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# Selection of suitable form of processing of cereal (pearl millet & moth bean) for preparation of whey- cereal based fermented beverage (Lassi) prepared by using ncdc-167 culture and 1 % fat content

**Monika Rani, Dabur RS and Priyanka**

**Abstract**
Fermentation not only makes milk more digestible, but is also a means of increasing the shelf-life and microbiological safety of the products. The demand for fermented milk products is increasing and it has been estimated that about 10% of total milk produced in India is used for preparation of traditional fermented milk products. The objective of the present study was to develop whey-cereal based fermented dairy products. Two different forms of cereal processing (soaked and germinated) of two cereals (pearl millet and moth bean) were used for development of the products and were evaluated for sensory acceptability as well as for growth indicators of NCDC-167 culture. Results revealed that germinated form of cereal processing of both the cereals were found more appropriate for preparation of whey pearl millet fermented dairy product (Lassi) and whey moth bean (Lassi) on the basis of sensory evaluation and starter culture growth indicators.

**Keywords:** Selection of suitable form of processing of cereal (pearl millet & moth bean) preparation of

## Introduction
Preservation of milk solids in form of fermented milk has been considered a very simple and immediately accessible convenient method (Sinha and Sinha, 2000) [20]. The consumer's interest in fermented milk products is gaining momentum due to the development of new food processing techniques, changing social attitudes; scientific evidence of health benefits of certain ingredients (Korhonen and Pihlanto, 2006; Stanton *et al.*, 2011) [11, 21]. Whey proteins have been known to have biological value superior to that of other naturally occurring proteins (e.g. egg, soya, beef, casein etc.). Millet grains, before consumption and for preparation of food, are usually processed by commonly used traditional processing techniques including decorticating, malting, fermentation, roasting, flaking, and grinding to improve their edible, nutritional, and sensory properties. Therefore, with value added strategies and appropriate processing technologies, millet grains especially the pearl millet can find a place in the preparation of several value added and health food products, which may then result in high demand from large urban population and non-traditional millet users (Mal *et al*., 2010) [12]. Therefore, to make the best use of dairy whey and low cost grains, the present study was proposed to develop delicious and nutritious traditional cereals based fermented dairy products from the combinations of whey, skim milk powder and low cost grain (pearl millet and moth bean) with the following objectives to know the processing effect of pearl millet and moth bean on sensory attributes of whey-cereal based fermented beverage (Lassi) prepared by using NCDC-167 culture and 1% fat content.

## Materials and methods
The constituents required for preparation of whey cereal based Lassi were standardized milk (4.5% fat & 8.5% SNF), paneer whey and cream was obtained from the department of Livestock Products Technology, LUVAS, Hisar. Skim milk powder (SMP) of Nova brand, pearl millet and moth bean, cumin, black pepper and salt, glass bottles of 200 ml capacity were bought from the local market. Starter cultures i.e. NCDC-167 (*Lactococcus lactis* ssp. *lactis*, *Lactococcus lactis* ssp. *cremoris* and *Lactococcus lactis* ssp. *lactis* biovar. *diactetylactis* in 1:1:1 ratio) were procured from Dairy Microbiology Division, National Collection of Dairy Cultures, ICAR-National Dairy Research Institute, Karnal (Haryana)-132001. Pectin (degree

of esterification 68-70 per cent) and Nisin was procured from Hi Media Laboratories Pvt. Ltd 23, Vadhani Ind, Est., LBS Marg, Mumbai-400086, and India.

**Preparation of cereals and whey**

One batch of cereals (Pearl millet and Moth beans separately) 25g was soaked and another batch of cereals (Pearl millet and Moth bean separately) 25g were germinated and cereal slurry was prepared by grinding. The whey obtained was passed through muslin cloth to remove suspended particles and obtained fresh whey was neutralized by using powder of sodium bicarbonate (@0.5gm/100ml of whey) to adjust pH to 6.8±0.2. Neutralized whey was standardised to 1 per cent fat and 18 per cent total solids level using fresh cream and skim milk powder and then kept in refrigerator till further use.

