

**THE CONCENTRATION OF CITRIC ACID IN
MILK AND ITS INFLUENCE IN MILK
REPLACER ON GROWTH RATE OF
BUFFALO CALVES**

Ram Sharan Dass

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NATIONAL DAIRY RESEARCH INSTITUTE
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Dissertation

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BY

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R.S. Das
(Ram Sharan Dass)

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CHAPTER I

INTRODUCTION

early ages. Citric acid may help in the early development of rumen mucosa of young calves (Prasad and Arora, 1975).

Milk replacer is a good alternate for milk for feeding of calves. The ingredients of milk replacer should be such to which the calf can adapt to its digestive system for feed efficiency and without any digestive disturbances. The latter ailments may lead to higher incidence of mortality which may affect the whole economy of feeding milk replacers. Therefore, the present study was conducted to try milk substitute containing citric acid with the following objectives:-

- i) to study the changes in concentration of citric acid in buffalo milk at different stages of lactation and to evaluate the effect of milk yield on the citric acid content of milk;
- ii) to study the growth rate and economy of feeding calves, fed milk replacers containing citric acid and comparing them with milk fed calves.

CHAPTER II

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Review of literature of the present study can be described under the following sub-headings:-

- I. Citric Acid in Cow and Buffalo Milk.
- II. Factors Affecting the Citric Acid content of Milk.
- III. Different Sources of Nutrients in Milk Replacer and Their Utilization by Young Calves.

I. Citric Acid in Cow and Buffalo Milk

Henkel (1891) and Somer et al. (1918) identified citric acid as a component of milk and observed that citric acid content of milk was not constant. The citric acid content in cow and buffalo milk was estimated by various workers as given in table 1.

Citric acid level in milk

Species	Average citric acid content(%)	Range (%)	Reference
Cow	-	0.8 - 0.130	Hess (1920)
„	0.18	0.07-0.33	Sherwood and Hammer (1926)
„	0.160	0.140-0.190	Hartman and Hilling (1933)
„	0.168	0.150-0.206	Arup (1938)
„	0.255	-	Anantakrishnan <u>et al.</u> (1943)

contd.....

Species	Average citric acid content(%)	Range (%)	Reference
Cow	0.180	0.122-0.258	Storgards (1948)
"	0.254	0.215-0.290	Fabris (1952)
"	-	0.184-0.209	Paul <u>et al.</u> (1954)
"	-	0.148-0.200	El-Negoumy (1954)
"	-	0.17 -0.20	Budarina (1958)
"	0.283	0.145-0.240	Balba <u>et al.</u> (1958)
"	0.217	-	Cataland (1959)
"	-	0.48 - 1.73	Tapernoux & Magat (1961)
"	-	108 - 210 mg/ 100 ml	Davidov & Kruglova (1960)
"	192 mg/ 100 ml	103 - 241 mg/ 100 ml	Anagama & Kami (1962)
"	-	230 - 270 mg/ 100 ml	D'Agostino <u>et al.</u> (1967)
"	192.5 mg/ 100 ml	-	Ismail <u>et al.</u> (1971)
"	-	130 - 216 mg	Prasad and Arora (1975)
Buffalo	0.196	-	Anantakrishnan <u>et al.</u> (1943)
"	-	0.170-0.181	Basu <u>et al.</u> (1949)
"	-	0.162-0.267	El-Negoumy (1954)
"	0.180	-	Paul <u>et al.</u> (1954)
"	0.245 gm/ 100 ml	0.200-320 gm/ 100 ml	Balba <u>et al.</u> (1958)
"	152.9 mg/ 100 g	-	Salam & Shibry (1966)

contd.....

Species	Average citric acid content (%)	Range (%)	Reference
Buffalo	0.219	0.158-0.290	Albonico <u>et al.</u> (1967)
„	247 mg/ 100 ml	-	Albonico <u>et al.</u> (1971)
„	208.5 mg/ 100 ml	-	Ismail <u>et al.</u> (1971)
„	175 \pm 1.40	-	Sindhu & Roy (1973)

II. Factors Affecting Citric Acid Content of Milk

1. Effect of Lactation Period

Reinart et al. (1959) observed no significant change in the citric acid content of milk during the different months of lactation. A decrease in the citric acid content was observed as the lactation advanced (Davidov et al., 1961). Abd-El-Salam and Shibny (1966) found maximum citric acid content in buffalo milk in the second month, and then there was a gradual decrease, followed by increase again in values in the 7th month of lactation. Yusa et al. (1969) observed an increase in the citric acid content of milk after 4 to 5 days of parturition, which gradually decreased with advancement in lactation period. Prasad and Arora (1975) reported that citric acid content of cow milk was not constant during different months of lactation.

2. Effect of Pregnancy

Zannelli et al. (1966) studied the effect of pregnancy on the citric acid content of milk. They found a decline in the citric acid content from 22½ mg/100 ml during early pregnancy to 172 mg/100 ml after 5½ months of pregnancy. The values rose again to 241 mg/100 ml at 6½ months of pregnancy, but these were 232 mg/100 ml at 8 months of pregnancy.

3. Effect of Temperature

Rogina (1935) observed no change in the citric acid content of boiled milk. The citric acid content of milk stored at 2 - 5.5°C has been shown to increase from 233 mg/100 ml to 241 mg/100 ml after 24 hours (Anagama and Kami, 1963). They recorded a decrease in the citric acid content of milk from 228 to 210 mg/100 ml when kept at a temperature of 23.5° - 27°C for 7 hours. Citric acid content of milk rose from 238 to 242 mg/100 ml followed by a decrease after 24 hours to 217 mg/100 ml by the same group of workers.

4. Seasonal Effect

Sherwood and Hammer (1926) found negative influence of season on the citric acid content of milk. Heinemann (1944) observed that the citric acid content of milk was higher (2.09%) in March and April and least (1.88%) in the month of November. Holwerda (1954) studied the citric acid content of milk which

was more (1.73 gm/litre) during April and minimum (1.38 gm/litre) from November to January. The values were showing relatively a higher trend from May (1.560) to October (1.690). He pointed out that this change due to season was not due to the change from outdoor feeding to indoor feeding. Reinart et al. (1959) observed lesser citric acid content in cow milk during grazing season than during winter season. Kamal et al. (1960) reported that the citric acid content in the milk of cows kept at low temperature (65°F) was higher than the cows maintained at high temperature (90°F). Anagama and Kami (1962) while estimating the citric acid content of milk observed least values (137 mg/100 ml) in July to August and higher values (221 mg/100 ml) in the month of March. The citric acid was found maximum (0.264%) in cow milk in winter season and minimum (0.180%) in summer season (Tikhomerova, 1963). Battisjoti (1965) observed lower values (156 - 166 mg/100 ml) of citric acid in cow milk from December to March and higher values (186 - 204 mg/100 ml) from April to November. Fulga et al. (1974) recorded higher values (190 - 220 mg/100 ml) of citric acid in cow milk from May to July and less values (137 - 167 mg/100 ml) from August to February.

