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Study on Formulation and Standardization of Nutraceutical Colostrum Milk Bar

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Abstract

With increasing urbanization, people are finding less time to prioritize health, leading to a growing demand for convenient, nutritious food options. This study focused on formulating a functional colostrum-based milk bar enriched with butterfly pea (Clitoria ternatea) extract and natural sweeteners like honey and jaggery, designed to cater to the dietary needs of the rising middle-class population. A total of six variants were developed using 5%, 10%, and 15% concentrations of both sweeteners. Among these, two specific formulations one with 15% butterfly pea extract and honey, and the other with 10% extract and jaggery, were combined with 100 mL of cow colostrum milk, saffron, and cardamom to produce a high-protein nutraceutical bar. Sensory evaluation indicated favorable consumer acceptance, particularly for the sweetened samples. The products were stored at 0–4°C, and periodic sensory and microbial assessments confirmed their stability. While the control (T1) lacked sweeteners, T2 (with honey) and T3 (with jaggery) demonstrated enhanced taste and overall appeal. The study concludes that the addition of botanical and natural ingredients significantly improves both the nutritional value and shelf life of the colostrum milk bar, making it a promising functional food product.

Key worlds: Colostrum Bar, Honey, butterfly pea flower.

Introduction:

Colostrum is the first secretion produced by mammals in the initial days following parturition. This nutrient-rich fluid plays a pivotal role in supporting the nutrition, immunity, and overall development of neonates (Kaplan et al., 2022). Unlike mature milk, colostrum contains significantly higher concentrations of total solids, proteins, immunoglobulins, fats, and growth factors, which are essential for the newborn's early physiological and immunological defence mechanisms (Silva et al., 2019). The composition of colostrum varies significantly across species, such as bovine, buffalo, sheep, and human colostrum, each having distinct biological functions and nutrient profiles (Silva et al., 2019; McGrath et al., 2016). In particular, bovine colostrum is noted for its high protein and fat content, with protein levels surpassing those found in mature bovine milk, which typically contains about 3.62% protein and 4.69% fat. The elevated protein content in colostrum is largely attributed to its abundance of immunoglobulins, especially IgG and IgA, which are critical for immune protection in early life. These immunoglobulin levels gradually decline as lactation progresses, reaching substantially lower levels in mature milk 0.72 mg/mL for IgG and 0.13 mg/mL for IgA underscoring the importance of timely colostrum intake by neonates (Silva et al., 2019)

. Numerous factors influence colostrum composition, including species, breed, nutritional management, parity, and post-collection processing methods. Given its dense nutritional and functional profile including bioactive compounds like enzymes and growth-promoting factors bovine colostrum has garnered considerable interest in both the pharmaceutical and functional food industries. Despite centuries of traditional use, the full spectrum of health benefits offered by bovine colostrum in humans is still under scientific exploration. Emerging

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research has highlighted its potential in enhancing immunity, gut health, and athletic performance. Additionally, studies on colostrum from other species, such as equine colostrum, have further supported its applications in food and human health. Hence, this review aims to examine the application of bovine colostrum in human nutrition, focusing on its rich nutrient composition and the functional properties of its bioactive compounds. It also presents current scientific evidence supporting its health benefits and potential for therapeutic and dietary supplementation.

Methods:

Flow sheet for preparation of butterfly pea flower extract

Take fresh Butterfly Pea flower extract
↓
Take 100 ml water and boil the petals for 3-4 minutes
↓
Strain the butterfly pea flower extract
↓
Store the extract in a beaker

Flow sheet for preparation of nutraceutical bovine colostrum milk bar

Receiving of Bovine Colostrum Milk (100 ml for each sample)
↓
Preparation of Butterfly pea extract
↓
Adding Honey / Jaggery and butterfly pea extract in bovine colostrum milk
(15%Honey and butterfly pea extract / 10%Jaggery and Butterfly pea extract)
↓
Sprinkle cardamom powder and kesar for additional flavours
↓
Heating pressure cooker with adding water at base
↓
Place the samples in pressure cooker, cover samples by dish and put lid without whistle for 20 minutes
↓
Take the samples out of pressure cooker
↓
Cool it down at room temperature
↓
Make pieces with the help of knife
↓
Serve or store it in refrigerator

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Determination of Colour by Hunter Colour Lab

To determine colour using Hunter lab, a sample is placed in the spectrophotometer and the reflectance or transmittance of light is measured. The data is then analysed using the L*, a*, and b* scales to determine the colour of the sample.