**Preparation of control**

Pearl millet control Lassi was prepared as per procedure developed by Modha and Pal (2011) [13] by using standardized milk (4.5% fat & 8.5% SNF) with some modifications.

For preparation of control, 500 ml standardized milk was preheated to 40 °C and then 25 gm germinated pearl millet slurry was added. Then the mixture was heated to 90 °C for 5 min and then immediately cooled to 37°C. Inoculated was done by addition of starter culture @ 3% (NCDC-167) followed by incubation at 37 °C for 6-8 h to obtain desirable acidity (0.7-0.8 % lactic acid).

350 ml water was heated to 60 °C in a separate pan, 5.20 gm pectin was added with continuous stirring followed by addition of 7.35gm salt, 0.75gm black pepper and 2.45 gm roasted cumin powder and then heated continuous to 80 °C temperature and then immediate cooled to 30 °C. Then the curd obtained was transferred to electric mixer jar having pectin and spiced water. The mixture was mixed for 2 minutes. The final products (Lassi) was cooled to 5 °C and stored in refrigerator (5-7 °C) for 2 hours before offered for sensory evaluation. Similarly, control of moth bean beverage (Lassi) was prepared replacing pearl millet with moth bean.

**Préparation of whey-cereal beverage (Lassi)**

Likewise, whey cereal beverage was prepared replacing milk with neutralized and standardised whey adding differently soaked and germinated pearl millet slurry was preheated to 40°C and and pectin @ 0.1 per cent of whey was added and then converted into two types of whey - pearl millet beverage (Lassi). Similarly, total two types whey-moth bean beverage (Lassi) were prepared replacing pearl millet with moth bean.

**Selection protocol**

Each developed Lassi samples along with control were offered for sensory evaluation (using 9 point hedonic scale) to six judges selected from faculty and post graduate students of Livestock Products Technology Department separately. Each type of Lassi samples along with control were also assessed for growth indicators of culture. Out of 2 treatments of each form of cereal processing, one best whey cereal combination was selected based upon sensory evaluation and growth indicators of NCDC-167 culture from both the cereal (pearl millet and moth bean)

**Sensory evaluation**

Sensory evaluation for attributes viz. colour and appearance, flavour, consistency and overall acceptability were conducted a semi trained panel consisting of faculty members and post graduate students using a 9-point Hedonic scale (Stone *et al*. 1974). The test samples were presented to the panellists after assigning the suitable codes. The water was served for rinsing the mouth between the samples.

## Results & Discussion

**Milk Composition**

Analysis results indicated that it contained 4.5 fat per cent and 8.5 per cent solid non-fat.

**Composition of whey**

Fresh whey was assessed for its composition and results are represented in table 1.

It is apparent from the results that whey contained moisture, total solids, lactose, protein, fat and ash were 93.3 per cent, 4.88 per cent, 6.7 per cent, 0.68 per cent, 0.65 per cent and 0.49 per cent respectively whereas, TA and pH were 0.48, and 4.33 respectively. These observations are in agreement with the earlier outcomes of Gupta (2008) [7] who also observed 6.50 per cent total solids, 0.5 per cent fat, 0.4 per cent protein, 0.5 per cent ash and lactose 5 per cent. TA and pH observations were similar to Bund and Pandit (2005) [3] who also observed paneer whey titratable acidity ranges between 0.4-0.6, and pH ranges from 4.0-5.0 respectively.

**Selection of Pearl Millet Form of Processing**

**Table 1:** Composition of whey

| Parameters | Per cent |
|---|---|
| Moisture (%) | 93.30 |
| Total solids (%) | 6.70 |
| Lactose (%)* | 4.88 |
| Protein (%) | 0.68 |
| Fat (%) | 0.65 |
| Ash (%) | 0.49 |

*Calculated by difference (T.S.-(Fat+Protein+Ash)

**Selection of suitable form of processing of cereals**

The appropriate form of cereal processing, with suitable starter culture and desirable fat per cent was optimized by comparing data obtained from sensory evaluation as well as growth indicators of starter cultures. The results are presented in tables (1-5) and figure 3.

