Concurrent studies of the flow of digesta in the duodenum and of exocrine pancreatic secretion of calves

4.* The effect of age

By J. H. TERNOUTH†, J. H. B. ROY AND SUSAN M. SHOTTON

National Institute for Research in Dairying, Shinfield, Reading RG2 9AT

(*Received* 1 *March* 1976 – *Accepted* 8 *April* 1976)

1. The effect of age on the flow of duodenal digesta and of pancreatic secretion was studied in preruminant calves fitted with duodenal re-entrant and pancreatic sac cannulas. In Expt 1 a comparison was made at 7, 24 and 63 d of age of Ayrshire calves given whole milk, and in Expt 2 Friesian calves given milk-substitute diets were studied during the period 16–37 d of age.

2. For the Ayrshire calves, *ad lib.* whole-milk intake increased with age, but whole-milk intake on a per kg metabolic body-weight ($W^{0.75}$) basis did not alter. As the intake increased with age, the whey fluids were passed out of the abomasum more rapidly.

3. As judged by the mean duodenal pH values and the values for 'chloride ion minus sodium ion' concentration in the digesta, and for apparent secretions (total quantity recovered at duodenal cannula during 12 h after feeding minus intake) the duodenal digesta became more acid as the Ayrshire calves became older. There was no change with age in the extent of degradation of the milk proteins as indicated by the protein-nitrogen:total N values for duodenal digesta.

4. The volume of pancreatic fluids secreted by the Ayrshire calves increased markedly with age, but when expressed on a per kg $W^{0.75}$ the volume did not increase after 24 d of age. In absolute terms and also in relation to whole-milk intake or $W^{0.75}$ there was a large increase in secretion of total protease, a very large increase in α-amylase (*EC* 3.2.1.1) but no change in trypsin (*EC* 3.4.4.4) activities with age.

5. In the Friesian calves secretion of pancreatic fluid and activities of α-chymotrypsin (*EC* 3.4.4.5), α-amylase, lipase (*EC* 3.1.1.3) and ribonuclease (*EC* 2.7.7.16, 2.7.7.17) but not trypsin or total protease increased with age.

6. The extent of the increase in secretion of pancreatic enzymes during the first hour after the Friesian calves were fed milk-substitute diets differed between enzymes.

7. The changes in the quantities of pancreatic enzymes secreted are discussed in relation to the changes in the digestibility of milk-substitute diets.

The neonatal ruminant is considered to be immature in terms of its ability to secrete acid and pepsin (*EC* 3.4.23.1) from the abomasal mucosa (Hill, Noakes & Lowe, 1970) but the effect of this immaturity upon the digestive efficiency of the abomasum does not appear to have been studied. Knowledge of the changes with age in the quantities of enzymes secreted by the pancreas of the calf is also limited (Zerebcov & Seryh, 1962; Morrill, Stewart, McCormick & Fryer, 1970).

In the calf, it has been found that the greater the quantity of milk fed, the longer is the time required for all the whey fluids to leave the abomasum (Ternouth, Roy & Siddons, 1974). Only when most of the whey fluids have left the abomasum is the secretion of acid able to lower the pH of the abomasal fluids so that rapid digestion of the casein clot can occur (Mylrea, 1966*a*, *b*; Ternouth, Roy & Siddons,

* Paper no. 3: *Br. J. Nutr.* (1975), **33**, 181.

† Present address: Department of Animal Husbandry, Faculty of Veterinary Science, University of Queensland, St Lucia, Queensland 4067, Australia.

1974). With the increase in voluntary milk intake of calves as they grow, it appears necessary for the rate of abomasal emptying to change to accommodate this increased intake.

To study these effects of age, six Ayrshire calves used in an earlier experiment (Ternouth, Roy & Siddons, 1974) were given whole milk, and the results compared in Expt 1 at a mean age of 7, 24 and 63 d. A re-analysis of results obtained from Friesian calves (Ternouth, Roy, Thompson, Toothill, Gillies & Edwards-Webb, 1975), to study the effects of age in this breed, is reported in Expt 2. A preliminary communication of some of these results has been published (Ternouth, Siddons & Toothill, 1971).

EXPERIMENTAL

Expt 1. Six Ayrshire calves with duodenal re-entrant and pancreatic sac cannulas (Ternouth & Buttle, 1973) were offered whole milk at 39° *ad lib.* by teat twice daily at 09.00 and 21.00 hours. The calves were given at least three feeds of whole milk before the beginning of the collection periods, which were between 6 and 8, 22 and 27 and 55 and 75 d of age. Polyethylene glycol (PEG) (approximately 1 g/l) was added to the milk fed at the beginning of the collection period. The methods used for collecting, sampling and returning the duodenal digesta and the pancreatic secretions have been described previously (Ternouth, Roy & Siddons, 1974). The age of the calves, their live weight and the quantity of milk consumed at each experimental meal are given in Table 1. Calves nos. 1 and 2 were slaughtered before they reached 50 d of age. The results for three collections (calf no. 1 25 d, calf no. 3 6 and 24 d) have been omitted because of abnormally low milk intakes. Before the analysis of the pancreatic samples could be completed, the final samples of calves nos. 5 and 6 (73–75 and 66–68 d respectively) were inadvertently lost.

