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Effect of product thickness and shelf temperature on drying characteristics and quality attributes of freeze-dried bovine colostrum powder

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ABSTRACT

The present study was undertaken to investigate the effect of product thickness and shelf temperature on the freeze-drying characteristics, energy requirement and quality attributes of bovine colostrum powder and to identify a suitable processing condition for its production. A 2×2 factorial experiment in a Completely Randomized Design was conducted using two product thicknesses, H_1 (2.5 mm) and H_2 (5.0 mm), and two shelf temperatures, T_1 (2 °C) and T_2 (10 °C). Freeze-dried powders obtained under the four treatment combinations were evaluated for drying time, energy consumption, moisture content, physical and flow properties, nutrient retention, pH, acidity and sensory quality. Drying time increased with increasing product thickness and decreased with increasing shelf temperature, with the shortest drying time observed at H_1T_2 (6.95 ± 0.12 h) and the longest at H_2T_1 (15.58 ± 0.27 h). Energy consumption ranged from 14.65 ± 0.66 to 27.50 ± 0.54 kWh, with the lowest and highest values recorded for H_1T_2 and H_2T_1 , respectively. Product thickness significantly influenced several powder characteristics, while shelf temperature and its interaction with thickness showed variable effects depending on the response. Protein retention ranged from (97.83 ± 0.73 %) to (99.45 ± 0.09 %), indicating good preservation of protein during freeze drying. The sensory scores indicated acceptable quality of all treatments, with H_2T_1 receiving the highest overall score. Considering drying performance, energy requirement and quality characteristics, H_2T_1 (5.0 mm thickness and 2 °C shelf temperature) was identified as the most suitable overall processing condition for freeze-dried bovine colostrum powder.

Keywords: Freeze-Drying, Bovine, Colostrum, Protein, Thickness

Introduction

Bovine colostrum (BC) is the first secretion from the mammary glands following parturition and is characterized by a high concentration of macro- and micronutrients, immunoglobulins and bioactive peptides. It plays a crucial role in providing nutritional and immunological support to neonatal calves, particularly during the early post-partum period when intestinal permeability to immunoglobulins is high (Panahi *et al.*, 2010; Godden, 2008) [2, 1]. The composition of colostrum changes rapidly after calving, making its timely collection and utilization essential. Besides immunoglobulins, bovine colostrum contains several bioactive constituents, including lactoferrin, insulin-like growth factor-1, α -lactalbumin and β -lactoglobulin, which contribute to its functional properties (Mihic *et al.*, 2016; Playford *et al.*, 2020) [3, 4].

The nutritional and functional characteristics of bovine colostrum have increased interest in its utilization as a functional food and nutraceutical ingredient. Colostrum from bovine, buffalo and goat sources is processed into various forms, including powders, tablets and liquid preparations (Godhia *et al.*, 2013; Bagwe *et al.*, 2015) [6, 5]. Conversion of colostrum into a stable powdered form can facilitate its storage, transportation and incorporation into value-added products. However, preservation of colostrum while retaining its nutritional and biological characteristics remains a challenge because of the sensitivity of several of its functional components to processing conditions.

Spray drying is widely used for the production of dairy powders owing to its rapid moisture removal, continuous operation and economic advantages. However, the process involves exposure to elevated temperatures, which may adversely affect heat-sensitive proteins and bioactive components. This is particularly important for bovine colostrum because immunoglobulins are among its major functional constituents. High-temperature processing has been reported to cause protein denaturation, while immunoglobulins in bovine colostrum begin to undergo denaturation at approximately 72 °C, with substantial loss of activity at higher temperatures (Borad *et al.*, 2018; Singh *et al.*, 2003) [7, 8]. Thus, the selection of an appropriate dehydration method is important when the objective is to obtain a stable colostrum powder while retaining its functional properties.