**Table 2:** Processing effect of pearl millet on sensory attributes of whey-pearl millet based fermented beverage (Lassi) prepared by using NCDC-167 culture and 1% fat content.

| Sensory attributes | Control | 1% Fat | |
|---|---|---|---|
| | | Form of Cereal Processing | |
| | | Soaking | Germination |
| Colour and Appearance | $8.00\pm0.68^{b}$ | $7.40\pm0.79^{a}$ | $7.50\pm0.80^{a}$ |
| Consistency | $8.20\pm0.57^{b}$ | $7.00\pm0.46^{a}$ | $7.20\pm0.38^{a}$ |
| Flavour | $8.20\pm0.29^{c}$ | $7.30\pm0.19^{a}$ | $7.80\pm0.28^{b}$ |
| Overall Acceptability | $8.20\pm0.35^{c}$ | $7.30\pm0.26^{a}$ | $7.70\pm0.17^{b}$ |

Mean ± SD, n=18

Means with different superscripts in a row differ significantly ($P\leq0.05$) in each particular group i.e. a,b,c for 1% group

**Selection of Suitable Form of Processing Of Cereals (Pearl Millet)**

**Processing effect of pearl millet on sensory attributes of whey-pearl millet based fermented beverage (Lassi) inoculated with culture NCDC-167**

Traditionally made fermented foods are not as per according to the present requirement due to lack of quality maintenance as well as also low in yield. By applying modern process technologies, they can be reinvented, for mass production to meet the consumer's new demand in response to changing life style. Soaking and germination improved the *in vitro* protein (14% to 26%) and starch (86% to 112%) digestibility in pearl millet (Archana *et al*., 2001) [2]. It also led to the reduction of anti-nutrients such as phytic acid, tannins, and polyphenols, which form complexes with protein (Hassan *et al*., 2006) [8]. Indian fermented milk product known for its refreshing taste, palatability and therapeutic values. It is prepared by fermentation of milk by using lactic acid bacteria. Indian fermented milk product differs from Western fermented milk product in its use of mixed starters of mesophilic lactococci and thermophilic culture. The present study was under taken to prepare whey-cereal based beverage by using mesophilic lactococci (NCDC-167).

Sensory scores of whey-pearl millet based fermented beverage (Lassi) prepared by using NCDC-167 starter culture and with two different fat content are presented in table 2.

**Table 3:** Processing effect of pearl millet on growth indicators of NCDC-167 culture used to prepare whey-pearl millet based fermented beverage (Lassi) with different fat content

| Growth factor | Control | 1% Fat | |
|---|---|---|---|
| | | Form of Cereal Processing | |
| | | Soaking | Germination |
| pH | 4.49±0.04$^{b}$ | 4.65±0.09$^{c}$ | 4.40±0.05$^{a}$ |
| Titrable acidity | 0.71±0.08$^{b}$ | 0.64±0.07$^{a}$ | 0.77±0.01$^{c}$ |
| Soluble nitrogen (%) | 0.24±0.08$^{b}$ | 0.14±0.02$^{a}$ | 0.22±0.09$^{b}$ |
| Acetaldehyde (ppm) | 1.25±0.03 | 1.23±0.01 | 1.24±0.04 |

Mean ± SD, n=18

Means with different superscripts in a row differ significantly ($P \leq 0.05$) in each particular group i.e. a,b,c for 1% group

**Whey-pearl millet based Lassi**

Panelist preferred colour and appearance and consistency of whey-soaked pearl millet based Lassi at par with whey-germinated pearl millet based Lassi. While flavour and overall acceptability of whey-germinated pearl millet based Lassi was preferred over whey-soaked pearl millet based Lassi by the panelist in scoring but rate at par as like moderately. It is evident from table 2 that judges preferred control beverage over experimental beverages for its colour and appearance, consistency, flavor and overall acceptability. On 9-point hedonic scale judges scores indicated that control beverage rated as liked very much for its colour and appearance, consistency, flavor and overall acceptability whereas experimental beverages irrespective of millet processing form were rated as liked moderately by the panelist almost for all the sensory attributes. Results revealed that processing of cereal had an influence on acceptability of beverage as flavour and overall acceptability of beverage was influenced as panelist scored germinated whey-millet Lassi significantly higher than soaked whey-millet Lassi.