The technique of Bartlett (1934) for dealing with missing values was used in the analysis of variance statistical programme.

Expt 2. The effect of age has been obtained from a re-analysis of the results of an experiment presented in a previous paper (Ternouth *et al.* 1975). That experiment was a comparison made during the period from 16 to 37 d of age of four milk-substitute diets given by teat to four Friesian calves cannulated as in Expt 1. Each diet was given for twelve consecutive meals, the experimental collections being made after the 6th and 12th meals. To minimize loss of enzyme activity, particularly of α-amylase (*EC* 3.2.1.1), lipase (*EC* 3.1.1.3) and ribonuclease (*EC* 2.7.7.16, 2.7.7.17), during the collection, the pancreatic fluids were collected in a stainless-steel container surrounded by a jacket of ice. All other experimental conditions were similar to those described in Expt 1.

Analyses

The chemical and enzyme assay techniques have been described by Ternouth, Roy & Siddons (1974) and Ternouth *et al.* (1975).

Table 1. *Expt* 1. *Live weight and whole-milk intake of six Ayrshire calves at various ages from* 6 *to* 75 *d old*

Calf no.	Live weight (kg)	Whole-milk intake: Age (d)	Intake (l)	Age (d)	Intake (l)
1	36·0*	6	2·80	8	3·64
	41·0	—†	—†	27	4·94
2	36·0*	6	3·27	8	3·79
	42·0	24	2·20	26	3·55
3	34·0*	—†	—†	8	2·93
	38·0	22	3·18	—†	—†
	51·0	55	5·23	57	4·64
4	44·0*	6	2·91	8	3·55
	51·0	22	5·09	24	5·27
	67·0	55	3·73	57	4·68
5	37·0*	6	3·41	8	4·05
	48·0	22	5·00	24	3·41
	68·0	73	4·30	75	5·07
6	34·0*	6	3·61	8	3·84
	42·0	22	5·07	24	3·87
	59·0	66	5·73	68	6·07

* Birth weight.

† Values for calf no. 1 at 25 d, and for calf no. 2 at 6 and 24 d were omitted because of abnormally low intakes.

Table 2. *Expt* 1. *Mean whole-milk intakes and duodenal digesta flow-rates of Ayrshire calves at three ages*

	Mean age (d)			SE of mean	Statistical significance of differences between ages		
	7	24	63		7 *v.* 24	24 *v.* 63	7 *v.* 63
No. of calves	6	6	4				
Whole-milk intake (l)	3·53	4·16	4·69	0·23	—	—	**
Duodenal digesta flow (ml) in the following periods (h) after feeding:							
0–3	2039	2626	3136	256	—	—	**
3–6	1811	1662	2015	214	—	—	—
6–9	1040	1081	1214	105	—	—	—
9–12	766	847	1009	109	—	—	*
Total flow-rate:							
ml/12 h	5655	6218	7374	273	—	**	***
ml/l whole-milk intake	1608	1536	1593	62	—	—	—
ml/kg $W^{0·75}$	386	366	335	15	—	—	—

$W^{0·75}$, metabolic body-wt.

* $P < 0·05$, ** $P < 0·01$, *** $P < 0·001$.

RESULTS

Expt 1. The intake of whole milk increased with age (Table 2), the mean live weights were 36·8, 43·7 and 61·3 kg at 7, 24 and 63 d and the daily whole-milk intakes were equivalent to 56, 60 and 56 g dry matter (DM)/kg metabolic body-weight ($W^{0·75}$) respectively for whole milk containing 125 g DM/kg. These intakes determined

Table 3. *Expt* 1. *Mean duodenal pH, and concentrations and total outflows of hydrogen, sodium, potassium and chloride ions during* 12 *h after feeding whole-milk for Ayrshire calves at three ages*

	Mean age (d)			SE	Statistical significance of differences between ages		
	7	24	63	of mean	7 *v.* 24	24 *v.* 63	7 *v.* 63
No. of calves	6	6	4				
pH	3·67	3·45	2·98	0·08	—	**	***
Concentration (mmol/l) of:							
Na^+	35·8	28·1	31·1	1·9	*	—	—
K^+	13·9	12·5	12·8	0·8	—	—	—
Cl^-	79·9	83·3	81·0	3·0	—	—	—
$Cl^- - Na^+$	44·0	55·1	49·9	2·8	*	—	—
Total outflow of:							
H^+: mmol	0·69	1·80	7·76	0·72	—	***	***
mmol/l whole-milk intake	0·26	0·51	1·63	0·14	—	***	***
mmol/kg $W^{0.75}$	0·069	0·118	0·352	0·035	—	***	***
Na^+: mmol	202·1	169·1	221·1	8·3	*	***	*
mmol/l whole-milk intake	57·9	43·9	50·0	4·2	—	—	—
mmol/kg $W^{0.75}$	13·6	10·1	10·2	0·6	***	—	***
K^+: mmol	77·0	76·0	92·3	6·6	—	—	—
mmol/l whole-milk intake	22·5	19·3	20·3	1·6	—	—	—
mmol/kg $W^{0.75}$	5·32	4·78	4·27	0·36	—	—	—
Cl^-: mmol	451·5	513·7	594·2	22·3	—	*	***
mmol/l whole-milk intake	128·4	128·8	128·9	8·0	—	—	—
mmol/kg $W^{0.75}$	30·7	30·2	27·2	1·2	—	—	—
Cl^- minus Na^+:							
mmol	249·3	344·8	373·3	21·6	**	—	**
mmol/l whole-milk intake	70·5	84·9	78·9	5·7	—	—	—
mmol/kg $W^{0.75}$	17·1	20·2	16·9	1·2	—	—	—