III. Different Sources of Nutrients in Milk Replacer and Their Utilization by Young Calves

1. Source of Protein in Milk Replacer

Some workers have shown the inefficient utilization of vegetable protein by calves when included in milk replacer (Williams

et al., 1950; Noller et al., 1956; Gorrill et al., 1967 and Colvin et al., 1968). Williams and Knodt (1950) reported that raw soyabeans were not satisfactory when used at a level of 40% in milk replacer. All the calves fed on milk replacer diet died between the period from 29th to 58th day of age. Stein et al. (1954) reported that 43% of the nonfat dry milk solids in milk replacer could be substituted with soyabean flour when fed with dried brewers yeast and dried skim milk. Gorrill et al. (1967) indicated that calves fed on a highly purified soyabean flour (71% crude protein) performed equally well as those kept on whole milk. When calves were fed soyabean flour (containing 50% crude protein) to furnish 60% of the dietary protein, their growth rates were poor and there was a higher incidence of diarrhoea. Growth rate was reduced when calves were fed a milk replacer containing soya protein (Antal, 1970). Similarly, growth rate of calves fed soyabean flour replacer from 7 to 10 days of age was approximately 20% less than that of the calves fed all milk replacer (Zaffira et al., 1972). Gorrill and Nicholson (1970a) found no adverse effect on growth rate of calves when 70% of the protein in their diet was from soyabean concentrate. Total intake of dry matter per kg gain to weaning was significantly less for calves given whole milk than for those given soyabean milk substitute. Sirotkin (1975) reported a reduction of 66% in the consumption of milk protein when soya milk was fed to calves.

Slade and Huber (1965) fed a diet containing three levels of fish flour. The diet contained 26% crude protein, 10% fat, 5% ash and 66% nitrogen free extract. In control diet, the source of protein was skim milk. The level of protein from fish flour was 20, 40 and 60% to that of total protein in the diet. The rate of gain was significantly less ($P < 0.1$) in the group fed diet containing 60% fish flour. All the calves died at 100% level of fish flour in milk replacer (Huber and Slade, 1967; Wendlandt et al., 1968). Genskow et al. (1969) substituted low ash fish protein concentrate for skim milk and whey at 0, 50 and 100% of total protein in the ration of calves and determined the effect of feeding fish protein on plasma free amino acid levels. Histidine and tyrosine in plasma declined markedly as the concentration of fish protein in milk replacer increased whereas methionine increased. Low availability of histidine might account for the depressed growth response to 100% fish flour in milk replacer, whereas increased plasma methionine and arginine level might indicate growth stimulation in response to 50% fish flour in replacer.

Williams and Knodt (1950) compared two diets containing two different levels of dried skim milk and observed that the diet containing 50% dried skim milk gave consistently better results than the 20% levels. Noller et al. (1956) reported that milk protein was superior to non-milk protein when used in milk replacer for young calves. Lassitier et al. (1960) and Huber and Miller (1964) fed the calves with milk replacers containing different

levels of protein and reported an increase in body weight gain with the increase of protein level in milk replacer. Lassitier et al. (1963) conducted two trials with two day old calves to study the minimum protein requirement in milk replacer to support normal growth. They indicated that calves gained normal growth when fed milk replacer ration containing 24% protein. The effect was, however, not serious on growth rate of calves when protein content of milk replacer was decreased to 19%. Harshbarger and Galwicks (1965) compared the effect of milk and three milk replacers containing different sources and varying amounts of protein on the gain in body weight on isonitrogenous basis. Average daily gain for groups 2, 3 and 4 as a per cent of group 1 were 79.0, 70.8 and 88.2% at 6 weeks of age, 92.0, 91.0 and 98.8% at 12 weeks of age. Antal (1971) reported that upto 90 days, gain in body weight was less when less milk protein was given to calves. Gorrill et al. (1973) found no difference in growth rate of calves when liquid diet was whole milk or milk replacer with milk protein.

Some workers have tried to increase the nutritive value of various proteins. Colvin and Ramsey (1968) reported that nutritive value of soyabean flour for young calves could be improved markedly by exposing the flour to an acid treatment for five hours at 37°C, prior to its inclusion in the milk replacer. Calves fed acid treated soyabean flour grew at nearly twice the rate of those receiving untreated soya flour. Results were similar with milk

replacers containing alkali treated fully cooked soyabean flour. Lister (1971) observed that when high temperature treated skim milk was added in milk replacer for calves there was no growth response to an increased protein level as compared with low temperature treated skim milk.

2. Nature of Fat in Milk Replacer

Lambert et al. (1955) demonstrated growth retardation when calves were fed with lipid free diet for three weeks. Olson and Williams (1959) studied the effect of 0, 5, 10, 20 and 30% stabilized lard in milk replacer diets of young calves. The average daily gains were found to be 0.76, 0.79, 0.85, 0.80 and 0.93 lb, respectively. Growth rate was about 9% faster in calves fed milk replacer containing 10% fat as compared to other fat levels in milk replacer (Lassitier et al., 1963). Brown et al. (1964) reported that the daily gain in body weight was 1.03, 1.09 and 1.02 lb/calf per day, respectively, with a milk replacer containing 5, 10 and 15% refined coconut oil. Kaunitz (1973) found no significant difference in feed intake and general health when monkeys were fed with milk replacer containing 3.05% coconut oil or 3.10% of soyabean oil. Cottyn et al. (1973) studied the influence of different fats and fat mixtures in milk replacer on efficiency of feed conversion. The best feed conversion efficiency 1.56 kg/kg gain was found to be with



PCB (Palm oil 15, coconut oil 25, and hydrogenated whole oil 60%).

Improved calf growth has been found when particle size of the fat added in milk replacer was reduced through homogenisation (Hodgson and Murdock, 1960; Roy et al., 1961; Warner et al., 1962), emulsifier addition (Warner et al., 1956; Hopkins et al., 1959; Roy et al., 1967; Tollec and Mathiev, 1971) or spray drying (Warner et al., 1962).

All these treatments increased the digestibility of fat. Apparent digestibility of tallow with and without emulsifier was found to be 93 and 87%, respectively (Tollec and Mathiev, 1971).

3. Source of Carbohydrates in Milk Replacer

Flipse et al. (1950) studied the nutritive value of lactose, glucose, corn syrup and starch as sources of carbohydrate in milk replacer. Calves appeared well nourished and thrifty when lactose, glucose and corn syrup were fed, but appeared unthrifty when fed on starch. Noller et al. (1956) reported that the average daily gains of the calves on the basal, whey and lactose ration were 0.87 ± 0.08 , 0.92 ± 0.05 and 0.87 ± 0.07 , respectively and lactose was not found to have any beneficial effect. Huber et al. (1963) reported that calves receiving milk replacer in which 14% of the solids were starch had inferior gains upto three weeks of age as compared to calves on control

(high lactose) or 9% starch ration. Kwißt Kawska (1970) studied the effect of milk diet containing 1.3% fat and supplemented with glucose, lactose, sucrose, or potato starch upto 20 days. Calves fed with glucose or lactose gave significantly greater gain than other groups.

