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## Development and physico-chemical characterization of sterilized chocolate milk drink stabilized with oat flour

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### Abstract

Chocolate flavoured milk is one of the most consumed liquid product in the milk beverage category due to its popularity among peoples of all age groups. The present study was done to develop sterilized Chocolate Milk Drink (CMD) by adding oat flour (0, 1, 2 and 3%) in different concentrations and maltodextrin 3% as a stabilizer, along with sugar and cocoa powder. Oat contains soluble fiber  $\beta$ -glucan, which acts as a stabilizer and possess technological significance in the preparation of milk beverages. All the ingredients were mixed followed by in-bottle sterilization at 121°C for 15 minutes at 15 psi and evaluated for different physico-chemical properties. The physico-chemical properties of CMD were affected with an increase in the level of oat flour for fat, protein, SNF, total carbohydrates, etc. The specific gravity was also increased with an increase in the level of oat flour addition. The findings of this study provide a scientific basis for formulation of nutritionally enriched, shelf-stable sterilized CMD using oat flour and maltodextrin as stabilizer.

**Keywords:** Chocolate milk drink, oat flour,  $\beta$ -glucan, stabilizer, soluble fiber

### Introduction

Flavoured milk, second most consumed liquid dairy product after plain milk, prepared by addition of flavour (natural or synthetic) and sweetener (sugar). It can be either pasteurized or sterilized. Flavoured milk drinks, particularly chocolate milk beverages, represent an important and growing segment of the value-added dairy industry. They serve as an effective vehicle for delivering high-quality milk proteins, calcium, phosphorus, riboflavin, and vitamins to consumers in a palatable and acceptable form. Among various categories, sterilized flavoured milk drinks are gaining commercial significance owing to their extended ambient shelf life without refrigeration, making them particularly suited to tropical developing markets such as India. The sterilization process, typically conducted at 121°C for 15 minutes, ensures commercial sterility by destroying heat-resistant spore-forming microorganisms including *Clostridium botulinum*, while also inactivating lipases, proteases, and oxidases that could otherwise compromise product quality during storage.

In the preparation of sterilized chocolate milk drinks, the physical stability of ingredients is the major technological challenge. Cocoa powder, added as flavouring and colouring agent, which are highly hydrophobic, are prone to sedimentation in the milk matrix during preparation and storage of flavoured milk, leading to phase separation, sediment formation and ultimately consumer rejection <sup>[1]</sup>. Additionally, the high heat treatment during sterilization can partially disrupt the colloidal calcium phosphate bridges within casein micelles, affecting the structural integrity of the protein system and potentially exacerbating sedimentation. the addition of hydrocolloids and polysaccharide-based stabilizers is, therefore, a widely adopted technological strategy to prevent sedimentation, enhance viscosity, improve mouthfeel, and maintain the colloidal stability of chocolate milk drinks during processing and storage.

Oat, recognized as one of the richest cereal sources of mixed-linkage (1 $\rightarrow$ 3) (1 $\rightarrow$ 4)- $\beta$ -D-glucan, a high-molecular-weight soluble dietary fiber polysaccharide. Oat grain typically contains 3-7%  $\beta$ -glucan on a dry weight basis, with the  $\beta$ -glucan concentrated primarily in the sub-aleurone layer and the endosperm cell walls <sup>[2]</sup>. The unique rheological behavior of

Oat  $\beta$ -glucan in aqueous systems, characterized by its capacity to form high-viscosity solutions even at low concentrations through the formation of entangled random coil polymer networks, makes it an effective natural stabilizer and viscosity-enhancing agent in dairy-based beverages. Beyond its technological functionality, oat  $\beta$ -glucan is associated with clinically validated health benefits, including reduction of postprandial blood glucose, attenuation of serum LDL cholesterol, and modulation of gut microbiota, properties recognized in health claims by the European Food Safety Authority (EFSA, 2011) and the US Food and Drug Administration (FDA). Maltodextrin is a partially hydrolysed starch-derived carbohydrate polymer characterized by a dextrose equivalent value of 3-20, produced by the controlled enzymatic or acid hydrolysis of native starch. In milk products preparations, maltodextrin works as a stabilizer by increasing the viscosity of the continuous aqueous phase, and reduces the sedimentation of cocoa powder.

The study was conducted to determine the effect of oat flour addition on the chemical composition and physico-chemical changes that takes place in chocolate milk drink.

## Materials and Methods

### Materials

For the preparation of CMD, skim milk containing 0.5% fat was obtained from local market. Cocoa powder, refined cane sugar, maltodextrin and oat flour were procured from local market of Karnal. All analytical chemicals and reagents were of analytical grade procured from HiMedia Laboratories Pvt. Ltd., Mumbai, India. Glass bottles for packaging and sterilization of CMD was procured from local market.

