

قسم : الصحة ومراقبة الأغذية - كلية الطب البيطرى - جامعة أسيوط.

رئيس القسم : أ.د / على يوسف لطفى

التقييم النوعى والكمى لجزئيات كازين اللبن

لأبقا والفريزيان والجاموس المصرى بطريقة الفصل الكهربائى

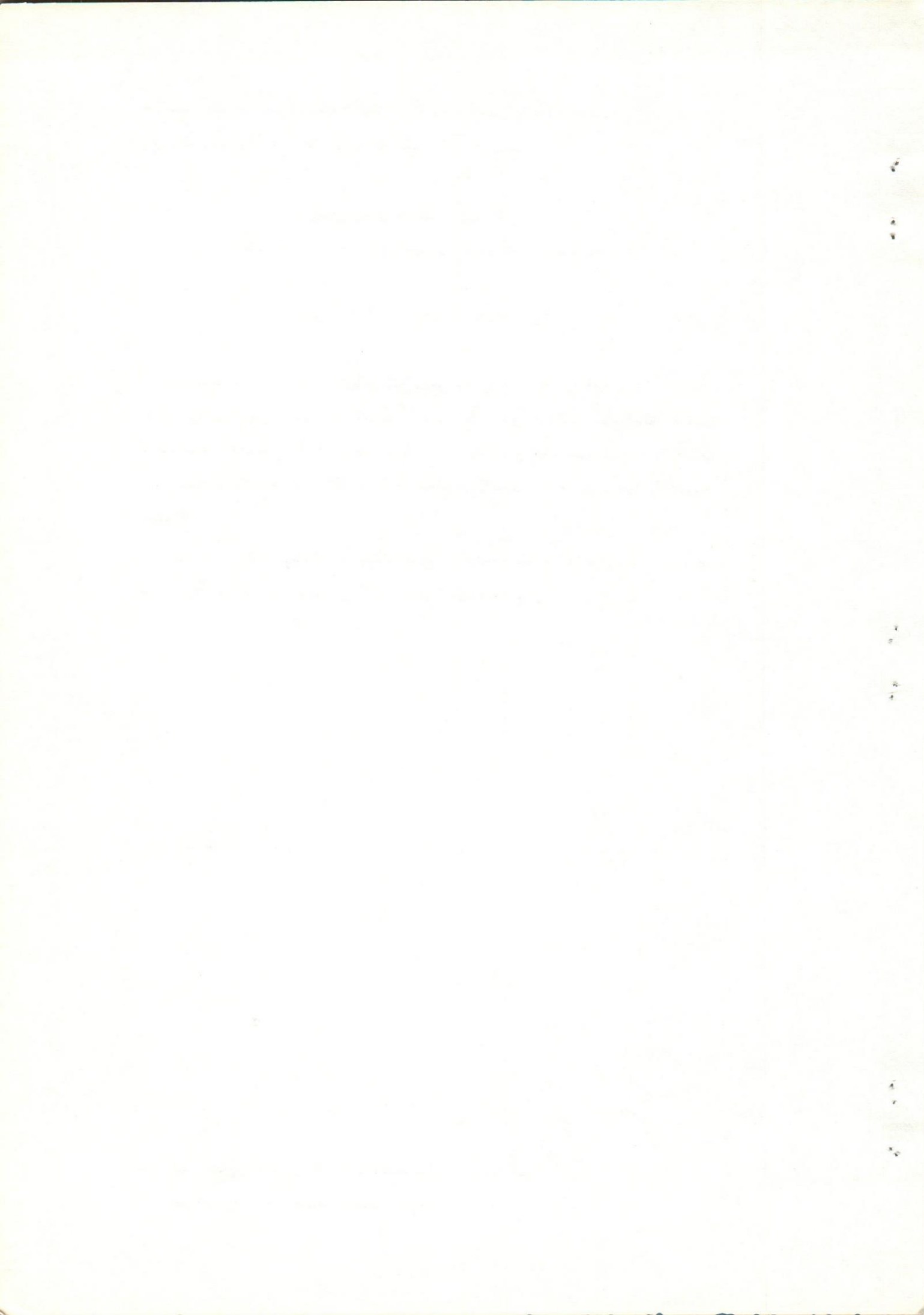
فـوزى ابو الخير ، راقية الحلوانى \*

تم تحديد نماذج الفصل الكهربائى للكازين فى ( ٣٦ ) عينة فردية وجماعية من ثلاث حيوانات مختارة من كل النوعين . وقد أوضح البحث أن كازين اللبن تجزأ الى ثلاثة أقسام مختلفة الكمية حسب الهجرة الكهربائية الى الفا وبيتا وجاما كازين . وقد أتضح أيضا وجود فروق معنوية فى نسبة جزئى الالفا والبيتا كازين لكل من اللبن الجاموسى والبقرى بينما تساوى تقريبا فى نسبة جزئى الجاما - كازين .