$W^{0.75}$, metabolic body-wt.
* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

for calves sucking teats are similar to the maximal daily intakes recorded by Roy (1970) for bucket-fed Friesian calves given reconstituted milks containing 138 g DM/kg.

The pattern of flow of duodenal digesta which was shown in Fig. 3 of Ternouth & Buttle (1973) is indicated in results given in Table 2. At the older ages more duodenal digesta were collected and returned to the calf, reflecting the increased whole-milk intake and also the increased volume of apparent secretion at 63 d (Table 4). The increased volume of duodenal digesta at older ages was found throughout the 12 h postprandial period, although it was more marked during the first 3 h after feeding. When the rate of PEG outflow was expressed on a square-root basis (Ternouth, Roy & Siddons, 1974), the mean period of time after feeding (h) for all PEG to leave the abomasum was unaffected by age, the values being 9·5, 9·7 and 9·0 (SE ± 0·85) at 7, 24 and 63 d of age respectively.

Table 4. *Expt* 1. *Mean apparent secretion*† *of fluid and 'chloride ions minus sodium ions' in the duodenal digesta during* 12 *h after feeding whole-milk for Ayrshire calves at three ages*

	Mean age (d)			SE of mean	Statistical significance of differences between ages		
	7	24	63		7 *v.* 24	24 *v.* 63	7 *v.* 63
No. of calves	6	6	4				
Volume of fluid:							
l	2·13	2·06	2·69	0·16	—	*	*
l/l whole-milk intake	0·61	0·54	0·59	0·06	—	—	—
l/kg $W^{0.75}$	0·145	0·123	0·121	0·009	—	—	—
'Cl^- minus Na^+':							
mmol	199·4	287·1	307·2	20·1	**	—	**
mmol/l whole-milk intake	56·2	71·0	64·8	5·6	—	—	—
mmol/kg $W^{0.75}$	13·7	16·8	13·9	1·1	—	—	—
mmol/l fluid secreted	94·7	143·1	119·3	18·1	*	—	—

$W^{0.75}$, metabolic body-wt.
* $P < 0.05$, ** $P < 0.01$.
† Total quantity recovered at the duodenal cannula during 12 h after feeding minus intake.

The mean pH and the total outflow of hydrogen ions in the 12 h period after feeding changed significantly with age, the duodenal digesta becoming more acid in the older calves (Table 3). The general pattern of flow during the 12 h collection period was similar to that described by Ternouth, Roy & Siddons (1974). During the first 4 h after feeding, no consistent differences with age were found in the pH of the duodenal digesta, but after 4 h, the duodenal pH values were lower as the calves became older.

The total outflow of sodium, chloride ions, and 'Cl^- minus Na^+' increased at the older ages, but this increase could be accounted for by the increased whole-milk intake or by the increased $W^{0.75}$ of the calves (Table 3). At the earliest age studied, the concentration of Na^+ was significantly higher than at older ages but the concentration of Cl^- did not change with age. These changes are reflected in the 'Cl^- minus Na^+' concentration which increased significantly between 7 and 24 d of age. The total quantity of apparent secretion (total quantity recovered at duodenal cannula during 12 h after feeding minus intake) of 'Cl^- minus Na^+' and its concentration also increased significantly between 7 and 24 d of age, but the total quantity did not differ with age when related to whole-milk intake or $W^{0.75}$ (Table 4).

