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Effects of drying techniques on functional and physicochemical properties of *Chenopodium album* (L.) powder

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Abstract

Drying is an ancient technique that is used to store and preserve the food when they are available in abundance, to be consumed for longer periods. *Chenopodium album* (L.) is a green leafy vegetable with a high nutritive and functional value. In order to avail all these properties of *bathua* throughout the year, the study was aimed to evaluate the compositional, functional and technological properties of *bathua* leaves powder by different drying methods. The compositional, functional and technological properties of the powder differed significantly with the drying process. *Bathua* powder prepared by freeze dried method had the highest bulk density value, *i.e.*, 0.59 g/ml than sun dried and tray dried powder. The functional properties of freeze dried *bathua* powder were highest with its antioxidant activity being 41.91% DPPH inhibition vis-à-vis to 32.72 and 28.72% DPPH inhibition for tray dried and sun dried powder, respectively. Also, the microbial counts of the *bathua* powder over a period of two months, though increased but was found to be within the limits prescribed by FSSAI for commercial dried fruits and vegetables, signifying drying as a better alternative of preserving *bathua* leaves. The total plate count was recorded to be least in freeze dried powder, *i.e.*, 3.20 $\log_{10}$ cfu/g after 60 days than tray dried and sun dried powder while coliform count was least in case of tray dried powder, *i.e.*, 1.74 $\log_{10}$ cfu/g after 60 days.

1. Introduction

The inclusion of neglected and under-utilized crops into present farming systems can result in nutrient-dense, climate-resilient, and sustainable agriculture, as these species have immerse possibilities to deal with poverty, hunger and malnutrition in low-income countries (Li *et al*., 2020). These green leafy vegetables can also help in addressing various nutrition related problems. As they are reported to be a good source of minerals, vitamins and fibre (Pushpangadan *et al*., 2014; Indhuleka *et al*., 2020). C. *album* is an under-utilized crop, popularly known as *Bathua* in Hindi, which originated in India and is widely cultivated as a weed or non-traditional vegetable in countries like India and Bangladesh. In different parts of India, *bathua* leaves and soft stems are consumed as vegetables, as it is rich in nutrients like protein, fibre, fat, minerals, vitamins and essential amino acids. In addition, *bathua* has been reported to exhibit several bioactive activities and functional properties like antioxidant, antibacterial, antihelmintic, antidiarrheal, hepatoprotective, *etc*. (Pathan and Siddiqui, 2022) as reported for other plants and vegetables (Saloni *et al*., 2022).

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Green leafy vegetables are grown in specific season and are highly perishable due to their higher water content. This leads to huge wastage of these seasonal vegetables due to surplus production, inadequate storage, transport and processing capacity. Thus, it is of utmost importance to utilize and explore the preservation techniques that can preserve these seasonal vegetables, so they can be used in off-season. Dehydration is an ancient technique that can preserve the food when they are available in abundance, and can be stored and consumed for longer periods (Pande *et al*., 2000). Dehydration of vegetables reduces the cost of packaging, handling, storage and transportation. In addition, improvement in shelf-life can be attained by protecting it from micro-organisms, and retarding the undesirable reactions in vegetables during storage. Different drying techniques are employed in drying of leafy vegetables such as sun drying, hot-air drying, cabinet drying, solar drying or freeze drying for herbal/ medicinal plant or leaves (Gupta *et al*., 2013).

The current study, therefore was undertaken with the aim to prepare *C. album* (*bathua*) powder by different drying techniques such as sun, tray and freeze-drying and also evaluating the effect of dehydration on nutritional composition. In addition storage stability of prepared; *bathua* powder was also examined under ambient conditions.

2. Materials and Methods

2.1 Materials

Fresh and matured locally grown variety of *C. album* (*bathua*) was procured from the local vegetable market of Varanasi, India in morning time. All the chemicals used in analysis were of analytical grade, were procured from reliable supplier and prepared freshly before analysis as per standard protocol. In addition, the glassware used in the study was of reputed brand, cleaned, dried and pre-calibrated. Packaging materials used for packaging of *bathua* powder was procured from local supplier Nanak Provision Store, Varanasi, India.

