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STUDIES ON THE PREPARATION OF PROTEIN ENRICHED SOFT DRINK FROM MUSTARD SEED CAKE

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A protein enriched soft drink, containing various levels of mustard seed protein, was prepared. Protein dispersibility of the heat processed beverage was found to be maximum at pH 6.8. Fat fortification also helped in stabilizing the beverage. Addition of 0.6 % carboxy methyl cellulose resulted in maximum protein dispersibility in the beverage.

Key words: Mustard seeds, protein isolate, soft drink.

INTRODUCTION

Beverages, indigenous or imported, are relished very much by the people of this region. Preference for different beverages, however, is based on social background and traditional customs. "Lassi" (Butter milk) has been a traditional drink of Pakistanis especially of the rural population for a long time. This beverage, being rich in nutrients, such as protein and minerals, is a refrigerant and suits the climatic conditions of this region. However, with an increase in population and short supply of milk, the consumption of this drink is dwindling. Consequently, the demand for cola type beverages, which contain only a few minerals and little energy, is increasing day by day. The situation demands that a drink with most of the qualities of "Lassi" should be developed.

Detoxified mustard seed meal produced by the method of shah *et al.* [10], which had amino acid profile similar to soy bean [4], was used for the production of a drink similar to "Lassi" in the course of these investigations.

MATERIALS AND METHODS

Mustard seeds (cultivar RL-18) were purchased from Ayub Agricultural Research Institute, Resalewala. Oil was extracted and meal was detoxified following the procedures of Shah *et al.* [10].

(a) *Protein isolation techniques.* Protein was isolated by the methods of Girault [5], Lonnerdal *et al.* [6] and Bhatia *et al.* [2]. These methods were modified in the light of the results obtained. Detoxified mustard seed meal (100 mesh) was suspended in water (1 : 7 w/v) and pH of the suspension was adjusted (9.5 pH), and protein isolate was prepared following the procedure of Shah *et al.* [10].

(b) *Preparation of beverage.* Wet protein isolate was used in the beverage formulations. A beverage, containing mustard seed protein isolate containing 1.5, 2.0, 2.5 and 3.0 %, and 4.0 % sucrose was prepared. This preparation was fortified with 3.5 % vegetable oil, 0.6 % mineral mix [7], vitamins (A 180 IU, B_1 50 μg, Riboflavin 200 μg) and containing carboxy-methyl-cellulose (CMC) 0.2, 0.4, 0.6, 0.8 and 1.0 %. The prepared samples were filled in (210 ml capacity) glass bottles, crown corked and processed in a vertical retort under 15 lb. pressure for 10 minutes. The beverage was analysed for its protein content following the A.O.A.C. [1] method. Amino acids of the mustard seed protein isolate were determined as described by Wilkinson *et al.* [12].

RESULTS AND DISCUSSIONS

(a) *Effect of isolation technique on yield and solubility of protein.* Techniques applied by Bhatia *et al.* [2], Lonnerdal *et al.* [6] and Girault [5] resulted in protein isolation upto 63.64, 60.08 and 47.45 % respectively (Table 1), were used for the protein isolation from detoxified mustard seed meal. Adjusting pH of peptization to 9.5 and that of precipitation to 5.0 proved to be better techniques for optimum protein extraction (64.82 %). This method was found to be simple, efficient and less time consuming as compared to the techniques of Lonnerdal *et al.* [6] and Girault [5] in which proteins were precipitated in three steps by gradually lowering the pH to 4.9 and 5.6 respectively. It may be observed from Table 1 that although the yield of protein isolate, through the Girault [5] technique, was the lowest (47.45 %), yet it had the highest solubility (82.14 %) at pH 6.8. However, in real terms this value represents only 38.98 % of the total protein present

Table 1. Effect of isolation technique on the yield and solubility of protein.