The pattern of nitrogen, protein and fat outflow in the duodenal digesta during the 12 h collection period is shown in Fig. 1. As there was no significant difference in the pattern between ages, a single mean value is shown. As a result of the coagulation of the casein, the concentrations of N, protein and fat were lower 2–4 h after feeding (Ternouth, 1971), and especially in the instance of fat considerably higher 6–10 h after feeding. The difference in outflow of N, protein and fat with age could be largely accounted for by differences in whole-milk intake. The extent

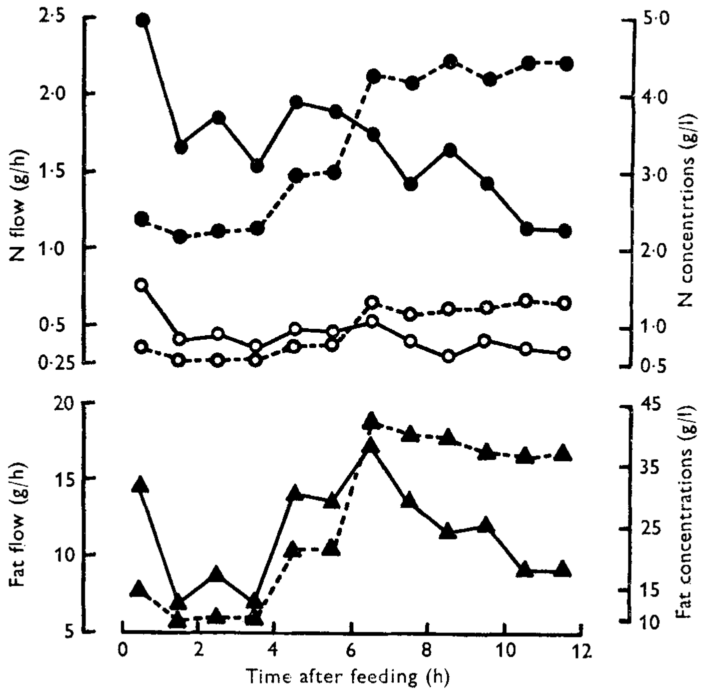

Fig. 1. Expt 1. Mean hourly total flow (g/h) (—) and concentration (g/l) (----) of nitrogen (●), protein-N (○) and fat (▲) through the duodenum of Ayrshire calves during 12 h after feeding whole milk.

of degradation of the protein, as indicated by the value for protein-N:total N, did not change with age (Table 5).

The changes in the volume of pancreatic secretion with age were large (Table 6). The pattern of secretion over a 12 h period (Ternouth & Buttle, 1973), indicated that calves had higher secretion rates as they became older, although the differences were small in the period 2–4 h after feeding, when the rate of secretion was lower than at any other time in the 12 h postprandial period. The volume of pancreatic secretion on a per l whole-milk intake basis increased with age, but no increase was apparent when secretion rate on a per kg $W^{0.75}$ basis was considered.

Total trypsin (*EC* 3.4.4.4) activity tended to increase with age (Table 7), although the non-significant change was almost entirely due to changes in the volume of pancreatic secretion rather than to a change in trypsin activity on a per ml pancreatic secretion basis. There was nearly a fourfold increase in the total protease activity between 7 and 24 d of age and a further twofold increase between 24 and 63 d. Changes in the α-amylase activity were so marked that the results were transformed into a logarithmic scale before being analysed statistically.

Expt 2. Changes in the pancreatic secretion of the four Friesian calves, given four milk-substitute diets between the ages of 16 and 37 d of age, are indicated by results given in Table 8. The regressions were calculated after removing the variability associated with differences between diets and between calves. With the exception

Table 5. *Expt* 1. *Mean duodenal outflow of protein, nitrogen and fat during* 12 *h after feeding whole milk for Ayrshire calves at three ages*

	Mean age (d)				Statistical significance of differences between ages		
	7	24	63	SE of mean	7 *v.* 24	24 *v.* 63	7 *v.* 63
No. of calves	6	6	4				
Total N:							
g	16·84	20·59	21·50	1·06	*	—	**
g/l whole-milk intake	4·72	5·12	4·76	0·35	—	—	—
g/kg $W^{0.75}$	1·15	1·20	0·97	0·06	—	—	—
Protein-N:							
g	3·92	5·55	6·61	0·54	—	—	**
g/l whole-milk intake	1·14	1·36	1·46	0·12	—	—	—
g/kg $W^{0.75}$	0·27	0·33	0·30	0·03	—	—	—
Protein-N: total N	0·240	0·266	0·313	0·021	—	—	—
Fat:							
g	119·6	141·1	143·6	12·3	—	—	—
g/l whole-milk intake	34·0	36·2	31·0	4·0	—	—	—
g/kg $W^{0.75}$	8·22	8·32	6·30	0·66	—	—	—

$W^{0.75}$, metabolic body-wt.
* $P < 0.05$, ** $P < 0.01$.

Table 6. *Expt* 1. *Volume of pancreatic secretion after feeding whole milk for Ayrshire calves at three ages*

