumen were about 250-252 cc/kg body weight in average, and no significant difference was observed in the three dietary groups (Table 3). As seen in Table 4, the level of 250 cc/kg body weight was considerably higher than the level of calves fed on milk, hay and starter (6). This indicated that enlargement of the reticulo-rumen cavity was encouraged by the feeding of concentrates alone.

The omasum, as observed grossly, appeared largest in Group HC and smallest in Group C. However, the inner capacity of this compartment was largest in Group NC. The capacity of the omasum was surprisingly small in Group C, being almost one-ninth of those in Groups NC and HC (Table 3).

Diet	Calf	Body weight (kg)		Daily	Stomach capacities (cc/kg B.W.)		Stomach tissue weight (% B.W.)	
Dict	No.	Initial	Final	gain (kg)	R+R *	Om.	R + R	Om.
	1	49.8	84.0	0.376	280.9	3.6	2.14	0.57
HC	4	50.0	79.5	0.324	223.9	3.8	2.26	0.64
	7	50.0	82.0	0.351	283.5	8.3	2.07	0.51
	Average			0.350	262.8	5.2	2.16	0.57
NC	5	51.0	78.5	0.302	241.3	6.8	2.08	0.32
NC	8	55.5	92.0	0.401	276.1	5.1	2.07	0.39
	Average			0.352	258.7	6.0	2.08	0.35
	3	47.8	72.0	0.273	225.6	0.1	1.61	0.26
С	6	43.0	67.5	0.270	394.2	1.0	1.73	0.25
	9	50.5	72.5	0.242	196.6	1.0	1.66	0.25
	Average			0.262	252.2	0.7	1.67	0.25

Table 3. Daily weight gain, and capacity and tissue weight of the forestomach.

#### Stomach tissue weights

There was a marked difference in the stomach tissue weights between Group C and the other two groups (Table 3), being not consistent with the result in the previous section. Namely, the weights in Group HC and NC were 2.16 and 2.08 per cent of body weight respectively, the values higher than those of calves fed on milk, hay and starter. More remarkable difference in the tissue weight was noted in the omasum (Table 3). The weight was highest in Group HC and lowest in Group C, and the difference between Group NC and C was relatively small. Even in Group C, the omasum tissue weight was apparently higher than that of calves fed milk only.

<sup>\*</sup> Rumen plus reticulum

Diet	Age in weeks	Stomach capacity (cc/kg body weight)	Stomach tissue weight (% body weight)	Maximum papillary length(mm)	References
Milk	12	63.0	0.73	1.3	(1)
Milk	16	_	_	< 1.0	(2)
Milk, hay and starter	4	86.5	1.04	2.5	(1)
Milk, hay and starter	8	101.5	1.85	6.2	"
Milk, hay and starter	12	114.0	1.78	6.8	"
Hay and concentrates (9:1)	16	_		4.1	(2)
Hay and concentrates (9:1)	16	_	_	7.5	(2)
Hay and concentrates	13	262.8	2.16	5.5	(3)
Dried native grass and concentrates	13	258.7	2.08	5.6	"
Concentrates	13	252.2	1.67	3.5	//

Table 4. Comparison of the capacity, tissue weight, and papillary length of the reticulo-rumen in the calves fed on various diets.

#### Papillary development in the rumen

The average length of the rumen papillae in the ventralmost part of the anterior dorsal blind sac appeared largest in Group NC and smallest in Group C (Table 5), though the difference between Groups HC and NC was statistically not significant. The maximum lengths of the papillae in Group NC and HC were also larger than that in Group C, except one calf in Group HC. Comparison of these data with those of the previous experiments (Table 4) indicated that the papillary development was greatly retarded in Group C, almost remaining in the level of four weeks old calves receiving milk, hay and starter. Even in the other two groups, development appeared to be in the level between eight weeks and 12 weeks old calves on the same diet (Table 4). Foliate or paddle like papillae were found in the rumens of Group HC and NC, but not in Group C (Table 5). In the latter group, they were mostly finger-like, showing that their growth was retarded.

The number of papillae (per sq. cm) was much larger in Group C than in the other groups. This confirmed the reciprocal relation between the number

<sup>(1)</sup> Tamate et al. (1962)

<sup>(2)</sup> Warner et at. (1959)

<sup>(3)</sup> Present study (Tables 3 and 5)

and length of the papillae, suggested by the senior author (6-7).