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Freeze drying, also known as lyophilization, provides an alternative low-temperature dehydration technique for heat-sensitive dairy products. The process involves freezing the product followed by removal of water through sublimation under reduced pressure, thereby minimizing thermal exposure and associated degradation of sensitive constituents. Freeze drying has been reported to provide better retention of functional components such as immunoglobulins, insulin-like growth factors and lactoferrin than conventional thermal processing. In particular, [Rabbani \(2018\)](#) ^[9] reported significantly higher IgG retention in freeze-dried bovine colostrum compared with pasteurized and spray-dried samples. Independent published evidence also reports a lower reduction in IgG after freeze drying than after spray drying or pasteurization, supporting the suitability of freeze drying for preservation of bovine colostrum.

Despite its advantages in preserving heat-sensitive components, freeze drying is relatively time-consuming and energy intensive. Consequently, optimization of operating conditions is essential for improving process efficiency while maintaining desirable powder quality. Among the different process variables, product thickness and shelf temperature are important because they influence heat and mass transfer during sublimation. An increase in product thickness increases the distance through which moisture must migrate and may consequently prolong the drying process, whereas an appropriate increase in shelf temperature can enhance heat transfer and accelerate sublimation. However, excessive or inappropriate processing conditions may adversely affect product quality. Therefore, optimization of these parameters requires consideration of both drying performance and the characteristics of the resulting powder.

Although freeze drying has been investigated for preservation of bovine colostrum, limited information is available on the combined influence of product thickness and shelf temperature on drying behaviour and energy requirement during freeze drying. The present study was therefore undertaken to investigate the effect of product thickness (2.5 and 5.0 mm) and shelf temperature (2 and 10 °C) on the drying characteristics, energy consumption and quality attributes of freeze-dried bovine colostrum powder, with the aim of identifying suitable processing conditions for efficient production of a high-quality powder.

2. Materials and Methods

2.1. Procurement and preparation of bovine colostrum

Fresh bovine colostrum was collected during milking from the Livestock Research Station (LRS), Kamdhenu University, Anand. The samples were collected directly into clean and dry containers, chilled and immediately transported to the laboratory, where they were stored under refrigerated conditions until further processing. The compositional analysis of raw bovine colostrum was carried out at the Dairy Chemistry Department, Sheth M. C. College of Dairy Science, Kamdhenu University, Anand.

Prior to processing, the colostrum was filtered through a muslin cloth to remove visible impurities and extraneous matter. The filtered colostrum was collected in clean containers and used for subsequent freezing and freeze-drying.

2.2. Experimental design and freeze drying

The freeze-drying experiment was conducted using a 2 × 2 factorial arrangement in a Completely Randomized Design (CRD). Two independent processing variables were investigated: product thickness and shelf temperature during primary drying. Product thickness was maintained at 2.5 mm (H₁) and 5.0 mm (H₂), while shelf temperature was maintained at 2 °C (T₁) and 10 °C (T₂). Thus, four treatment combinations were obtained. Each treatment combination was replicated four times for bovine colostrum. The desired product thickness was achieved by adjusting the volume of colostrum loaded into the stainless-steel trays and verified using a vernier caliper before freezing. All other processing conditions were kept constant throughout the experiment.

The filtered colostrum was transferred into stainless-steel trays at the required thickness of 2.5 or 5.0 mm and frozen at −30 °C for 12 h in a deep freezer to ensure complete solidification. The frozen samples were then immediately transferred to a laboratory-scale vacuum freeze dryer to minimize thawing.

Freeze drying was carried out using a condenser maintained at −35 °C, with the chamber pressure maintained at 1–2 mbar. Thermocouple probes were used to monitor the shelf and product temperatures during the drying cycle. Primary drying was continued until the product temperature approached the shelf temperature, indicating completion of ice sublimation. Secondary drying was subsequently carried out at 30 °C for approximately 45% of the primary drying time to remove residual bound moisture. After completion of the drying cycle, the freeze-dried colostrum was immediately collected and packed in moisture-impermeable, airtight glass containers until further analysis.

