Aspects of the value and the limitations of milk protein as a food material

By WILLIAM MANSON, *The Hannah Research Institute, Ayr*

It is generally accepted that milk and milk-based products constitute a valuable source of dietary protein. In the last few years considerable advances have been made in understanding the chemistry of milk proteins and in particular in the elucidation of their primary chemical structures so that it is now possible to examine this general conclusion concerning the worth of milk proteins, their value and limitations as foodstuffs in the light of an almost complete knowledge of their basic chemistry, a situation which, it may be noted in passing, exists for no other dietary protein. It is also feasible at least to begin to consider some of the implications of the utilization of milk proteins in non-traditional forms.

Before proceeding to considerations of this kind, mention should be made of a major limitation to the further utilization of milk protein which has little direct connection with an evaluation of its nutritional worth. This concerns the acceptability of the form in which it is presented as a foodstuff. While this paper is concerned largely with the intrinsic properties of milk proteins and not with the production of new acceptable food forms, it should be remembered that the requirement of acceptability does exist and is indeed of overriding importance.

In any assessment of the significance of milk protein as food consideration should be given to the amount of it consumed. In 1977 the milk protein produced for human consumption in this country amounted to slightly more than 440 000 tons which it may be calculated was sufficient to supply everyone in the country with about 25% of his total requirement for protein and accounted for about 65% of his animal protein intake. These figures are reasonably representative of the situation in Western Europe as a whole and in the United States. They are not universally applicable, since in India and Pakistan it has been estimated (Smith, 1970) that only about 8–10% of the protein intake originates in milk and elsewhere in Asia the figure is as low as 1–2%. It is therefore the populations of the developed areas which rely on milk and milk-based products for protein and this they clearly do to a highly significant extent. Whatever their nutritional value may be there is no disputing the present commercial value of milk proteins. Any increase in their utilization in this type of market will have to take place in the form of food products of some sophistication. The questions may be asked, therefore, what are these proteins and how are they constituted?

Milk protein is, with the possible exception of that derived from eggs, the only major dietary component which in Nature is produced specifically for the nourishment of young. In this functional respect it differs from other dietary animal proteins, vegetable proteins and unconventionally produced proteins. However it is only with man that these proteins are included in the diet throughout the entire life span, a point which will be mentioned again later.

With the exception of some egg yolk proteins, milk protein differs from proteins derived from other sources in the further respect that it is largely phosphoprotein in character. Its nutritional value may be considered therefore under two headings, as a carrier of amino acids and a provider of phosphorus. The constituents of milk protein are listed in Table 1. From this it will be seen that the phosphoprotein fraction, the caseins, accounts for almost 80% of the total protein. The remaining

Table 1. *The main constituents of milk protein expressed as a percentage of the total protein*

Protein	Total milk protein (% by wt)
Caseins	78
β-lactoglobulin	14
α-lactalbumin	3
Immunoglobulins	2
Blood albumin	3

soluble proteins are collectively known as the whey proteins. The casein complex consists of three main components designated α_s-casein, which is itself a mixture of several phosphoproteins (Annan & Manson, 1969), β-casein and κ-casein in the approximate proportion of 50% : 35% : 15%, respectively. The contributions of these fractions to the overall value of milk protein when considered as a supplier of essential amino acids are listed in Table 2, where they are also compared with the composition of a hypothetical ideal standard protein as defined by the World Health Organization and the Food and Agriculture Organization (FAO/WHO, 1965). It will be seen that all of the essential amino acids are present in milk

Table 2. *The essential amino acid contents of individual caseins and of whole milk protein expressed as g amino acid produced by hydrolysis of* 100 *g protein*