	Mean age (d)				Statistical significance of differences between ages		
	7	24	63	SE of mean	7 *v.* 24	24 *v.* 63	7 *v.* 63
No. of calves	6	6	4				
Period after feeding (h):							
0–3							
ml	62·4	84·9	109·3	4·3	**	**	***
ml/l whole-milk intake	18·8	22·2	24·2	1·6	—	—	—
ml/kg $W^{0.75}$	4·45	5·26	4·91	0·26	—	—	—
3–6:							
ml	73·9	109·6	167·8	12·6	—	**	***
ml/l whole-milk intake	22·9	26·2	35·6	2·5	—	*	**
ml/kg $W^{0.75}$	5·42	6·49	7·30	0·61	—	—	—
6–9:							
ml	70·2	112·6	158·1	18·9	—	—	**
ml/l whole-milk intake	22·1	27·6	33·9	4·0	—	—	—
ml/kg $W^{0.75}$	5·18	6·68	6·95	0·95	—	—	—
9–12:							
ml	74·2	107·2	161·0	9·6	*	**	***
ml/l whole-milk intake	22·5	25·7	35·2	2·3	—	*	**
ml/kg $W^{0.75}$	5·39	6·29	7·13	0·52	—	—	—
0–12:							
ml	280	419	598	36	*	**	***
ml/l whole-milk intake	86·3	101·7	128·9	8·2	—	*	**
ml/kg $W^{0.75}$	20·5	24·7	26·3	1·8	—	—	—

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.
$W^{0.75}$, metabolic body-wt.

Table 7. *Expt 1. Mean secretion of enzymes (mg/12 h) by the pancreas during 12 h after feeding whole-milk for Ayrshire calves at three ages*

	Mean age (d)			SE of mean	Statistical significance of differences between ages		
	7	24	63		7 v. 24	24 v. 63	7 v. 63
No. of calves	6	6	4				
Trypsin (*EC* 3.4.4.4):							
mg	133·1	169·9	191·6	47·1	—	—	—
mg/l whole-milk intake	39·0	36·7	49·6	9·6	—	—	—
mg/kg $W^{0.75}$	9·54	9·73	8·74	2·72	—	—	—
Total Proteases:							
mg	399	2701	5065	785	—	—	**
mg/l whole-milk intake	164	580	1248	150	—	*	***
mg/kg $W^{0.75}$	51·0	162·8	227·1	39·8	—	—	*
α-Amylase (*EC* 3.2.1.1) (mg)	8·9	46·8	269·2	na	**	**	***

$W^{0.75}$, metabolic body-wt; na, SE not available as values transformed for analysis.
* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

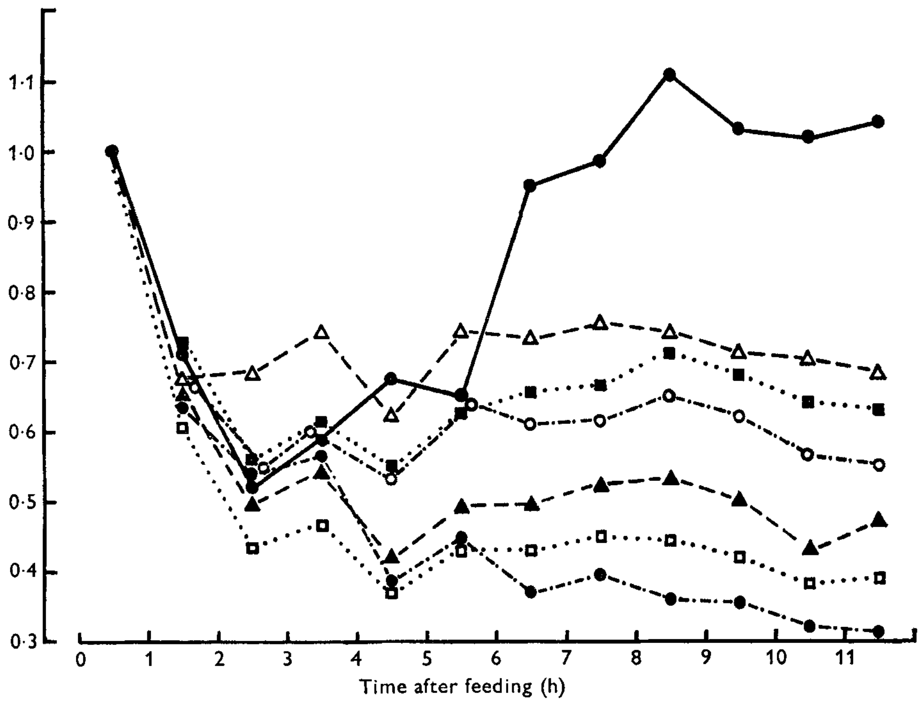

Fig. 2. Expt 2. Mean quantities of pancreatic fluid (●—●), α-amylase (*EC* 3.2.1.1) (△), ribonuclease (*EC* 2.7.7.16, 2.7.7.17) (■), α-chymotrypsin (*EC* 3.4.4.5) (○), trypsin (*EC* 3.4.4.5) (▲), lipase (*EC* 3.1.1.3) (□) and total protease (●) activities secreted by four Friesian calves during 12 h after feeding relative to those secreted during the first hour after feeding milk-substitute diets (for details of diets, see Ternouth, Roy, Thompson, Toothill, Gillies & Edwards-Webb (1975)).

