The relative nutritive value of irradiated spray-dried blood powder and heat-sterilized blood meal as measured in combination with whey protein

T.E.H. Downes*, L.D. Nourse, F.K. Siebrits
Animal and Dairy Science Research Institute, Private Bag X2, Irene, 1675 Republic of South Africa
J.W. Hastings
Radiation Technology Division, Nuclear Development Corporation of South Africa (Pty) Ltd.

Received 15 November 1985

A method of processing blood meal in which the nutritive value of the protein is preserved is described, since appreciable losses occur in the nutritive value of the protein when prepared by heat sterilization with drying at atmospheric pressure in steam jacketed vessels. Blood was spray dried and irradiated at an intensity of 10 kGy. Collectively the heat of spray drying and irradiation was effective in killing both the virus plaque-forming units and the bacteria, thus producing a commercially acceptable sterile product of higher nutritive value. The relative nutritive values (RNV) of 50:50 protein mixtures were 0,56 for whey protein concentrate plus heat-sterilized blood meal and 0,90 for whey protein concentrate plus irradiated spray-dried blood powder. Whey protein concentrate used as a control has a RNV of 1,0.

Omdat die hitte-sterilisasieproses in die vervaardiging van bloedmeel die voedingswaarde van die proteïen aansienlik verlaag, is 'n alternatiewe manier van prosessering getoets. Bloed is gesproeidroog en bestraal met 'n intensiteit van 10 kGy en daar is gevind dat die proses voldoende was om bakterieë en virusse te vernietig sonder strawwe hittebehandeling. Op dié manier was dit moontlik om die relatiewe voedingswaarde van die bloedproteïen te verhoog van 0,56 vir gewone bloedmeel en weiproteïen, tot 0,90 wanneer in 50:50 mengsels sproeigedroogde bloed- en weiproteïen gevoer word, teenoor die kontrole van weiproteïen alleen van 1,0.

* To whom correspondence should be addressed

Introduction

It has been shown that vat-dried blood meal (Prepared by heat sterilization with drying at atmospheric pressure in steam-jacketed vessels) has a nutritive value much less than would be expected from an analysis of the amino acid content of fresh blood because the biological value of both lysine and methionine were reduced to between 0 – 43% of total lysine and methionine values (Waibel, Cuperlovic, Hurrell & Carpenter, 1977). It would appear that a similar deterioration pertains in the commercially prepared blood meals in South Africa. Unpublished work carried out at the Animal and Dairy Science Research Institute by G.A. Smith showed that 30 – 40% of the nutritive value was lost when compared to spray-dried blood powder (SDBP) in commercial feed preparations containing heat-processed blood powder (L.D. Nourse, unpublished observations 1975 – 1981). A negative protein efficiency ratio (PER) results when blood protein is used as a sole source of protein in a diet due mainly to a low isoleucine content. When fed in a 50:50 protein mixture with whey protein a positive PER of 2,99 was obtained (Downes, Cruywagen, Smith & Pelster, 1882).

In the commercial process blood meal is sterilized because of the temperature conditions used. Spray-dried blood powder is not subjected to high temperatures during preparation. Under commercial abattoir conditions it is not possible to collect blood only from healthy animals, unless an elaborate system of blood collection is installed, so an alternative method of sterilization involving irradiation was tried. The purpose of this project was thus to test the efficiency of spray drying and irradiation on the relative nutritive value (RNV) and sterility of blood protein.

Materials and Methods

Preparation of spray-dried protein sources

Whey protein was produced by removing a large proportion of the lactose by ultrafiltration and spray drying the resultant protein concentrate. Blood was spray dried at an air-inlet temperature of 160°C and outlet temperature of 80°C. During spray drying the temperature of the blood does not exceed the outlet temperature of the dryer due to the rapid evaporation of water in the warmer parts of the dryer. The powder is therefore exposed to a temperature of 80°C for a few seconds during the process.