2.2 Preparation of dried *C. album* powder

Fresh and matured *bathua* leaves was sorted manually by hand. Sorted *bathua* leaves were washed with RO water and excess water being removed by placing in steel wire mesh basket. *Bathua* leaves were further blanched with hot water at 85°C for 5 min. Sun dried (unblanched) *bathua* leaves served as control sample. Further *bathua* leaves was dried by different drying techniques. Detailed flow-diagram of *bathua* powder drying with different method and products is shown in Figures 1 and 2, respectively.

2.2.1 Sun drying

Washed *bathua* leaves were placed on high density polythene (HDPE) plastic film under the clear sunlight during day time. On completion of drying leaves were collected and grinded into fine powder by using low temperature grinder (Balaji Processpack Pvt. Ltd., India). Grinded *bathua* powder was packed in low density polythene (LDPE) pouches and stored at room temperature in dry and cool place for further analysis.

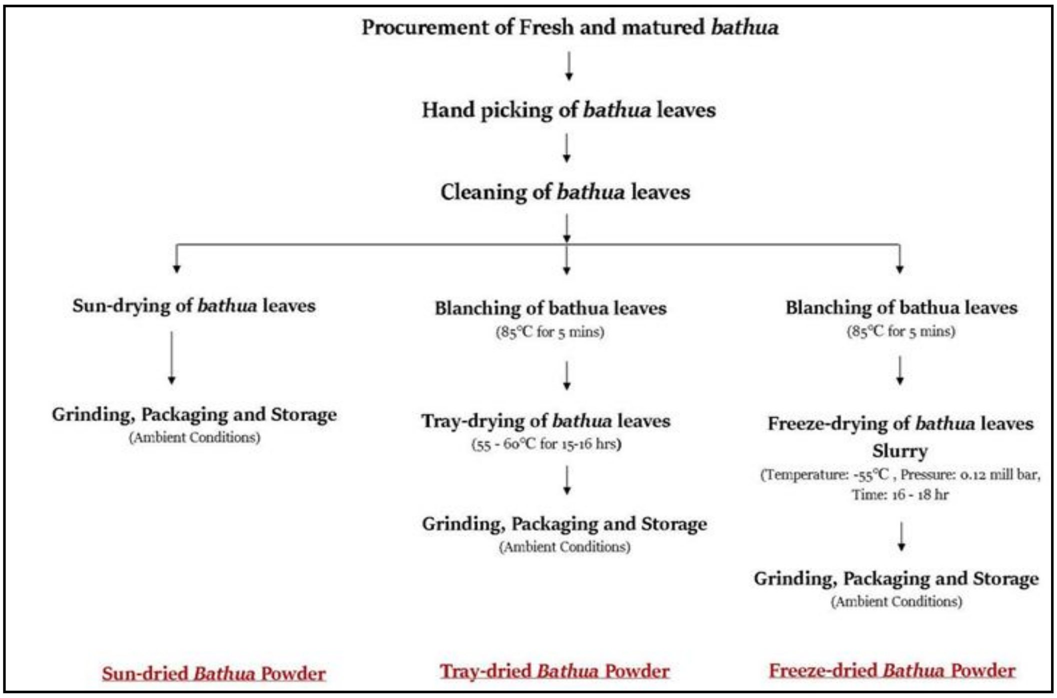


Figure 1: Flow diagram of *C. album* (*bathua*) leaves powder by different drying techniques.

