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### To evaluate the nutritional virtues of geriatric food by employing bioassay's study of geriatric health beverage

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#### Abstract

The research was carried out to evaluate the nutritional virtues of geriatric food by employing bioassay's studies of geriatric health beverage. Geriatric health beverage was prepared by using cow milk, ragi malt (2, 4 and 6%) whey protein concentrate (3, 5 and 10%), lactoferrin (15, 20 and 25 ppm), bacteriocins (10, 15 and 20 ppm), and probiotics (*L. acidophilus* LA5 and *B. bifidum* BB12 (1:1) (Mixed culture at, 2, 3 and 4%) which was To evaluate the nutritional virtues of geriatric food by employing bioassay's study of geriatric health beverage. Finally, based on these characteristics, geriatric health beverage was prepared. The study concluded that the geriatric health beverage prepared using cow milk, ragi malt (4 percent), whey protein concentrate (3, 5, and 10 percent), Lactoferrin, Bacteriocin, sugar (6, 8, 10%) and probiotics (2, 3, 4%) provided excellent nutritional values by employing bioassay studies.

Keywords: Cow milk, WPC, ragi malt, lactoferrin, bacteriocin, probiotics, rats

#### Introduction

Malnutrition in varying degrees is seen in the elderly, and this is also related to vitamin deficiency states. Malnutrition is also related to under nutrition, specific nutrient deficiencies, and nutritional imbalance. Malnutrition is the term used to describe not only pathologic states that arise from a dietary deficiency of essential nutrients or caloric excess, but also any significant deviation in dietary pattern that may result in an undesirable risk factor or that can be detected by physical examination, or biochemical or physiological tests (Thompson *et al.*, 1980).

The geriatric food should be nutritionally well-balanced in protein, fat, carbohydrate, essential vitamins and minerals.

#### **Bioassay studies**

In the development of geriatric health food, blends were formulated from germinated ragi malt as geriatric foods for elderly. Weanling rats weighing 23-24 g were given feeds containing germinated or non-germinated flour for a period of 28 days. The germinated flour diet appeared to be more palatable to the rats, leading to higher feed intakes. Germination increased alpha-amylase activity, which altered the nutrient composition of the grains and improved polysaccharide digestibility. It is suggested to use ragi malt as geriatric foods for elderly and infant. (Ikujenlola and Fashakin, 2005)<sup>[2]</sup>.

A rat bioassay was conducted to preclinically evaluate the nutritional quality of two supplementary foods (SF) based on ragi malt for feeding elderly. The SF were prepared by extrusion cooking and then modified to taste either sweet or salty. Groups of male weanling rats were fed SF for 4 weeks to evaluate the protein quality. Results showed that the body weight gain of rats fed with SF were significantly higher than those fed with the skim milk power (SMP) diet (control). The bio-availability of calcium was not significantly different (p>0.05) from values of the control groups. It is concluded that these SF are nutritionally comparable to SMP (Baskaran and Bhattacharya, 2004)<sup>[1]</sup>.

#### **Materials and Methods**

Fresh cow milk procured from Student Experimental Dairy Plant of Dairy Science College, Bangalore was used for the preparation of geriatric health beverage.

Skim milk powder "Nandini" brand skim milk powder manufactured by Karnataka Milk Federation was procured from the local market to standardize the geriatric health beverage.

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Ragi malt Good quality Ragi malt was procured from the local market was used in the preparation of geriatric health beverage.

Whey Protein Concentrate. Fresh spray dried Whey Protein Concentrate having 70% protein was procured from Mahan Proteins Ltd., Mathura, Uttar Pradesh, was used for the enrichment of geriatric health beverage.

Lactoferrin Lactoferrin was isolated from cheese whey in the PG laboratory of department of dairy technology.

Bacteriocin Bacteriocin was isolated from lactic acid bacteria in the PG laboratory of department of dairy technilogy.

Sugar Good quality cane sugar was procured from the local market and used in the preparation of geriatric health

#### beverage.

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Stabilizers Food grade Carboxy Methyl Cellulose (CMC) was procured from Lucid Colloids Ltd. Jodhpur and was used as stabilizer.

#### Preparation of geriatric health beverage

Whey protein concentrate with suitable protein content is to be reconstituted in water as a 10% solution. Heat treat the solution to 80° for 30 min. and inoculate with lactobacillus acidophilus or lactobacillus bifidus after adjusting pH (Puranik, 1996)<sup>[3]</sup>, with slight modification which has been made to optimize the functional geriatric health beverage. The flow diagram for preparation as follows.