No. of Number of Papillary growth Group Length (mm) Number Shape in calves samples Average Maximum  $(/cm^2)$ gross 2 1.09 1 4.4 220 foliate HC 2 2.09 5.5 198 foliate 4 7 2 1.29 2.8 287 foliate 235 Av. 1.45 5.5 2 5 1.62 4.8 340 foliate NC 8 2 227 1.85 5.6 foliate Av. 1.71 5.6289 3 2 0.79 2.9 284 finger-like C 2 6 0.79 2.7 569 finger-like 2 9 0.823.5 368 finger-like 0.08 407 Av. 3.5

Table 5. Papillary growth in the rumen (anterior dorsal blind sac).

#### Histology of the forestomach epithelia

The following terminology of the forestomach epithelia will be used in this study (19). Namely the five layers of the epithelium will be termed as stratum corneum, stratum transitionale, stratum granulosum, stratum spinosum, and stratum basale. Stratum disjunctivum and stratum lucidum in the older terminology (20) correspond to stratum corneum and stratum transitionale of the new system, respectively. Stratum germinativum consists of the inner three layers, stratum granulosum, spinosum and basale.

The rumen epithelia of the calves in Groups HC and NC showed little difference in their histology. Namely the corneum cell in both groups showed typical secondary or permanent swelling with little affinity to acid dyes employed, whereas those in stratum transitionale were stained intensely by them (Figs. 1-2). The swelling was not so numerous as those observed in the rumens of 12 weeks old calves fed on milk, hay and starter (7). In Group C, however, the corneum cells were stained more intensely by the acid dyes, specially by Orange G in Crossman's trichrome stain (Fig. 3). Most of the cells retained nuclei, seldom showing secondary swelling. The minimum thickness of stratum germinativum was larger in Group C than in Groups HC and NC, except for one calf in Group C (No. 6). In this calf the rumen epithelium was thinner and stratum corneum was better defined than those of the two calves in the group. The thickness of stratum cornem was not different in the three dietary groups (Table 6).

The histology of the epithelia in the reticulum and omasum was generally

similar to those observed in the rumen. Namely the distinction between stratum corneum and stratum transitonale was less evident in Group C than in Groups HC and NC (Figs. 4-8). The swellings of the epithelial cells in these two layers were fewer in Group C. The minimum thickness of stratum germinativum and of stratum corneum in the omasum showed no difference in the three dietary groups (Table 6).

Dietary	Calf	I	Rumen	Omasum		
Group	No.	Stratum corneum	Stratum germinativum	Stratum corneum	Stratum germinativum	
	1	7.9	20.8	6.1	17.7	
HC	4	7.2	21.2	4.8	19.8	
	7	6.9	19.8	4.5	18.4	
	Average	7.3	20.6	5.1	18.6	
NC	5	7.2	21.9	5.8	17.1	
	8	6.9	20.8	6.1	17.4	
	Average	7.1	21.4	6.0	17.3	
	3	7.9	22.6	4.5	19.1	
С	6	6.1	26.5	5.2	18.4	
	9	6.2	27.8	5.2	14.8	
	Average	6.7	25.6	5.0	17.4	

Tadle 6. Minimum thickness of the epithelia of the rumen and omasum  $(\mu)^*$ 

It will be concluded that the relatively thicker, and less keratinized epithelia of the forestomachs in Group C may be regarded as indications of retarded growth of the organs, compared with those in Groups HC and NC. In other words, feeding of concentrates without roughage was unfavourable for the growth of the calf forestomach epithelia. Similar parakeratotic condition of the epithelia was recently reported by Vidacs and Ward (21). They concluded that such condition may result from the alternation of volatile fatty acids absorption. A heavy parakeratosis of the rumen epithelium produced by the insufficient administration of the acids was reported by the senior author (7).

#### Discussion

The results of this study showed that the better growth of the forestomachs was obtained in the animals fed on hay or on dried native grass in addition to concentrates (Groups HC and NC). The calves in these groups also presented better weight gains than those which received concentrates alone (Group C). Of the various criteria employed for the evaluation of the stomach growth, papillary growth in the rumen and the stomach tissue deposition were

<sup>\*</sup> All values are averages of 10 measurements each.

proved to be most satisfactory. Histological observations also provided evidences that the epithelial growth in the reticulo-rumen was retarded by the feeding of concentrates alone. The capacity increase on the reticulo-rumen of the same animals probably due to the effect of prolonged stay of the feed in the organ under slower fermentation in the cavity. Similar increase in the capacity has been reported in the calves that consumed a large amount of shavings (10).