4. Minerals and Vitamins in Milk Replacer

Studies of several workers indicated that the growth rate of calves improved when minerals and vitamins were added in their diet. Neathery et al. (1958) fed the calves with whole milk or milk replacer alone or with vitamin B₁₂. Performance with both types of rations was increased when vitamin B₁₂ was added, but not significantly. Supplementation of vitamin B₁₂ concentrate (APF No 3) to a plant protein calf starter was without effect on the growth of calves to 90 days of age (Russof et al., 1951). Tardani and Dee Monte (1967) studied the effect of supplementation of graded amount of pyridoxine in the milk substitute on the growth of Friesian calves. The milk substitute was having 1.87 mg pyridoxine per kg. Supplements of pyridoxine for three groups were 6, 10 or 20 gm/100 kg body weight. Average daily gains were 724, 746, 736 and 716 gm and intake of DM/kg gain were 2.04, 1.96, 2.09 and 2.15 kg. There was no effect on weight gain or feed efficiency when pyridoxine was increased from 2.4 to 5.5 mg per kg milk substitute between 60 to 90 kg live weight (Krichgesnar et al.,

1965). Genskow et al. (1968) fed the calves with a milk replacer containing low ash defatted fish meal as the only source of protein along with lactose, dextrose, emulsified lard, salt, minerals, vitamin A and D and chlorotetracycline. Blood level indicated that diet was deficient in vitamins E, A, D and B complex. Supplementation of these vitamins improved weight gain and prevented deaths. Makdani et al. (1970) fed the calves with dichloroethane extracted fish protein concentrate (DCE-FPC) with 0, 46 and 92 mg/kg added vitamin E. Growth responses improved as vitamin E in the DCE-FPC ration increased, while extensive muscular lesions were observed in calves fed no supplemental vitamin E.

Wilson (1964) compared the body weight gain of calves fed with a mixture of 15 mg sodium selenite and 200 mg CuSO_4 , with the group given no trace element. Average daily weight gain in the next 38 days were 0.81 and 1.09 lb, respectively. Supplementation of CaHPO_4 or CaCO_3 and trace element to a milk replacer given to calves prevented signs of deficiency which were seen when the supplements were not given (Urbanyl et al., 1970). Felsman et al. (1973) conducted two experiments to study the effect of four levels of copper sulphate and two levels of chlorotetracycline on the performance of calves. The levels of dietary copper sulphate were 125, 250 and 550 ppm in experiment I, while the levels were 0, 300, 600 and 900 in experiment II.

Each of the copper level was fed with and without 22 mg chlorotetracycline/kg feed. Results indicated a significant ($P \leq 0.05$) increase in average daily gain due to chlorotetracycline. But increase in average daily gain with copper sulphate was only in experiment II.

5. Role of Antibiotics in Milk Replacer

Several workers have shown an improved performance in growth and reduced incidence of diarrhoea when fed antibiotics (Bartley et al., 1951; Rusoff, 1951; Voelker and Cason, 1951; Reed and Velu, 1956; Murdock and Hodgson, 1961; Mudgal and Ray, 1965; Tangi and Adan, 1964; Jousselein et al., 1965 and Ramsey and Witassec, 1972). Supplementation of antibiotics to a milk substitute had greatest growth promoting effect during the first four weeks of replacer feeding (Bloom and Knodt, 1952). Magruder et al. (1960) reported that feed efficiency was 41% greater in antibiotic supplemented group than control.

6. Effect of Enzyme Supplementation in Milk Replacer

Williams and Knodt (1951) reported that addition of papain powder and pancreatic powder to the milk replacer reduced the growth rate and feed consumption. Fries et al. (1958) conducted an experiment to study the effect of supplementing milk replacer with enzymes on the growth of calves. Milk

replacer was predigested with malt diastase, papain and a combination of these two enzymes. There was no significant improvement in growth or feed consumption with any of the treatments. Lassitier et al. (1959) reported that 0.5% pepsin depressed slightly the rate of growth of all calves.

7. Digestibility of nutrients in Milk Replacer

A number of workers (Noller et al., 1956; Raven and Robinson, 1960; Radostitis and Bell, 1969; Gorril and Nicholson, 1970 and Matre, 1973) compared the digestibility of nutrients of milk and various milk replacers fed to calves. Noller et al. (1956) reported that the mean coefficients of apparent digestion of ram whole milk were DM = 94.8, CP = 90.1, EE = 97.8 and NFE = 97.1%. The average apparent dry matter digestibility increased from 25% in 10 to 14 days old calves to an average of 75.4% in 26 to 38 days old calves. The digestibility of all the nutrients in milk replacer was low at 6 days of age but increased consistently from 6 days onwards to 24th day (Raven and Robinson (1960) and Kzenov (1960) reported that the digestibility of dry matter in whole milk was 25% at 6 days of age and 75.4% at 24 days of age. Matre (1973) indicated that milk replacer was not as well digested as in whole milk.

8. Growth Responses of Calves Fed Milk

Young (1953) reported that calves fed whole milk gained 1.04 and 1.17 lb/day with milk replacer. There was no significant difference in growth between calves fed whole milk and those fed milk replacer.

There was no significant difference in weight gain or intake of DM/kg gain when calves were fed with milk or milk substitute (Krisgard et al., 1964). Randel (1968) observed that growth rate was higher in the group given whole milk at least to 3 weeks of age, but after that there was no difference in body weight gain of calves fed with whole milk or milk replacer. More consistent gain in body weight was observed with milk replacer when whole milk feeding was discontinued at 17 days of age rather than at 10 days of age (Bush et al., 1968). Radcliff and White (1970) reported that whole milk feeding was superior in terms of weight gains as compared to milk replacer feeding. There was no significant difference on weight gains of calves fed milk or milk replacer (Gorril and Nicholson, 1970b). Press and Krolizec (1971) reported that calves given milk substitute gained 13% more than those given whole milk. Ozaplak et al. (1975) reported similar gains in calves fed milk and milk substitute in which milk fat was entirely replaced by rapeseed oil and casein was partially replaced by whey/albumin and which contained starch/sugar.

A number of workers (Burt, 1966; Ackerman et al., 1969; Perks et al., 1968; Khouri, 1969; Willet et al., 1969; Wilson, 1969; Davis and Woodward, 1970; Nicotov, 1973 and Fowler et al., 1974) compared once a day versus twice a day feeding of milk replacer to dairy calves. They reported that there was no significant difference in average daily gain, heart girth, wither

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height gain or consumption of milk replacer. Once a day feeding as compared to twice a day feeding did not cause any increase in number of digestive disorders.

CHAPTER III

EXPERIMENT I

EXPERIMENT I

The objective of this experiment was to study the variations in citric acid content of buffalo milk during the different stages of lactation and to find the effect of milk yield on citric acid level in milk. This experiment will help in computing milk replacer diets for buffalo calves from birth to three months of age.