Similar results were reported by Sudha *et al.* (2016) [23] who observed that beverage made from milk of millet sprouts were acceptable in terms of sensory property (score7.1-7.3) while the beverage prepared from the milk of soaked millet was unacceptable which may be due to high sedimentation value and wheying off percentage. Modha and Pal (2011) [13] observed effect of form of addition of pearl millet solids and out of total 9 combinations tried (Raw flour, 24 and 48 h germinated flour in 2 stages before and after fermentation and re-fermentation of raw flour), flour of 24 h germinated grains when added to milk solids before fermentation was found better than other forms with overall acceptability score of 7.2 on 9-point Hedonic scale. And also found that raw pearl millet flour and wet ground slurry in all forms provided astringent flavour to the beverage, thus the scores were low. Similarly, Hassan *et al.*, 2006 [8] reported that germination followed by fermentation was more effective in increasing the protein digestibility of pearl millet.

On the basis of above results, it is concluded that germination had significant influence on flavour and overall acceptability of beverage, whey germinated pearl millet based Lassi was scored significantly higher and was selected for further experiments.

**Processing effect of pearl millet on growth indicators of NCDC-167 culture**

Data of growth indicators of starter culture (NCDC-167) used to prepare whey-pearl millet based fermented beverage (Lassi) with two different fat content is presented in table 3.

**Whey-pearl millet based Lassi**

Whey-soaked pearl millet based Lassi had shown higher pH value in comparison to control beverage and whey-germinated pearl millet based Lassi. However, significant difference was observed in pH of control beverage and whey-germinated pearl millet based Lassi. Results are in agreement with Ocheme and Chinma (2008) [14] who reported that in case of malted millet grains flour beverage the pH decreased. Titrable acidity results had shown reverse trend to pH in case of whey-soaked pearl millet based Lassi. While titrable acidity value was highest among all sample in case of whey-germinated pearl millet based Lassi. It indicates that starter culture growth was better in whey-germinated pearl millet in comparison to control and whey-soaked pearl millet. Results are in agreement with Ocheme and Chinma (2008) [14] who reported that in case of malted millet grains flour beverage total titrable acidity (TTA) significantly increased. This increase in acidity in malted samples could also be as a result of hydrolysis of some complex organic molecules. Moreover, the significant difference between control and germinated also existed between control and germinated regarding TA and pH. This may be because of higher total solids per cent in whey germinated sample as compared to control. Higher total soilids contributes to more acidity (Chawla, 1985) [5].

Soluble nitrogen (per cent) was lowest in whey-soaked pearl millet based Lassi in comparison to control as well as whey-

germinated pearl millet based Lassi. Results revealed that proteolytic activity was highest in control which was at par with malted pearl millet sample which may be due to increase in protein in whey-germinated pearl millet during malting. Results are in close concord with Inyang and Idoko (2006) [9] who reported increase in protein with increasing malting levels, similar to the observation with ogi made form malted corn (Okoli and Adeyemi, 1989) [15]. According to Chavan *et al*., 1989, quality of cereal proteins as well as the content may also be improved by fermentation. Germination may also result in increase in protein probably due to dry matter loss as well as a result of mobilization of storage nitrogen of millet to produce the nutritionally high quality proteins which the young plant needs for its development as suggested by Tsai *et al.* (1975) [24]. Acetaldehyde content of all the samples of Lassi was observed at par. Sensory results are also confirmed that flavor of whey-germinated pearl millet based Lassi and control Lassi was at par.

