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RESEARCH ARTICLE

Development of phytonutrient enriched avocado milkshake powder and its quality evaluation

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Abstract: Consumers are progressively searching out food with good taste that can be instantly prepared, easily consumable, and wholesome. Avocado is nutritious fruit but its preservation is a challenge for food researchers. A new freeze-dried milkshake powder is developed by using avocado. This product provides healthful mixture of dairy and fruit components with high nutritional and longer shelf-life. Butter fruit milkshake powder (BFMS) formulation was prepared by mixing avocado pulp, pasteurized milk, maltodextrin, and sugar in the ratio of 84%, 28%, 10%, and 6% respectively. BFMS were analyzed for proximate composition, physiochemical, and flow properties. Results showed that the inclusion of milk and maltodextrin increase protein, fat, ash content, and preserve phenolic, flavonoid, and â-carotene content of the avocado pulp. The freeze-dried BFMS powder has light color, lower water activity, and instant water-solubility (30 sec). For powder rheology, BFMS provided better flowability and decreased compressibility. BFMS is beneficial for all age groups, especially for armed forces because of instant solubility, lightweight texture, and is highly nutritious.

Keyword: Avocado, BFMS, Freeze drying, Milkshake, Phytonutrient

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Introduction

To refrain from disease and maintenance of good health regular consumption of fruits and vegetables is essential because it contains abundant quantities of vital nutrients, minerals and bioactive components such as flavonoid, phytosterols, and antioxidants that prevent the risk of chronic disease and maintain metabolic homeostasis (Slavin and Lloyd, 2012; Santos et al. 2019).

Avocado (*Persea americana*) is a tropical and subtropical fruit belongs to the Lauraceae family. It originated from Central America, and especially from West Indian, Mexico, and Guatemala, (Maitera et al. 2014). Consumers are highly interested in avocado because it contains numerous types of nutrients and phytochemicals. Avocado is called butter fruit because approximately ¾ energy comes from fat, most of which are MUFAs(Monounsaturated fatty acids), that increase the level of HDL (High-density lipoprotein) that is good for the heart (Souza et al. 2011). Avocado contains polyhydroxylated fatty alcohols (PFA) that suppress inflammatory response and protect against UV-induced damage in skin cells (Rosenblat et al. 2011). The avocado pulp contains lipophilic acetogenins that exhibit the highest antioxidant capacity (Bhuyan et al. 2019). Additionally, avocado is rich in vitamins such as tocopherol, ascorbic acid, pyridoxine, beta-carotene, and minerals like K and Mg (Fulgoni et al. 2013; Stêpieñ et al. 2020). According to Ortiz-Avila et al. (2015) consumption of avocado gives protection from coronary heart disease, hypersensitivity, anti-mycobacterial activity, oxidative damage and improves immune systems.

However, an avocado grows only in a particular region, and its shelf life is also very short, so there is a need for researchers and food technologists to discover a method to preserve avocado with its nutritional properties. In view of the above, spanish avocado honey, avocado jelly and avocado puree as fat replacer in oat meal cookies have been prepared (Wekwete et al. 2005; Rodríguez et al. 2019;López-Ramírez et al. 2020). The challenge is that ripening caused lipid oxidation, which causes off-flavor and nutritive loss. Many enzymes are also formed in ripening like phenolases that catalyze oxidative reactions, aids the conversion of phenolic compounds to another class of compounds that is ortho-quinones responsible for the color brown to avocado. During ripening other enzymes, such as LOX (lipoxygenases) and lipases also corroborate for chemical alterations in the biological compounds present in the avocado (Jacobo-Velázquez and Hernández-Brenes, 2010). These modifications deteriorate organoleptic properties, shelf-life and quality of avocado.

Industrially, drying process is the most efficient and practical method for food preservation, which increases the shelf life of food. Although drying increases shelf life of food products, it also can affects the presence and stability of nutritional and bioactive compound such as vitamin C, polyphenol, and carotenoid due to their sensitivity towards heat (Rawson et al. 2011). Different research have happened in past years on finding for an efficient avocado preservation method: freezing, drying pasteurization, and extraction of oil (Palou et al. 2000; Soliva et al. 2001; Soliva-Fortuny et al. 2004).

Compared to the other methods, lyophilization is the best dehydration method and cost-effective, which allows the preservation of the high amount of essential compounds such as vitamins, minerals, flavor, aroma, and color. Freeze dried products has pore structure, little shrinkage, good taste and retain aroma, and best rehydration quality (Souza et al. 2015).