MATERIALS AND METHODS

Selection of the animals

Six newly calved buffaloes were selected and were fed standard farm ration. Colostrum samples were taken on the day of calving from these animals and milk samples were taken at one week intervals. Milk yield of each animal was also recorded at the time of sampling. The concentrations of citric acid in buffalo milk were estimated according to the method of Marrier and Boulet (1958) with slight modifications. One ml of colostrum as well as milk was diluted 10 times with 10% trichloroacetic acid to precipitate the protein which was removed by centrifugation at 2000 rpm for 10 minutes. Deproteinisation was essential to avoid precipitate formation after colour development with pyridine and acetic anhydride according to the original method. Corresponding blank contained the same amount of 10% trichloroacetic acid. Samples after colour development were read at 420 mμ in spectronic-20.

RESULTS

The data for citric acid concentration in milk and milk yield on the day of sampling is given in appendix I.

The citric acid content in milk was maximum (219.1 mg/100 ml) in the second month of lactation and there after there was a continuous decrease upto 7 months of lactation as shown in table 1 (Fig. 1). The overall mean citric acid content was 180.9 mg/100 ml of milk in 7 months. Statistically, analysis of data (table 2) indicated that the effect of different months of lactation on the citric acid level in buffalo milk was significant ($P < 0.01$). The variation in citric acid content from animal to animal was also significant. It was also observed that milk yield has some effect on the citric acid content of milk.

DISCUSSION

Several factors have been affecting the citric acid content in milk. Davidov and Kruglova (1960) reported that citric acid concentration in milk was highest in winter (210 mg/100 ml) and least in summer (208 mg/100 ml). Anagama and Kami (1962) observed the same pattern of citric acid in milk and suggested that lower values of citric acid in summer could be due to the high environmental temperature. Anagama and Kami (1963) found a decrease in concentration of citric acid in milk

Table 1

Least square means for the concentration of citric acid at different months of lactation

Months of lactation	Citric acid concentration mg/100 ml	SE
1	187.4	± 8.52
2	219.4	± 9.02
3	209.0	± 7.63
4	193.3	± 7.84
5	166.0	± 9.62
6	156.2	± 7.06
7	135.0	± 6.22

when it was exposed to sunlight for 5 - 30 minutes. A slight increase in the citric acid concentration at mid day than in the morning and a similar increase in the first milk than in the stripping was also noted by Anagama and Kami (1964). Thus it is clear that concentration of citric acid in milk changes due to the interaction of different factors.

The results in this experiment showed that concentration of citric acid in buffalo milk goes on decreasing with the advancement of lactation and these results are in accordance with the findings of Kamal et al. (1960); Davidov et al. (1961) and Yusa et al. (1969). The higher value of citric acid in buffalo milk in second month of lactation found in this study was comparable to the findings of Salam and Shibny (1966). But in this study, no increase in citric acid concentration in the 7th month of lactation was observed as reported by them. It might be due to low milk yield in this month as milk yield was found to have positive correlation with citric acid in this study.

As the main purpose of this experiment was to study the changes in concentration of citric acid in milk, so that similar amount of citric acid can be added in milk replacer diet of buffalo calves. This study revealed that citric acid level in the milk of 6 buffaloes ranged from 135.0 mg to 219.1 mg/100 ml and the milk yield ranged from 1.5 kg to 6.8 kg. On DM basis,

Table 2

Analysis of variance - citric acid

Source of Variation	DF	SS	MS	F
Between animals	5	78724.3	15744.86	17.9
Between months	6	157814.5	26302.41	30.05**
Error	156	136530.1	875.2	
Total	167	373068.9		

** P / 0.01

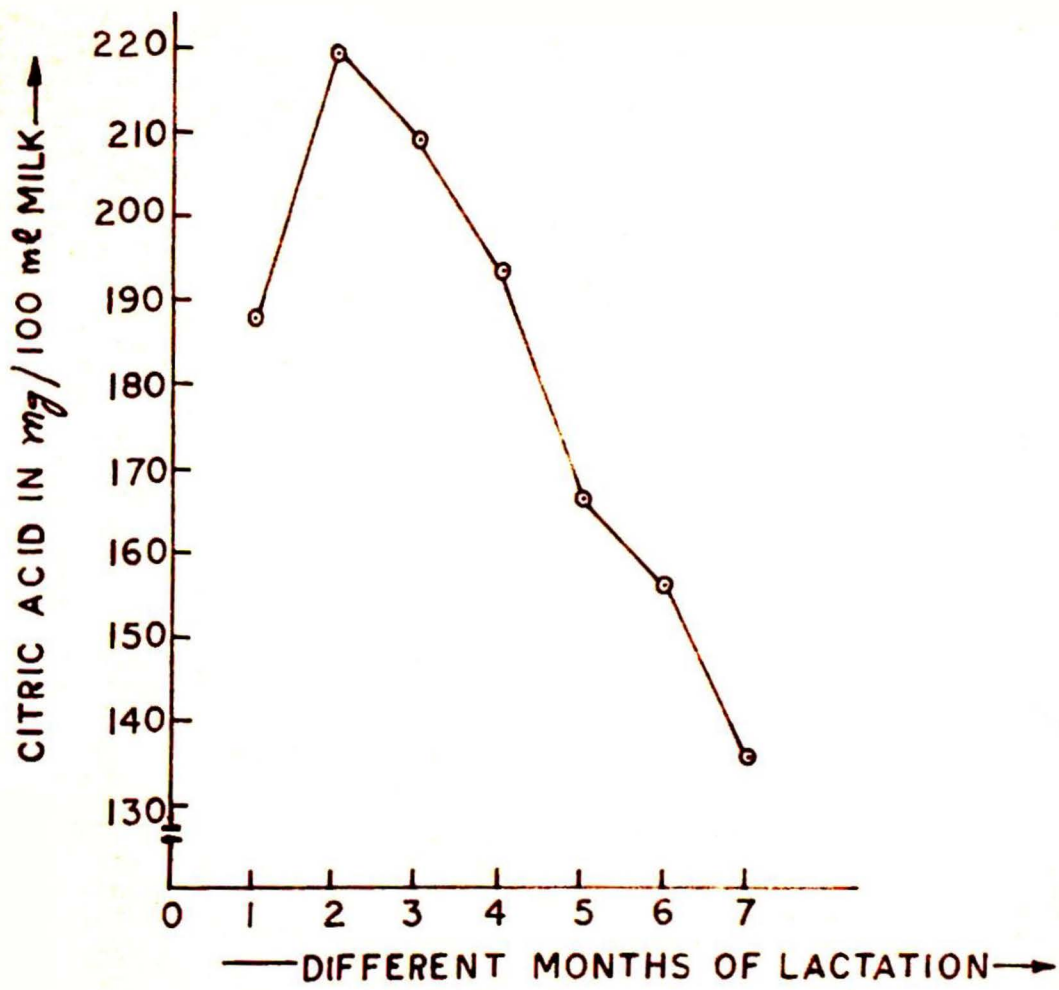


FIG. I. CITRIC ACID IN MILK IN DIFFERENT MONTHS OF LACTATION.

citric acid would be in the range of 0.9 - 1.46% showing an average of 1.36% in first three months of lactation, but average citric acid content in cow milk on dry matter basis was found to be 1.9% (Prasad, 1974). The difference in citric acid concentration in buffalo and cow milk was in accordance with the earlier findings of various workers (Ananthkrishnan et al., 1943; Ismail et al., 1971).