**Processing Effect of Moth Bean on Sensory Attributes of Whey-Moth Bean Based Fermented Beverage (Lassi) Inoculated With Ncdc-167**

Moth bean is commonly grown in arid areas of India and is consumed either as such after cooking or after germination and cooking. The nutritive value of grain legumes depends primarily on their nutrient and the presence or absence of anti-nutrient and toxic factors. Some processing technique, such as soaking, germination, and cooking are highly effective for the reduction of anti-nutritional factors and for improving its organoleptic quality. This study involves the effect of different household processing on sensory attributes and starter culture growth factors in developing whey-moth bean fermented beverage (lassi).

Sensory scores of whey-moth bean based fermented beverage (lassi) prepared by using NCDC-167 culture and different fat content are presented in table 10.

**Table 5:** Processing effect of moth bean on growth indicators of NCDC-167 culture used to prepare whey-moth bean based fermented beverage (lassi) with 1% fat content

| Growth indicators | Control | 1% Fat | |
|---|---|---|---|
| | | Form of Cereal Processing | |
| | | Soaking | Germination |
| pH | 4.50±0.01$^{b}$ | 4.56±0.09$^{c}$ | 4.44±0.02$^{a}$ |
| Titrable acidity | 0.76±0.08$^{b}$ | 0.67±0.03$^{a}$ | 0.85±0.07$^{c}$ |
| Soluble nitrogen (%) | 0.21±0.04$^{b}$ | 0.14±0.06$^{a}$ | 0.18±0.05$^{b}$ |
| Acetaldehyde (ppm) | 1.23±0.01 | 1.19±0.02 | 1.21±0.03 |

Mean ± SD, n=18

Means with different superscripts in a row differ significantly ($P \leq 0.05$) in each particular group i.e. a,b,c for 1% group

**Whey moth bean based Lassi (1% fat)**

Judges scored colour and appearance and consistency of whey-soaked moth bean based Lassi sample was scored at par with of whey-germinated moth bean based Lassi sample. The score of whey-germinated moth bean based Lassi was significantly higher for its flavour than whey-soaked moth bean based lassi. Panelists judged overall acceptability of whey-germinated moth bean based Lassi sample significantly higher in comparison to whey-soaked moth bean based Lassi sample. Sensory panelists scored control sample significantly higher than the experimental samples for its colour and appearance, consistency, flavour and overall acceptability.

Similar results were also observed by by Salve and Mehrajfatema (2011) [17] who also found that incorporation of germinated moth bean flour in cakes up 5% replacing Maida could enhance the sensorial as well as nutritional quality characteristics of cake. They also reported that germinated moth bean fortified cake was found to be comparatively more sensorily more acceptable highe than that of control as well as non-germinated moth bean fortified cake. Similarly, Saha and Dunkwal (2009) developed value added spread instant mix by using germinated moth bean and β-carotene rich vegetables and results revealed that the calculated overall mean organoleptic scores for the control instant mix (procured from the local market) was to be ranging between 7.0 to 7.2 against 8.2 to 8.7 scores for develop instant mix on nine point hedonic ranking scale.

**Processing effect of moth bean on growth indicators of NCDC-167**

Data of growth indicators of starter culture (NCDC-167) used to prepare whey- moth bean based fermented beverage (lassi) with different fat content is presented in table 11.

**Whey Moth bean based Lassi (1% fat)**

Whey-soaked moth bean sample had significantly highest value of pH as compared to sample prepared from whey-germinated moth bean sample followed by control and whey-germinated moth bean sample attained significantly lowest value as compared to sample prepared from whey-soaked moth bean sample but statistically at par with whey-germinated moth bean sample. Titrable acidity was recorded significantly lower for whey-soaked moth bean sample in comparison to whey-germinated moth bean sample as well as control sample. However, there was significant difference among control sample and whey-germinated moth bean sample. Low pH and high titratable acidity of the germinated maize flour are due to organic acids (Akinrele, 1970) [1], and lower pH in foods suggests longer shelf-life. Among treatments, germinated form had significantly highest titrable acidity. Jood *et al.* (2012) [10] studied the effect of germination and probiotic fermentation on pH and titratable acidity of sorghum based food mixtures. The reason behind it may be that during germination starch is hydrolysed into sugars which is readily utilized by the organisms and converted to lactic acid.