### Preparation of chocolate milk drink

The oat based CMD was developed by following the procedure of Chatterjee and Patel <sup>[3]</sup> with slight modifications. The formulation of CMD were prepared as detailed in Table 1. For preparation of CMD, skim milk was preheated to 45°C followed by addition of sugar @7%, oat flour in different concentrations (1, 2 and 3%) and maltodextrin in different concentrations (1, 2 and 3%) and cocoa powder @2%. The control sample was prepared using maltodextrin @3% as a stabilizer and without oat flour. The mix was further pre-heated to 65°C followed by homogenization in two stage homogenizers at 2000 and 500 psi in the first and second stage, respectively to form homogenous mixture followed by sterilization in glass bottles at 121°C for 15 min at 15 psi. After sterilization, CMD was cool to room temperature prior to physico-chemical analysis.

## Physico-chemical analysis

### Fat

Fat content of oat added CMD was estimated by Gerber method (BIS 1224, Part II, 1977). The fat percentage on the graduated scale of the butyrometer can be read directly and noted. The fat analysis was done in triplicate and the values were presented in percentage.

### Solids-not-fat

Solids-Not-Fat (SNF) of CMD was determined by the standard procedure. The SNF content of CMD was related

to its fat percentage and specific gravity at 27°C, by the Richmond's formula.

$$\% \text{ SNF} = \text{CLR}/4 + 0.2F + 0.55$$

Where, CLR = corrected lactometer reading

F = fat percentage of milk

### Protein

Protein content was determined by Kjeldhal method using a conversion factor of 6.38 (AOAC, 2005).

### Ash

Ash (mineral) content of CMD was determined by incineration at 550°C for 5 hours in a muffle furnace (AOAC, 2005). Calculation was done by the following formula:

$$\% \text{ Ash content by weight} = 100(W_1 - W) / (W_2 - W)$$

Where, W = weight in g of the crucible.

W<sub>1</sub> = weight in g of the crucible containing the ash.

W<sub>2</sub> = weight in g of the crucible with the dried material taken for the test.

### Carbohydrates

The total carbohydrates were calculated by the "By - difference" method. After determining the percentage of total solids, protein, fat and total ash content in the developed product it was calculated as follows:

Total carbohydrates, including sucrose, dextrose and dextrin's, maltose or lactose, percent by weight = {A - (B+C+D)} %

Where, A = percentage by weight of total solids

B = percentage by weight of total protein

C = percentage by weight of fat

D = percentage by weight of total ash

### Total solids

The total solid of CMD was determined by the Gravimetric method by over drying at 102 ± 1°C to constant weight (BIS 1479, Part I, 1981).

### Specific gravity

Specific gravity of CMD was determined by finding out the weights of a certain volume of sample and of the same volume of distilled water at the same temperature taken in a specific gravity bottle.

$$\text{Relative specific gravity of milk at } T^{\circ}\text{C} = \frac{(W_1 - W)}{(W_2 - W)}$$

$$\text{Absolute specific gravity of milk at } T^{\circ}\text{C} = \frac{(W_1 - W)}{(W_2 - W)} \times d_w$$

Where, W = Weight of empty specific gravity bottle with stopper (g)

W<sub>1</sub> = Weight of specific gravity bottle with stopper and sample at T°C (g)

W<sub>2</sub> = Weight of specific gravity bottle with stopper and distilled water at T°C (g)

d<sub>w</sub> = Density of water at t°C (Standard bottle)

### Titrateable acidity

The titrateable acidity CMD were determined by the method described in (AOAC, 1995). A known weight of sample (10ml) was titrated against the standard 0.1 N NaOH solution using phenolphthalein as an indicator and the

acidity was calculated by using following formula and expressed in terms of per cent lactic acid.

### Statistical analysis

All data were subjected to one-way analysis of variance (ANOVA) using SPSS software. Treatment means were compared using Duncan's Multiple Range Test at a significance level of  $p < 0.05$ . Results are expressed as mean  $\pm$  standard deviation (SD) of three independent replicates ( $n=3$ ). Within each row, means bearing different lowercase superscript letters differ significantly ( $p < 0.05$ ).

### Results and discussion

The physico-chemical analysis of CMD was conducted and the results are presented in Table 2. In most of the parameters, significant differences ( $p < 0.05$ ) were observed in all the samples with the variation in concentration of oat flour and maltodextrin.