Determination of moisture content

A piece of each sample was taken and weight was measured. Then placed it in hot air oven for 5 hours at 110℃. The initial weight of samples was 10g each T1, T2 and T3 respectively. The final weight was taken after 5 hours was T1 – 4.28g, T2 – 3.23g and T3 – 2.49g. The total moisture content was calculated by formula and readings were noted down.

Determination of Ash content

To determine ash content muffle furnace was preheated. The weight of empty crucible was taken. The samples' weight was taken is 5g each T1, T2 and T3 respectively. The samples in crucible were placed in Muffle Furnace. The samples were placed in it for 5 hours at 500-600℃. After 5 hours the weight of ash with crucible was taken and ash content was calculated.

Determination of fat content

The fat content was determined by Soxhlet apparatus method. The moisture free samples were wrapped in Whatman no. 1 filter paper and placed in the thimble. Placed condenser on it. The solvent was taken in round bottom flask. The process of fat determination was completed after the 8 cycles of distillation and condensation of solvent. After that the weight of round bottom flask was taken with the solvent. Solvent was evaporated and then the weight of fat was calculated.

Determination of Protein content

The fat free sample was further used for the determination of protein content by kjeldahl method. It was determined by 3 steps like digestion, distillation and titration respectively. The chemical required was prepared like catalyst (potassium sulfate + copper sulphate), 4% boric acid, 40% NaOH, and 0.1N HCl. 0.2g of each sample were taken for digestion by adding 10ml conc. H_2SO_4. The samples were heated for 1-2 hours at 420℃. The samples were cooled down. Then it is followed by distillation process by adding 10 ml conc. H_2SO_4 in tube, adding 10ml NaOH in the alkali container and 25ml boric acid in conical flask. Then the distillation process

carried out for 9 minutes. After that the titration was carried out with HCl by adding bromocresol green and methyl red indicator. It was completed by disappearing of green colour and it turns into light pink colour.

Determination of pH content

The pH meter was standardized by dipping the pH rod in 0.1N Conc. HCl for 24 hours. Then for calibration of pH meter Buffer solutions was prepared i.e. pH 7 and pH 4 for acidic samples and pH 9.20 for base samples. pH meter was calibrated and readings of sample T1, T2 and T3 was taken.

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Determination of microbial growth in samples

The PDA media was prepared by using autoclave. Then serial dilution of samples was carried out. The media was poured in laminar air flow in proper hygienic conditions. Then the serially diluted samples i.e. 10-4 for each sample was inoculated by using micropipette by pour plate method. Then it was placed in incubator for 24-48 hours. After that the growth of micro organisms was seen is more. It couldn't count.

Results:

Chemical composition of bovine colostrum bar

The chemical composition of bovine colostrum bars provides essential insights into their nutritional quality and shelf stability. Three variations of the product—T1 (control), T2 (with honey), and T3 (with jaggery)—were analyzed for key parameters including moisture, ash, fat, protein content, and pH. The results are summarized in Table 1.

