Aspects of protein quality in calf nutrition. Problems and possibilities of milk protein substitutes

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The high cost of milk proteins limits the profits of veal production. The situation is made worse by an increasing demand for beef. For this reason the rearing of calves for veal production is expected to decline to one-third within the next 10 years. One method of solving some of these problems would be the partial or total replacement of milk proteins with suitable substitutes. This course of action may, however, generate new problems. The alternative protein sources might become more and more expensive. Also, there are many technical and physiological difficulties connected with the use of milk-protein substitutes. We shall have an increasing overproduction of milk protein which cannot be consumed by man alone; it is best utilized in veal production.

Protein retention and protein requirements in the calf

The requirements of the calf for digestible protein depend mainly on the age of the animal and the daily protein deposition in the tissues. The amounts of nitrogen retained for each kg gain in body-weight are relatively constant. Results of balance studies in our Institute (Zucker, Gropp, Giessler & Barry, 1969; Gropp, 1971) showed average values of 31.5 g N retained/kg body-weight gain. Further studies using the urea : creatinine ratio and urinary concentrations as an index of N retention gave an average value of 32 g N retained/kg body-weight gain. Only extremely fast-growing calves, which showed body-weight gains above 2 kg/day, retained not more than 28 g N/kg body-weight gain (Bochmcke, Gropp & Rieder, 1972; Brüggemann, Tiews, Gropp & Bochmcke, 1973). These results are similar to those reported by Brisson, Cunningham & Haskell (1957) and Roy (1970). Recent results from our laboratory show that no differences could be detected in the composition of muscle tissue from calves with growth rates between 1.0 and 1.6 kg/d (see Table 1).

We found that the minimum crude-protein content of milk-substituted diets which gave optimum development was 240 g/kg for calves up to 100 kg body-weight, and below 200 g/kg for calves weighing more than 100 kg (Zucker, Erbersdobler & Gropp, 1968; Zucker et al. 1969). These values are similar to those reported in more detail by Roy (1970), and also by Raven (1967) and van Weerden & van Hellemo (1967). The amino acid requirements of the veal calf have not been determined, but we can assume that—with the exception of lysine—they are similar to those of other monogastric animals; the requirement for lysine is apparently higher (van Loen & Balfoort, 1968; Zucker et al. 1968). This may be the result of either a real
Table 1. The water and protein content of muscle tissue (g/kg) (muscularis supraspinatus) from groups of calves with growth rates between 1.3 and 1.5 kg/d.

(Mean values and standard deviations for eight calves in each group)

<table>
<thead>
<tr>
<th>Composition of the muscle tissue</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>Daily body-weight gains (g) Mean</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>1280</td>
<td>1248</td>
</tr>
<tr>
<td></td>
<td>1469</td>
<td>1448</td>
</tr>
<tr>
<td>Water Mean</td>
<td>751</td>
<td>734</td>
</tr>
<tr>
<td></td>
<td>759</td>
<td>739</td>
</tr>
<tr>
<td>Protein (N × 6.25) Mean</td>
<td>196</td>
<td>202</td>
</tr>
<tr>
<td></td>
<td>202</td>
<td>200</td>
</tr>
<tr>
<td>SD</td>
<td>4</td>
<td>8</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>5</td>
</tr>
</tbody>
</table>

Table 2. The composition (g/kg) and the amino acid content of dried skim milk, dried whey and the fish-protein concentrate and the soya-bean-protein concentrate