Conclusion: The citric acid in milk substitute needs to be mixed at the rate of 1.36% in case of buffalo calves.

EXPERIMENT II

The purpose of this experiment was to compare the effect of feeding milk and milk replacer on the growth rate of buffalo calves.

Selection and Distribution of Calves

Fourteen buffalo calves were weaned at birth from their dams for the purpose of this study. Two calves died during the experiment and were excluded from the final statistical analysis of the data. Rest of the 12 calves were divided randomly on the basis of body weight into two groups of 6 each, one served as a control group I and the other as an experimental group II. Description of the calves is given in table 3.

Table 3

Sr.No.	Group I (control)		Group II (experimental)	
	Number	Birth weight (kg)	Number	Birth weight (kg)
1.	784	24.5	1167	24.0
2.	791	35.5	1199	35.0
3.	801	31.0	1188	30.0
4.	821	36.5	1155	37.5
5.	830	35.0	1173	35.0
6.	845	36.5	1175	37.5
	Mean:	33.2	Mean:	33.2

The average birth weights of calves were 33.2 kg in both the groups. The weight of all the weaned buffalo calves were taken before feeding colostrum.

Housing and Management of the Calves

All the calves were housed in a well ventilated byre so constructed as to keep them comfortable. Calves were kept in a wooden cage. The floor was washed daily with tap water and then phenyl was sprayed. The walls of byre and wooden pens were white washed weekly. They were managed for preventive treatment as shown in table 4.

Feeding Schedule of Calves

Calves were fed 2 kg colostrum per day per calf for the first four days of age when the birth weight of the calf was upto 35 kg. Additional half kg colostrum was given for every 5 kg increase in birth weight. After the colostrum period, calves in both the groups were given dam milk upto the age of 14 days. Milk was reduced gradually from fifth day onwards upto 9 days of age, in order to avoid the incidence of diarrhoea. This decrease was followed by a gradual increase upto 14 days of age.

Table 4Package of Practices in Buffalo Calves

Age (Days)	Treatments 1, 2, 3,	Preventive against
1	Orally Auromycin nutritional formula 2 spoonful	Calf Scour
1	Sealing naval vessels	Naval Ill
2	Vitamin A concentrate (1 ml vitablend)	Night blindness
3	Piperazine adipate, 2 spoonful Enterovioform 1 tablet followed by 1 Oz liq. paraffin after 6 hours.	Ascariasis and Dysentery.
4	a. Buff. antiserum 50 ml s/c b. Strinacin 1/2 to 1 tablet	To raise antibody titre in blood Diarrhoea/Calf Scour
7	As on day 3	Ascariases and Dysentery.
8	Sulmet course for 4 days	Coccidiosis.

1 Mineral supplement daily

2 TM-5 or Aurofac daily

3 Rovimix in oil once a week (10,000 i.u. of vitamin A)

The calves in control group were given whole milk, heated and cooled to body temperature upto the age of 15 weeks according to the schedule given in table 5.

Table 5

Body weight	Milk (kg)	
	Morning	Evening
Upto 35 kg	1.0	1.0
36 to 40 kg	1.5	1.0
41 to 45 kg	1.5	1.5
46 to 50 kg	2.0	1.5
51 to 55 kg	2.0	2.0
56 to 60 kg	2.5	2.0
61 kg and above	2.5	2.5

As mentioned earlier feeding schedule of experimental calves was same as in the group I upto the age of 14 days. On the fifteenth day, calves were fed milk replacer along with milk. There after milk replacer quantity was increased with the gradual decrease in the whole milk as per the feeding schedule given in table 6.

Table 6

Age (Days)	Milk	Milk replacer (gm)
0-14	Milk as per control calves	-
15-18	„ „ „	50
19-22	„ „ „ -0.5 kg	100
23-26	„ „ „ -1.0 kg	200
27-30	„ „ „ -1.5 kg	300
31-34	„ „ „ -2.0 kg	400
Body weight (kg)		
40	Minimum one kg milk	500
45	„ „	550
50	„ „	600
55	„ „	650
60	„ „	700
65	„ „	750
70	„ „	800
75	„ „	850
80	„ „	900
85	„ „	950
90	„ „	1000

Calves were provided a minimum of 1.5 kg milk upto 2 months of age. After that only 1.0 kg milk was given upto 15 weeks of age.

The milk replacer given to the calves was of the composition as given in table 7.

Table 7

Wheat flour	8 kg
Fish meal	12 kg
Linseed meal	40 kg
Coconut oil	6 kg
Linseed oil	6 kg
Molasses	6 kg
Citric acid	1.5 kg
Butyric acid	0.660 litre
Aurofae	300 gms
Rovimix	15 gms
Mineral mixture	3.0 kg

Three kg mineral mixture added in milk replacer was of the following composition.

Dicalcium phosphate	1.65 kg
Chalk	0.3312 kg
Sodium chloride	0.9000 kg
Ferrous sulphate	0.0150 kg
Copper sulphate	0.0021 kg
Cobalt chloride	0.0015 kg
Potassium iodide	0.0003 kg
Sodium flouride	0.0003 kg
Zinc sulphate	0.0075 kg
Manganese oxide	0.0021 kg

Weighing of Calves

All the calves were weighed at weekly intervals. To compare growth rate and feed efficiency, daily feed intake and weekly gains were recorded for all the animals upto 15 weeks of age.

RESULTS

The results of the effect of feeding milk replacer containing citric acid to buffalo calves on the growth rate, efficiency of feed conversion and economics of feeding milk replacer are presented.

The data for growth at weekly intervals, average initial body weights, average final weights and gain in weight between two groups are given in table 8. The data for individual calf is given in appendix II. The mean birth weights of calves in both the groups were 33.2 kg. The average final weight after 15 weeks of age were 76.5 and 63.1 kg in groups I and II, respectively. Average gain in weight during the same period were 44.3 kg in group I and 30.25 kg in group II.