Amino acid	Whole milk	Casein				FAO‖ Standard
		whole	α_s*	β†	κ‡	
Lysine	7·9	7·7	8·7	6·7	6·9	4·2
Tryptophan	1·3	1·3	1·7	0·9	1·1	1·4
Threonine	4·9	4·1	2·5	4·5	8·8	2·2
Cystine	1·0	0·2	0·0	0·0	1·3	2·1
Valine	6·9	7·0	5·5	9·3	6·8	4·2
Methionine	2·2	3·1	3·2	3·7	1·6	2·2
Isoleucine	6·5	6·3	6·1	5·5	9·0	4·2
Leucine	10·1	9·8	9·4	12·0	5·5	4·8
Phenylalanine	4·9	5·5	5·6	6·2	3·5	2·8
Tyrosine	4·7	6·2	7·7	3·0	8·6	2·8
Phosphorus	0·6	0·8	1·05	0·65	0·16	—

Values calculated using information from:
*Mercier, Grosclaude & Ribadeau Dumas (1971).
†Manson & Annan (1971).
‡Mercier, Brignon & Ribadeau Dumas (1973).
‖FAO/WHO (1965).

https://doi.org/10.1079/PNS19780032 Published online by Cambridge University Press

protein as a whole and with the exception of cysteine all are present in amounts in excess of those of the standard protein. The deficiency of cysteine, although partly reduced by the amount of methionine present, renders milk protein somewhat deficient in sulphur-containing amino acids. As a consequence the utilization of some of the more abundant amino acids such as lysine and threonine is reduced. It is also apparent that almost all of the cysteine of milk protein is present in the whey fraction, in β-lactoglobulin and α-lactalbumin in particular. Without these components the biological value of milk protein would be significantly reduced and a larger part of the more abundant essential amino acids would be rendered unusable. Whey protein is therefore a valuable product and strenuous efforts are being made to increase its recovery in a commercially viable fashion from processes such as cheese manufacture where it was traditionally regarded as a waste product.

For the full potential value of milk protein to be realized it requires fortification with sulphur-containing amino acids and this may conveniently be done by mixing it with proteins reasonably rich in cysteine or methionine but deficient in other essential amino acids. As may be seen from Table 3, wheat protein has such an amino acid composition as has rice protein. Thus by judicious mixture of cereals

Table 3. *The essential amino acid content of various dietary proteins expressed as g amino acid released on hydrolysis of* 100 *g protein*

	Protein source				
Amino acid	Milk	Egg*	Soya†	Wheat‡	Fishmeal
Lysine	7·9	6·9	6·0	2·4	7·0
Tryptophan	1·3	1·6	1·3	1·5	1·2
Threonine	4·9	5·0	3·7	3·2	4·2
Cystine	1·0	2·3	0·9	3·5	1·0
Valine	6·9	7·4	5·0	4·4	5·2
Methionine	2·2	3·3	1·0	1·5	2·6
Isoleucine	6·5	6·9	4·9	4·3	4·6
Leucine	10·1	9·4	8·1	7·9	7·3
Phenylalanine	4·9	5·8	5·6	6·0	4·0

Values calculated using information from:
*Smith (1970).
†Meyer (1967).
‡Ewart (1967).

with relatively small amounts of milk protein a product properly balanced for amino acid intake can be prepared. Enhancement of the biological value of nutritionally poor proteins is probably the single most valuable property of milk protein and if fully exploited it could have far-ranging consequences. Apart from the obvious advantage of permitting the more efficient use of locally produced cereal proteins such as rice, it permits the more efficient use of land since although intensive milk production and wheat growing produce similar amounts of protein per hectare the difference in quality of the products is such that the yields per

hectare of the essential amino acids lysine and threonine from dairy production are respectively three times and twice those from cereal production (Blaxter, 1968). Blending of cereal and milk proteins could also reduce the amount of fossilized energy expended in agricultural processes since it would permit the utilization of some of the essential amino acids which are otherwise lost but for which there is an energy cost in their production.