Table 8. *Expt 2. Regression equations*† *relating the volume of fluid (ml/12 h) or enzyme activity (mg (or kU)/12 h) secreted by the pancreas of four Friesian calves aged between 16 and 37 d when fed on milk-substitute diets*

(Values in parentheses are standard errors of the regression coefficients)

	Secretion				Secretion (/l milk)				Secretion (/kg $W^{0.75}$)			
	a	b	RSD	Statistical significance	a	b	RSD	Statistical significance	a	b	RSD	Statistical significance
Pancreatic volume	117·2	6·42 (±1·12)	43·1	***	31·83	1·75 (±0·54)	20·57	**	8·482	0·248 (±0·059)	2·281	***
α-Amylase (*EC* 3.2.1.1)	−7·10	0·972 (±0·185)	7·11	***	−2·47	0·282 (±0·054)	2·09	***	−0·246	0·046 (±0·009)	0·351	***
α-Chymotrypsin (*EC* 3.4.4.5)	209·7	36·2 (±8·82)	339·0	***	40·51	10·27 (±2·75)	105·7	**	19·19	1·567 (±0·436)	16·78	**
Lipase (*EC* 3.1.1.3) (kU‡/12 h)	−4·68	0·980 (±0·177)	6·82	***	−1·68	0·276 (±0·059)	2·25	***	−0·113	0·0457 (±0·0088)	0·338	***
Ribonuclease (*EC* 2.7.7.16, 2.7.7.17)	3·37	1·087 (±0·158)	6·07	***	−1·01	0·372 (±0·076)	2·09	***	0·422	0·0464 (±0·0075)	0·287	***
Total Protease	7791	−73·7 (±40·4)	1553	—	2013	−17·6 (±9·8)	376	—	446	−5·27 (±2·02)	77·8	*
Trypsin (*EC* 3.4.4.4)	172·4	3·54 (±1·81)	69·5	—	43·49	1·024 (±0·527)	20·25	—	10·81	0·116 (±0·089)	3·43	—

$W^{0.75}$, metabolic body-wt, RSD, residual standard deviation.

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

† Covariance regressions estimated after variability due to calves and diets had been removed; regressions in the form $y = a + bx$, where y is the amount of secretion and x is the age of the calf (d).

‡ One unit of activity is the amount of enzyme which liberates 1 μmol acid/min.

of the total proteases, which tended to decrease with age, the total activities of all the other pancreatic enzymes and the pancreatic volume increased with age, except that the increase for trypsin was not significant. The mean daily rate of increase in secretion of pancreatic fluids was +2·26%. The corresponding values for α-amylase, α-chymotrypsin (*EC* 3.4.4.5), lipase and ribonuclease were +5·36, +4·94, +4·72 and +3·45% respectively. For the non-significant regressions for trypsin and total proteases, the values were +1·34 and −1·25% respectively. Although the calves were allowed to suck their diets *ad lib.* at each meal, there was no significant change in the intake of milk-substitute with age over the experimental period (Ternouth *et al.* 1975) so that the regressions of pancreatic secretion expressed per kg whole-milk intake and per kg $W^{0.75}$ were similar.

Fig. 2 shows the mean quantities of pancreatic fluid and enzyme activities secreted by Friesian calves during 12 h after feeding relative to those secreted during the first hour after feeding. The quantities of each enzyme remained constant between 3 and 12 h after feeding, whilst the volume of pancreatic fluid increased to a level greater than 1·0 between 6 and 12 h after feeding.

DISCUSSION

The changes associated with age found in these experiments could have been confounded by residual effects of the surgical procedure, but as no abnormalities of gastrointestinal function, e.g. diarrhoea, were noted at any time and all the calves were consuming relatively large quantities of milk within 3 d of the surgical operation, these residual effects were considered to be small.

It was evident from the results of Expt 1 that the calf adapted to increased whole-milk intake with increasing age so that additional time was not required for the abomasal emptying of larger quantities of whole milk. Ternouth, Roy & Siddons (1974) found that as the quantity of whole milk was increased at a given age, the period of time required for all of the PEG to pass out of the abomasum is also increased. In the present experiment, the additional quantities of whole milk consumed by the older calves passed out of the abomasum within similar periods of time. The mean values for complete PEG outflow (h) of 9·5, 9·7 and 9·0 for the three ages may be compared with values of 8·8, 9·6 and 10·8 respectively, obtained by applying the regression equation, OT = 6·2+0·864 MI (Ternouth, Roy & Siddons, 1974), where OT is the mean period of time for PEG outflow (h), and MI is the mean whole-milk intake (l) of the calves at the three ages. The faster rate of abomasal emptying in the older calves is associated with a relatively greater weight of abomasal and intestinal tissue due to increases in both gut length and the thickness of the gut wall (Warner, Flatt & Loosli, 1956; Wardrop & Coombe, 1960; Large, 1964; Siddons, 1968).

