

THE DIGESTIBILITY OF THE ALBUMINOUS CONSTITUENTS OF HUMAN MILK AND THAT OF VARIOUS SUBSTITUTES FOR IT.

BY F. W. TUNNICLIFFE,

Professor of Materia Medica and Pharmacology in King's College, London.

(From the Pharmacological Laboratory, King's College, London.)

THE most important of the proteid constituents of milk is casein. From 70 to 90 % of the total proteids of milk consists of this substance¹. In milk, casein exists as a soluble calcium compound, which is not precipitated by boiling, but from which casein is deposited by the addition of acid or by the action of rennet². This phenomenon is known as the curdling of milk and is a preliminary to its gastric digestion, both the acid in the stomach and the rennet being capable of producing it. By artificially influencing the nature of its curd we can influence the digestibility of the milk. In human gastric digestion and in the experiments detailed below both acid and rennet are present. The curdling of milk is a necessary preliminary to its digestion in an acid medium, and by influencing this process we can also influence the digestibility of the milk.

In cow's milk the addition of a few drops of acid or rennet occasions in a few minutes the precipitation of a hard toughish coagulum, whereas if we treat human milk or one of the prepared milk foods, for instance Fairchild's Peptogenic Milk, in the same way, a more or less finely flocculent precipitate of casein is thrown down. In the case of human milk the floculi are very fine indeed. *Ceteris paribus* the fineness of the precipitate is an important fact in its digestibility.

¹ Our own figure is 93·5 %. Stutzer (*Milch als Kinder-Ernährung*, Bonn, 1895) gives without stating authority 40 %. Lehmann and Hempel (*Pflüger's Archiv*, vol. LVI. 1894, p. 577) 70 %.

² Hammarsten, Zur Kenntniss des Kaseins u. der Wirkung des Lab-Fermentes, *Abhandl. König. Gesellschaft zu Upsala*, 1877. Arthus and Pagés, *Archives de Phys.* 1890, p. 331.

This is partly to be explained mechanically, a finely divided casein offering, per unit weight, more surface for the action of the digestive juices than a casein deposited in lumps. Lumps further act as a mechanical irritant to the stomach and intestines, stimulating peristalsis. The lumps thus become hurried down the intestinal tract and are voided undigested.

We can influence the degree of fineness in which casein is precipitated in various ways. One of these is to dilute the milk and thus expose to the digestive juices a more dilute solution of casein. Another method is to diminish the amount of soluble calcium salts¹ present in the milk by the addition of *e.g.* an oxalate or citrate. A further method² consists in altering the reaction of the milk to be curdled and the strength of the acid used to curdle it. The stronger the acid the more tough and compact the curd and hence the greater the trouble to digest it. This fact is of interest in connection with the difficulty often experienced in digesting milk by patients suffering from hyperchlorhydria, which can often be entirely removed by the addition of an alkali to the milk.

The factor however of greatest importance in determining the digestibility of a casein is the nature of the casein itself. The casein obtained from human milk is chemically different to that obtained from the milk of any other animal. Wroblewsky³, working under Drechsel's guidance, found the percentage compositions of cow's and human casein to differ especially with regard to their sulphur and phosphorus contents. As the result of peptic digestion experiments these authors found that in the case of human milk no insoluble nuclein compound was split off; on the other hand in the case of cow's casein an insoluble nuclein residue invariably remained even after indefinitely prolonged peptic digestion. From these experiments Wroblewsky concludes that human and cow's casein are chemically different substances. According to Siegfried⁴ all the phosphoric acid in human milk is bound directly to "albumen," whereas in cow's milk only half of it is so bound.

The following experiments were undertaken to ascertain to what extent this chemical difference between human and cow's casein affects their respective digestibilities and also to what extent their differences

¹ Arthus and Pagés, *loc. cit.*

² Courant, *Ueber die Reaktion der Kuh- u. Frauen-Milch.* Inaug. Dissertation, Bonn, 1891, p. 39.

³ *Beiträge zur Kenntniss des Frauens Kaseins.* Inaug. Dissertation, Bern, 1894.

⁴ *Zeitschr. für physiol. Chemie*, vol. xxii. p. 575.

in this respect can be compensated for by artificial means as adopted in various proprietary milk foods.