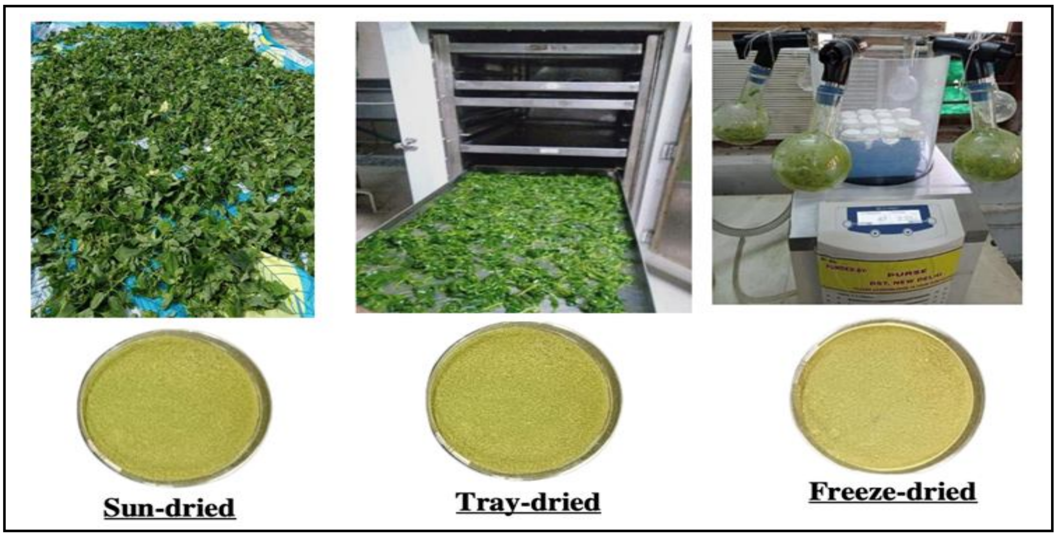


Figure 2: Methods used for drying of *C. album* (*bathua*) leaves and *C. album* (*bathua*) powder prepared from different drying techniques, *i.e.*, sun, tray and freeze drying.

2.2.2 Tray drying

Blanched *bathua* leaves were uniformly spread on stainless steel trays and placed in air circulated tray drier (Balaji Processpack Pvt. Ltd., India) at 55-60°C until complete drying (15-16 h) of *bathua* leaves. Dried leaves were collected, *bathua* grinded and packed in LDPE pouches.

2.2.3 Freeze drying

Blanched *bathua* leaves were grinded into fine slurry and transferred to the freeze dryer flask. Flask was attached to the freeze dryer (Christ, Germany) probe and dried the *bathua* leaves slurry at –55°C at 0.12 milli bar for 16-18 h. Freeze dried *bathua* leaves slurry was collected and grinded to fine powder. The powder was packed in low density polythene (LDPE) pouches and stored at room temperature in dry and cool place for further analysis.

2.3 Compositional analysis of *bathua* powder

Fat, protein, total solids, crude fibre, ash and carbohydrates content of *bathua* powders (sun, tray and freeze dried) were determined by AOAC (2000) methods.

2.4 Reconstitution properties of *bathua* powder

2.4.1 Bulk density

Bulk density of *bathua* powder was evaluated by the method given by Jan *et al*. (2015) with slight modification. In brief, 50 g *bathua* powder was taken in 100 ml tarred graduated plastic cylinder. The powder filled cylinder was tapped twice on floor to remove void space in the powder and volume (in ml) was recorded after tapping the cylinder. Bulk density of *bathua* powder was estimated by following formula:

$$\text{Bulk denisty (g/ml)} = \frac{\text{Weight of sample (g)}}{\text{Volume occupied by sample (ml)}}$$

2.4.2 Water solubility index (WSI)

Water solubility index (WSI) of *bathua* powder was determined as per the method suggested by Asaduzzaman *et al*. (2013) with slight modification. One g *bathua* powder was mixed with 10 ml distilled water and this mixture was centrifuged (Cowbell Super Centro, India) at 4000 rpm for 30 min. The supernatant was collected and oven dried at 70 ± 5°C till complete drying. Dried samples were cooled in desiccator for 30 min and weighed. Percentage of WSI of *bathua* powder was determined by following formula:

$$\text{WSI (in \%)} = \frac{W2 - W3}{W1} \times 100$$

W1 = Dried sample weight (g); W2 = Weight of aluminum (Al)-dish and dried liquid (g); W3 = Weight of empty Al-dish (g)

2.4.3 Swelling power

Swelling power of *bathua* powder samples was evaluated by the method prescribed by Shafi *et al*. (2016) with slight modification. In brief, 500 mg of *bathua* powder was dispersed into 50 ml distilled water and this mixture was heated at 90°C for 30 min. The heated mixture was further centrifuged at 3000 rpm for 15 min and supernatant obtained after centrifugation was decanted into aluminum dish, and oven dried at 100 ± 2°C till constant weight. Swelling power of *bathua* powder was represented as g per g of the dried sample.