#### Fat content

The fat content of CMD ranged from 0.40 to 0.62 %, all the samples possess significant difference ( $p < 0.05$ ) with each other. The fat content in all the samples of CMD increased due to the addition of increased level of oat flour, which may be due to the presence of ~6% fat in oats. As oat contains fat in it, addition of oat slightly increased the final fat content in the product. Cocoa powder also contributes a very small fraction (~10-12% fat), which was constant across treatments. The present study are in accordance with the findings of Ramanathan and Sivakumar <sup>[4]</sup> for the preparation of dahi by incorporating oat flour, and they reported that with an increase in the concentration of oat flour from 1%, 2%, 3% and 4%, the gradual increase in the fat content of dahi was observed.

#### Solids-Not-Fat (SNF)

The addition of oat flour in CMD significantly increased ( $p < 0.05$ ) the SNF content from 8.52% (CMD03) to 10.34% (CMD30). The increase in SNF content of CMD attributed to the compositional changes across treatments due to addition of oat flour and maltodextrin addition. Oat contains, a complex matrix of starch (approximately 55-60%), dietary fiber including  $\beta$ -glucan (~4-7%), protein (~12-17%), lipids (~5-9%), and minerals (~2-3%) to the formulation. Maltodextrin, with a dry matter content exceeding 95%, is composed almost entirely of carbohydrate polymers of varying chain lengths (degree of polymerization, DP), predominantly dextrans, which are completely soluble in the aqueous milk matrix and therefore quantitatively and directly increases the SNF and total solids of the product <sup>5</sup>.

#### Protein content

The addition of oat flour significantly increased ( $p < 0.05$ ) the protein content of CMD from 3.11 to 3.44%. The increase in protein content may be attributed to the increase in the concentration of oat flour as it contains significant of protein in it (~15%). The results are in consistent with the findings of Ramanathan and Sivakumar, <sup>[4]</sup> in which they reported that the addition of oat flour in dahi in the concentration of 1%, 2%, 3% and 4% increased the total protein content in the final product to 3.6%, 3.8%, 3.9% and 4.0%, respectively. Mohamed *et al.* <sup>[6]</sup> reported for increase in protein content of cheese with an increase in the concentration of oat flour from 2.5%, 5.0% and 7.5%.

#### Total carbohydrate

The total carbohydrate content significantly increased ( $p < 0.05$ ) from 10.81% (CMD03) to 12.27% (CMD30) in CMD without oat flour and CMD with 3% oat flour, respectively. The increase in the content of oat flour increased the total carbohydrate content of CMD. The total carbohydrates in CMD comprises of carbohydrates and starch content from oat and maltodextrin along with 7% sugar. De *et al.*, <sup>[7]</sup> reported for increase in the carbohydrate content of *kheer* with an increase in the content of cereals in its manufacturing. Mohamed *et al.*, <sup>[6]</sup> also reported similar findings that the increase in the level of oat flour addition in the preparation of cheese significantly increased the carbohydrate content in the product.

#### Ash content

The addition of oat flour in CMD significantly increased ( $p < 0.05$ ) its ash content from 0.72 to 0.87% in CMD03 to CMD30, respectively. Oat flour contains ~1.5 to 2.5% ash which might have slightly contributed to the total ash content in the CMD. The present findings are in accordance with Mohamad *et al.* <sup>[6]</sup> in which the ash content of the processed cheese spreads fortified with oats increased with an increase in the concentration of oats. Sahan *et al.* <sup>[8]</sup> reported that the increase in the concentration of oat  $\beta$ -glucan hydrocolloidal composite in the yoghurt increased the ash content.

#### Total solids

The total solids content of CMD was increased due to an increase in the concentration of oat flour ranged from 0 to 3% in different samples. The total solid content varies from 23.54% and 27.54% in CMD03 and CMD30, respectively. The increase in total solids content may be attributed to the increased level of oat flour addition from 0% to 3%. The total solids of CMD depends upon different factors such as the total solids content of milk, sugar percentage, oat flour concentration and other additives added during the preparation of the product. Higher solids content in CMD increase the viscosity of the product. The addition of oat flour in dahi in different concentrations (1%, 2%, 3% and 4%) increased the total solids content of dahi from 25.6 to 29.58%. The addition of oat flour in the preparation of yoghurt significantly increased the total solids content with an increase in concentration of oat flour <sup>[9]</sup>.

#### Acidity

The addition of oat flour in different concentrations did not significantly affect ( $p < 0.05$ ) the acidity of the CMD samples ranging from 0.14 to 0.16% titratable acidity from 0% to 3%, respectively. Oat flour increased viscosity of CMD at higher concentration (3%).

#### Specific gravity

The specific gravity of CMD was significantly increased ( $p < 0.05$ ) with an increase in the concentration of oat flour from 1.042 to 1.078 in CMD03 and CMD30, respectively. The specific gravity of CMD increased with an increase in the concentration of oat flour as oat contributes to total solids content of the product. The present results showed similarity with the findings of Mittal and Bajwa <sup>[10]</sup> in which they reported that the specific gravity of low calorie milk drink was increased with an increase in the concentration of inulin.