Fig 1: Flow diagram for the preparation of probiotic geriatric health beverage

#### **Results and Discussion**

#### Initial and final weight of the weanling rats

The effect of diet on average body weight and gain in body weight of weanling rats fed with skim milk powder, malted geriatric health beverage, geriatric health beverage with WPC and probiotics,

It could be seen from the table that, initially the weights of all the groups of weanling rats were almost homogeneous and in the subsequent days there was variation in average weight and gain in body weight, depending on the diet fed. The average weight of rats fed on test diets C0, G1 and G2 were 28, 30 and 35 g, respectively during 1<sup>st</sup> week, 35.5, 39.8 and 44.2 g, respectively in 2<sup>nd</sup> week, 43.1, 48.9 and 58.5 g, respectively in 3<sup>rd</sup> week and 50, 65 and 70 g, respectively in 4<sup>th</sup> week, respectively.

Statistical analysis of results revealed that, there was significant difference in body weight of rats ( $p \le 0.05$ ) during initial stages with three types of diets. However, as feeding

continued the growth of animals was significantly differed in average body weight of rats at the end of 4<sup>th</sup> week. Thus the malted geriatric beverage with added WPC and probiotics definitely influenced the body weight of rats.

| Bioassay diet        | Week                    |                         |                               |                               |                    |  |  |
|----------------------|-------------------------|-------------------------|-------------------------------|-------------------------------|--------------------|--|--|
|                      | Initial                 | 1 <sup>st</sup>         | 2 <sup>nd</sup>               | 3 <sup>rd</sup>               | 4 <sup>th</sup>    |  |  |
| C0                   | 23                      | 28                      | 35.5                          | 43.1                          | 50                 |  |  |
| G1                   | 22                      | 30                      | 39.8                          | 48.9                          | 65                 |  |  |
| G2                   | 24                      | 35                      | 44.2                          | 58.5                          | 70                 |  |  |
| CD                   | 0.197                   | 0.189                   | 1.585                         | 1.384                         | 1.056              |  |  |
| C0<br>G1<br>G2<br>CD | 23<br>22<br>24<br>0.197 | 28<br>30<br>35<br>0.189 | 35.5<br>39.8<br>44.2<br>1.585 | 43.1<br>48.9<br>58.5<br>1.384 | 5<br>6<br>7<br>1.0 |  |  |

\*Average weekly weight of six rats in each group

C0: Animals fed with skim milk powder

G1: Animals fed with malted geriatric health beverage

G2: Animals fed with geriatric health beverage with WPC and probiotics



Fig 2: Effect of Diets on average weight of Weanling Rats

## Bone weight (g), Bone mineral density (g/cm<sup>3</sup>), FER, Calcium (%) in bone rudiments of rats.

Bone weight of rats were taken before and after feeding for 28 days and the average results presented in table 1. The values of the table clearly show that there was a variation in the bone weight before and after feeding control and test diets. It could be seen from the table that, before feeding, the observed bone weight was 0.18 g whereas after feeding with control sample it increased to 0.29 g. Further there was significant increase in the bone weight with respect to the test diets. Malted geriatric health beverage and malted geriatric health beverage with WPC and probiotics increased the bone weight to 0.33 g and 0.43 g, respectively.

Bone mineral densities in weanling rats was determined before feeding as well as after feeding for 28 days with control and test diets and the results are presented in Table 18. It could be observed from the table that, bone mineral density before feeding was 0.9 g/cm<sup>3</sup>. The corresponding values at the end of 28 days by feeding with malted geriatric health beverage and malted geriatric health beverage with WPC and probiotics were 2.2 and 2.9 g/cm<sup>3</sup>. respectively.

It is clear from the table that, there is an increase trend in the FER values of three types of diets i.e. control, malted geriatric food and malted geriatric food with WPC and probiotics. The FER values for the three diets were 0.29, 0.34 and 0.37,

respectively. Calcium concentrations in bones before feeding as well as after feeding for 28 days of control and test diets (G1 and G2) has been shown in Table 18. The results indicated that there is a increase in the concentration of calcium of bone rudiments before and after feeding. Geriatric malted beverage (ragi malt), geriatric malted food with WPC an

 Table 2: Bone weight (g), Bone mineral density (g/cm<sup>3</sup>), FER,

 Calcium (%) in bone rudiments of rats.