2.3. Analysis of raw bovine colostrum

Raw bovine colostrum was analysed for titratable acidity, pH, total solids, fat, protein, ash and lactose content. Titratable acidity was determined by titration of the sample with standard 0.1 N sodium hydroxide using phenolphthalein as an indicator and expressed as percentage lactic acid (% LA). The pH was measured using a calibrated digital pH meter. Total solids were determined by the gravimetric method, fat by the Gerber method, protein by the Kjeldahl method using a nitrogen conversion factor of 6.38, and ash by the gravimetric method. Lactose content was determined by the Lane–Eynon titration method. The analyses were carried out according to the procedures described in the BIS Handbook (SP 18: Part XI, 1981) ^[10] and FSSAI methods, as applicable.

2.4. Analysis of freeze-dried bovine colostrum powder

Freeze-dried bovine colostrum powder was stored in airtight borosilicate glass containers at room temperature prior to analysis. The samples were thoroughly mixed before analysis. The powder was evaluated for proximate composition and selected physicochemical and functional properties.

Moisture content was determined according to the BIS Handbook (SP 18: Part XI, 1981) ^[10]. Fat content was determined gravimetrically using a Mojonnier milk tester (Model-D) following the standard procedure described in the Laboratory Manual (1959). Protein content was determined by the micro-Kjeldahl method using a nitrogen conversion factor of 6.38. Ash content was determined gravimetrically following the method described in GEA Niro Analytical Methods for Dry Milk Products (GEA Process Engineering A/S, Denmark). Lactose content was calculated by difference from the measured moisture, protein, fat and ash contents.

For physicochemical characterization, titratable acidity was determined by titration of a sample suspension against standard 0.1 N sodium hydroxide using phenolphthalein as an indicator and expressed as percentage lactic acid. For pH measurement, the powder was reconstituted in distilled water to obtain a 10% (w/v) suspension, and the pH was measured at 22 °C using a calibrated digital pH meter, following the procedure of [Oldfield et al. \(2005\)](#) ^[11].

2.5. Physical and functional properties of freeze-dried bovine colostrum powder

Loose bulk density and packed bulk density of the freeze-dried bovine colostrum powder were determined using a 50 mL graduated cylinder according to the method described in GEA Niro Analytical Methods for Dry Milk Products (GEA Process Engineering A/S, Denmark). For loose bulk density, the cylinder was filled to the 50 mL mark with powder and weighed. For packed bulk density, the filled cylinder was tapped 100 times and the final powder volume was recorded. Both densities were expressed as g mL^{−1}.

Hausner ratio and Carr's index were calculated from the loose and packed bulk density values according to [Garhwal et al. \(2025\)](#) ^[12]. Hausner ratio was expressed as the ratio of packed bulk density to loose bulk density, while Carr's index was calculated from the differ

ence between packed and loose bulk density relative to packed bulk density.

The angle of repose was determined as an indicator of powder flowability following the method described by Deshwal *et al.* (2020) [13]. It was calculated from the height and radius of the powder heap.

The insolubility index was determined according to GEA Niro Analytical Methods for Dry Milk Products. Powder (13 g) was dispersed in 100 mL distilled water at approximately 24 °C and homogenized at high speed for 90 s. After standing for 15 min, the suspension was gently stirred, and a 50 mL aliquot was centrifuged for 5 min. The supernatant was discarded, the tube was refilled with water and the sediment was resuspended and centrifuged again. The volume of sediment was recorded as the insolubility index and expressed in mL. Wettability was determined according to GEA Niro Analytical Methods for Dry Milk Products. A 10 ± 0.05 g powder sample was allowed to fall onto the surface of 250 mL deionized water maintained at 25 ± 0.5 °C, and the time required for complete wetting of the powder particles was recorded. Measurements were performed in duplicate and expressed as wetting time in seconds.