Table no 01: Chemical composition of Bovin Colostrum Bar

Sr. No	Parameter (%)	Sample T1	Sample T2	Sample T3
1	Moisture	57.1	67.7	75.1
2	Ash	0.82	0.98	1
3	Fat	7	2.2	3.9
4	Protein	0.36	0.29	0.1
5	pH	6.46	6.71	6.21

*Each value is an average of three determinations

Where: **T1**: Control sample (no sweetener), **T2**: Sample with Honey, **T3**: Sample with Jaggery

The moisture content ranged from 57.1% in T1 to 75.1% in T3. The highest moisture content was observed in the jaggery-enriched sample (T3), followed by the honey-enriched sample (T2), while the control (T1) had the lowest value. The increase in moisture content in sweetener-enriched samples can be attributed to the hygroscopic nature of honey and jaggery, which tend to absorb and retain more water. Elevated moisture may improve texture and palatability but could also affect shelf life by increasing susceptibility to microbial spoilage. Ash content, which reflects the total mineral content, showed a slight increase across the samples, with values of 0.82%, 0.98%, and 1.00% for T1, T2, and T3 respectively. The addition of jaggery in T3 resulted in the highest ash content, consistent with the fact that jaggery is known to contain significant amounts of minerals such as iron, calcium, and magnesium. This suggests that T3 may offer additional nutritional benefits in terms of mineral fortification.

Fat content varied significantly among the samples. The control (T1) had the highest fat content at 7%, while T2 and T3 had 2.2% and 3.9% respectively. The reduction in fat in the treated samples could be due to the dilution effect caused by the addition of liquid sweeteners, which likely decreased the relative fat concentration in the final product. This reduction may be beneficial from a calorie-conscious perspective.

The protein content, measured on a fat-free basis, was found to be highest in T1 (0.36%), followed by T2 (0.29%) and lowest in T3 (0.10%). The observed decline in protein with the addition of sweeteners may again be due to a dilution effect, as the added honey and jaggery do not contribute significantly to protein content. However, all samples still retain some of the inherent protein value of bovine colostrum, which is rich in bioactive peptides and immunoglobulins. The pH values ranged from 6.21 to 6.71, indicating near-neutral to slightly acidic conditions.

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T2 exhibited the highest pH (6.71), possibly due to the buffering capacity of honey, while T3 had the lowest (6.21). A lower pH, as seen in T3, may improve microbial stability but could also influence taste and texture. Maintaining pH within a moderate range is important for both sensory acceptance and microbiological safety.

The addition of natural sweeteners like honey and jaggery to bovine colostrum bars significantly affects their chemical composition. While jaggery enhances mineral content and moisture, it reduces protein and fat concentrations. Honey contributes similarly but maintains a slightly higher pH. These variations suggest that formulation choices can be optimized based on the desired balance between nutrition, shelf life, and consumer preference. Among the samples, T3 offers better mineral content and moisture, whereas T1 retains higher fat and protein values.

Colour analysis of Prepared Product by Hunter colour lab.

Colour is a critical sensory parameter influencing the visual appeal and consumer acceptance of food products. The color values of the bovine colostrum bars were evaluated using the Hunter Lab color scale, which includes **L*** (lightness), **a*** (red-green spectrum), and **b*** (yellow-blue spectrum). The results are summarized in table No 02.

Table no 02: Colour analysis of Prepared Product by Hunter colour lab.

Sr. No	Sample	L*	a*	B*
1	T1	70.66	-7.30	7.90
2	T2	72.79	-4.36	11.71
3	T3	62.35	3.33	19.42

*Each value is an average of three determinations

Among all samples T2 (honey-sweetened sample) had the highest L* value (72.79), indicating the lightest color. T1 (control) followed with 70.66. T3 (jaggery-sweetened) showed the *lowest L value** (62.35), indicating a darker appearance. The darker shade in T3 can be attributed to the natural deep brown color of jaggery, which likely influenced the overall product tone during processing. Honey, being lighter in color, resulted in a brighter and more appealing appearance. The *a values** determine the red-green hue T1 and T2 showed negative a* values (−7.30 and −4.36, respectively), indicating a greenish tone, possibly due to the presence of butterfly pea extract which contains anthocyanins that can impart blue to greenish shades depending on pH. T3 exhibited a positive a* value (3.33), signifying a reddish tone, likely contributed by the caramel-like hue of jaggery during heating and mixing.