Method.

Since the best method at our command was digestion *in vitro* we were anxious to imitate physiological conditions as far as possible and to contrive so that the results were comparable *inter se*.

The cow's milk was obtained from a good London dairy. In submitting the artificial milk foods to digestion the directions on the label were followed exactly. The human milk, of which we obtained two litres, was kindly collected for us by one of the externe nurses of Queen Charlotte's Lying-in Hospital, London. It was derived from twenty-nine women in periods of lactation varying from six weeks to five months. The different portions of milk were well mixed before submitting the whole to analysis and digestion. The following table (Table I.) gives the composition of the milks as used for the experiments, with the exception of the cow's milk which was diluted with an equal quantity of New River tap water.

Three classes of digestive experiments were made: (1) Peptic, (2) Pancreatic, (3) Peptic and subsequently Pancreatic.

Peptic digestion. The average amount of milk in an infant meal is from 60 to 90 c.c., which in the case of human milk would represent about 1 gramme of total albuminoids. By preliminary experiments we satisfied ourselves of the efficiency of Fairchild's Pepsin and of the approximate accuracy of the data upon the label. We used therefore for our peptic experiments a digestive solution of the following composition. Pepsin (Fairchild's) 0·06 gramme, hydrochloric acid (33 %) 6·00 c.c., water 600 c.c. Of this solution 6 c.c. were added to 50 c.c. of the milk under experiment. The normal infant stomach is usually empty from one to two hours after the ingestion of a meal¹; but as a somewhat fine index was required the time for peptic digestions in these experiments was fixed at one hour. The temperature at which they were made was 38° C. At the end of the hour the mixtures were brought rapidly to the boil. The amount of albuminoids digested in each case was ascertained as follows. After the boiling the contents of each flask was washed on to a prepared filter which was provided with an ice and salt jacket. Filtration was allowed to continue until complete.

¹ Epstein, von Puteren, Wohlmann and others, quoted by Soltau Fenwick, *Disorders of Digestion in Infancy*, London, 1897.

TABLE I.

Showing the percentage composition of the milks etc. used for the experiments.

	Specific gravity	Water	Total solids	Ash	Total nitrog. matter	Total albuminoids	Casein	Fat	Milk sugar	Mal. lase	Dex. trine	Sol. starch	Starch	Cane sugar
Human milk	1.032	88.42	11.58	0.19	1.18	1.06	0.99	2.55	6.28					
Cow's milk	1.032	87.67	12.33	0.74	3.07	2.74	2.56	3.15	4.76					
Patent milk I	1.036	85.65	14.53	0.42	1.86	1.64	1.32	4.00	8.22					
" II	1.022	92.41	7.59	0.38	1.84	1.70	1.48	1.37	3.15	0.32	0.57			
" III	1.019	94.49	5.51	0.10	0.85	0.80		0.33	0.39			0.58	0.90	1.46
" IV	1.030	87.99	12.01	0.30	1.38	1.23	1.18	3.65	6.71					
" IV (a)	1.030	87.79	12.21	0.31	1.53	1.45	1.37	3.65	7.18					

The temperature during the whole time of filtration never exceeded 3° C. This was deemed important since filtration proceeded very slowly at ordinary temperatures. Owing to bacterial activity further decomposition of the respective mixtures might easily have taken place. When filtration was complete the residue was washed with a little water to free it more thoroughly from soluble albuminoids, and was subsequently removed to a Kjeldahl flask and the nitrogen estimated in the usual manner. The figure thus obtained was calculated to proteids and the result subtracted from the original insoluble proteid content. The remainder gave the quantity of proteid digested. The results of the peptic digestion are given in the respective tables.

Pancreatic digestion. For the pancreatic digestion a standard zymine solution was made up on the lines of that used by Biffi¹, with the exception that instead of powdered pancreas, Fairchild's zymine powder was used. This solution had the following composition: zymine powder (Fairchild's) 1.0 gramme, chloroform water 100 c.c., saturated sodium carbonate solution 1.0 c.c. This solution was allowed to stand for 12 hours at 40° C. and then filtered. In the pancreatic experiments 50 c.c. of the milks or feeding-bottle mixtures were taken; the time allowed was three hours; the temperature 38° C.; 10 c.c. of the above solution was added to each. During the whole time of digestion the reaction was kept alkaline. At the end of three hours the nitrogenous substances undigested were precipitated by potash alum according to Schlossmann's method, and nitrogen estimations made as above. The amount thus obtained calculated as proteid was then subtracted from the total albuminoids originally present, the remainder representing the quantity of albuminoids digested².