لم يلاحظ اختلافات معنوية عند تحليل العينات الفردية والجماعية لكل نوع على حده .  
تم توضيح أهمية التحليل الكهربائى لكازين اللبن البقرى والجاموسى .

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QUALITATIVE AND QUANTITATIVE EVALUATION OF CASEIN FRACTIONS  
FROM EGYPTIAN BUFFALOE'S AND FRIESIAN COW'S BY ELECTROPHORETIC TECHNIQUE  
(With 3 Tables & One Figure)

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SUMMARY

The electrophoretic pattern of casein complex of milk from Egyptian buffalo's and Friesian cow's were determined in 36 individual and composite samples using paper electrophoretic technique. Three distinct fractions were obtained named as alpha, beta-, and gamma fractions. Variations in the concentration of casein fractions is distinctly present in both cow's and buffalo's milk. Insignificant differences could be detected between the concentration of the different fractions in the individual and composite milk samples, while significant differences detected between cow's and buffalo's milk in the concentration of alpha and beta casein fractions only. The obtained results add new informations about milk proteins from Egyptian buffalo's and Friesian cow's which has a great significance from the nutritional and technological point of view.

INTRODUCTION AND LITERATURE

Knowledge about the colloidal state of milk is of great importance from the nutritional and technological point of view. Casein from cow's milk has been thoroughly investigated (ROSE 1970, MCKENZIE 1971, DILL *et al.*, 1972, DAVIES and LOW, 1977). Data concerning the fractionation of milk proteins from buffalo's are relatively scanty in current literature. Only few reports were available (MAJUMDER and GANGULI 1972, NAGASAWA *et al.* 1973, ABO-EL-SALAM 1975, ADDOE *et al.*, 1977). It has been reported by ASLANYAN (1965) that alpha-casein fraction slightly decrease and beta-casein increased while gamma casein remained constant in buffalo's milk towards the end of lactation period.

It was found that diets affect the percentage of different fractions in casein complex (DAVIDOV 1969). Each casein fraction characterized by its own physical and chemical character. The amino acid content of each fraction differs also (DIADCHINKO 1959, KAZANSKI *et al.*, 1960, NOSEIR *et al.*, 1976).

Kappa casein fraction is noted for its ability to stabilize alpha-casein (THOMPSON *et al.*, 1969, JOSHI and GANGULI 1970). The technical properties of cheese depend mainly on the concentration of casein fraction in milk from which was made. Alpha-casein contain 1 per cent phosphor and 0.72 per cent sulphar, while beta-casein contain 0.55 and 0.86 per cent, respectively. Both fractions well be coagulated completely by rennit. Gamma-casein contain 0.11 per cent phosphor and 1 per cent suipher, rennit has no effect on the coagulation of gamma casein fraction (UNIXOV 1970).

RANDOLPH *et al.*, (1974) reported that mastatic milk contained relatively higher concentrations of bovine serum albumun, proteose peptone, immunoglobulin, alpha and Kappa-casein and lower concentrations of beta-lactoglobulin, beta-casein, there was no changes in the distribution of alpha-lactalbumin.

DAVIES and LOW (1977) found that of the total casein in cow's milk the alpha-casein seemed to be less influenced by stage of lactation, with an average of about 48.9 per-cent being found for the early, mid and late lactation. Beta-casein was low during early (38.3 per cent) and late lactation (32.8 per cent) as compared with mid lactation (37 per-cent). While BERNATONS *et al.*, (1973) reported that the casein fractions were affected by parturition rather than the stages of lactation, while the reverse is true for serum proteins.

It has been reported by SAFINAS EL-SHIBINY (1978) that alpha-casein was found to represent about 40 per-cent of the whole casein fractions of goats milk.