2.5 Functional analysis of *bathua* powder

2.5.1 Total phenolic content (TPC)

TPC of *bathua* powder was estimated by Folin-Ciocalteu's method prescribed in AOAC (2000). 100 mg of *bathua* powder was mixed with one ml absolute methanol and extraction of phenolic compound was carried for 2 h in shaker incubator at room temperature, followed by centrifugation at 3000 rpm for 10 min. 0.2 ml of the extract was transferred to 10 ml volumetric flask containing 4 ml distilled water. Further, 0.5 ml Folin-Ciocalteu's reagent was added, after 1 min, 2 ml of 20% sodium carbonate solution was added and the volume was made-up with distilled water. Absorbance of sample was taken at 760 nm after 30 min of incubation against blank solution. TPC of *bathua* powder was represented as mg of gallic acid equivalents (GAE)/g.

2.5.2 DPPH inhibition antioxidant activity

DPPH (2,2-Diphenyl-1-picrylhydrazyl free radical) inhibition antioxidant activity of *bathua* powder was determined by the method given by Yu *et al*. (2018) with slight modification. Extract of *bathua* powder was prepared as per protocol used in previous test (Total phenolic content). 0.1 ml of extract was added to 3.9 ml of 0.06 mM/l methanolic DPPH solution. Absorbance of sample and control (methanol) was taken at 515 nm. DPPH inhibition antioxidant activity was calculated by following formula and results expressed as % inhibition:

$$\text{DPPH inhibition (\%)} \frac{\text{Absorbance of blank} - \text{Absorbance of sample}}{\text{Absorbance of blank}} \times 100$$

2.5.3 Tannin content

Tannin content of *bathua* powder was determined by method described by AOAC (2000). In brief, 1 ml extract of *bathua* powder was taken and 2-3 drops of 5% (w/v) ferric chloride aqueous solution was added. Appearance of greenish colour on addition of ferric chloride indicated presence of tannins in the sample. For, quantitative estimation of tannins AOAC (2000) method was adopted with slight modification. 5 ml of *bathua* powder extract was mixed with 12.5 ml indigo-carmine solution and 375 ml of distilled water. This mixture was titrated against $KMnO_4$ solution (Y ml) till blue color of mixture turns to yellowish color with slight pink tint. This titration volume includes tannins as well as other related compounds. To determine the volume of $KMnO_4$ (X ml) used to titrate non-tannin compound, another aliquot 50 ml of *bathua* powder extract was mixed with 25 ml gelatin solution (25 g gelatin was soaked in saturated NaCl solution for 1 h and mixture was warmed to dissolve gelatin, followed by cooling the mixture, and make up the final volume to 1000 ml with saturated NaCl solution). 50 ml acidic NaCl solution (25 ml conc. H_2SO_4 was added to 975 ml saturated NaCl solution) and 5 g of powder kaolin. The mixture was shaken for 15 min and filtered through Whatman No. 1 filter paper. 12.5 ml of filtrate was mixed with 12.5 ml indigo-carmine solution and 375 ml distilled water. This mixture was titrated against $KMnO_4$ solution until color changed to faint pink. The volume of $KMnO_4$ used for titration of true tannin was calculated.

2.6 Microbial analysis

1:10 dilution of phosphate buffer was prepared by mixing of one g powder into 9 ml sterile phosphate buffer in aseptic condition. Further, dilutions of the sample in buffer were made and poured in a sterile petri-plate. For the enumeration of total plate count, coliform and yeast and mold count, 1 ml from selected dilutions were poured in duplicate plates and mixed with sterile cooled plate count agar (PCA), violet red bile (VRBA) agar and potato dextrose agar (PDA), respectively. For coliform count, double layer of agar was made. Colonies in the plates were counted and was expressed as Log_{10}CFU/ g (APHA, 1992).