A relatively small amount of roughage intake, hay or dried native grass, was effective in stimulating good rumen growth. Similar results was reported by the Cornell workers (8). They reported that best papillary growth was obtained by the feeding of 90 per cent concentrates-10 per cent hay (Table 6). The present authors were inclined to believe that high rate feeding of hay, such as recommended by the Ohio workers (14), was not essential for the early development of the reticulo-rumen, provided that the fermentation in the organ was successfully encouraged by proper feeding.

The data in this study appeared to indicate that the feeding of dried native grass as roughage was as good as hay in the raising of young calves. As seen in Table 2, the crude protein content and crude fat content of the dried native grass in this experiment were considerably lower than those of hay. The fact that the intakes of roughages were low should be also taken into consideration. In conclusion, however, the present authors suggest that the dried native grass or native grass pasture may be of practical values in such feeding systems as employed in this study, i.e., high concentrates-low roughage one. Further studies along this line were hoped.

#### Summary

Eight Holstein calves were allotted, at eight weeks of age, to three dietary groups: hay and concentrates (Group HC), dried native grass and concentrates (Group NC), and concentrates alone (Group C). The animals were slaughtered at 13 weeks of age, and development of their forestomachs as well as their growth was observed.

The calves that received hay or dried native grass with concentrates presented better weight gains than those which received concentrates alone. Better development of the forestomachs was clearly demonstrated by the better papillary growth in the rumen and by the weight increase of the forestomach tissues. No difference was observed in the capacity increase of the organs in the three dietary groups.

Retarded growth of the forestomach epithelia resulted from the feeding of concentrates alone. This was indicated by the thicker, and more parakeratotic epithelia of the forestomachs in the calves in Group C. A possible relation of this parakeratosis with the alternation of the volatile fatty acids absorption

in the rumen was briefly discussed.

The present author suggest that the feeding of dried native grass or native grass pasture may be of paractical values in the high concentrates-low roughage feeding systems for raising young dairy calves.

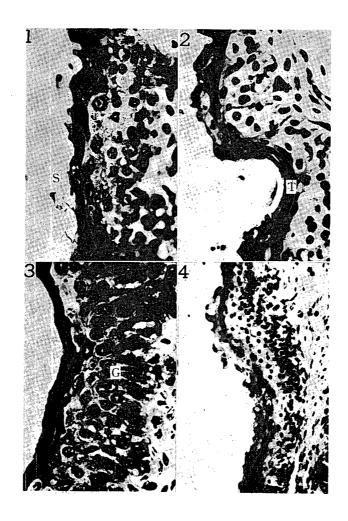
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## Plate 1. Explanation of Figures

- Fig. 1. Rumen of Calf No. 7 in Group HC. Crossman's trichrome. × 400. Secondary swellings (S) in stratum corneum is noted.
- Fig. 2. Rumen of Calf No. 8 in Group NC. Hematoxylin-eosin.  $\times 400$ . Secondary swelling and stratum transitionale (T) are evident.
- Fig. 3. Rumen of Calf No. 9 in Group C. Crossman's trichrome.  $\times 400$ . Stratum germinativum (G) is thicker, and corneum is absent.
- Fig. 4. Reticulum of Calf No. 1 in Group HC. Hematoxylin-eosin. ×200. Stratum corneum is conspicuous.

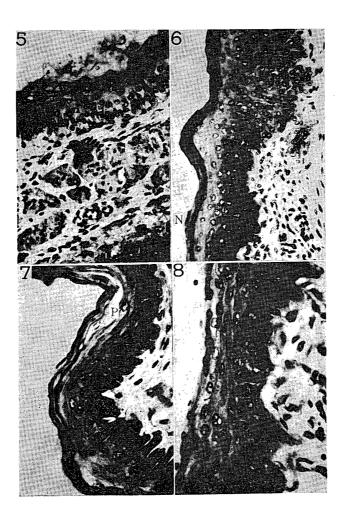


Plate 2. Explanation of Figures

- Fig. 5. Reticulum of Calf No. 8 in Group NC. Crossman's trichrome.  $\times 200$ . Secondary swelling of stratum corneum is evident.
- Fig. 6. Reticulum of Calf No. 6 in Group C. Crossman's trichrome. ×400. Retention of nuclei (N) in outer transitionale is noted.
- Fig. 7. Omasum of Calf No. 8 in Group NC. Crossman's trichrome.  $\times 400$ . Primary (P) and secondary swellings are noted.
- Fig. 8. Omasum of Calf. No. 9 in Group C. Crossman's trichrome. ×400. Stratum corneum is hardly seen.