<table>
<thead>
<tr>
<th>Dried skim milk*</th>
<th>Dried whey*</th>
<th>Fish-protein concentrate</th>
<th>Soya-bean-protein concentrate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (N × 6.25)</td>
<td>340</td>
<td>120</td>
<td>801</td>
</tr>
<tr>
<td>Fat (diethyl ether extract)</td>
<td>7</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Water</td>
<td>40</td>
<td>40</td>
<td>48</td>
</tr>
<tr>
<td>Ash</td>
<td>80</td>
<td>90</td>
<td>145</td>
</tr>
<tr>
<td>Calcium</td>
<td>12</td>
<td>9</td>
<td>46</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>10</td>
<td>7</td>
<td>24</td>
</tr>
<tr>
<td>Iron (ppm)</td>
<td>10</td>
<td>5-10</td>
<td>261</td>
</tr>
<tr>
<td>Amino acids (g/kg protein†)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lysine</td>
<td>92</td>
<td>89</td>
<td>83</td>
</tr>
<tr>
<td>Methionine</td>
<td>25</td>
<td>18</td>
<td>31</td>
</tr>
<tr>
<td>Cystine</td>
<td>12</td>
<td>21</td>
<td>13</td>
</tr>
<tr>
<td>Tryptophane</td>
<td>13</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Threonine</td>
<td>48</td>
<td>61</td>
<td>44</td>
</tr>
<tr>
<td>Leucine</td>
<td>105</td>
<td>96</td>
<td>78</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>66</td>
<td>61</td>
<td>47</td>
</tr>
<tr>
<td>Valine</td>
<td>67</td>
<td>54</td>
<td>49</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>45</td>
<td>45</td>
<td>39</td>
</tr>
<tr>
<td>Tyrosine</td>
<td>44</td>
<td>31</td>
<td>32</td>
</tr>
<tr>
<td>Arginine</td>
<td>41</td>
<td>23</td>
<td>55</td>
</tr>
<tr>
<td>Glycine</td>
<td>20</td>
<td>25</td>
<td>55</td>
</tr>
</tbody>
</table>

nd, not determined.

*Products of best quality containing only traces of furosine.
†(N × 6:25).

demand for protein synthesis or because lysine has other important functions in the metabolism of the calf. It is interesting that the lysine content of cow's milk protein is remarkably high.

Table 2 compares the composition and the amino acid content of milk products with that of other protein sources to be discussed later.

The digestion of food proteins and aspects of protein quality

The details and characteristics of the digestion of food proteins in the preruminant calf have been reviewed extensively by Porter (1969), Roy (1970) and Radostits &
Bell (1970). These authors emphasize that one of the most important factors in the digestion of food proteins and in the compatibility of the liquid diet is the slow release of the proteins from the abomasum. Normally only the free amino acids and other soluble nitrogenous compounds are released immediately from the stomach, followed later by the whey proteins and finally by the caseins (Tagari & Roy, 1969). It is detrimental, therefore, to the calf if a significant proportion of the food proteins leaves the stomach too rapidly. Many of the milk-protein substitutes do not clot sufficiently in the abomasum, and are consequently released from the stomach quickly. This is most important for the very young calf which secretes predominantly rennin instead of pepsins in the stomach. As only casein can be regarded as a specific substrate for the action of rennin, there may be many complications if casein is substituted by other proteins.

Fig. 1 shows the influx of lysine and arginine into the portal vein after a test meal of liquid diets containing milk protein or a mixture of milk proteins and a fish-protein concentrate. The diet containing fish protein was less palatable and only consumed with delay. In the test meals therefore a 1:1 mixture with a skim milk preparation was used. The lysine concentrations increased more rapidly (within the first hour) after the meal containing the fish-protein concentrate. This might be partly the result of the higher concentration of free lysine in the tested fish protein, which represented about 40% of the total protein in the test meal.

On the other hand similar results were obtained with arginine indicating that in the calf given fish meal, part of the protein (either the whey or the fish protein) was released earlier from the stomach.

In the following 8 h, there were differences in the lysine concentrations and, to a lesser extent, in the arginine concentrations between the two groups, suggesting that the course of digestion of the two types of milk substitute diet differed. In the growth experiment the calves received the original milk-substituted diet containing fish protein and not the mixture. The growth rates obtained with the diet containing the fish protein were fairly good, with body-weight gains of 1.15 kg/d and a feed conversion ratio of 1.71 kg food intake/kg body-weight gain for the fish-protein group compared with 1.2 kg body-weight gain and 1.59 kg food intake/kg body-weight gain for the milk-protein group. The values are means from eighteen calves, whose initial body-weights were about 67 kg at the beginning of the 11-week experiment.

There are difficulties when giving diets containing heat-damaged milk proteins. Roy and his co-workers (Roy, 1970) found that heat damaging of the milk proteins results in a reduction in the nutritive value of the milk diet for the young calf and increases the incidence of diarrhoea. These nutritional defects may result in part, from an increase in the clotting time of the casein by rennin. Also, heating proteins in the presence of glucose or lactose results in the formation of lysine-sugar complexes, in large amounts, in the initial stage of the so-called Maillard reaction (Brüggemann & Erbersdobler, 1968; Erbersdobler, 1970). During the heat treatment of milk products lactulosyllysine (galactose-fructosyllysine) is the main product formed, in which the sugars are linked at the ε-amino group and the α-amino group.
Fig. 1. The amounts of lysine (a) and arginine (b) in the portal plasma of a female veal calf, weighing 100–110 kg, 1–9 h after a test meal of liquid diets containing 800 g milk substitute* (24% protein) in 4 l water. —— meal with (g/kg) 950 milk protein and 50 whey protein; ——— meal with (g/kg) 480 milk protein, 120 whey protein and 400 fish protein.