3. Statistical analysis

The data related to chemical, reconstitution and functional properties of *bathua* powder prepared from different drying techniques were analyzed using one way-ANOVA by SPSS 16.0 software (SPSS INC, Chicago, IL, USA) and all the measurements were done in triplicate.

4. Results

4.1 Proximate analysis of *bathua* powder prepared from different drying method

The moisture content of sun, tray and freeze dried *bathua* powder was 9.28 ± 0.03%, 8.54 ± 0.08% and 9.83 ± 0.04%, respectively. Fat content of sun, tray and freeze dried *bathua* powder were 1.22 ± 0.16%, 1.10 ± 0.12%, and 0.88 ± 0.07%. *Bathua* powder prepared from sun, tray and freeze drying had protein content of 25.41 ± 0.27%, 26.10 ± 0.29%, and 27.57 ± 0.18%, respectively, indicating *bathua* powder to be a good source of protein. *Bathua* powder prepared from different drying techniques, *i.e.*, sun, tray and freeze drying, had 5.18 ± 0.09%, 5.22 ± 0.24%, and 5.85 ± 0.16% crude fibre, respectively. Carbohydrate content of *bathua* powder samples were 42.56 ± 0.07%, 41.93 ± 0.04%, and 40.43 ± 0.08% in sun, tray and freeze drying, respectively. *Bathua* powder prepared from different drying methods, *i.e.*, sun, tray and freeze drying, had 16.35 ± 0.17%, 17.11 ± 0.07%, and 15.44 ± 0.12% ash content, respectively. Results reported by Kaur and Kaur (2018); Singh *et al.* (2007) for *bathua* powder leaves are in agreement with the present study.

Table 1: Proximate analysis of *bathua* powder prepared from different drying method

Parameters	Sun dried	Tray dried	Freeze dried
Moisture (%)	9.28 ± 0.03^{b}	8.54 ± 0.08^{a}	9.83 ± 0.04^{b}
Fat (%)	1.22 ± 0.16^{b}	1.10 ± 0.12^{a}	0.88 ± 0.07^{a}
Protein (%)	25.41 ± 0.27^{b}	26.10 ± 0.29^{a}	27.57 ± 0.18^{c}
Crude fibre (%)	5.18 ± 0.09^{b}	5.22 ± 0.24^{a}	5.85 ± 0.16^{c}
Carbohydrate (%)	42.56 ± 0.07^{a}	41.93 ± 0.04^{c}	40.43 ± 0.08^{b}
Ash (%)	16.35 ± 0.17^{b}	17.11 ± 0.07^{b}	15.44 ± 0.12^{a}

Values are reported as mean ± SD (n=3); a, b, and c values with different superscripts differ significantly ($p<0.05$) throughout the row.

4.2 Reconstitution properties (bulk density, swelling power and water solubility index) of *bathua* powder prepared from different drying methods

Results of bulk density, swelling power and WSI of *bathua* powder prepared from different drying techniques are given in Table 2. Bulk density of *bathua* powder was 0.46 ± 0.02, 0.54 ± 0.04 and 0.59 ± 0.03 g/ml for sun dried, tray dried and freeze dried powder, respectively. Swelling power for sun dried, tray dried and freeze dried *bathua* powder was 8.27 ± 0.23, 8.04 ± 0.18 and 7.91 ± 0.13 g/g, respectively. Sun dried *bathua* powder had highest swelling power compared to tray and freeze dried *bathua* powder. Waseem *et al.* (2021) reported 7.05 ± 0.30 g/g swelling powder for cabinet dried spinach powder. WSI of sun dried, tray dried and freeze dried *bathua* powder was 5.12 ± 0.36, 4.93 ± 0.33 and 4.54 ± 0.29%, respectively.