Stock cultures of *Bacillus subtilis* ATCC 6633 were maintained on Nutrient Agar (Difco) slants, which were incubated at 37°C for 2 days and stored at 4°C until needed. Spore suspensions of these were prepared according to Busta (1966) and added in a concentration of between 8×10^3 and 1×10^4 per ml before spray drying. These organisms were chosen as marker organisms because of their very high resistance to destruction by heat treatment (van der Heever, personal communication). The blood powder produced was then irradiated at levels of 10, 20, 45 and 50 kGy and tested for survival of the *Bacillus* organisms. It is, however, known that viruses are very resistant to irradiation but more sensitive to heat treatment. For this reason blood was contaminated with 1×10^3 Bluetongue virus (vaccine strain 8) and $1 \times$

10^4 Banzi virus plaque-forming units per ml before spray drying and irradiation. The blood powders were then tested for bacterial and virus sterility before and after irradiation.

Bacterial enumeration

Standard plate counts were made in accordance with the standard method of the International Dairy Federation(IDF, 1981) and the plates incubated for 72 h at 30°C. Spore suspensions were subjected to heat treatment of 80°C for 10 min and the total aerobic spores counted on nutrient agar (Biolab) after 72 h at 30°C. Brewer's Thioglycollate medium (Merck) was used for total anaerobic spore counts. Plates were Incubated in BBL anaerobic jars (Gas Pak Anaerobic System) at 37°C for 72 h. The coliform count was determined on violet red bile-lactose agar (Biolab) after incubation of plates at 30°C . Faecal streptococci were counted on citrate-azide-tween-carbonate agar (Merck) and incubated at 30°C for 72 h. For *Salmonella* 10 g of the blood powder was placed in lactose broth (Merck) for 4 h at 30°C. 10 ml of this broth was then dispensed into 90 ml of Selenite enrichment broth (Merck) and incubated at 34°C for 72 h. Every 24 h a loopful was streaked onto xylose-lysine-deoxycholate (Merck), and Salmonella-Shigella media (Merck), incubated at 37°C for 48 h and observed for typical colonies.

The products were tested for virus survival by titration in cell cultures and intracerebrally in day-old mice.

Determination of relative nutritive value (RNV)

The multi-point slope ratio assay of Hegsted, Neff & Worcester (1968) as described by Siebrits, Esterhuyse & Kemm (1986) with male Wistar rats was used. Body composition was determined by the method of Bell & Stern (1977), without the removal of intestinal contents. Body protein gain was chosen as a measure of response to dietary protein. At 28 days of age 65 rats were fasted for 24 h, after which 11 were asphyxiated and used to determine the initial body composition. The remaining 54 rats were divided into 13 groups of equal mass, one group consisted of six rats and received a protein-free diet, while the remaining 12 groups of four rats each were allocated to the four sources of protein under test, the diets from each source being formulated to contain 3,0; 6,0 and 9,0% protein. The proteins used were whey protein alone (control), and 50:50 protein combinations of protein from whey with either abattoir blood meal, SDBP or irradiated SDBP. The diets were formulated to be isocaloric at the 3,0; 6,0 and 9,0% protein levels. To obtain the 50:50 protein mixtures of the blood proteins and whey proteins, 282 g of SDBP (84,06% protein) and 518 g of whey protein concentrate (45,8% protein) were mixed. Similarly an amount of 325,5 g of abattoir blood meal (73,0% protein) was mixed with 524,5 g whey protein concentrate. The composition of the various diets is presented in Table 1.

These experimental diets were fed *ad libitum* from 29 to 41 days of age, whereafter the rats were again fasted for 24 h, asphyxiated, and their body composition determined. The experimental period lasted 14 days instead of 21 days as the rats on the poor protein diets became very weak.

The rats were individually housed in metabolism cages, enabling daily collection of faeces and spillage. The differences between the regression lines were determined by analysis of covariance as described by Siebrits, *et al.* (1986).

Results and Discussion

Bacterial counts were carried out in two independent laboratories and the results were similar. Spray drying was not effective in killing all of the bacteria present or added to the blood before it was spray dried as shown in Table 2 under the column of zero irradiation where it is seen that faecal *Streptococci* and *B. subtilis* are still present. Coliforms and *Salmonella* sp. do however appear to be absent after spray drying. Radiation was, however, effective in removing all the bacterial strains present in or added to the blood, even at the lowest dosage tried, as shown in Table 2.