I should now like to turn briefly to some of the detail of the chemistry of the phosphoprotein complex of bovine milk. The primary structures of the two most abundant components, α_s-casein and β-casein, are quite distinct in character and possess only one small region in common. This is shown in Fig. 1 and contains a significant proportion of the phosphorus of each protein in the form of a highly specialized structure. Furthermore it is now known that the parts of the casein molecules which contain this extended structure resist digestion by intestinal proteolytic enzymes and fragments remain available for absorption in the form of relatively small peptides rich in phosphorus. The nutritional significance of this phosphate and of such phosphopeptides is uncertain but phosphoproteins occur in quantity in Nature only in materials produced as nutrients for a developing animal organism, such as fowl and fish eggs and the milks of many species. Evidence is now available which suggests that the phosphorus is arranged in these proteins in a similar manner, in groups of phosphoseryl residues typified by the structure illustrated. This arragement of phosphoseryl residues is chemically reactive and it is also well suited to the binding and the transport of small positively-charged ions such as Na^+, K^+, Ca^{2+}, Mg^{2+} and Fe^{2+}. It is probable that the biological function of the caseins involves these properties and is concerned with the transfer of phosphorus from milk to suitable sites within the developing organism where it may be used for a specific purpose. Cellular phosphoprotein has for example been shown to be involved in the transport of iron across mitochondrial membranes in the form of complexes between iron and phosphorylated sites in the protein (Donella, Pinna & Moret, 1972).

In human milk the major phosphoprotein is a β-casein which contains a similar arrangement of phosphoseryl residues to that of bovine β-casein. However, human milk contains only about one-seventh of the phosphoprotein of bovine milk so that the use of bovine milk in human nutrition involves the supply of phosphopeptides of structures chemically similar to those from human milk but in much larger quantities than arise from a similar weight of human milk protein. If the same procedure were to be followed in human nutrition as occurs naturally with those species which rely heavily on phosphoprotein for the initial nutrition of their young, no phosphoprotein would occur in the human diet after infancy. The requirement for phosphoprotein would appear to be confined in the natural state to early development and consequently the value of supplying large amounts of these

-Glu-Ser(P)-Leu/Ile-Ser(P)-Ser(P)-Ser(P)-Glu-Glu-

Fig. 1. Sequence of amino acid residues common to α_{s_1}-casein and β-casein.

https://doi.org/10.1079/PNS19780032 Published online by Cambridge University Press

specialized phosphorus-containing structures throughout adult life is questionable. However, the preservation of such structures throughout processes for the manufacture of milk protein where the product is intended for use by infants is worthy of care since if they are destroyed a potentially valuable property may be lost.

Much of what has been discussed thus far concerns milk protein in its native, or near native, state. Most milk protein consumed is no longer in its native state but has been subjected to heat treatment in one form or another. It is important therefore to determine by how much the intrinsic properties of milk protein may be effected by heat treatment. A number of chemical reactions take place among the milk proteins themselves or between them and other milk constituents when subjected to heat. The most widely recognized of these is that occurring between the ε-amino group of lysyl residues and aldehydes produced from milk carbohydrates, the Maillard reaction. It is unnecessary to discuss the mechanism of this reaction further here but some of its implications are of interest apart from the obvious one that lysine modified in this way is not available for absorption and utilization in the same way as unmodified lysine. It has been claimed, for example (Bleumink & Young, 1968), that bovine milk contains an atopic allergen which was identified as β-lactoglobulin in which lysyl residues had been modified as a result of heating in the presence of lactose. When lysyl residues are modified in this way trypsin is no longer able to cleave the peptide bond between the lysyl group and the next amino residue so that the normal digestion of the protein is impaired and abnormal peptides are produced. Attempts are made to minimize this reaction during heat treatment of milk protein and these are effective when judged by the proportion of lysyl residues remaining at the conclusion of the treatment. But the amount of modification of lysyl residues required to produce effective quantities of an allergen is likely to be very small.