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There are a number of reports testifying the possibility of utilizing electrophoretic technique for detection of adulteration of milk. MAJUMDER and GANGULI (1972) studied the possibility of detection of adulteration of cow's milk with buffalo milk by starch gel electrophoresis. The author's mentioned that adulteration of cow's milk with buffalo's milk to the extent of 5 per cent, can easily be detected.

As buffalo's milk is an important source of protein in Egypt, moreover, casein fraction is one of the main factors which affect the technical behaviour and utilization of milk, in addition casein fraction has an important function in modern clinical laboratory diagnosis. Therefore investigation of the electrophoretic pattern of casein from Egyptian buffalo's and Friesian cow's was aimed to fulfill such lacking information.

### MATERIAL and METHODS

A total of 36 individual and composite milk samples from Friesian cow's and Egyptian buffalo's were used in this experiment. All milk samples from individual cow's and buffalo's were taken three times monthly (9 trials in each) during the fourth, fifth and sixth month of lactation with 10 days interval. Composite milk samples were collected from three selected animals from the two species (According to the date of last delivery). The dairy animals belong to Karade-Mehalet Moussa experimental dairy farms, animal production research institute, Kafr El-Shiekh Governorate, Ministry of Agriculture.

Both experimental animals were clinically healthy throughout the period of the experiment. Milk samples were conveyed immediately to the laboratory where heated at 40°C then defatted by centrifugation. Casein was precipitated from 100 ml skim milk by 20 per cent acetic acid till PH 4.6. The obtained precipitate was washed three times with dist. water then dissolved in 25 ml veronal buffer to which 40 per cent urea was added. The solution was filtered, then 0.05 ml from the filtrate was applied on Whatman filter paper strips (No. 1) 3 x 40 cm. Casein proteins have been partitioned in veronal buffer (PH 8.6) contain 40 per cent urea to facilitate the breaking of the hydrogen bonds which prevent aggregation and complex formation.

The protein zones were finally stained with amido black dyes. The quantitative measurement of each fraction was done according to the method described by KYGENOV and BARABANCHIK (1973). Statistical analysis of the data was conducted according to SNEDICOR (1955) using "t" test.

### RESULTS

Obtained results are presented in tables 1 & 2 and Fig. 1.

### DISCUSSION

The mean values, and standard errors for the concentrations of casein fractions from cow's and buffalo's milk are given in table 1 and 2, together with the values obtained from analysis of the individual and composite samples from the two animal species. The data expressed in percentage to the total casein fractions. Electropherogram (Fig. 1) reveals three casein fractions, alpha, Beta and gamma from cow and buffalo milk. Similar findings were obtained by VLADOVETS (1959), SAMULESON, (1962), ASLANYAN (1965), and ADDOE *et al.*, (1977).

Casein may be divided into several fractions. The number of the fractions that may be found depend on the method of analysis used which in turn may be governed by the particular purposes for which the determination was made. The paper electrophoretic technique is fairly satisfactory for separating milk proteins (KYGENOV and BARABANCHIK 1973).

## ELECTROPHORESIS OF CASEIN

Table (1): Distribution of casein fractions from cow's milk

Casein fractions	Individual Samples			Mixed Samples			N = 9 $M_1 - \bar{M}_1$
	Min.-Max.	$M_1$	SE	Min.-Max.	$M_1$	SE	
Alpha	52.2-60.0	56.44	1.13	54.6-60.0	75.87	0.89	-1.43
Beta	35.2-38.2	37.38	0.47	34.6-39.4	37.27	0.56	+0.11
Gamma	3.4-10.4	6.18	0.97	2.2- 7.4	4.87	0.60	+1.31

Table (2): Distribution of casein fractions from buffalo's milk

Casein fractions	Individual Samples			Mixed Samples			N = 9	
	Min.-Max.	$M_2$	SE	Min.-Max.	$M_2$	SE	$M_2 - \bar{M}_2$	$M_1 - M_2$
Alpha	34.8-41.8	37.84	0.77	35.2-41.2	38.24	0.72	-0.41	-18.6*
Beta	53.0-58.2	55.49	0.57	52.4-58.6	56.53	0.78	-1.04	+18.11*
Gamma	4.6- 8.6	6.87	0.66	2.2- 8.4	5.22	0.76	+1.45	+ 0.49

\* Significant at 0.01 level  
Data represent average values for 9 trials for each.