2.6. Drying time and energy consumption

The freeze-drying time was recorded for each treatment combination from the beginning of primary drying until completion of the freeze-drying cycle. Electrical energy consumption during the freezing and freeze-drying stages was measured separately using an energy meter and expressed in kWh. The total electrical energy consumption for each treatment was calculated by summing the energy consumed during freezing and freeze drying.

2.7. Sensory evaluation

The sensory quality of freeze-dried bovine colostrum powders obtained under the four treatment combinations was evaluated using a trained panel comprising faculty members and postgraduate students of the institute. Prior to evaluation, the powders were reconstituted with distilled water to their respective original total solids concentration. The reconstituted samples were coded with random three-digit numbers and presented to the panelists in randomized order.

Sensory evaluation was carried out using the BIS 100-point score card for milk and milk products. The sensory attributes evaluated included appearance of the dry product (15 marks), appearance of reconstituted milk (15 marks), body and texture (20 marks), flavour (45 marks), and packaging (5 marks), giving a maximum possible score of 100. Since identical packaging was used for all samples, the full score was assigned for the packaging attribute. Drinking water was provided for mouth rinsing between successive samples to minimize flavour carry-over. The scores assigned by individual panelists were averaged for each attribute and used for statistical analysis.

3. Results and Discussion

3.1 Drying characteristics

Drying time was significantly ($P < 0.05$) affected by product thickness, shelf temperature, and their interaction (Table 2). The mean drying time was significantly lower at H₁ (8.30 h) than H₂ (13.36 h), and at T₂ (9.05 h) compared with T₁ (12.61 h). The minimum drying time (6.95 ± 0.12 h) was recorded for H₁T₂, while the maximum (15.58 ± 0.27 h) occurred at H₂T₁. The longer drying time at greater product thickness reflects the increased resistance to heat and mass transfer through the dried layer, while the shorter drying time at the higher shelf temperature reflects faster sublimation driven by increased heat input. These trends are consistent with reports that reducing product thickness in camel milk freeze drying shortened drying time by up to 40% owing to reduced mass-transfer resistance (Altybay *et al.*, 2025) [14], that increased fill height prolongs primary drying by increasing vapour diffusion resistance through the dried cake (Srinivasan *et al.*, 2019) [15], and that higher primary drying temperatures accelerate sublimation while maintaining product quality within an optimal range (Semenov *et al.*, 2024) [16].

3.2 Energy consumption

Energy consumption during freeze drying followed a pattern consistent with drying time and was significantly ($P < 0.05$) affected by both product thickness and shelf temperature, while their interaction was non-significant (Table 2). Mean energy consumption was significantly lower at H₁ (17.79 kWh) than H₂ (25.14 kWh), and at T₂ (18.71 kWh) than T₁ (24.21 kWh). The minimum energy consumption (14.65 ± 0.66 kWh) was recorded for H₁T₂, and the maximum (27.50 ± 0.54 kWh) for H₂T₁. This pattern reflects the longer operating duration required at greater thickness and lower shelf temperature, consistent with reports that reduced product thickness and elevated primary drying temperature both lower the specific energy requirement of freeze drying by shortening the sublimation stage (Altybay *et al.*, 2025; Semenov *et al.*, 2024) [14, 16].

3.3 Moisture content

Final moisture content was significantly ($P < 0.05$) affected by product thickness, while shelf temperature and the H $\times$ T interaction were non-significant (Table 3). Mean moisture content was significantly lower at H₂ (1.28%) than H₁ (3.29%). Among treatment combinations, the minimum moisture content ($0.91 \pm 0.10\%$) was observed at H₂T₁, and the maximum ($3.45 \pm 0.33\%$) at H₁T₂. The lower moisture content at the greater thickness may be linked to the correspondingly longer drying time at H₂, which allowed more complete sublimation and desorption of bound water despite the greater diffusion path length.