**Table 6:** Processing effect of moth bean on sensory attributes of whey moth bean based fermented beverage (Lassi) prepared by using NCDC-167 culture and 1% fat content

| Sensory attributes | Control | 1% Fat | |
|---|---|---|---|
| | | Form of Cereal Processing | |
| | | Soaking | Germination |
| Colour &Appearance | 8.00±0.68$^{b}$ | 7.50±0.83$^{a}$ | 7.60±0.94$^{a}$ |
| Consistency | 8.20±0.57$^{b}$ | 7.10±0.55$^{a}$ | 7.20±0.61$^{a}$ |
| Flavour | 8.20±0.29$^{c}$ | 7.20±0.28$^{a}$ | 7.50±0.37$^{b}$ |
| Overall Acceptability | 8.20±0.35$^{c}$ | 7.30±0.27$^{a}$ | 7.60±0.36$^{b}$ |

Mean ± SD, n=18

Means with different superscripts in a row differ significantly ($P \leq 0.05$) in each particular group i.e. a,b,c for 1% group

Soluble nitrogen per cent was significantly higher in whey-germinated moth bean sample than whey-soaked moth bean sample. While control sample and whey-germinated moth bean sample had statistically similar values. Germination had a little effect on crude protein prepared by different levels of incorporation of content except little increases, which could be due to biosynthesis during germination (Sattar *et al.*, 1989; Venderstoep, 1981) [18, 25]. The protein content decreased from

23.02 to 22.9% in 12 hour water soaked and 22.84% in 12 hour salt soaked moth bean seeds. It is also possible thought that the increase in protein was due to changes resulting from the uptake of water during sprouting. Germination produced a small increase in protein utilization in both legumes (soybeans and lupin seeds) (Donangelo *et al.*, 1995) [6]. Similarly, Singh *et al*. (2015) [19] summarized that the protein concentration increased and the amino acid profile is balanced by germination and fermentation.

Therefore, out of soaking and germination, from 1 percent whey cereal based lassi, germination was selected keeping in consideration the sensory scores and growth indicators and for further product development.

## Conclusion

The increasing awareness for nutrition, health and quality food consciousness of consumers and the keen competition in the market, compel the food industry to search for such ingredients, which can impart specific functionalities to food products, while preserving and enhancing the nutritional quality of foodstuffs, in order to sell their products profitably. Whey-cereal based beverage (lassi), pearl millet and moth bean were used and cereals were processed by soaking germinating before making slurry of cereals to develop whey-cereal Lassi. The two different cereal slurry with two different processing treatment were mixed with standardized whey to develop whey-cereal based beverage (lassi) seperately. Whey fat content was adjusted to 1 percent by using fresh cream and total solids were enhanced to the tune of 18 percent by adding skim milk powder and inoculated with starter culture viz.NCDC-167 for making curd from the mixture of whey and cereals slurry. Then curd obtained was used for preparing whey-cereal based fermented beverage (lassi) by stirring with addition of pectin and spiced water. Selection of suitable form of cereal processing (soaking & germoination) was done on the basis of sensory attributes and growth indicators starter cultures. Present study results indicated that whey-cereal Lassi samples prepared with germinated form with both the cereals i.e. pearl millet as well as moth bean were found most suitable in comparison to prepared using soaked form, because germination form improved the sensory scores and growth indicators (depicting growth characteristics of starter culture). Sensory panelist scores (for flavor and overall acceptability) were recorded highest in samples inoculated with starter culture hving germinated form of cereal slurry.

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