*The milk substitutes contained (g/kg): 180 fat, 30 cooked starch, 20 minerals, vitamins and feed additives and 670 dried skim milk + 100 dried whey, or 340 dried whey + 230 fish-protein concentrate (see Table 2). One meal contained the milk substitute with the milk proteins above; the other meal contained 1:1 by wt of the two milk substitutes. The milk substitute with the fish-protein concentrate was supplemented with 1 g L-lysine HCl/kg.
is bound in the protein structure. Fructosyllysine or lactulosyllysine, which are very unstable to acid hydrolysis, can be estimated by analysing a compound formed by hydrolysis with hydrochloric acid. We found this useful indicator some years ago (Erbersdobler & Zucker, 1966; Erbersdobler, 1970). Heyns, Heukeneshoven & Brose (1968) and Finot, Bricout, Viani & Mauron (1968) have determined the structure of this compound, and named it furosine (ε-N-(2-furoyl-methyl)-L-lysine).

The following scheme shows the initial steps of the Maillard reaction.

Several intermediates

\[
\text{Protein} \rightarrow \text{LYSINE} + \text{GLUCOSE} \rightarrow \text{(Galactose)}
\]

\[
\text{(Galactose)} \rightarrow \text{FRUCTOSYL-LYSINE} \rightarrow \text{FUROSINE} \rightarrow \text{Brown-coloured polymers}
\]

\[
\text{HCl} \quad \text{(Indicator)}
\]

Table 3 shows some results of determinations of the lysine and inactivated-lysine contents of some milk products used as milk substitutes.

These lysine-sugar complexes are not utilized by the animals and, if protein-bound, are poorly digested and absorbed (Brüggemann & Erbersdobler, 1968; Erbersdobler, 1971; Erbersdobler & Dümmer, 1971; Ford & Shorrock, 1971; Finot, 1973). Large amounts of non-digestible peptides containing the lysine-sugar complexes may, therefore, pass into the lower gut. However the microflora of the digestive tract are able to utilize these undigestible protein fragments (Erbersdobler, Gunsser & Weber, 1970), and consequently changes in the composition of the intestinal flora, and signs of incompatibility, may result (Gedek, 1969; Erbersdobler, 1972).

**Table 3. The lysine and furosine contents and the calculated amounts of inactivated lysine g/kg protein* in some commercial milk products**

<table>
<thead>
<tr>
<th></th>
<th>No. of samples</th>
<th>Lysine†</th>
<th>Furosine</th>
<th>Inactivated lysine†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spray-dried skim milk</td>
<td>31</td>
<td>85</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>Roller-dried skim milk</td>
<td>25</td>
<td>81</td>
<td>8</td>
<td>11</td>
</tr>
<tr>
<td>Spray-dried whey</td>
<td>25</td>
<td>83</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>Roller-dried whey</td>
<td>14</td>
<td>57</td>
<td>29</td>
<td>41</td>
</tr>
<tr>
<td>Whey concentrates dried§</td>
<td>26</td>
<td>75</td>
<td>15</td>
<td>21</td>
</tr>
<tr>
<td>Diets for veal calves</td>
<td>31</td>
<td>81</td>
<td>5</td>
<td>7</td>
</tr>
</tbody>
</table>

*(N x 6.25).

†Partially unavailable (see the amounts of inactivated lysine).

‡Calculated from the amount of lysine bound in furosine (the yields of furosine from lysine-sugar complexes are 40%).

§Partially deprived with lactose.
Results of feeding experiments with calves indicated that when a heat-damaged milk product made up only part of the food, lower body-weight gains and a reduced feed conversion ratio resulted (Fender, 1969; see also Erbersdobler, 1972). Fender demonstrated clearly that a dried skim-milk which was heat-damaged (70% of the lysine inactivated) was not tolerated in amounts exceeding one-third of the diet in young calves. Greater quantities produced a severe incidence of diarrhoea. Similar results were obtained by Weiss, Baur & Schiefer (1967).