| Parameters<br>Feeding period |         | Bone<br>weight (g) | Bone mineral<br>density (g/cm <sup>3</sup> ) | FER   | Calcium<br>(%) |       |
|------------------------------|---------|--------------------|--|-------|----------------|-------|
| Before                       |         | 0.18               | 0.9  | -     | 8.5            |       |
| After                        | Type of | C0                 | 0.29   | 1.5   | 0.29           | 11.29 |
|                              | diet    | G1                 | 0.33   | 2.2   | 0.34           | 12.56 |
|                              |         | G2                 | 0.43   | 2.9   | 0.37           | 13.74 |
|                              |         | CD                 | 0.059  | 0.326 | 0.145          | 0.245 |

\*All values are average of 3 trials

Control (C0) - Skim milk powder as protein source  $G_1$  - Malted geriatric health beverage

 $G_2$  - Malted geriatric health beverage with WPC and probiotics

FER - Feed Efficiency Rati

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Probiotics influenced the concentration of calcium in the bone rudiments. As it is evident from the values that among all the diets, concentration of calcium was significantly more in the rat bones which were fed with malted geriatric beverage and malted geriatric beverage with WPC and probiotics compared to control diet. In case of control before feeding, the percentage of calcium in the bone was 8.5 and at the end of 28 days of feeding, the corresponding values increased to 11.29, 12.56 and 13.74 per cent in rats fed with control diet, malted geriatric health beverage, and malted geriatric health beverage with WPC and probiotics, respectively.

#### Effect of diet on average weight gain of weanling rats

During initial stage of experiment, the average weights of all groups of test animals were almost similar and in the subsequent weeks there was variation among the groups depending on the diet fed. The group of rats fed with malted geriatric health beverage with WPC and probiotics gained highest average weights (70 g) followed by malted geriatric health beverage (65 g), control skim milk powder (50 g), after 4 weeks of feeding. The growth pattern observed in this experiment was in accordance with the observations reported by Tripathy *et al.*, (2003) <sup>[1]</sup> in case of WPC supplemented wheat and ragi based products.

As could be observed, upon addition of WPC and probiotics in malted geriatric health beverage, there was a significant improvement in average gain in weight of weanling rats. This could be attributed to nutritional and functional properties of whey proteins in terms of its amino acid profile and biological quality. This added effect in terms of better growth could also be due to the complimentary effect of malted ingredients with enhanced bioavailability of whey proteins and mineral profile in presence of probiotics.

# Effect of weight (g), bone mineral density (g/cm<sup>3</sup>), FER and calcium (%) in bone rudiments of rats

As it is evident from the experimental results, that there was significant increase in bone weight and bone mineral density in all the three bone rudiments of rats viz. tibia, femur and humerus which were fed with WPC and probiotics supplemented geriatric health beverage (0.29, 0.33 and 0.43 g and 1.5, 2.2 and 2.9 g/cm<sup>3</sup> respectively). This increase in bone weight and bone mineral density was 10-15% more when compared to the animals which were maintained on control diet and geriatric health beverage without WPC and probiotics. This indicates that WPC with probiotics incorporated in the diet of rats enhanced the absorption of calcium, which favoured the calcification of bones and deposition in laboratory animals and consequently the bone mineral density (BMD) increased. This increased bone mineral density in earlier stages is important to prevent osteoporosis in later stages of life. FER is expressed as gain in weight in grams to the food consumed for a specified period of time. In the present study, FER significantly increased in case of rats fed with WPC and probiotics supplemented geriatric health beverage.

As reported, the availability of calcium was more in geriatric health beverage supplemented with WPC and probiotics (13.74%), compared to geriatric health beverage without WPC and probiotics (12.56%) and control diet (8.5%). The increase in bio-availability of calcium in beverage enriched with ragi malt, WPC and probiotics could be attributed to the better bio-availability of calcium in the diet. Bio-availability of calcium from milk and milk products, especially from WPC in presence of probiotics is more (Weaver, 1994)<sup>[5]</sup>.

#### Conclusion

Bioassay studies on weanling rats revealed that rats fed with malted geriatric health beverage with WPC, probiotics and bacteriocins showed maximum gain in body weight and also highest bone mineral density. The biological parameters derived from the bioassay studies revealed that group of rats fed with WPC, probiotics and bacteriocins resulted in highest feed efficiency ratio. It was also observed that bio-availability of calcium was more in WPC and probiotics supplemented geriatric health beverage. The results of the bioassay study indicated that WPC and probiotics can be effectively incorporated into geriatric health beverage for the enhancement of bio-availability of minerals.

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