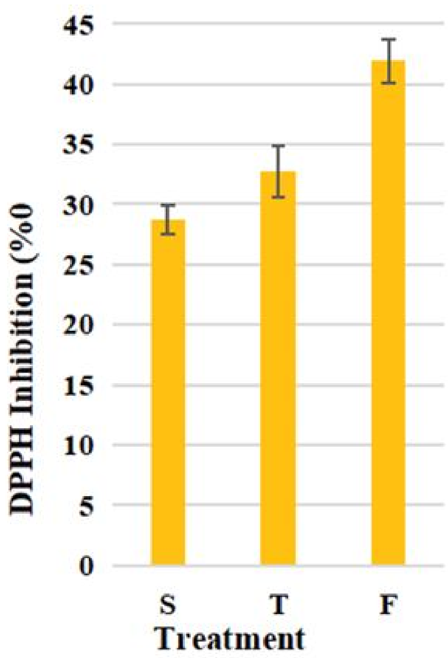
Table 2: Bulk density, swelling power and water solubility index of *bathua* powder prepared from different drying methods

Parameters	Sun dried	Tray dried	Freeze dried
Bulk density (g/ml)	0.46 ± 0.02^{a}	0.54 ± 0.04^{b}	0.59 ± 0.03^{b}
Swelling power (g/g)	8.27 ± 0.23^{a}	8.04 ± 0.18^{a}	7.91 ± 0.13^{a}
WSI (%)	5.12 ± 0.36^{a}	4.93 ± 0.33^{a}	4.54 ± 0.29^{a}

Values are reported as mean ± SD (n=3); values with different superscript differ significantly ($p<0.05$) throughout the row.

4.3 Functional properties (total phenolic content and antioxidant activity) and tannin content of *bathua* powder prepared from different drying methods

Results of TPC, DPPH inhibition activity and tannin content are represented in Figure 3. The TPC of sun, tray and freeze dried *bathua* powder were 2.04 ± 0.09, 2.52 ± 0.15 and 2.91 ± 0.05 mg GAE/g, respectively. The antioxidant activity in terms of percentage DPPH (2,2-Diphenyl-1-picrylhydrazyl free radical) inhibition activity were 28.72 ± 0.21, 32.72 ± 0.18 and 41.91 ± 0.32% for sun, tray and freeze dried *bathua* powder, respectively. Tannin content of sun, tray and freeze dried *bathua* powder were 64.08 ± 2.09, 71.62 ± 3.14, and 86.46 ± 2.27 mg/100 g and was significantly different for different drying technique. Gupta *et al.* (2013) reported tannin content 94.8 ± 7.61% for oven dried *C. album* powder (60°C for 10-12 h).



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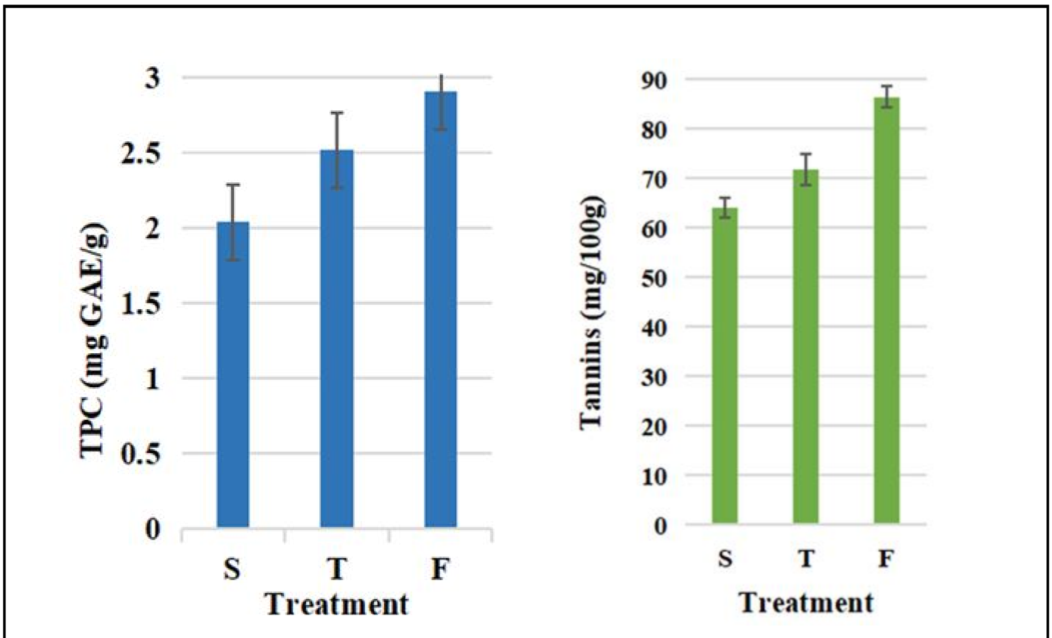