Ternouth, Roy & Siddons (1974) used the 'Cl^- minus Na^+' concentration as a second indicator of the quantity of abomasal acid secretion. However, Sasaki (1968) found a considerable increase in the concentration of bicarbonate ions and a decrease in the concentration of Cl^- in the saliva of the older preruminant calf, and this may preclude the use of the 'Cl^- minus Na^+' values in the duodenal effluent of the 63-d-old calf. Between 7 and 24 d of age, there was a significant increase in the 'Cl^- minus Na^+' concentration and a significant reduction in the mean pH although there was an increased buffering capacity resulting from increased protein intake. Between 24 and 63 d of age, a further reduction in pH occurred. It is thus evident that at least for the first 4 weeks of life, the abomasal secretion of acid increases. Hill (1956) has reported changes in the numbers of parietal cells present in the abomasal mucosa of the neonatal lamb but comparable values do not appear to be available for the calf. The outflow of almost 75% of the total N as nitrogenous material soluble in dilute trichloroacetic acid solution (20 g/l) at all three ages suggests that the quantities of abomasal enzymes and acid secreted are adequate, even at a young age, to maximize the abomasal proteolysis of whole milk.

The absolute volume of pancreatic secretion and the volume in relation to milk intake was found to increase with age, both between 7 and 63 d of age in Ayrshire calves and between 16 and 37 d of age in Friesian calves; these results are in keeping with those obtained by Gorrill, Thomas, Stewart & Morrill (1967) and McCormick & Stewart (1967). However, on a $W^{0.75}$ basis there was no increase in secretion with age in Ayrshire calves. The pattern of secretion within a 12 h period (see Fig. 3 of Ternouth & Buttle (1973)) suggests that the increase in secretion is due to greatly increased glandular response to some form of stimulation.

Reynolds (1972) found that intravenous infusion of secretin in adult sheep, caused an immediate fourfold increase in the volume of pancreatic secretion but no change in the quantities of trypsin, α-chymotrypsin and α-amylase secreted apart from a temporary initial increase (a wash-out effect). In the period 6–12 h after feeding in our experiments, the increased secretion of pancreatic fluid but not enzymes (Fig. 2) is due to stimulation by secretin. If the constant levels of enzyme secretion between 3 and 12 h after feeding are considered to be 'basal' for the calf in a post-prandial state, then the only stimulus to increased secretion during the whole 12 h post-prandial period was the act of feeding. During the first hour after feeding, the level of secretion was 39, 51, 66, 104, 138 and 185% higher than the 'basal' rate for α-amylase, ribonuclease, α-chymotrypsin, trypsin, lipase and total protease activities respectively. In the adult sheep, non-parallel secretion of enzymes as a result of vagal stimulation has been found by Reynolds (1972). He found a 1·5-fold increase in the flow of pancreatic fluid and a 3–5-fold increase in trypsin, α-chymotrypsin and α-amylase concentrations. The very high rates of secretion of certain enzymes may indicate their importance in the preruminant calf (total protease, lipase and trypsin). α-Chymotrypsin may be of less importance because it hydrolyses the same peptide bonds as pepsin and rennin (*EC* 3.4.23.4). In contrast to the high values for single-stomached animals, the value for α-chymotrypsin:trypsin in ruminant animals generally is close to 1·0, while the value for preruminants is 0·39–0·54 (Gorrill & Thomas, 1967; Gorrill *et al.* 1967; Schingoethe, Gorrill, Thomas & Yang, 1970). The higher value for this ratio in the older, weaned calf may be associated with the more uniform pancreatic secretion in the ruminanting animal (Taylor, 1962; J. H. Ternouth, J. H. B. Roy, I. J. F. Stobo, S. M. Shotton & C. M. Gillies, unpublished results).

When fats are re-incorporated into skim milk, the digestibility is considerably increased by homogenization and the use of emulsifying agents (Hopkins, Warner & Loosli, 1959; Roy, Shillam, Thompson & Dawson, 1961; Thomke, 1963*a*; Raven & Robinson, 1964; Toullec & Mathieu, 1969; Raven, 1970). Although there is no indication that there is insufficient lipase in the young calf fed whole milk, lipase activity may be limiting when the quantity of substrate available to be hydrolysed is reduced by blending fats with skim milk, i.e. by a direct reduction in the interfacial area, or indirectly by a shortage of emulsifying agent to remove the hydrolysed fatty acids from the interfacial area. Under these conditions the increased digestibility of fat with increasing age (Thomke, 1963*b*; Raven & Robinson, 1964) accords with the increased secretion of lipase found in the present experiment.

Although comparisons between the two experiments need to be treated with caution, Friesian calves appeared to secrete considerably more total protease (g activity/12 h) (6·0 cf. 2·8) but less α-amylase (mg activity/12 h) (16·0 cf. 57·5) than Ayrshire calves at a mean age of 24 d. As the diets fed to the two breeds of calves differed both before and during the experiments, it cannot be concluded that these are true breed differences. However, it could be more than mere coincidence that Friesian in comparison with Ayrshire calves have higher digestibilities of milk-protein at the same age, are less susceptible to diarrhoea when fed on skim milk and have higher faecal DM contents (Roy, 1970; Roy, Stobo & Gaston, 1970; Ternouth, Roy, Stobo, Ganderton, Gillies & Shotton, 1974). Shillam & Roy (1963) and Roy, Stobo, Gaston, Shotton & Ganderton (1973) have also reported lower apparent digestibilities of N in 1-week-old Ayrshire calves than in older calves.

