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## Standardization of Greek yoghurt using *Lactobacillus acidophilus* (LA-5) and *Cassia auriculata* flower powder

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#### Abstract

Greek yoghurt is also known as concentrated yoghurt or strained yoghurt. The present study is to develop functional Greek yoghurt using *Lactobacillus acidophilus* (LA-5) by incorporating *Cassia auriculata* flower powder which is a shrub used for medicinal purposes. The initial trials of functional Greek yoghurt with *Lactobacillus acidophilus* (LA-5) was prepared by incorporation of *Cassia auriculata* flower powder in different concentrations viz., 0.5%, 1%, 1.5%, 2%, 2.5%, 3%, 3.5% and 4%. By using semi-trained panelists the sensory evaluation was done using nine point Hedonic scale. As a result the incorporation of 3.0% of *Cassia auriculata* flower powder in Greek yoghurt was determined as the acceptable level and subjected for further research. Based on the sensory parameters like colour and appearance, flavour and taste, body and texture and overall acceptability, the inclusion level of 3.0% of *Cassia auriculata* flower powder was selected as the optimum inclusion level for developing Greek yoghurt with good organoleptic properties as compared to the control Greek yoghurt.

Keywords: Greek yoghurt, Cassia auriculata, standardization, probiotics, sensory evaluation

#### 1. Introduction

Milk is a complete food which acts as an excellent source of protein, vitamins and minerals. India is the biggest milk producer in the world and it has the per capita availability of 394 g of milk available per person. Approximately 50-55% of the milk in India is processed into conventional value added dairy products like cultured, heat desiccated, heat acid coagulated and fat rich dairy products (Arulkumar *et al.*, 2023) <sup>[1]</sup>. Functional foods are referred to food products that not only offer essential nutritions but also have a positive impact on human health. Plant supplements comprise vitamins, minerals, and other biologically active compounds that possess the capacity to improve the functionality of the immune system (Swelam *et al.*, 2021)<sup>[2]</sup>.

Yoghurt is a fermented dairy product which is prepared by lactic acid fermentation of milk with the action of yoghurt culture which consists of *Streptococcus salivarius* subsp. *thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* (Raghunath *et al.*, 2020) <sup>[3]</sup>. Probiotics are active microorganisms which intended to give health beneficial effects to the host. Due to the incorporation of *Lactobacillus acidophilus* like probiotics have resulted in balancing intestinal micro flora, relieve constipation, prevent H. pylori infection and enhance immune system (Sreeja *et al.*, 2013) <sup>[4]</sup>. Greek yoghurt is also said to be concentrated yoghurt or strained yoghurt which is originated from the Middle Eastern parts of the world. Greek yoghurt is traditionally prepared by draining whey from the plain yoghurt by hung it in the cheese cloth bag for 6 to 10 hours in refrigeration condition. In modern method of production centrifugation and ultrafiltration were employed (Jaoude *et al.*, 2010) <sup>[5]</sup>. Greek-style yoghurt offers lots of nutritional and health benefits over conventional yoghurt. Greek yoghurt had increased total solids content, creamy texture and rich in protein content by 2-2.5 times of regular yoghurt (Chandan *et al.*, 2013) <sup>[6]</sup>.

As per World Health Organization (WHO), around 80% of the people worldwide use herbal medicines for all their basic medicinal needs. In many regions of the world, medicinal plants have been utilized for thousands of years as traditional medicine for treating a wide range of human health issues. In India, the use of plants as a medicine was regarded as an essential aspect in the medicine field (Sahoo *et al.*, 2021)<sup>[7]</sup>.*Cassia auriculata* is a shrub with bright yellow coloured flowers belong to Fabaceae family which is found in India and several other parts of Asia. *Cassia auriculata* is also known as Tanner's cassia which a Telangana state flower known by different names such as Aavaram in Tamil, Tangedu in Telugu, Avraike in Kannada.