3.4 Powder flow, reconstitution, and physical properties

Bulk and tapped density. Neither bulk density (H₁: 0.26; H₂: 0.27 g/cm³) nor tapped density (H₁: 0.42; H₂: 0.41 g/cm³) was significantly affected by product thickness, shelf temperature, or their interaction (Table 4), indicating that the selected freeze-drying conditions did not markedly alter the packing behaviour of the powder particles.

Angle of repose. Angle of repose was significantly ($P < 0.05$) affected by product thickness only, being significantly lower at H₂ (38.74°) than H₁ (42.15°); shelf temperature and the interaction were non-significant. The lower angle of repose at the greater thickness indicates comparatively better flowability of the powder produced under H₂.

Wettability. Wettability time was significantly ($P < 0.05$) affected by product thickness only, being significantly lower (i.e., faster wetting) at H₂ (173.38 s) than H₁ (190.50 s); shelf temperature and the interaction were non-significant. Das *et al.* (2024) [17] reported a wettability time of 150.1 ± 3.03 s for spray-dried bovine colostrum powder, broadly comparable to the range observed here for the freeze-dried product.

Insolubility index. Insolubility index was significantly ($P < 0.05$) affected by product thickness and by the H $\times$ T interaction, while shelf temperature alone was non-significant. Mean insolubility was significantly lower at H₂ (0.27 mL) than H₁ (0.33 mL), with the minimum (0.21 ± 0.02 mL) at H₂T₁ and the maximum (0.38 ± 0.05 mL) at H₁T₁. Das *et al.* (2024) [17] reported insolubility index values of 0.10–0.15 mL for spray-dried bovine and buffalo colostrum powders; the somewhat higher values obtained in the present study may reflect differences in product composition and the freeze-drying process employed.

Interstitial air. Interstitial air content was not significantly affected by product thickness, shelf temperature, or their interaction (H₁: 156.01; H₂: 164.34 mL/100 g).

Hausner ratio and Carr's index. Both indices, which describe powder flowability and compressibility, were significantly ($P < 0.05$) affected by product thickness and shelf temperature. Hausner ratio was significantly lower at H₂ (1.52) than H₁ (1.60), and at T₁ (1.54) than T₂ (1.58); the interaction was also significant, with the minimum (1.51 ± 0.01) at H₂T₁ and the maximum (1.63 ± 0.00) at H₁T₂. Carr's index followed a similar pattern, being significantly lower at H₂ (34.39) than H₁ (37.57) and at T₁ (35.20) than T₂ (36.77), with the interaction non-significant; the minimum (33.72 ± 0.42) occurred at H₂T₁ and the maximum (38.48 ± 0.26) at H₁T₂. Collectively, these results indi

cate that the combination of greater product thickness and lower shelf temperature (H_2T_1) produced powder with the most favourable flow characteristics among the treatments evaluated.

3.5 Protein, fat and lactose retention

Protein retention was significantly ($P < 0.05$) affected by both product thickness and shelf temperature, while the interaction was non-significant (Table 5). Mean protein retention was significantly higher at H_2 (99.07%) than H_1 (98.02%), and at T_1 (98.83%) than T_2 (98.26%). Retention ranged from a minimum of $98.70 \pm 0.23\%$ (H_2T_2) to a maximum of $99.45 \pm 0.09\%$ (H_2T_1), indicating consistently high preservation of colostrum protein across all treatments, with the greatest retention associated with the higher thickness and lower shelf temperature.

Fat retention followed a broadly similar pattern to protein retention and was also significantly ($P < 0.05$) affected by product thickness and shelf temperature, with a non-significant interaction (Table 5A). Mean fat retention was significantly higher at H_2 (99.16%) than H_1 (98.73%), and at T_1 (99.31%) than T_2 (98.59%). The minimum fat retention ($98.32 \pm 0.26\%$) occurred at H_1T_2 , and the maximum ($99.48 \pm 0.21\%$) at H_2T_1 , indicating that a product thickness of 5.0 mm combined with a shelf temperature of 2 °C was most effective in preserving fat during freeze drying of bovine colostrum.