From Fig. 2, it is apparent that growth curve for absolute weight gain per week was superior in calves fed milk replacer diet upto 10 weeks of age. But after that absolute weight gain

Table 8
Showing gain in body weight

Calf number	Birth weight (kg)	Age (weeks)															Weight gain
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	
<u>Control group</u>																	
784	24.5	25.8	28.0	29.0	31.0	33.5	32.5	38.0	39.0	44.0	45.0	48.5	54.0	57.0	58.0	66.0	41.5
791	35.5	35.5	30.5	30.5	32.5	34.9	36.1	38.0	40.0	42.5	43.0	46.5	51.0	54.5	58.0	64.5	29.0
801	31.0	30.0	32.0	34.5	34.8	35.5	38.0	39.0	41.4	45.0	50.5	55.5	60.5	62.0	69.0	75.4	45.5
821	36.5	35.8	35.0	35.0	34.5	36.7	39.4	42.5	45.5	48.5	50.5	55.5	60.8	65.0	71.5	76.5	40.0
830	35.0	31.8	33.5	34.5	37.5	39.5	44.0	47.5	50.0	51.0	57.5	64.0	67.3	71.5	79.5	84.5	52.5
845	36.0	35.5	36.5	38.0	40.0	41.0	45.0	48.5	52.0	57.0	62.0	68.5	73.0	79.5	86.0	92.0	56.5
Average	33.2															76.5	44.3
<u>Experimental group</u>																	
1155	37.5	35.8	36.0	38.5	39.0	39.5	40.5	43.0	44.5	44.5	46.0	49.0	53.5	55.0	56.0	58.0	20.5
1167	24.0	28.0	32.3	34.0	36.0	38.5	41.0	44.0	45.0	46.9	48.0	50.5	51.2	51.6	54.4	57.0	33.0
1173	35.0	38.5	41.5	44.5	47.0	48.5	54.5	55.4	59.7	60.0	61.1	62.5	66.2	62.4	70.0	71.0	36.0
1175	37.5	37.5	40.0	40.5	43.0	43.4	49.2	53.4	54.4	59.9	59.5	61.7	63.5	65.8	70.5	71.8	34.3
1188	30.0	31.6	33.0	36.5	38.8	41.0	41.0	43.5	46.2	47.5	48.7	48.5	48.7	52.0	53.1	56.0	26.0
1199	35.0	36.2	37.5	40.0	43.4	46.5	46.0	48.3	51.6	56.0	56.4	57.4	56.5	61.0	63.0	65.0	30.0
Average	33.2															63.1	30.25

Table 9

The absolute and relative growth rate of calves
fed on whole milk diet for 15 weeks

Age in weeks	Weight (kg) m	Increase m (gm)	Growth rate per day (gm)	Natural log of weight log m/100	Increase in log m	Relative growth rate per cent per day
0	33.2	- 800	-114	5.8060	-0.0244	-0.34
1	32.4	200	28	5.7816	0.0062	0.09
2	32.6	1000	143	5.7878	0.0302	0.43
3	33.6	1400	200	5.8180	0.0410	0.59
4	35.0	1900	271	5.8590	0.0528	0.77
5	36.9	2200	314	5.9118	0.0580	0.85
6	39.1	3100	443	5.9698	0.0762	1.13
7	42.2	2400	343	6.0460	0.0553	0.79
8	44.6	3400	486	6.1013	0.0735	1.08
9	48.0	3400	486	6.1748	0.0686	1.00
10	51.4	5900	714	6.2434	0.0928	1.40
11	56.4	4700	671	6.3362	0.0799	1.10
12	61.1	3800	543	6.4261	0.0603	0.90
13	64.9	5400	771	6.4764	0.0802	1.19
14	70.3	6200	886	6.5566	0.0845	1.26
15	76.5	-	-	6.6411	-	-
Average			427			0.81

Table 10

The absolute and relative growth rate of calves
fed on milk replacer for 15 weeks

Age in weeks	Weight (kg) m	Increase m (gm)	Growth rate per day (gm)	Natural log of weight log m/100	Increase in log m	Relative growth rate per cent per day
0	33.2	1400	200	5.8060	0.0415	0.60
1	34.6	2100	300	5.8475	0.0590	0.86
2	36.7	2300	329	5.9065	0.0608	0.89
3	39.0	2200	314	5.9673	0.0548	0.80
4	41.2	1700	243	6.0221	0.0405	0.59
5	42.9	2500	357	6.0626	0.0567	0.81
6	45.4	2500	357	6.1193	0.0534	0.79
7	47.9	2300	329	6.1727	0.0470	0.69
8	50.2	2300	329	6.2197	0.0449	0.65
9	52.5	800	114	6.2646	0.0149	0.22
10	53.3	1600	229	6.2795	0.0297	0.42
11	54.9	1700	243	6.3092	0.0304	0.44
12	56.6	2000	286	6.3396	0.0348	0.50
13	58.6	2500	357	6.3744	0.0417	0.60
14	61.1	2000	286	6.4161	0.0323	0.47
15	63.1	-	-	6.4484	-	-
Average			280			0.62

Table 11

Representing average growth rate, feed conversion efficiency and economics of feeding milk replacer.

Group number	Number of animals	Average number of days fed	Average gain in weight (kg)	Average growth rate per day (kg)	Mean total milk consumed (kg)	Mean milk replacer consumed (kg)	Mean total dry matter consumed (kg)	Feed gain ratio (kg)	Cost of milk @ Rs.2.20 per kg	Cost of milk replacer @ Rs.3.00 per kg	Total cost (Rs.)	Cost per kg gain (Rs.)
I	6	105	44.30	0.422	327.7	-	52.4	1.18	720.54	-	720.54	16.27
II	6	105	30.25	0.288	157.9	53.8	73.68	2.43	347.38	161.40	508.78	16.83

per week was higher in calves fed whole milk and growth curve of calves fed milk replacer could not keep pace with that of control group. The relative growth rate per week was estimated to be 0.81 and 0.62% per day in groups I and II, respectively as shown in tables 9 and 10. The average gain in body weight per day in calves was 0.441 kg in group I and 0.288 kg in group II (Table 11). The average calculated 'b' values were 2.90 for group I and 2.00 for group II as shown in table 12 ($P < 0.01$).

Table 12

Regression coefficients i.e. 'b' values of growth at weekly interval

Number of Animals		'b' value	
Group I	Group II	Group I	Group II
784	1155	2.64	1.51
791	1167	1.96	2.01
801	1173	2.91	2.36
821	1175	2.63	2.46
830	1188	3.40	1.64
845	1199	3.86	2.03
Average:		2.90	2.0

Calculated "t" value 4.066** ($P < 0.01$)