Isolation technique	Parameter tested	
	Protein yield (%)	Protein solubility (%)
Girault (1973) method	47.45	82.14
Lonnerdal *et al.* (1977) procedure	60.08	71.34
Bhatia *et al.* (1966)	63.84	62.78
Technique applied in the present investigation	64.82	78.13

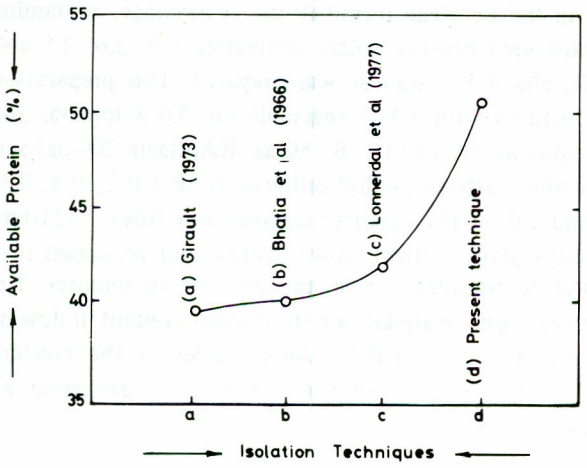

Fig. 1. Effect of isolation technique on the availability of protein.

in the meal. It is evident from Fig. 1 that the available protein was considerably higher by the improved method although the solubility of isolated protein was apparently lower (78.12 %) than the isolate prepared by Girault [5] procedure (82.14 %). The other two techniques also yielded lesser amount of soluble proteins than the improved method.

(b) *Quality evaluation of protein isolate.* Amino acid profile of the proteins isolated from detoxified mustard seed meal was comparable with that of soybeans except lysine (Table 2). Soybeans has been reported to contain 6.08 % lysine [4] while it was 4.80 % in mustard seed protein (MSP). However, MSP contained more methionine (2.40 %) as compared to soybean protein (1.42 %). Pattern of other essential amino acids in both the cakes was almost comparable (Table 2).

Table 2. Amino acid composition of mustard seed and soybean proteins (% of protein in g/100 g)

Amino acid	Mustard seed protein	Soybean* protein
(A) *Essential*		
Lysine	4.81	6.08
Methionine	2.40	1.42
Valine	5.04	5.23
Leucine	7.19	7.82
Isoleucine	4.26	4.83
Phenylalanine	4.27	5.01
Threonine	4.29	4.27
Histidine	2.75	2.54
Arginine	6.62	7.04
(B) *Non-essential*		
Cystine	1.64	1.60
Aspartic acid	6.98	11.50
Serine	4.36	5.55
Glutamic acid	14.99	18.51
Proline	5.62	5.62
Glycine	4.93	4.45
Tyrosine	3.24	3.79
Alanine	4.37	4.54

*F.A.O. Report-24, Rome (1970)

(c) *Stability of the protein-enriched beverage:* (i) *Effect of pH on the stability of beverage.* Protein solubility increased from 65 to 90 % when the pH was raised from 6.5 to 7.0. However, increase in pH resulted in a decrease in the protein solubility on heat processing of the beverage. Protein solubility was observed to be 88.62 and 67.54 % at pH 6.5 and 7.0 respectively (Table 3). Solubility of protein in the processed beverage was optimum at pH 6.8. Thus both solubility as well as availability of protein was found to be maximum at pH 6.8. These results do not agree with the findings of Priepki *et al.* [9] and Elahi *et al.* [3] who reported higher pH values for these types of beverages. However, these researchers used the proteins from vegetable sources other than mustard seeds.

(ii) *Effect of heat processing of the beverage.* Heat processing of the beverage resulted in a decrease in the solubility of protein (Fig. 2). This may be attributed to coagulation of proteins on processing. The decrease in the solubility was greater in samples containing higher percentage of protein (3.0 %). Similar loss in the stability of peanut-based beverage has been reported by Elahi *et al.* [3].

Table 3. Effect of change in pH on the protein solubility and protein retension on heat processing of the beverage.

Parameter tested	pH					
	6.5	6.6	6.7	6.8	6.9	7.0
Solubility (%)	65.43	68.48	73.10	78.61	84.12	90.06
Retension* after processing (%)	88.62	84.78	82.30	78.02	72.51	67.54
Total available protein (%)	57.98	58.06	58.70	61.33	60.89	60.83

*Amount of protein remaining soluble on heat processing of the beverage.