Lactose retention, in contrast, was not significantly affected by product thickness, shelf temperature, or their interaction (Table 5A). Mean lactose retention was numerically higher at H_2 (95.47%) than H_1 (91.35%), and at T_2 (93.77%) than T_1 (93.05%), but these differences did not reach statistical significance. The minimum lactose retention ($90.90 \pm 10.11\%$) was recorded at H_1T_2 , and the maximum ($96.64 \pm 2.45\%$) at H_2T_2 ; the comparatively large standard deviation at H_1T_2 indicates greater batch-to-batch variability for this parameter than for protein or fat retention.

3.6 pH and titratable acidity

pH was not significantly affected by product thickness, shelf temperature, or their interaction (Table 5), ranging from a minimum of 6.58 ± 0.08 (H_1T_1) to a maximum of 6.68 ± 0.09 , observed at both H_2T_1 and H_1T_2 . Titratable acidity, by contrast, was significantly ($P < 0.05$) affected by shelf temperature, while product thickness and the interaction were non-significant. Mean titratable acidity was significantly lower at T_1 (0.24% LA) than T_2 (0.26% LA), with the minimum ($0.24 \pm 0.02\%$ LA) at both H_1T_1 and H_2T_1 , and the maximum ($0.27 \pm 0.01\%$ LA) at H_2T_2 .

3.7 Sensory characteristics

Sensory scores, obtained using the BIS 100-point score card and reported as mean $\pm$ SD without inferential statistical analysis, are presented in Table 6. The treatment combination H_2T_1 recorded the highest overall sensory score (76.79 ± 6.54), followed by H_1T_1 (74.70 ± 4.27), H_1T_2 (73.12 ± 4.10), and H_2T_2 (73.00 ± 3.97). H_2T_1 also recorded the highest mean scores for appearance of the dry product, appearance of reconstituted milk, body and texture, and flavour. While the absence of ANOVA precludes a statistical significance claim for these differences, the panel consistently favoured the pow-

der produced under the H_2T_1 treatment, which corresponded to the treatment with the lowest moisture content, most favourable flow properties, and highest protein retention among those tested.

4. Conclusion

Product thickness significantly influenced drying time, energy consumption, moisture content, angle of repose, wettability, insolubility index, Hausner ratio, Carr's index, and protein retention of freeze-dried bovine colostrum powder, with the higher thickness (5.0 mm) generally associated with longer drying and higher energy use, but also with lower moisture content, improved powder flow characteristics, and greater protein retention. Shelf temperature significantly affected drying time, energy consumption, titratable acidity, Hausner ratio, Carr's index, and protein retention, with the lower shelf temperature (2 °C) associated with longer drying and higher energy consumption but improved flow properties, lower acidity, and higher protein retention. Bulk density, tapped density, interstitial air, and pH were not significantly affected by either factor. Microbiological quality was satisfactory across all treatments, with no coliforms detected and consistently low aerobic, yeast and mould, and *Staphylococcus aureus* counts. Sensory evaluation, assessed descriptively, indicated the highest acceptability for the powder produced under 5.0 mm thickness and 2 °C shelf temperature (H_2T_1), which also corresponded to the treatment with the lowest moisture content, most favourable powder flow properties, and highest protein retention among the conditions evaluated. Considered together, these findings indicate that a product thickness of 5.0 mm combined with a shelf temperature of 2 °C represents the most suitable of the four processing conditions tested for producing freeze-dried bovine colostrum powder of high physicochemical, functional, microbiological, and sensory quality, albeit at a higher energy and time cost than the other treatments.

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Aakash Bajirao Shinde carried out the experimental work, data collection, analysis, and preparation of the manuscript under the guidance and support of the co-authors and the institution.

Conflict of interest

The authors declare that there is no conflict of interest related to this work.