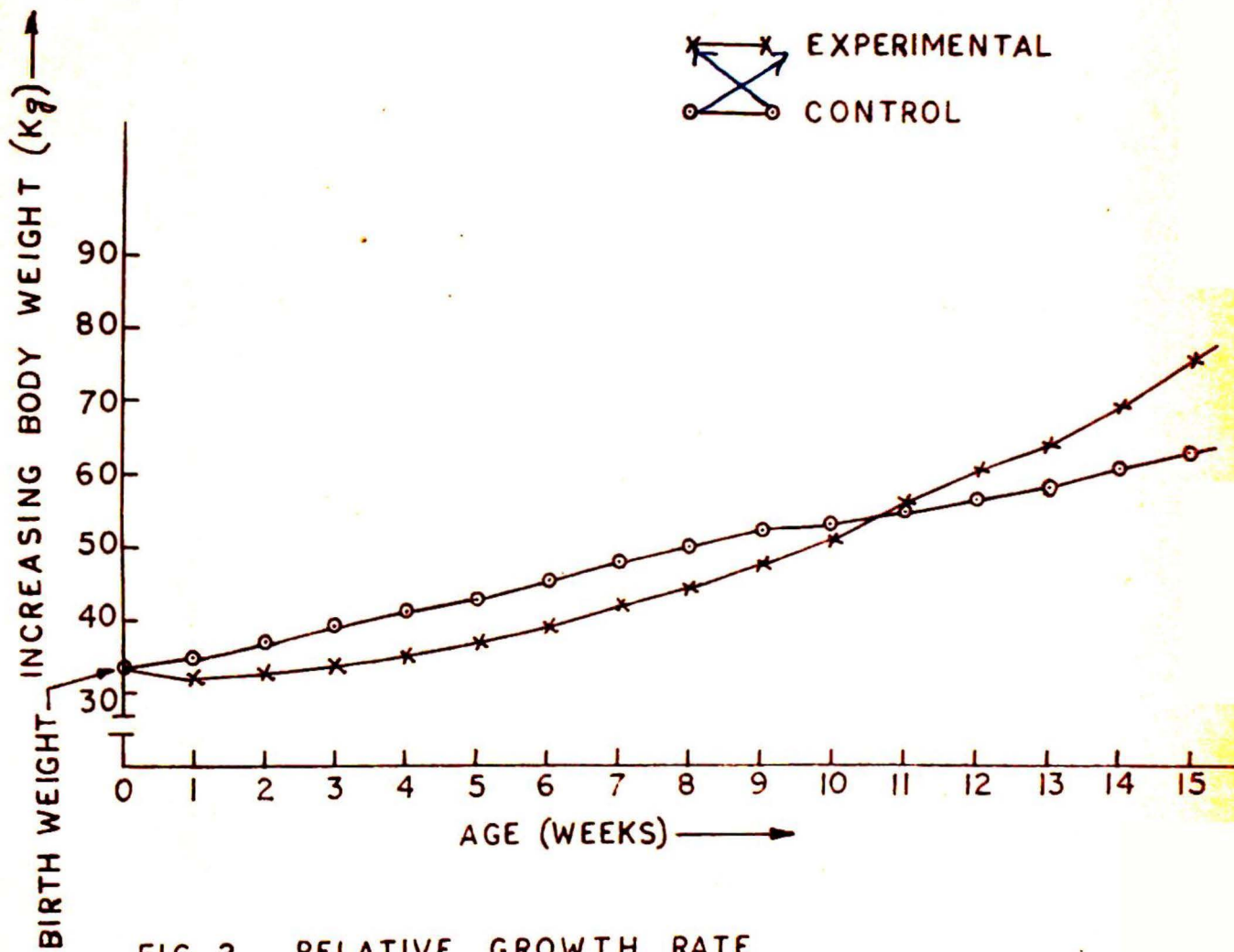


FIG. 2. RELATIVE GROWTH RATE

The dry matter intake was more (73.68 kg) in group II as compared to group I (52.4 kg), although milk consumption was 51.5% less in group I than group II. The feed:gain ratios were estimated to be 1.18 and 2.43 kg in groups I and II, respectively (Table 11). Total average cost of rearing the calves upto 15 weeks of age was estimated to be Rs.720.54 on whole milk and Rs.^{508.78}~~357.38~~ on milk replacer diet when the cost was Rs.2.20 per litre for milk and Rs.3.00 per kg for milk replacer. The cost per kg gain was found to be Rs.16.27 and Rs.16.83 in groups I and II, respectively.

DISCUSSION

Growth is such a phenomenon which can be defined in various ways. According to Mitchell (1962), animal growth is summation of those co-ordinated biological and chemical processes that are initiated with the fertilization of ovum and are terminated with the attainment of the body size, conformation, physiological capabilities, characteristics of the species and the hereditary background of the individual. Growth is characterised by the deposition of protein and fat which is supposed to be an integral part of all the cells. Growth of an animal depends not only upon the nature of diet, but other factors like environmental, managerial and hormonal etc. But when all other factors are

controlled, it is only the nutrition which might affect the growth rate. Animal growth is divided into two phases.

(i) Self accelerating phase in which both the absolute and relative growth rates are linear per unit time. In this phase instantaneous rate of growth, dw/dt depends on the growth already attained $\frac{dw}{dt} = kw$.

(ii) Self inhibitory phase: This phase follows the self accelerating phase and starts only after puberty. In this phase relative growth is not proportional to initial body weight.

This study was concerned only with the self accelerating phase of the calves.

The results obtained in this study showed that calves fed milk replacer grew at a slower rate than those fed whole milk. These results were supported by the earlier findings of Eskedal et al., 1957; Murley et al., 1957; Neathery et al., 1973; Bakshi, 1974 and Arora et al., 1975.

Some workers have reported the higher gain in body weight when calves were fed milk replacer as compared to whole milk (Young, 1953; Darwish et al., 1968 and Press and Kroliczec, 1971), while others found no difference in body weight gain when calves were fed whole milk or milk substitute (Krisgard et al., 1964; Gorrill and Nicholson, 1970b; Paliev et al., 1973).

The slow rate of gain in calves fed milk replacer in this study might indicate the less biological value of crude protein, fat, NFE and dry matter than in calves fed whole milk which was in agreement with the earlier reports of various workers. Noller et al. (1956) observed less retention of N_2 , calcium and phosphorus when calves were fed milk replacer as compared to milk. The assimilation of N_2 was poor in calves provided milk replacer than the calves fed whole milk (Klzeonov, 1965). Matre (1973) reported that fat in milk replacer was not as well digested as in whole milk. The digestibility of dry matter in milk replacer diet was significantly less as compared to whole milk diet (Noller et al., 1956; Raven and Robinson, 1960; Klzeonov, 1965).

But the above mentioned results were not in accordance with the work of Gorrill and Nicholson (1970a) who found no difference in nutrient utilization of calves fed milk or milk substitute. The retention of N_2 was relatively higher in calves fed milk replacer as compared to whole milk (Press and Kroliczec, 1971). So it may not be possible to reconcile the contradictory findings in different reports, although milk replacer has been found to be inferior to whole milk for growing calves by most of the workers.

This milk replacer contained coconut and linseed oil as a source of fat. Coconut oil was used because it has got more saturated

fatty acids than unsaturated fatty acids, while linseed oil provided the linoleic acid along with some other unsaturated fatty acids. Coconut oil has been reported to be 66% digestible when fed to calves, while on homogenisation it is 90% digestible (Hopkin, 1969). Linoleic acid, provided by the linseed oil helped in the proper development of hair on the shining coat of calves (Eskedal et al., 1957). Brown et al. (1964) reported that optimum level of coconut oil in milk replacer was 10 per cent. Poor growth found in this experiment in calves fed milk replacer might be due to high level of linseed oil which is very rich in unsaturated fatty acids. Probably calves could not utilize these acids and showed the high incidence of loose faeces. It was possible that high citric acid (1.5%) mixed in milk replacer might not be useful in relation to energy and might have been a depressing factor which needs to be elucidated further.