The authors wish to thank Dr J. Toothill, Mr E. Florence and Miss C. M. Gillies for undertaking some of the chemical analyses and enzyme assays. J.H.T. wishes to thank the Australian Dairy Produce Board for their financial assistance.

REFERENCES

Bartlett, M. S. (1934). *Jl R. statist. Soc.* Suppl., **1**, 252.
Gorrill, A. D. L. & Thomas, J. W. (1967). *J. Nutr.* **92**, 215.
Gorrill, A. D. L., Thomas, J. W., Stewart, J. E. & Morrill, J. L. (1967). *J. Nutr.* **92**, 86.
Hill, K. J. (1956). *Q. Jl exp. Physiol.* **41**, 421.
Hill, K. J., Noakes, D. E. & Lowe, R. A. (1970). In *Physiology of Digestion and Metabolism in the Ruminant*, p. 166 [A. T. Phillipson, editor]. Newcastle upon Tyne: Oriel Press.
Hopkins, D. T., Warner, R. G. & Loosli, J. K. (1959). *J. Dairy Sci.* **42**, 1815.
Large, R. V. (1964). *Anim. Prod.* **6**, 169.
McCormick, R. J. & Stewart, W. E. (1967). *J. Dairy Sci.* **50**, 568.
Morrill, J. L., Stewart, W. E., McCormick, R. J. & Fryer, H. C. (1970). *J. Dairy Sci.* **53**, 72.
Mylrea, P. J. (1966*a*). *Res. vet. Sci.* **7**, 333.
Mylrea, P. J. (1966*b*). *Res. vet. Sci.* **7**, 394.
Raven, A. M (1970). *J. Sci. Fd Agric.* **21**, 352.
Raven, A. M. & Robinson, K. L. (1964). *J. Soc. Dairy Technol.* **17**, 5.
Reynolds, J. D. (1972). Transport and secretion of pancreatic enzymes in the sheep. BSc (Hons) Thesis, University of New South Wales, Australia.
Roy, J. H. B. (1970). *The Calf*, vol. 2, 3rd ed. London: Butterworths.
Roy, J. H. B., Shillam, K. W. G., Thompson, S. Y. & Dawson, D. A. (1961). *Br. J. Nutr.* **15**, 541.
Roy, J. H. B., Stobo, I. J. F. & Gaston, H. J. (1970). *Br. J. Nutr.* **24**, 459.
Roy, J. H. B., Stobo, I. J. F., Gaston, H. J., Shotton, S. M. & Ganderton, P. J., (1973) *Anim. Prod.* **17**, 109.
Sasaki, Y. (1968). *Jap. J. zootech. Sci.* **39**, 333.

Schingoethe, D. J., Gorrill, A. D. L., Thomas, J. W. & Yang, M. G. (1970). *Can. J. Physiol. Pharmac.* **48**, 43.
Shillam, K. W. G. & Roy, J. H. B. (1963). *Br. J. Nutr.* **17**, 171.
Siddons, R. C. (1968). *Biochem. J.* **108**, 839.
Taylor, R. B. (1962). *Res. vet. Sci.* **3**, 63.
Ternouth, J. H. (1971). Studies of the role of the abomasum and pancreas in digestion in the young calf. PhD Thesis, University of Reading.
Ternouth, J. H. & Buttle, H. L. (1973). *Br. J. Nutr.* **29**, 387.
Ternouth, J. H., Roy, J. H. B. & Siddons, R. C. (1974). *Br. J. Nutr.* **31**, 13.
Ternouth, J. H., Roy, J. H. B., Stobo, I. J. F., Ganderton, P. J., Gillies, C. M. & Shotton, S. M. (1974). *Br. J. Nutr.* **32**, 37.
Ternouth, J. H., Roy, J. H. B., Thompson, S. Y., Toothill, J., Gillies, C. M. & Edwards-Webb, J. D. (1975). *Br. J. Nutr.* **33**, 181.
Ternouth, J. H., Siddons, R. C. & Toothill, J. (1971). *Proc. Nutr. Soc.* **31**, 89A.
Thomke, S. (1963*a*). *Züchtungskunde* **35**, 215.
Thomke, S. (1963*b*). *Züchtungskunde* **35**, 265.
Toullec, R. & Mathieu, C. M. (1969). *Annls Biol. anim. Biochim. Biophys.* **9**, 139.
Wardrop, I. D. & Coombe, J. B. (1960). *J. agric. Sci., Camb.* **54**, 140.
Warner, R. G., Flatt, W. P. & Loosli, J. K. (1956). *J. agric. Fd Chem.* **4**, 788.
Zerebcov, P. I. & Seryh, M. M. (1962). *Izv. timiryazev. sel'.-khoz. Akad.* no. 4, p. **214**.

Printed in Great Britain