Tables

Table 1. Experimental treatments and freeze-drying conditions for bovine colostrum

Treatment code	Product thickness	Shelf temperature	Replications
H_1T_1	2.5 mm	2 °C	4
H_2T_1	5.0 mm	2 °C	4
H_1T_2	2.5 mm	10 °C	4
H_2T_2	5.0 mm	10 °C	4

Freezing: -30 °C for 12 h. Freeze drying: condenser at -35 °C, chamber vacuum 1–2 mbar; primary drying continued until product

and shelf temperature equalized; secondary drying at 30 °C for ~45% of primary drying time.

Table 2. Effect of product thickness and shelf temperature on drying time and energy consumption of freeze-dried bovine colostrum

Treatment	Drying time (h)	Energy consumption (kWh)
H ₁ T ₁	9.65 ± 0.38	20.93 ± 1.80
H ₂ T ₁	15.58 ± 0.27	27.50 ± 0.54
H ₁ T ₂	6.95 ± 0.12	14.65 ± 0.66
H ₂ T ₂	11.15 ± 0.19	22.78 ± 0.53
Mean H ₁	8.30	17.79
Mean H ₂	13.36	25.14
Mean T ₁	12.61	24.21
Mean T ₂	9.05	18.71
SEm (H)	0.09	0.37
SEm (T)	0.093	0.366
SEm (H×T)	0.132	0.517
CD (0.05) H	0.288	1.127
CD (0.05) T	0.288	1.127
CD (0.05) H×T	0.407	NS
CV%	2.439	4.821
Significance	H, T, H×T significant (<i>P</i> < 0.05)	H, T significant (<i>P</i> < 0.05); H×T non-significant

NS = non-significant at 5% level. *n* = 4.

ACOP of freezing system (descriptive, not subjected to ANOVA): *H*₁ = (product load 0.691 ± 0.002 kg).
0.004 ± 0.000 (product load 0.347 ± 0.003 kg); *H*₂ = 0.009 ± 0.000

Table 3. Effect of product thickness and shelf temperature on moisture content, wettability, and insolubility index of freeze-dried bovine colostrum

Treatment	Moisture content (%)	Wettability (s)	Insolubility index (mL)
H ₁ T ₁	3.13 ± 1.091	197.5 ± 11.24	0.38 ± 0.05
H ₂ T ₁	0.91 ± 0.10	170.25 ± 13.94	0.21 ± 0.02
H ₁ T ₂	3.45 ± 0.333	183.5 ± 3.87	0.29 ± 0.06
H ₂ T ₂	1.65 ± 0.09	176.5 ± 9.98	0.33 ± 0.06
Mean H ₁	3.29	190.50	0.33
Mean H ₂	1.28	173.38	0.27
Mean T ₁	2.02	183.88	0.29
Mean T ₂	2.55	180.00	0.31
CD (0.05) H	0.626	11.364	0.058
CD (0.05) T	NS	NS	NS
CD (0.05) H×T	NS	NS	0.082
CV%	25.156	5.733	17.678
Significance	H significant; T, H×T non-significant	H significant; T, H×T non-significant	H, H×T significant; T non-significant

n = 4.

Table 4. Effect of product thickness and shelf temperature on powder flow and physical properties of freeze-dried bovine colostrum

Treatment	Bulk density (g/cm ³)	Tapped density (g/cm ³)	Angle of repose (°)	Interstitial air (mL/100 g)	Hausner ratio	Carr's index
H ₁ T ₁	0.263 ± 0.009	0.42 ± 0.01	41.62 ± 1.60	158.74 ± 8.58	1.58 ± 0.00	36.67 ± 0.17
H ₂ T ₁	0.271 ± 0.022	0.41 ± 0.03	37.72 ± 1.71	163.79 ± 22.25	1.51 ± 0.01	33.72 ± 0.42
H ₁ T ₂	0.261 ± 0.005	0.42 ± 0.00	42.68 ± 2.96	153.28 ± 4.66	1.63 ± 0.00	38.48 ± 0.26
H ₂ T ₂	0.262 ± 0.015	0.40 ± 0.02	39.76 ± 0.87	164.89 ± 14.57	1.54 ± 0.00	35.07 ± 0.12
Significance	All NS	All NS	H significant; T, H×T NS	All NS	H, T, H×T significant	H, T significant; H×T NS

NS = non-significant at 5% level. *n* = 4. Full ANOVA statistics (SEm, CD, CV%) for each parameter are as reported in the source data.