The phytoconstituents which found in the flowers of *Cassia auriculata* are phenols, flavonoids, terpenoids, alkaloids, saponins and glycosides which exhibit therapeautic properties (Guruprasad and Reddy, 2016) <sup>[8]</sup>.The flowers of *Cassia auriculata* were used for the treatment of diabetes, throat infection, nocturnal emissions and to treat urinary discharges and its plant parts were used in treatment of liver toxicity, microbial infection, inflammation, pyrexia and ulcers (Meena *et al.*, 2019) <sup>[9]</sup>. *Cassia auriculata* which is an Ayurvedic medicine used as "Avarai Panchaga Chooranam" and used as a main ingredient in Kalpa herbal tea (Yesurai *et al.*, 2012) <sup>[10]</sup>.

In Greek yoghurt incorporation of *Cassia auriculata* flowers which contains bioactive phytochemicals which would increase the nutritional quality of the developed product results in health benefiting properties such as antimicrobial, antifungal, antioxidant, anti-inflammatory and antidiabetic property (Singh *et al.* 2013)<sup>[11]</sup>.

Nowadays there is an increase in demand for healthful foods that have beneficial properties along with reduced calorie content. Today's generation is increasingly drawn to have innovative, nutritious food distinguished by good taste, lower in fat and notable health benefits. Therefore developing of Greek yoghurt using *Lactobacillus acidophilus* (LA-5) and incorporated with *Cassia auriculata* flower powder improves the human health functions as mentioned above.

#### 2. Materials and Methods

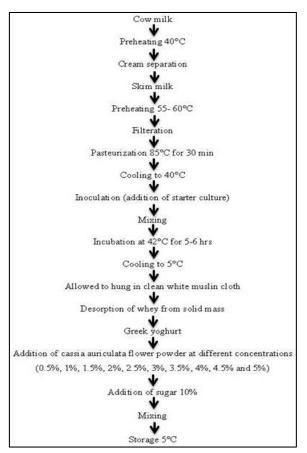
*Cassia auriculata* flowers were collected from the villages of Namakkal, Tamil Nadu. Initially the flowers of *Cassia auriculata* were cleaned and shadow dried. Then it is spread over in the aluminium trays and kept it in contact with hot air and dried by using the cabinet tray dryer at the temperature of 50 °C for 6-8 hrs. The dried flowers were powdered by using the mechanical mixture and stored in an air tight container.

Design for the different treatments of Greek yoghurt as follows

Treatments	Details			
Control	Greek yoghurt			
$T_1$	Greek yoghurt with 0.5 % of Cassia auriculata flower powder			
$T_2$	Greek yoghurt with 1.0 % of Cassia auriculata flower powder			
<b>T</b> 3	Greek yoghurt with 1.5 % of Cassia auriculata flower powder			
$T_4$	Greek yoghurt with 2.0 % of Cassia auriculata flower powder			
T5	Greek yoghurt with 2.5 % of Cassia auriculata flower powder			
$T_6$	Greek yoghurt with 3.0 % of Cassia auriculata flower powder			
<b>T</b> <sub>7</sub>	Greek yoghurt with 3.5 % of Cassia auriculata flower powder			
T <sub>8</sub>	Greek yoghurt with 4.0 % of Cassia auriculata flower powder			

As per the manufacturing procedure of De, (2004) <sup>[12]</sup> the plain yoghurt was prepared. Desai, (2022) <sup>[13]</sup> described the procedure for production of Greek yoghurt from plain yoghurt through traditional method using cheese cloth.

**2.1 Preparation of Greek yoghurt using** *Lactobacillus acidophilus* (LA-5) and *Cassia auriculata* flower powder Flow chart for the preparation of Greek yoghurt using *Lactobacillus acidophilus* (LA-5) and *Cassia auriculata* flower powder