The *b values** reflect the yellow-blue spectrum. All samples had positive b* values, confirming a yellowish hue. The b* value was highest in T3 (19.42), indicating a pronounced yellowish-brown color, consistent with the jaggery addition. T2 also had a notable yellow tone (11.71), while T1 had the least yellow (7.90), aligning with its unsweetened composition. The Hunter Lab color analysis revealed that the addition of sweeteners significantly affects the visual properties of bovine colostrum bars; Honey (T2) improved lightness and provided a mild yellow tint, potentially enhancing visual appeal. Jaggery (T3) contributed to a darker, reddish-yellow appearance, consistent with its natural color and caramelization during processing. Control sample (T1) maintained a moderate brightness with a slight greenish tone from butterfly pea extract. These color differences can influence consumer preferences, with lighter and warmer tones generally being more appealing in dairy-based products. Hence,

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sweetener choice not only alters taste and nutrition but also significantly affects the aesthetic quality of the final product.

Sensory Evaluation of prepared products

Sensory evaluation is a key method for assessing consumer perception and acceptability of food products, providing insights into appearance, taste, flavour, texture, and overall liking. In this study, sensory analysis of bovine colostrum bars was conducted using the **9-point hedonic scale**, one of the most commonly employed tools for measuring food acceptability. Semi-trained panellists in the evaluation of prepared products and result are presented on table no 03.

Table No 03: Sensory Evaluation of prepared product.

Sr. No	Sample	Appearance	Taste	Colour	Texture	Overall Acceptability
1	T1	9	8	8.5	8	7.5
2	T2	9	8	9	9	9
3	T3	9	8.5	8.5	8.5	8.5

*Each value is an average of three determinations

All three samples received the highest score of 9 for appearance, indicating that the products were visually appealing and well-presented, likely due to uniform shape, color, and clean finishing. Taste is a primary factor in product acceptance. T3 (jaggery) slightly outperformed the others in this parameter with a score of 8.5, possibly due to the rich, caramelized sweetness of jaggery. T1 and T2 both received 8, suggesting that the addition of honey or no sweetener maintained an acceptable but less intense flavor profile. T2 received the highest rating for color (9), indicating that honey contributed positively to the product's visual appeal, possibly giving a light golden hue. T1 and T3 both scored 8.5, showing good but slightly less vibrant or appealing color. The texture was best rated in T2 (9), suggesting that honey helped maintain a soft and cohesive structure, likely due to its natural viscosity and binding properties. T3 followed with 8.5, while the control (T1) scored slightly lower (8), possibly due to the lack of a sweetening and softening agent.

T2 (with honey) achieved the highest overall acceptability score of 9, indicating that panelists found this formulation to be the most favorable in terms of combined sensory attributes. T3 also performed well with 8.5, suggesting a strong consumer preference for naturally sweetened products. T1 had the lowest score (7.5), likely due to its plain taste and firmer texture. The sensory evaluation clearly demonstrates that the addition of natural sweeteners improves the sensory appeal of the bovine colostrum bar. Among all formulations, Sample T2 (with honey) was the most preferred by the panel for its appealing color, soft texture, and balanced taste. These findings indicate that honey not only enhances the nutritional profile but also significantly boosts consumer acceptability of the product. Incorporating such natural ingredients can be a key factor in developing successful nutraceutical products for health-conscious markets.