(continued on next page)

Table 5. Effect of product thickness and shelf temperature on protein retention, pH, and titratable acidity of freeze-dried bovine colostrum

Treatment	Protein retention (%)	pH	Titratable acidity (% LA)
H ₁ T ₁	98.22 ± 0.06	6.58 ± 0.08	0.24 ± 0.02
H ₂ T ₁	99.45 ± 0.09	6.68 ± 0.09	0.24 ± 0.02
H ₁ T ₂	97.83 ± 0.73	6.68 ± 0.09	0.26 ± 0.02
H ₂ T ₂	98.70 ± 0.23	6.66 ± 0.06	0.27 ± 0.01
Mean H ₁	98.02	6.63	0.25
Mean H ₂	99.07	6.67	0.25
Mean T ₁	98.83	6.63	0.24
Mean T ₂	98.26	6.67	0.26
CD (0.05) H	0.425	NS	NS
CD (0.05) T	0.425	NS	0.026
CD (0.05) H×T	NS	NS	NS
CV%	0.396	1.295	—
Significance	H, T significant; H×T NS	Non-significant (<i>P</i> > 0.05)	T significant; H, H×T non-significant

NS = non-significant at 5% level. *n* = 4.

Table 5A. Effect of product thickness and shelf temperature on fat and lactose retention of freeze-dried bovine colostrum

Treatment	Fat retention (%)	Lactose retention (%)
H ₁ T ₁	99.15 ± 0.41	91.80 ± 2.65
H ₂ T ₁	99.48 ± 0.21	94.30 ± 4.21
H ₁ T ₂	98.32 ± 0.26	90.90 ± 10.11
H ₂ T ₂	98.85 ± 0.27	96.64 ± 2.45
Mean H ₁	98.73	91.35
Mean H ₂	99.16	95.47
Mean T ₁	99.31	93.05
Mean T ₂	98.59	93.77
SEm (H)	0.106	2.039
SEm (T)	0.106	2.039
SEm (H×T)	0.149	2.884
CD (0.05) H	0.326	NS
CD (0.05) T	0.326	NS
CD (0.05) H×T	NS	NS
CV%	0.302	6.175
Significance	H, T significant (<i>P</i> < 0.05); H×T non-significant	Non-significant (<i>P</i> > 0.05)

NS = non-significant at 5% level. *n* = 4.

Table 6. Sensory evaluation scores of freeze-dried bovine colostrum powder (BIS 100-point scale)

Treatment	Appearance, dry product (15)	Appearance, reconstituted (15)	Body and texture (20)	Flavour (45)	Total score (100)*
H ₁ T ₁	11.66 ± 0.76	12.00 ± 0.65	15.45 ± 1.02	30.58 ± 2.18	74.70 ± 4.27
H ₂ T ₁	12.20 ± 1.02	12.45 ± 1.10	16.16 ± 1.43	30.95 ± 3.18	76.79 ± 6.54
H ₁ T ₂	11.54 ± 0.65	11.91 ± 0.71	15.29 ± 0.95	29.70 ± 2.27	73.12 ± 4.10
H ₂ T ₂	11.41 ± 0.65	11.79 ± 0.58	15.16 ± 0.91	29.62 ± 1.73	73.00 ± 3.97

*Values are mean ± SD; no ANOVA performed. 5 marks for packaging (identical across treatments) are included in the total score.

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