The two virus strains added to the blood, namely the

Table 1 Composition of various diets (%)

	Protein source									
	Source A & B			Source C			Source D			
Dietary component	3%	6%	9%	3%	6%	9%	3%	6%	9%	0%
SDBP + whey protein concentrate	5,06	10,12	15,18							–
Blood meal + whey protein concentrate				5,31	10,62	15,93				–
Whey protein concentrate							6,55	13,10	19,65	–
Starch	79,44	74,88	69,82	79,69	74,38	69,07	78,45	71,90	65,35	85
Maize oil	10	10	10	10	10	10	10	10	10	10
Vitamins and mineral premix	5	5	5	5	5	5	5	5	5	5

Source A & B — SDBP and irradiated SDBP plus whey protein concentrate
Source C — Abattoir blood meal and whey protein concentrate
Source D — Whey protein concentrate

Table 2 Bacterial counts in spray-dried blood protein (SDBP) before and after irradiation at four levels of treatment

	Radiation dosage (kGy)				
	0	10	20	45	60
Bacterial group	Counts (per g sample)				
Total aerobes	$1,0 \times 10^5$	<10	<10	<10	<10
Total aerobic spores	9×10^3	<10	<10	<10	<10
Total anaerobic spores	<10	<10	<10	<10	<10
Faecal streptococci	$3,7 \times 10^2$	<10	<10	<10	<10
Coliforms	<10	<10	<10	<10	<10
Salmonella sp.	0	0	0	0	0
B. subtilis	$6,0 \times 10^4$ $7,0 \times 10^4$	0	0	0	0

Table 3 The relative nutritive values (RNV) of the diets of proteins processed by the various treatments in combination with whey protein

	Parameter		
Protein source	Slope	*SE*	RNV
Whey protein concentrate	0,8541[a]	0,4735	1,00
SDBP + whey protein concentrate	0,7148[b]	0,4531	0,84
Irradiated SDBP + whey protein concentrate	0,7701[b]	0,2993	0,90
Abattoir blood meal + whey protein concentrate	0,4799[c]	0,4162	0,56

Different superscripts denote significantly ($P<0,05$) different slopes
SE—Standard error of regression coefficient

hardy Bluetongue virus (vaccine strain 8) and the more fragile Banzi virus, were completely inactivated during the spray-drying process because neither could be detected in SDBP, before and after irradiation, by titration in cell cultures or intracerebrally in day-old mice. This result indicates that this heat treatment of 80°C for a few seconds was sufficient to inactivate the virus plagues.

The results obtained on the determination of RNV of different proteins are presented in Table 3. Whey protein had a significantly better RNV ($P<0,05$) than the SDBP, both with and without irradiation. The RNV of the abattoir blood meal protein was significantly lower ($P<0,05$) than any of the other protein sources.

The slightly higher RNV of the irradiated SDBP protein over non-irradiated SDBP protein was not significant but demonstrates that irradiation does not alter the nutritive value. The criticism discussed by McLauglan & Keith (1975) against including a zero-protein group in the calculation of the respective slopes due to curvilinearity does not affect the present results since no significant curvilinearity was found and thus it was decided to include the data of the protein-free group. This also increased the range of protein intake since the palatability of the abattoir blood meal was low.

The indications are that palatability is a problem with the whey-abattoir blood meal diet as the rats showed immediate rejection when changed to this diet, with only limited acceptance thereafter. This did not occur with the SDBP diets. It also resulted in a lower protein intake as illustrated in Figure 1.

The severe heat treatment of abattoir blood meal resulted in such a dramatic reduction in the nutritional value that 2,25 g of blood meal protein plus 2,25 g of whey protein resulted in a loss of body protein whereas 2,25 g of whey protein alone gave a gain in body protein (Figure 2).