Peptic and Pancreatic digestions. In the living infant the milk ingested is submitted first to the action of the gastric juice, and the residue is subsequently exposed to the pancreatic juice. An attempt was made to imitate this *in vitro*. Quantities of milk (50 c.c.) were submitted to the action of the artificial peptic solution for 1 hour, were then filtered quickly at a low temperature (see above), the residue rinsed into a flask with 50 c.c. of water and made alkaline. The ensuing

¹ *Virchow's Archiv*, 1898, Band CLII, Heft 1.

² In some earlier experiments we attempted to estimate the albuminoids digested by the artificial pancreatic solution in a manner similar to that adopted in the case of the peptic digestion. The great difficulty of filtering the mixtures after pancreatic digestion, especially in the case of human milk and those milk preparations approaching it in digestibility, led us however to adopt the above method.

mixture was then submitted to the action of the pancreatic solution for three hours and subsequently treated as described under pancreatic digestion.

The results of these experiments are given below in tabular form. Each result is the mean of three or four experiments, the extremes never differing by more than 0.2 %. In the case of the last patent food we made two separate series of experiments with two samples obtained at different times, on account of the great difference in the results obtained with this and the other patent foods.

Methods.

All nitrogen estimations were made by Gunning's modification of Kjeldahl's method. The general factor for the calculation of proteids was 6.37, except in the case of human milk when it was 6.34 (Munk, *Virchow's Archiv*, vol. CXXXIV. p. 501). Total albuminoids were estimated by a slightly modified Ritthausen's method (*Journ. für prakt. Chemie*, N. F. vol. xv. 1877, p. 329). Casein was estimated by Schlossmann's method.

TABLE II.

Showing the digestibilities in vitro of the proteids of human and cow's milk, and those of certain artificial milk foods.

Name of milk	Digested after 1 hour's peptic digestion	Digested after 3 hours' pancreatic digestion	Digested after 1 hour's peptic and 3 hours' pancreatic digestion	Remarks
I. Human milk 6th week to 5th mo.	48.12 %	21.07 %	75.46 %	
II. Cow's milk 50 %	47.44 %	27.00 %	47.44 %	
III. Patent milk I	54.88 %	40.24 %	54.88 %	
IV. Patent milk II	55.29 %	24.11 %	56.47 %	
V. Patent milk III	26.25 %	32.50 %	40.00 %	This food contained no milk; it was deficient chemically, & contained starch & cane sugar
VI. Patent milk IV				
Sample A	28.46 %	32.52 %	35.96 %	} This milk was the dearest of all those examined and bore the name—"human milk"
Sample B	28.96 %	33.10 %	36.19 %	

CONCLUSIONS.

The above tables require but little comment. Physiologically the results confirm those of Wroblewsky and Siegfried showing that there is an essential difference between human and cow's casein, and especially that this difference affects the nucleo-proteid moiety of the casein molecule, or that part of it which is digested by the pancreatic in distinction to the part digested by the gastric juice. From the point of view of experimental dietetics the results show the importance, in estimating the digestibility of proteid substances *in vitro*, of submitting the residue of gastric digestion to the artificial pancreatic juice. It will be obvious from the above tables that in so far as concerns the simple gastric or simple pancreatic digestion, the digestibility of the proteid constituents of certain milk foods and indeed simple cow's milk itself closely approximates to or even exceeds that of human casein. When however we regard the total digestibility after peptic and pancreatic digestion we see that the substitutes for human milk fall considerably short of human milk itself. It will be obvious also from the table that certain milk foods do dietetically possess a considerable advantage over simple, unmanipulated cow's milk. From the point of view of public health the above results accentuate the nutritive advantage to the infant of mother's milk as opposed to any substitute for the same, in other words of breast-feeding as opposed to bottle-feeding.