The data presented revealed that the average concentrations of alpha, beta-casein from cow's milk constitute 56.44 and 37.38 per cent, respectively, while the concentration of these fractions in buffalo's milk constitutes 37.84 per cent. It was noted that beta casein fractions from buffalo milk constitute the major component of the total casein fractions, while alpha-casein from cow's milk constitute the majority of the total casein fractions. The high concentration of beta-casein fraction in buffalo's milk if compared with cow's milk may be attributed to species differences and physical condition of the animals.

It was also noted that gamma-casein fraction is relatively high in buffalo milk (6.67%) than cow milk (6.18), however statistical analysis revealed insignificant differences.

In view of the obtained results, it could be concluded that the variations between cow and buffalo milk were confined mainly in the value of alpha and beta-casein fractions, since statistical analysis showed high significant differences ( $P = 0.01$ ). Furthermore, there are slight variations in the concentration of different casein fractions in the individual and mixed milk samples from the two animal species. Therefore, mixing of the sample does not affect the concentration of the casein fractions.

Table(3) contain the results of fractionation of casein obtained by several author's. The obtained results showed that casein from cow and buffalo milk either in individual or mixed samples was composed of the same fractions.

It was clear from the data of the different author's that the value of casein fractions differs from one to another. The figure obtained in the present study was slightly different from those obtained by other author's this may be due to difference in the method of electrophoresis, and breed differences at which the experiment was made.

Concerning casein fractions from cow's milk. The obtained results may agree with those reported by DILL *et al.*, (1972). Regarding the fractions from buffalo milk the present results were favourable with the finding of ADDEO *et al.*, (1977) although the author's analysed casein fractions from milk of different animal breeds.

Table (3)  
Concentrations of casein fractions obtained by several author's

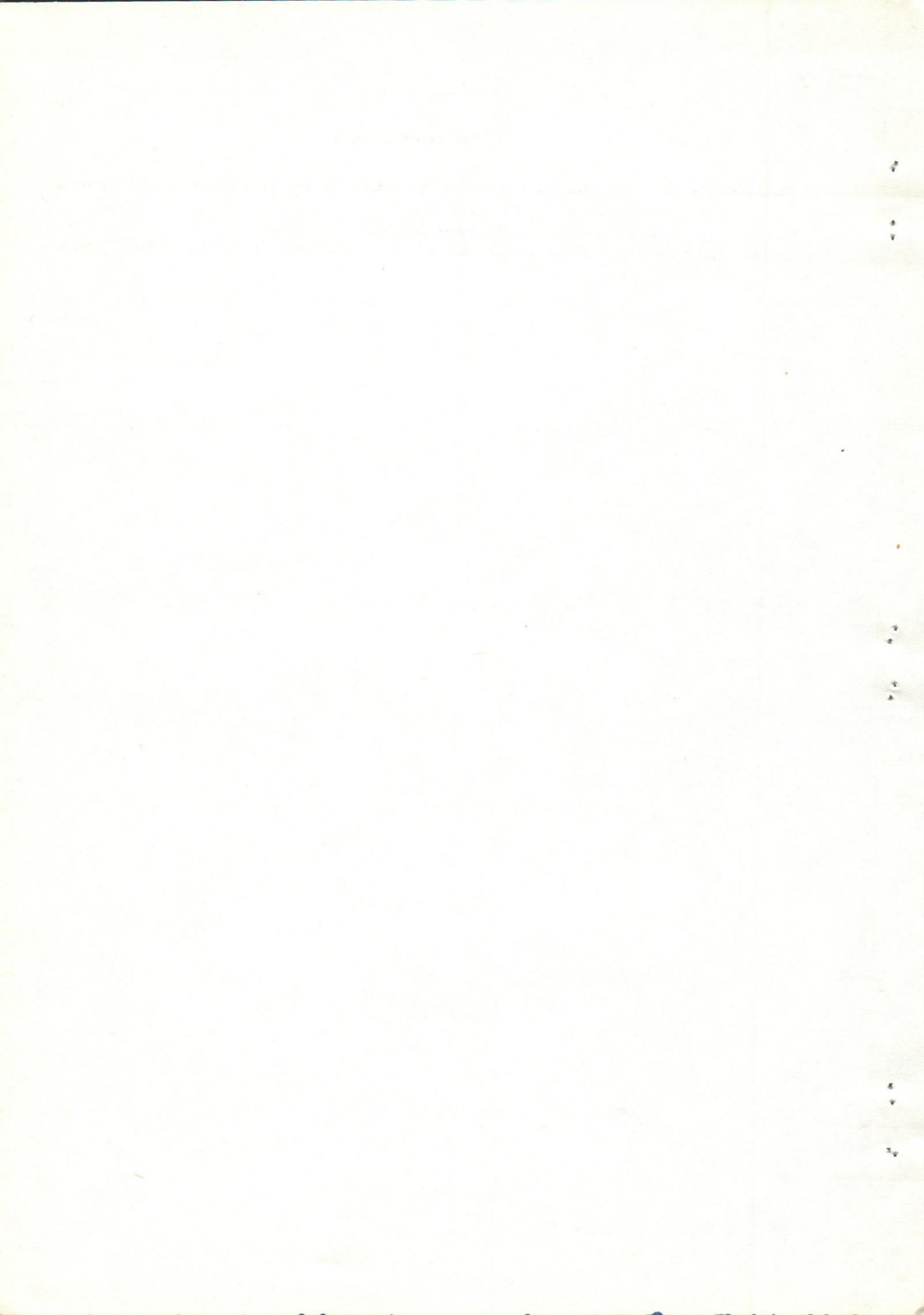
Animal species	Casein fractions in per-cent to the total casein fractions			Author
	Alpha	Beta	Gamma	
Cow	38.00	53.00	9.00	DAVIDOV (1969)
	60.47	35.85	3.76	DILL <i>et al.</i> , (1972)
	49.57	45.08	5.76	RANDOLPH (1974)
	47.00	38.00	15.00	ADDEO <i>et al.</i> , (1977)
	36.6	56.9	6.5	VLADOVETS and ZHDANOVA (1954)
	52.6	40.2	7.2	KYGENOV and BARABANCHIK (1973)
Buffaloe	20.2	75.5	4.3	ASLANYAN (1965)
	34.0	51.0	15.0	ADDEO <i>et al.</i> , (1977)