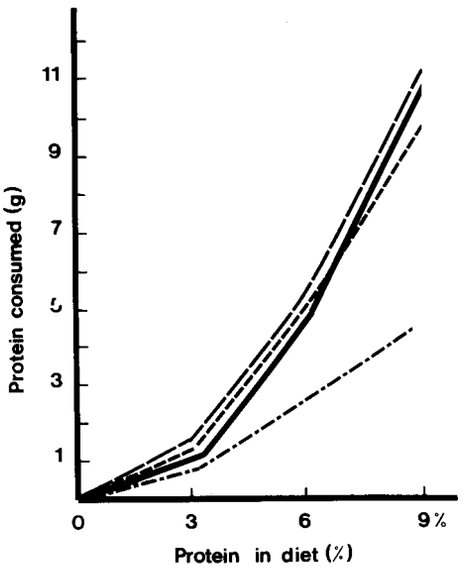

Figure 1 Influence of protein source on protein consumption (—— Whey + irradiated SDBP; ------ whey + SDBP; — — — whey; — - — whey + abattoir blood meal)

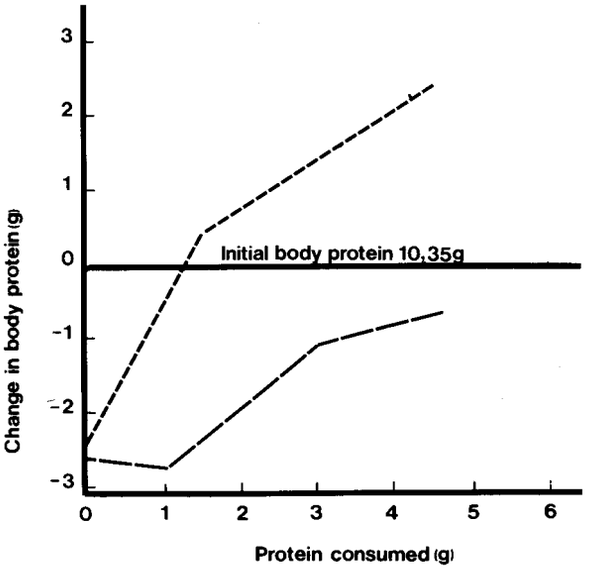

Figure 2 Comparison between blood meal + whey protein and whey protein alone in relation to protein consumption and changes in body protein (------ Whey protein; — - — blood meal + whey protein)

Conclusions

The results of this investigation revealed that spray drying plus irradiation at 10 kGy, results in a commercially sterile product, and due to the great improvement in the relative nutritive value in using this technique, the regulations prohibiting the use of non heat-sterilized blood meal in animal nutrition should be changed. This is of importance due to the anticipated shortage of protein in South Africa in the future. The cost of production of irradiated SDBP would be higher than that of conventional blood meal, but the increase in RNV would result in an increase of the nutritive value of the product, which according to present estimates, would more than compensate for the increased cost of production. Furthermore abattoir blood meal, although useful as a source of protein for ruminants, has very little value in the nutrition of monogastric animals, but spray-dried blood could be very useful for the latter.

A further possibility is that the blood be separated into plasma and blood cells by centrifugation and then treated separately by the same process. The plasma powder would have a value comparable to soya isolate or sodium caseinate in the production of processed meats.

In the separation into plasma and blood cell fractions, 60% plasma with a solids content of 8% and 40% corpuscles with a solids content of 38% is obtained. Therefore, from 100 kg of blood 4,8 kg of plasma powder would be produced plus 15,2 kg corpuscle powder. Blood cell powder has a lower isoleucine content (0,6% in the protein) than whole blood protein (1,04% in the protein) so that in using blood cell powder for animal nutrition more isoleucine-rich sources of protein will be needed to balance the essential amino acid composition of the diet.

Acknowledgements

Dr B.J.H. Barnard of the virology department of the Veterinary Research Institute, Onderstepoort, is thanked for his advice, the supply of virus material and subsequent analysis of the product, while Dr I.F. Mostert, Senior Research Officer of the Microbiology Section of the ADSRI is also thanked for the preparation of the culture of *B. subtilis* and the bacterial analysis of the product. The assistance of Mr J.C. Pelster, also of the ADSRI, in preparing the blood and whey protein powders was very much appreciated.

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