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For the preparation of Greek yoghurt, skim milk is obtained by cream separation of cow milk and then it is preheated to 60 °C. The skim milk is pasterurized at the temperature of 85 °C for 30 minutes and then rapidly cooled to 40 °C. At this stage the yoghurt starter culture of Streptococcus thermophilus and a probiotic culture of Lactobacillus delbrueckii spp. bulgaricus (Chr. Hansen, Denmark) and Lactobacillus acidophilus (LA-5) from (Chr. Hansen, Denmark) were added to the milk. The inoculated milk is kept in incubator at 42±1 °C until the setting of yoghurt curd. After reaching certain pH the yoghurt is placed in a muslin cloth and hung in the refrigeration condition for 6 hours to remove whey thereby the total solids content increases. After that the concentrated voghurt is incorporated with different concentrations of Cassia auriculata flower powder and sugar. Sugar was added at the rate of 10 percent (w/v) in the Greek yoghurt and the powdered Cassia auriculata flowers were roasted in ghee then added at the concentrations of 0.5%, 1%, 1.5%, 2%, 2.5%, 3%, 3.5% and 4% in Greek yoghurt. All the contents

were blended mechanically and stored at the temperature of  $5\pm1$  °C.

#### **3. Results and Discussions**

### 3.1 Sensory evaluation for the inclusion of optimum level of *Cassia auriculata* flower powder in the Greek yoghurt

Table 1 represents the Mean±SE values for the control Greek yoghurt and various treatments of *Cassia auriculata* flower powder incorporated Greek yoghurt in terms of color and appearance, flavor and taste, body and texture, and overall acceptability scores. It is observed that there is a no significant difference ( $p \ge 0.05$ ) was observed between the treatments (T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>6</sub>) and control. In Greek yoghurt the sensory scores were favorable up to addition of 3.0% (T<sub>6</sub>) of *Cassia auriculata* flower powder. Beyond that level the sensory scores will be significantly decreased. Therefore a maximum tolerable level of inclusion of 3.0% of *Cassia auriculata* flower powder was selected and added to the Greek yoghurt for further research studies.

 Table 1: Mean± SE values for the sensory evaluation of Greek yoghurt using Lactobacillus acidophilus (LA-5) and incorporating with various levels of Cassia auriculata flower powder

Parameters	Colour and Appearance	Flavour and Taste	Body and Texture	Overall acceptability
Treatments				
Control	8.83±0.11ª	8.85±0.11 <sup>a</sup>	8.81±0.11 <sup>a</sup>	8.83±0.15ª
$T_1$	8.79±0.12ª	8.76±0.13 <sup>a</sup>	8.75±0.15 <sup>a</sup>	8.78±0.13ª
$T_2$	8.77±0.12ª	8.73±0.12 <sup>a</sup>	8.72±0.12 <sup>a</sup>	8.74±0.12ª
T3	8.74±0.11ª	8.71±0.11 <sup>a</sup>	$8.68 \pm 0.08^{a}$	8.71±0.11ª
$T_4$	8.71±0.11ª	8.68±0.11 <sup>a</sup>	8.63±0.12 <sup>a</sup>	8.67±0.11 <sup>a</sup>
T5	8.69±0.15 <sup>a</sup>	8.65±0.21 <sup>a</sup>	8.61±0.12 <sup>a</sup>	$8.65 \pm 0.09^{a}$
$T_6$	8.65±0.11ª	8.67±0.17 <sup>a</sup>	8.59±0.13ª	8.63±0.11 <sup>a</sup>
T <sub>7</sub>	7.89±0.14 <sup>b</sup>	7.83±0.12 <sup>b</sup>	7.62±0.22 <sup>b</sup>	7.65±0.15 <sup>b</sup>
T <sub>8</sub>	7.51±0.12°	7.42±0.12 <sup>c</sup>	7.44±0.17°	7.42±0.18°

Different superscripts in a column differ significantly (*p*<0.01)

#### **3.2** Colour and Appearance

From the above table 1 the mean±SE value of colour and appearance vary from 7.51±0.12 to 8.83±0.11. Statistical analysis reported that there is a no significant  $(p \ge 0.05)$ difference was observed in the colour and appearance between control and different treatments (T1, T2, T3, T4, T5 and  $T_6$ ). Then there is a significant (p < 0.01) difference was noticed between treatments ( $T_7$  and  $T_8$ ), when compared with control and other six treatments. With an increase in the amount of addition of Cassia auriculata flower powder results, decline in the scores of colour and appearance. The bright golden yellow colour of the product is due to the presence of flavonoids present in Cassia auriculata flower (Meena et al., 2019)<sup>[9]</sup>. In accordance with (Vijay et al., 2022) <sup>[14]</sup> due to the increase in the addition of *Moringa oleifera* leaf powder in Greek yoghurt results in the greenish colour which lowers the sensory scores.