Substitution of milk proteins

The substitution of milk proteins with other protein sources in liquid diets for veal calves were not at first successful. The results of earlier optimistic reports are difficult to compare with ours, as in these experiments the level of food intake was low and values for body-weight gains were only 300–500 g/d (e.g. Gorill, Thomas, Steward & Morill, 1967; Huber & Slade, 1967; Colvin & Ramsey, 1968).

Higher body-weight gains and a better performance by the calves was obtained when partially- or completely-hydrolyzed proteins were used (Bonsembiante & Parigi-Bini, 1967; Burgstaller, 1972). As this processing technique is rather expensive, generally, protein sources of poor quality are hydrolyzed. It was found, however, that only small amounts of these hydrolyzates could be tolerated in the diet.

The use of fish flour has also been investigated. After the first, not very encouraging, experiments, better results were obtained with fish concentrates which had been defatted and specially treated (van Hellemond, 1967; van Weerden, 1970; Hallgreen, Sjöberg & Stelleman, 1973). Results of experiments done in our Institute have been discussed above. The digestibility and protein quality of the products available now seem to be somewhat inferior to the milk proteins. Also most fish-protein concentrates contain appreciable amounts of iron. They are, for this reason, mainly suitable for calf rearing and not for feeding to veal calves (see also Gropp, Beck & Erbersdobler, 1973).

Still more problems arise when vegetable proteins are used. The soya-bean products contained inhibitors of digestive enzymes and other growth-depressing factors (Colvin & Ramsey, 1968; Liener, 1973). The isolated soya-bean protein contains appreciable amounts of these heat-labile factors. In our studies with rats (Erbersdobler, Weber & Gunssser, 1972) we found that a commercially-isolated soya-bean protein was digested to a lesser extent than the same product after mild heat-treatment. Porter (1969) reported values for the apparent digestibility in calves of only 75% for soya-bean protein. Difficulties also may result from the production of anti-soya-bean-protein antibodies in calves given milk substitutes containing certain soya-bean products (van Adrichem, 1967; Smith & Wynn, 1971). After several weeks other metabolic disturbances and diarrhoea may result from these incompatibility reactions.

More recent experiments have shown, however, that acid- or alkali-treatment of soya-bean products or enzymic pre-digestion of samples markedly improved their food value when they were included in diets given to pre-ruminant calves (Colvin & Ramsey, 1968, 1969). In our Institute, a soya-bean-protein concentrate from
Israel has been studied and it was found that a large proportion of the milk-proteins can be substituted with this type of specially-prepared product. The nutritive value of the soya-bean meal has been improved, mainly by extracting the carbohydrates of high molecular weight and finely grinding the meal. However, the protein digestibility and the protein quality of this product was still inferior to that of milk proteins (Chassin, 1969; Nitsan, Volcani, Gordin & Hasdad, 1971).

Results shown in Table 4 suggest that rations containing 34% or 68% of the total protein derived from the soya-bean-protein concentrate produced good growth rates which were only 8% below the body-weight gains obtained with milk proteins alone. Similar results were obtained from measurements of the feed conversion ratio. The small differences in performance may result from the somewhat lower digestibility of the soya-bean product studied, or from residues of enzyme inhibitors and other growth-depressing factors. Nitsan et al. (1971) were able to detect enzyme inhibitor activity in this product and, therefore, other deleterious factors, like the heat-labile phytohaemagglutinins (Liener, 1973) which occur in the raw soya-bean, may also be found. The results, however, were very satisfactory, and, compared with those from earlier studies were very encouraging.

Conclusions

Cow’s-milk protein is of very high quality and optimum digestion of protein is obtained when it is given in the diet. However, it is easy to damage milk protein by processing which results in changes in the process of digestion and possibly signs of incompatibility. Milk protein in milk-substitute diets can now be substituted with specially-treated protein concentrates from fish products or vegetable sources. However, the most promising studies were done mainly with calves with body-weights greater than 60 kg and not with the highly-sensitive, very young calf. In our country two special milk-substituted diets have been used for veal calves, one for the young
calf up to 90-100 kg, and one with a lower protein content for older calves. The milk-protein substitutes are most suitable for this second type of milk-substituted diet.

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Printed in Great Britain