A second type of reaction is that which leads to the production of lysinoalanine. It has been shown (Manson & Carolan, 1972) that when the caseins are warmed with dilute alkali, phosphate is eliminated from phosphoseryl residues and dehydroalanyl residues are produced (Fig. 2). These in turn react with the ε-amino

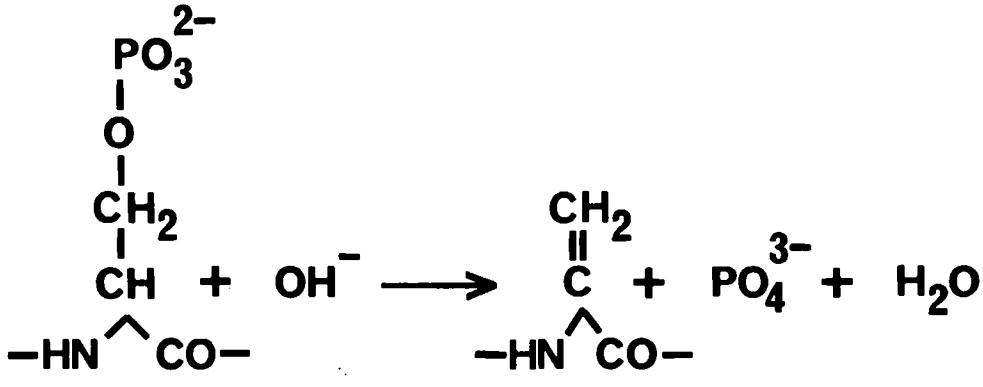

Fig. 2. Formation of dehydroalanyl residue by alkaline degradation of phosphoseryl residue.

https://doi.org/10.1079/PNS19780032 Published online by Cambridge University Press

groups of lysyl residues with the formation of lysinoalanine (Bohak, 1964). This may also be produced by condensation of lysyl residues with dehydroalanyl residues generated by the action of alkali on cysteinyl residues and from seryl residues linked O-glycosidically to carbohydrate as in κ-casein. Thus processes which involve the heating of proteins in alkali, such as in the production of spun soya protein for the preparation of analogue foods and in the manufacture of sodium caseinate, must be expected to produce lysinoalanine unless steps are taken to prevent this. In the case of protein preparations used for supplying texture to foods the formation of lysinoalanine may be desirable since the physical properties of the protein are advantageously altered by the cross-linking introduced in its formation. Where the product may form the basis of a high-quality nutritional preparation its formation is not desirable since the availability of lysine and possibly cysteine is reduced. Lysinoalanine has been regarded as toxic (Woodward, 1973) although subsequent reports have failed to confirm this (Van Beek, Feron & De Groot, 1974) and present opinion does not regard lysinoalanine as being positively toxic.

Before leaving the subject of heat treatment notice should be taken of the growing volume of reports, which suggest that milk is a coronary health hazard (Segal, 1977) and that the real site of this hazard resides in the antigenicity of heated milk proteins (Davies, Davies and Richards, 1969; Davies, Johnson, Rees, Elwood & Abernethy, 1974; Annand, 1971, 1972). While no direct chemical evidence is yet available to support or refute this hypothesis it is apparent from a study of the reactions already mentioned that milk proteins are particularly susceptible to modification by reaction with other milk constituents when heated even under the mildest conditions. Further study is required to determine which reactions result in valuable products and which represent limitations on their use as a food source.

I should like now to return briefly to the subject of acceptability of milk based products. Treatment of milk protein and of the caseins in particular with proteolytic enzymes has been shown frequently to give rise to digests which are bitter in taste (Murray & Baker, 1952; Harwalker & Elliott, 1965; Petritschek, Lynen & Belitz, 1972; Sullivan & Jago, 1972). This rendered them unpalatable and precluded their use for the therapy of disorders of protein digestion and absorption for which they were otherwise well suited. In one such digest which was designed to provide a source of nitrogen suitable for children suffering from cystic fibrosis it was found possible to eliminate this bitter taste by further digestion with hog kidney peptidases and also to extract material in which the bitterness was concentrated (Clegg, 1971). Further investigation of this material (Clegg, Lim & Manson, 1974) showed that all of the bitter taste could be attributed to a peptide composed of residues 53–79 of β-casein. Other peptides having bitter tastes have also been isolated by various treatments of casein but all have one chemical property in common, they are all strongly hydrophobic. This supports the contention put forward by Ney (1971) that the most important factor determining whether a peptide will be bitter in taste is its degree of hydrophobicity. From a

study of the sequences of amino acid residues occurring in the individual caseins it is possible to predict which regions can give rise to bitter-tasting peptides. It may be concluded that processes used for the preparation of partly digested dairy products from whole casein, unless capable of reducing parts of the β-casein molecule to very small fragments, will of necessity give rise to unpalatable and therefore unacceptable products.