## Tables and figures

**Table 1:** Treatment design for CMD formulations

| Ingredients          | CMD03 | CMD12 | CMD21 | CMD30 |
|----------------------|-------|-------|-------|-------|
| Skim milk (mL)       | 100   | 100   | 100   | 100   |
| Sugar (% w/v)        | 7.0   | 7.0   | 7.0   | 7.0   |
| Cocoa Powder (% w/v) | 2.0s  | 2.0   | 2.0   | 2.0   |
| Oat Flour (% w/v)    | 0.0   | 1.0   | 2.0   | 3.0   |
| Maltodextrin (% w/v) | 3.0   | 3.0   | 3.0   | 3.0   |

Values represent concentration (% w/v) per 100 mL of standardized milk base. All treatments contain 7% sugar and 2% cocoa powder.

**CMD03**- chocolate milk drink with 0% oat flour + 3% maltodextrin

**CMD12** - chocolate milk drink with 1% oat flour + 2% maltodextrin

**CMD21** - chocolate milk drink with 2% oat flour + 1% maltodextrin

**CMD30** - chocolate milk drink with 3% oat flour + 0% maltodextrin

**Table 2:** Physio-chemical analysis of chocolate milk drink

| Parameter               | CMD03                      | CMD12                       | CMD21                      | CMD30                      |
|-------------------------|----------------------------|-----------------------------|----------------------------|----------------------------|
| Fat (%)                 | 0.40 ± 0.01 <sup>a</sup>   | 0.46 ± 0.02 <sup>b</sup>    | 0.54 ± 0.02 <sup>c</sup>   | 0.62 ± 0.03 <sup>d</sup>   |
| SNF (%)                 | 8.52 ± 0.16 <sup>a</sup>   | 9.05 ± 0.24 <sup>ab</sup>   | 9.72 ± 0.19 <sup>bc</sup>  | 10.34 ± 0.36 <sup>c</sup>  |
| Protein (%)             | 3.11 ± 0.09 <sup>a</sup>   | 3.23 ± 0.07 <sup>ab</sup>   | 3.35 ± 0.05 <sup>bc</sup>  | 3.44 ± 0.06 <sup>c</sup>   |
| Total Carbohydrates (%) | 10.81 ± 0.12 <sup>a</sup>  | 11.21 ± 0.14 <sup>b</sup>   | 11.71 ± 0.22 <sup>c</sup>  | 12.27 ± 0.29 <sup>d</sup>  |
| Ash (%)                 | 0.72 ± 0.02 <sup>a</sup>   | 0.75 ± 0.03 <sup>ab</sup>   | 0.80 ± 0.04 <sup>b</sup>   | 0.87 ± 0.03 <sup>c</sup>   |
| Total solids (%)        | 23.54 ± 0.24 <sup>a</sup>  | 24.70 ± 0.56 <sup>b</sup>   | 26.12 ± 0.72 <sup>c</sup>  | 27.54 ± 0.47 <sup>d</sup>  |
| Specific Gravity        | 1.042 ± 0.004 <sup>a</sup> | 1.049 ± 0.006 <sup>ab</sup> | 1.063 ± 0.005 <sup>c</sup> | 1.078 ± 0.009 <sup>d</sup> |
| Acidity (%)             | 0.14 ± 0.01 <sup>a</sup>   | 0.14 ± 0.01 <sup>a</sup>    | 0.15 ± 0.01 <sup>a</sup>   | 0.16 ± 0.01 <sup>ab</sup>  |

Values are expressed as mean ± standard deviation (SD). Means within a row bearing different superscript letters (a, b, c and d) differ significantly ( $p < 0.05$ ).

**CMD03**- chocolate milk drink with 0% oat flour + 3% maltodextrin

**CMD12** - chocolate milk drink with 1% oat flour + 2% maltodextrin

**CMD21** - chocolate milk drink with 2% oat flour + 1% maltodextrin

**CMD30** - chocolate milk drink with 3% oat flour + 0% maltodextrin

## Conclusion

The present study demonstrated the development and physico-chemical characterization of CMD incorporated with oats and maltodextrins. The CMD was prepared with the addition of oat flour in different concentrations ranging from 1%, 2%, 3% and 4%. The presence of oat flour significantly affected the chemical composition such as fat, protein, SNF, carbohydrates and total solids of CMD. Specific gravity was significantly increased with an increase in the level of oat flour in CMD. Acidity was not much affected by the level of oat flour addition. The findings from this study suggested that oat flour addition can increase the nutritional properties of the chocolate milk drink or dairy beverage along with high fiber content which lacks in milk and milk based products.

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