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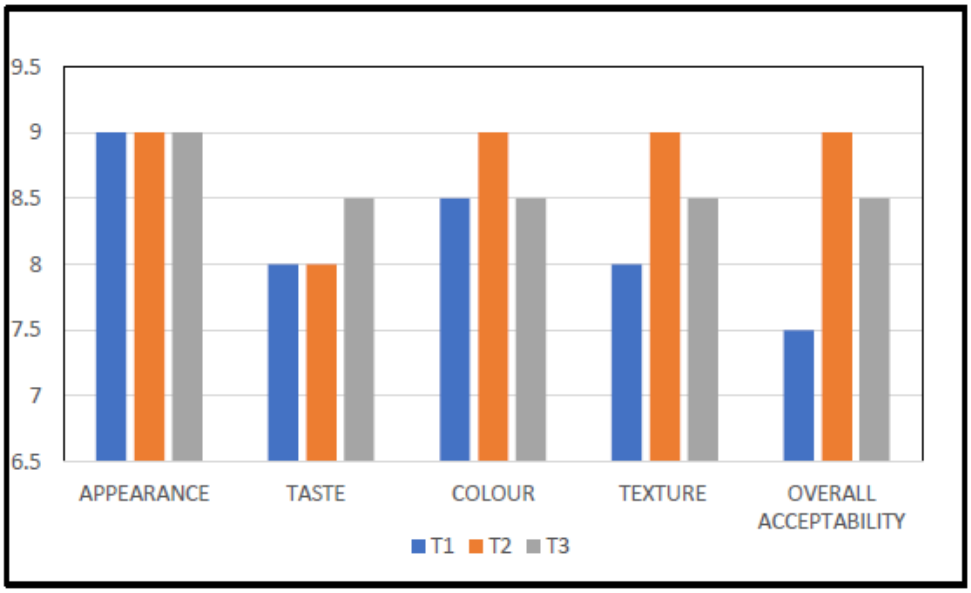

Fig No 01: Sensory Evaluation of prepared product

Total plate count of colostrum milk bar

Microbial analysis is an essential aspect of food quality and safety evaluation. The total plate count (TPC) of the Colostrum Milk Bar samples was conducted to assess microbial load at serial dilution levels (10^{-1} to 10^{-4}). The results, expressed in colony-forming units per milliliter (CFU/mL), are presented in the table below:

Table No 04: Total plate count of colostrum milk bar in CFU / ml

Sr. No	Sample plate	10^{-1}	10^{-2}	10^{-3}	10^{-4}
1	T1	TNTC	300	250	200
2	T2	TNTC	300	260	220
3	T3	TNTC	250	280	180

*Each value is an average of three determinations

Where : TNTC - Too Numerous To Count

T1 (Control sample): The control, without any added natural sweeteners, exhibited high microbial load, with counts too numerous to count (TNTC) at the 10^{-1} dilution and high counts even at higher dilutions. This suggests a lack of antimicrobial agents in the formulation, making it more prone to microbial growth.

T2 (With honey): The sample containing honey also showed TNTC at 10^{-1} and high counts at other dilutions, with 300 CFU/mL at 10^{-2}, 260 CFU/mL at 10^{-3}, and 220 CFU/mL at 10^{-4}. Although honey is known for its natural antimicrobial properties due to low water activity, acidity, and hydrogen peroxide content, the results indicate that its effect on microbial reduction in this matrix may be limited or offset by its sugar content, which can support microbial growth under certain conditions.

T3 (With jaggery): The jaggery-containing sample also showed TNTC at 10^{-1}, but comparatively lower microbial counts at higher dilutions (250, 280, and 180 CFU/mL at 10^{-2}, 10^{-3}, and 10^{-4}, respectively). While the microbial count at 10^{-3} was slightly higher, the overall trend suggests better control over microbial growth than the control. Jaggery contains polyphenolic compounds and minerals that may contribute to its antimicrobial activity.

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Conclusion

The study focused on developing a nutraceutical bovine colostrum bar using butterfly pea extract, honey, and jaggery as natural additives. Among the three samples, T2 (with honey) showed superior sensory attributes including appearance, texture, and overall acceptability. Chemical analysis revealed that T2 had balanced moisture, fat, and pH levels, while T3 (with jaggery) had the highest ash and moisture content. Color evaluation showed T2 had the most appealing lightness and tone. Microbial analysis confirmed all samples were within safe limits during storage. Overall, the addition of honey enhanced both the nutritional and sensory quality of the colostrum bar, making it a promising functional food product.

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