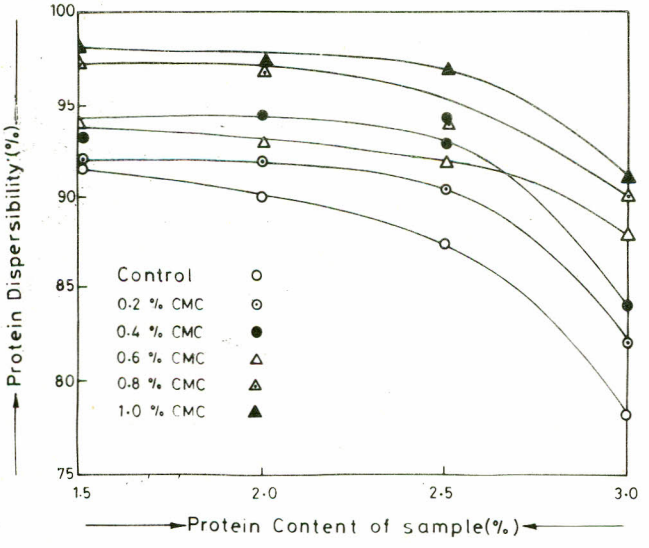

Fig. 2. Combine effect of emulsifier (CMC) and processing on stability of protein suspension in the beverage.

(iii) *Effect of fat fortification.* Fat fortification effect, as regards protein stability, was found to be more pronounced in the samples with higher protein contents (3.0 %) than the beverage containing lower amount of protein 1.5 % (Fig. 3). Protein solubility in the fat-free and fat-fortified samples, with 1.5 % protein was almost equal, while the fat-fortified beverage containing 3.0 % protein showed almost 5 % more soluble protein than the fat-free sample with the same level of protein. These results are in agreement with the observations of Mustakas [8], Priepki *et al.* [9] and Tornberg and Hermansson [11], who reported that lipid protein beverage had a good suspension stability.

(iv) *Effect of emulsification.* Addition of emulsifier beyond 0.6 % did not show any significant emulsifying effect both in fat-free and fat-fortified samples containing

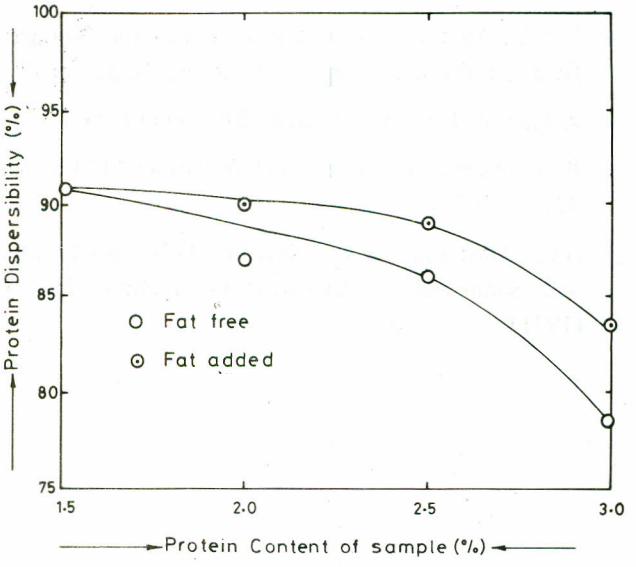

Fig. 3. Effect of fat on stability of the beverage.

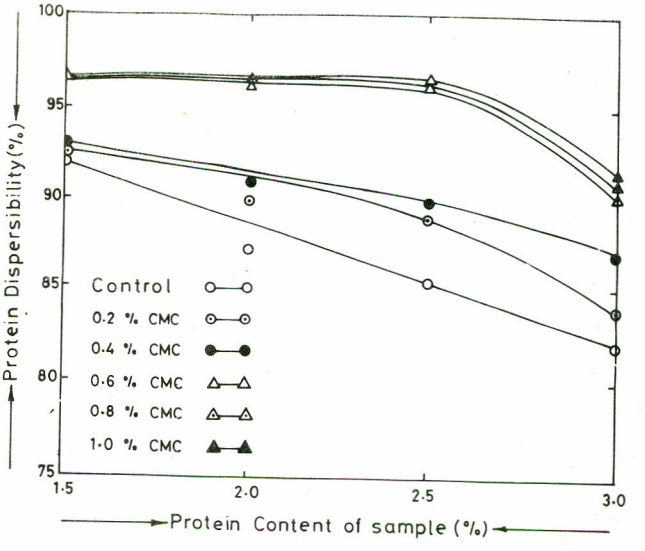

Fig. 4. Combine effect of fat and emulsifier (CMC) on protein stability of the beverage.

1.5 % and 2.0 % protein. However, fat-free samples showed improved dispersibility with the addition of CMC even beyond 0.6 % in case of 2.5 % and 3.0 % protein beverages (Fig. 4).

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