#### 3.3 Flavour and Taste

The mean±SE value of flavour and taste ranged from 7.42±0.12 to 8.85±0.11. The present research found that in terms of taste and flavour scores, there is no significant ( $p \ge 0.05$ ) difference was observed between control and treatments (T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>6</sub>). As a result, T<sub>7</sub> and T<sub>8</sub> differed significantly (p < 0.01) from the other six treatments and the control Due to the addition of Cassia auriculata flower powder the taste and flavour scores were decreased significantly due to the presence of tannins and polyphenols which imparts bitter flavour. The results are in agreement with Świąde *et al.*, (2020)<sup>[15]</sup>, which infused tea in set yoghurt

which developed a bitter flavour due to the presence of polyphenols. In accordance with Dhawi *et al.*, (2020) <sup>[16]</sup> reported that the flavour scores of fenugreek and *Moringa oleifera* seed flour fortified yoghurt decreased with increase in concentration of fenugreek and *Moringa oleifera* seed flour.

#### **3.4 Body and Texture**

Based on statistical analysis the scores of body and texture had a no significant  $(p \ge 0.05)$  difference were observed between control and different treatments (T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and  $T_6$ ). The Mean  $\pm$ SE value of body and texture decreased from  $8.81\pm0.11$  to  $7.44\pm0.17$ . In comparison to the other six treatments and the control, T7 and T8 showed a significant (p<0.01) difference. Due to the addition of *Cassia auriculata* flower powder there is significant degree of change in body and texture. It supports the findings of Walkunde et al. (2008) <sup>[17]</sup>, who found that control yoghurt scored highest on body and texture as compared with the yoghurt samples incorporated with guava pulp. It shows that Greek yoghurt added with Cassia auriculata flower powder reported a decline in scores of body and texture due to the presence of fiber content in the flower powder. Vijay et al., (2022)<sup>[14]</sup> reported that in yoghurt addition of Moringa olifera which contains fiber contains reduced the body and texture scores drastically.

#### 3.5 Overall acceptability

The statistical analysis of the overall acceptability reported that the Mean±SE values ranged from 7.42±0.18 to 8.83±0.15. Whereas there is no significant difference ( $p \ge 0.05$ )

was observed between the control and different treatments (T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>6</sub>) of Greek yoghurt. The treatment T<sub>7</sub> and T<sub>8</sub> were significantly (p<0.01) differing in colour, taste and texture than the control and other six treatments. However an increase in the inclusion of *Cassia auriculata* flower powder results in the bright yellow colour and bitter taste when added with sugar. Karki *et al.*, (2020) <sup>[18]</sup> analyzed the sensory evaluation of yoghurt incorporated with *Moringa olifera* in different concentrations by using 9 point hedonic scale. The findings are in agreement with Silva *et al.* (2021) <sup>[19]</sup>, who reported the inclusion of bovine colostrum significantly lowers the scores of overall acceptability.

Vijay *et al.*, (2022)<sup>[14]</sup> due to the increase in the addition of *Moringa oleifera* leaf powder in Greek yoghurt results in decrease in sensory scores of colour, flavour, texture and overall acceptability due to the increase in concentration which gives bitter flavour. Based on the overall acceptability, 3% of *Cassia auriculata* flower powder incorporated Greek yoghurt met the tolerable level of acceptability and selected as the optimum inclusion level in the final developed product. The incorporation level of 3.0% of *Cassia auriculata* flower powder in Greek yoghurt is standardized and subjected for further research studies.

#### 4. Conclusion

From the above study it has been concluded that addition of *Cassia auriculata* flower powder up to 3% level in Greek yoghurt using *Lactobacillus acidophilus* (LA-5) without altering any sensory properties.

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