Finally and by way of summing up, it is apparent that milk protein has unique nutritional characteristics which are at the moment presented in a number of acceptably traditional food forms. If a proper regard is paid to the chemical properties of these proteins there is no scientific reason why this number of acceptable food forms should not be dramatically increased and an even greater contribution be made to the diets of affluent and poor communities alike.

REFERENCES

Annan, W. D. & Manson, W. (1969). *J. Dairy Res.* **36**, 259.
Annand, J. C. (1971). *Atherosclerosis* **13**, 137.
Annand, J. C. (1972). *Atherosclerosis* **15**, 129.
Blaxter, K. L. (1968). *Science Journal* p. 53.
Bleumink, E. & Young, E. (1968). *Int. Arch. Allergy appl. Immun.* **34**, 521.
Bohak, Z. (1964). *J. biol. Chem.* **239**, 2878.
Clegg, K. M. (1971). Production of Pre-digested Protein Food. British Patent no. 1338936.
Clegg, K. M., Lim, C. L. & Manson, W. (1974). *J. Dairy Res.* **41**, 283.
Davies, D. F., Davies, J. R. & Richards, M. A. (1969). *J. Atheroscler. Res.* **9**, 103.
Davies, D. F., Johnson, A. P., Rees, B. W. G., Elwood, P. C. & Abernethy, M. (1974). *Lancet* i, 1012.
Donella, A., Pinna, L. A. & Moret, V. (1972). *FEBS Lett.* **26**, 249.
Ewart, J. A. D. (1967). *J. Sci. Food Agric.* **15**, 119.
FAO/WHO (1965). *Protein Requirements,* FAO Nutrition Meetings Report, Series no. 37, Rome.
Groves, M. L. & Gordon, W. G. (1970). *Archs. Biochem. Biophys.* **140**, 47.
Harwalker, V. R. & Elliott, J. A. (1965). *J. Dairy Sci.* **48**, 784.
Manson, W. & Annan, W. D. (1971). *Archs. Biochem. Biophys.* **145**, 16.
Manson, W. & Carolan, T. (1972). *J. Dairy Res.* **39**, 189.
Mercier, J.-C., Brignon, G. & Ribadeau Dumas, B. (1973). *Eur. J. Biochem.* **35**, 222.
Mercier, J.-C., Grosclaude, F. & Ribadeau Dumas, B. (1971). *Eur. J. Biochem.* **23**, 41.
Meyer, E. (1967). *Proceedings of the International Conference on Soyabean Protein Foods,* USDA Publication ARS–71–35, p. 142.
Murray, T. K. & Baker, B. E. (1952). *J. Sci. Food Agric.* **3**, 470.
Ney, K. H. (1971). *Z. Lebensmittelunters. u.-Forsch.* **147**, 64.
Petritschek, A., Lynen, F. & Belitz, H. D. (1972). *Lebensmittel Wissenschaft u Technologie* **5**, 47.
Ribadeau Dumas, B., Grosclaude, F. & Mercier, J.-C. (1975). *Modern Problems in Paediatrics* **15**, 46. Basle; S. Karger.
Segal, J. J. (1977). *Brit. J. prev. soc. Med.* **31**, 81.
Smith, J. A. B. (1970). In *Protein as Human Food,* [R. A. Lawrie, editor]. London: Butterworth.
Sullivan, J. J. & Jago, G. R. (1972). *Aust. J. Dairy Technol.* **27**, 98.
Van Beek, L., Feron, V. J. & De Groot, A. P. (1974). *J. Nutr.* **104**, 1630.
Woodward, J. C. (1973). *Fed. Proc.* **32**, 884.

Printed in Great Britain