Figure 3: Representation of total phenolic content, antioxidant activity, tannin of *bathua* powder prepared from different drying methods (Note: Values are used as Mean ± SD).

4.4 Changes in microbial quality (total plate count, yeast and mold count, total coliform count) of b*athua* powder prepared from different drying methods during storage period

Bathua powder prepared from different drying techniques were studied for 2 months of storage period at an interval of 15 days for microbial count, *i.e.,* total plate count, yeast and mold count and coliform count. Microbial count (total plate count, yeast and mold count and coliform count) increased significantly during the entire storage period (Table 4).

Table 4: Total plate count, yeast and mold count, total coliform count during storage period of 60 days

Intervals (in days)	0	15	30	45	60
Total plate count (log_{10} CFU/g)					
Sun dried	2.60 ± 0.01^a	3.02 ± 0.04^b	3.20 ± 0.01^c	3.32 ± 0.02^d	3.46 ± 0.02^e
Tray dried	1.93 ± 0.02^a	2.54 ± 0.01^b	2.99 ± 0.04^c	3.13 ± 0.06^d	3.26 ± 0.04^e
Freeze dried	1.90 ± 0.02^a	2.49 ± 0.02^b	2.90 ± 0.01^c	3.15 ± 0.04^d	3.20 ± 0.02^d
Yeast and mold count (log_{10} CFU/g)					
Sun dried	0.90 ± 0.01^a	1.26 ± 0.01^b	1.38 ± 0.02^c	1.57 ± 0.02^d	1.83 ± 0.01^d
Tray dried	0.30 ± 0.02^a	0.90 ± 0.01^b	1.23 ± 0.04^c	1.46 ± 0.02^d	1.74 ± 0.02^d
Freeze dried	0.70 ± 0.02^a	1.04 ± 0.02^b	1.30 ± 0.01^c	1.40 ± 0.04^d	1.76 ± 0.02^d
Coliform count (log_{10} CFU/g)					
Sun dried	0.70 ± 0.01^a	1.40 ± 0.01^b	1.89 ± 0.02^c	1.92 ± 0.01^c	2.09 ± 0.02^d
Tray dried	Nil	Nil	0.76 ± 0.01^a	1.18 ± 0.04^b	1.21 ± 0.02^c
Freeze dried	Nil	0.41 ± 0.01^a	1.11 ± 0.02^b	1.49 ± 0.02^c	1.55 ± 0.04^c

Values are reported as mean ± SD (n=3); Values with different superscripts differ significantly ($p<0.05$) throughout the row.