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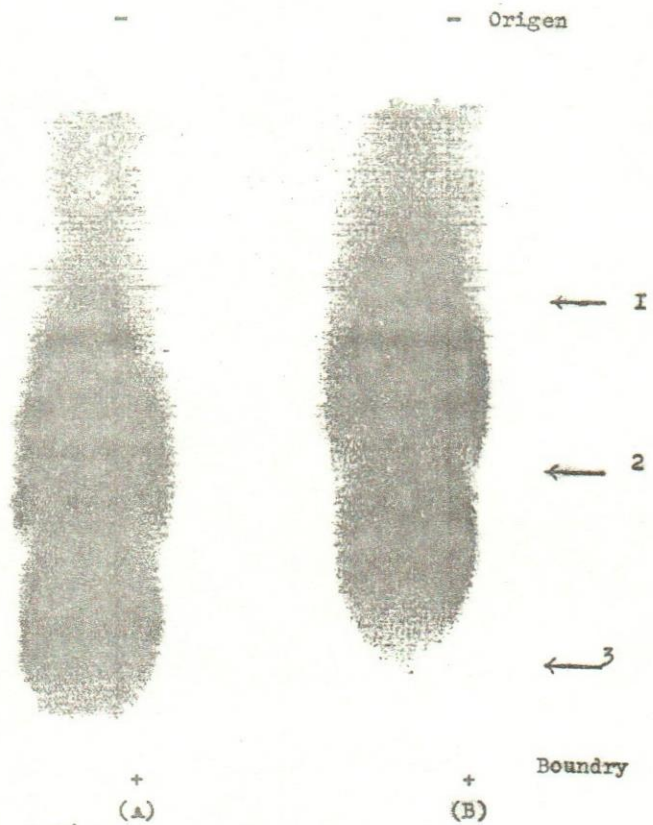


Fig . 2 Distribution of casein fractions of cow (A) and buffale milk (B) .

- 1. Alpha casein
- 2. Beta- casein
- 3. Gamma- casein