CHAPTER V

SUMMARY AND CONCLUSIONS

SUMMARY AND CONCLUSIONS

The studies were conducted in experiment I to estimate the citric acid content of buffalo milk in different months of lactation and to evaluate the effect of milk yield on the citric acid levels. The other experiment was undertaken with a view to study the effect of feeding milk replacer containing citric acid on the growth rate, feed conversion efficiency and its economics.

Six newly calved buffaloes were taken in experiment I and were fed on standard farm ration. Milk samples were obtained and analysed at an interval of one week upto 7 months of lactation. It was found that citric acid content goes on decreasing with the progress in lactation. It was maximum (219.1 mg/100 ml milk) in second month and minimum (135.0 mg/100 ml milk) in 7th month of lactation. Milk yield was found to have positive correlation with citric acid level of milk.

Fourteen birth weaned buffalo calves were taken for second experiment and were divided into two groups. Calves in group I were fed whole milk whereas calves in group II were fed milk replacer upto 15 weeks of age. Two calves died during the experimental period and their data were excluded.

Average gainⁱⁿ body weights in 15 weeks of age were 44.30 and 30.25 kg in groups I and II, respectively. The gain was appreciably higher in calves fed milk than milk replacer group.

Absolute growth was superior upto 10 weeks of age in calves fed milk replacer but after that growth was better in calves fed milk, upto 15 weeks of age. Fifty one per cent reduction in the consumption of milk was practised in calves fed milk replacer. The total milk consumed was 327.7 and 157.7 litres in groups I and II, respectively. The dry matter intake was 52.4 kg in group I and 73.68 kg in group II. Feed:gain ratio was 1.18 in group I and 2.43 kg in group II.

Total cost of rearing the calves upto 15 weeks of age was Rs.720.54 and Rs.508.78 in groups I and II, respectively. But the cost per kg gain was more or less the same.

Conclusions:

Performance of calves fed milk replacer was not optimum. Poor gain in body weight might be due to higher level of exogenous unsaturated fatty acids and lesser level of saturated fatty acids. Citric acid was added 1.5% in this milk replacer on DM basis which is slightly higher than the level of 1.36%, observed in this study. So to obtain the better growth rate in buffalo calves, this milk replacer should be modified and improved further.

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sampling upto 28 weeks of lactation.

Animal No.	11		12		13		14		15		16	
	a	b	a	b	a	b	a	b	a	b	a	b
5	1.8	230	3.0	224	2.5	240	2.5	246	3.3	128	3.2	118
45	3.2	168	3.3	186	4.2	178	4.3	186	1.5	186	2.0	118
47	3.5	276	4.2	298	4.0	284	3.8	314	5.2	216	4.5	244
69	3.0	216	3.8	216	3.5	208	3.4	194	3.4	194	3.2	186
132	1.5	186	3.5	178	4.0	170	3.5	140	3.4	132	3.2	132
262	2.8	208	3.5	216	3.2	202	3.2	194	3.5	202	3.5	178

APPENDIX I (contd...)

Representing citric acid content in mg/100 ml of milk and milk yield on the day of sampling upto 28 weeks of lactation.

Animal No.	Weeks																							
	17		18		19		20		21		22		23		24		25		26		27		28	
	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b
5	3.2	186	3.4	186	2.8	194	2.5	186	3.5	194	3.5	194	3.2	208	3.3	202	2.7	224	3.0	178	3.2	145	2.9	138
45	2.5	186	2.4	178	5.0	140	4.8	148	3.8	186	3.5	178	3.3	178	3.8	118	4.3	156	3.6	164	3.5	140	3.3	132
47	6.0	202	6.8	186	6.8	178	6.0	170	4.7	170	6.0	162	6.0	224	5.2	140	4.7	140	4.6	148	4.5	140	4.3	132
69	3.5	178	4.0	170	4.0	178	3.9	162	3.8	148	3.7	156	3.9	156	3.5	118	3.0	126	3.0	120	2.5	108	2.4	118
132	4.0	120	3.7	140	3.5	140	3.2	132	2.5	108	3.2	120	2.8	88	3.0	132	3.0	104	3.1	102	2.8	102	2.0	101
262	4.0	162	4.0	156	4.0	156	3.8	148	3.7	156	3.5	148	4.0	156	3.5	118	3.5	128	3.0	98	3.0	140	3.0	132

a = Milk yield on the day of sampling.

b = Citric acid content in mg/100 ml of milk.

APPENDIX II

Representing the data for total milk, milk replacer and dry matter consumed, feed:gain ratio and cost per kg gain in weight of individual animal.

Calf number	Birth weight (kg)	Milk consumed (kg)	Dry matter in milk (kg)	Cost @ Rs.2.20 per kg	Milk replacer consumed (kg)	Dry matter in milk replacer (kg)	Cost @ Rs.3.00 per kg	Total dry matter consumed (kg)	Total cost (Rs.)	Final weight	Gain in weight (kg)	Rate of gain per day (kg)	Feed gain ratio (kg)	Cost per kg gain (Rs.)
794	24.5	290.5	46.48	639.10	-	-	-	46.48	639.10	66.0	41.5	0.395	1.12	15.4
791	35.5	303.5	48.56	667.70	-	-	-	48.56	667.70	64.5	29.0	0.276	1.67	23.2
801	31.0	328.0	52.48	721.60	-	-	-	52.48	721.60	75.4	45.5	0.433	1.15	15.8
821	36.5	329.5	52.72	724.90	-	-	-	52.72	724.90	76.5	40.7	0.388	1.29	17.7
830	35.0	359.0	57.44	789.80	-	-	-	57.44	789.80	84.5	52.7	0.502	1.09	14.09
845	36.0	355.5	56.88	782.10	-	-	-	56.88	782.10	92.0	56.5	0.538	1.01	13.84
1155	37.5	160.5	25.68	353.10	55.600	50.04	166.80	75.72	519.90	59.7	22.2	0.211	3.72	23.42
1167	24.0	167.75	26.84	369.05	40.450	36.40	121.35	63.24	490.40	57.0	33.0	0.314	1.92	14.88
1173	35.0	162.5	26.00	357.50	57.750	51.97	173.25	77.97	530.75	71.0	36.0	0.343	2.16	14.74
1175	37.5	152.5	24.40	335.50	59.150	53.23	159.69	77.63	495.19	71.8	34.3	0.327	1.29	13.38
1188	30.0	150.0	24.00	330.00	53.500	48.15	144.45	72.15	444.45	56.0	26.0	0.348	2.79	17.09
1199	35.0	146.0	23.36	321.80	56.300	50.67	152.01	74.03	473.21	65.0	30.0	0.286	2.46	15.67

VERIFIED
Manager
Signature