5. Discussion

Bathua powder obtained by tray drying had lowest moisture content as compared to sun and freeze drying, while there was no significant difference ($p \leq 0.05$) in the moisture content of sun and freeze dried powder. Moisture values obtained from current study were in-line with the study of Kaur and Kaur (2018) and Singh *et al*. (2007), as both the studies reported the moisture content in range of 6.3 to 10.3% for dried *bathua* leaves. Sun dried *bathua* powder had highest fat content, while fat content of tray and freeze dried *bathua* powder was non-significantly different ($p \leq 0.05$). The results are almost similar with Kaur and Kaur (2018) in which fat content of *bathua* leaves varied from 0.73 to 1.40%. Fat content of *bathua* powder was quite less compared to spinach powder as Joshi *et al*. (2021) reported much higher fat content of *spinach* (4.05-4.65%). Highest protein content was obtained in freeze dried *bathua* powder and lowest protein content was obtained in case of sun drying. Results of protein content of *bathua* powder were in agreement with findings reported by Kaur and Kaur (2018). Shin *et al*. (2015) reported protein content of *Ipomoea aquatica* Forsk powder in range of 20.28 - 36.18%. Freeze dried *bathua* powder had statistically ($p \leq 0.05$) highest crude fibre while lowest was obtained in sun dried samples. Results of the current study are satisfied by Singh *et al*. (2007); Vishwakarma and dubey (2011). Gupta *et al*. (2013), also reported 4.68% fibre in dehydrated *Brahmi* (*Centella asiatica*). Highest carbohydrates content was found in tray dried *bathua* powder while lowest carbohydrate was obtained with sun drying. Kaur and Kaur (2018) reported carbohydrates content of *bathua* powder in range of 44-54%.

Bulk density of sun and freeze dried did not differed significantly ($p \leq 0.05$). Bulk density of tray dried *bathua* powder was lowest compared to tray and freeze dried *bathua* powder. Higher bulk density of tray and freeze dried *bathua* powder might be attributed to the lesser trapped air due to continuous air circulation during tray drying and vacuum conditions maintained in freeze drying (Mirhosseini and Amid 2013). Bulk density results of Ankita and Prasad (2013); Joshi *et al*. (2021) for *spinach* powder were quite similar with those recorded in the present study. On the other hand, bulk density for spray dried instant soluble sage (*Salvia fruticosa* miller) powder was 0.324-0.352, lesser compared to *bathua* powder [a]ahin-Nadeem *et al*. (2013). Sun dried *bathua* powder had highest WSI compared to tray and freeze dried *bathua* powder, it may be probably due to more pore spaces between the powder particles in sun dried *bathua* powder which increase the affinity for water molecules in powder and ultimately improved solubility (Ankita and Prasad, 2013). Value of WSI of *bathua* powder was closer to the WSI values (4.23%) reported for cabinet dried *spinach* powder at 45°C (Waseem *et al*., 2021). Ankita and Prasad (2013) reported the value of WSI for *spinach* powder in the range of 2.62 to 3.18% for different drying conditions (50, 60, 70 and 80°C) that were quite lower compared to the prepared *bathua* powder. Highest TPC content was observed in freeze dried *bathua* powder compared to sun and tray dried *bathua* powder, it may be due a higher amount of phenolic compounds get retained in freeze dried *bathua* powder.

Highest antioxidant activity was observed in freeze dried powder compared to sun and tray dried *bathua* powder. Poonia and Upadhayay (2015) reported 84.89% DPPH radical scavenging activity for *bathua* leaves extract (undried), thus concluding that blanching and drying causes significant reduction in antioxidant activity of *bathua* leaves.

The microbiological counts, *i.e.,* total plate count, coliform count, yeast and mold count for the powder prepared by different methods were under acceptable limit of commercially dried fruit and vegetables as per FSSAI standard (2019) (FSSAI standards: total aerobic count: $4 \times 10^4 - 1 \times 10^5$cfu/g; yeast and mold count: $1 \times 10^2 - 1 \times 10^4$cfu/g). Leafy vegetables are more prone to microbial spoilage due to high moisture content (Kar *et al*., 2013), but the study indicates that the microbial growth was under control after drying.

6. Conclusion

The study was carried to study the effect of different drying conditions on the proximate, technological and functional properties of *bathua* leaves powder. All the three methods used employed to make the powder freeze dried method was efficient enough in maintaining the technological and functional properties of the powder. The study also indicated that the drying of *bathua* leaves can serve as a better method to maintain the quality and the shelf-life of *bathua*, as it has been reported to have limited shelf-life under ambient conditions. So, drying of *bathua* leaves can serve as a better alternative of consuming the leaves over a longer period of time along with ensuring an overall better quality.

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Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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