Due to the growing dairy product business, there has been opportunity of an assimilation of the dairy product with the fruit beverages and introduce 'Juiceceuticals' that is hybrid dairy produce with fruit provide flavor and health to the consumer (Khurana and Kanawjia, 2007). Instant dairy mixes like lassi powder, dry ice cream mix, and milk powder do not contain phytonutrients. There is enormous scope for developing freeze-dried and spray-dried dairy mixes fortified with phytonutrients derived from fruit pulp or juice. Hence, the objective of the present study was to investigate if it would be possible to produce butter fruit milkshake powder (BFMS) by maintaining its nutrient richness and flow behavior employing a scalable industrial process like freeze-drying.

Materials and Methods

Preparation of Butter fruit pulp (BFP)

Fresh and fully ripe avocado (*Persea americana*) was obtained from the local market (Mysore, India), stored in a cold chamber at 4ºC until further use. To obtain avocado pulp, stored avocado was thawed, peeled, and the non-edible parts: the seed was removed and cut into small pieces. These small pieces are blended in a high-speed kitchen blender (Sunflame's blender) and obtained homogenous avocado pulp.

Formulation of Butter fruit milkshake powder (BFMS)

Butter fruit milkshake powder (BFMS) was prepared by mixing avocado pulp, pasteurized toned milk (Amul), sugar (ISI mark) and maltodextrin in the ratio of 84, 28,10,6 % respectively shown Figure 1. MD was added to improve the drying process. The mixture was blended in kitchen blender in order to obtain a homogenous paste. The homogenous paste was dried in a lyophilizer (NWS-275, India) under pressure equal to 0.035 mbar in 2 cm layers on a petri dish at -20 ÚC. The resultant butter fruit milk powder (BFMP) was collected in LDPE bags. The bags were closed and sealed and keep in the refrigerator until further study.

Proximate analysis of BMFS

Moisture content was analyzed by oven-dried method with the AOAC method no.930.15. The protein was determined by the Kjeldahl method that estimate of total nitrogen in the sample with AOAC method no. 981.10, fat content with SOCS PLUS SCS4 according to AOAC official method no.948.22, and crude fiber of sample was estimated by AOAC (2005) method no.,962.09. Muffle furnish was used in the determination of Ash content at 550°C for overnight. Carbohydrate was determined from the difference. All the proximate analysis was performed in triplicate.

Physio chemical analysis

TSS, pH and Titratable acidity

Total soluble solid, pH, titratable acidity of BFMS was carried out by the methods described Ranganna, 2001. TSS of BFMS was determined by hand refractometer at 20°C.Titratable acidity determined by titration against 0.1N NaOH soluble using few drops of phenolphthalein indicator and Cyber scan 510 pH meter was used determination of pH.

Color

The Color of BFMS influence consumer acceptability. Color values determined by Color Flex, Hunter Lab. Result was comes in terms of L*(Black=0, White=100), a* (Greenness=-a, Redness=+a) and b* (Blueness=-b, Yellowness=+b).

Water activity

Water activity (a_w) of the freeze-dried samples was determined by water activity meter (Aqua-Lab, model 3TE, Decagon Devices Inc., Pulluman, WA, USA) with 25°C temperature.

Wettability

The powder's wettability was estimated by the procedure described by Fuchs et al. (2006) with slight modifications. 0.1 g of powder sample was spread over the surface of 100ml deionized water at room temperature without stirring. The time was noted until the last particle of powder submerged.

Bulk density

Bulk density was estimated as the method explained by Sahin-Nadeem et al. (2013). Twenty gram of BFMS was taken into measuring cylinder (100 ml) and the volume of the BFMS was noted.

Bulk Density = (Mass of BFMS) / (Volume of BFMS)

Tapped density

The Tapped density of the BFMS was calculated by the method defined by Ozdikicierler et al. (2014). 20 g of the sample placed in a 100 ml mixing cylinder, and the volume was noted after the sample was tapped 20 times on rubber pad from a height of 15 cm.

Tapped Density = (Mass of BFMS) / (Tapped Volume)

Nutraceutical characteristics of BFMS

Phenolic and Flavonoids content

Total phenolic and flavonoid content in butter fruit milk shake powder was estimated by the procedure described by Tyagi et al. (2020).

Phenolic and flavonoid compounds from BFMS was extracted with $CH_3OH/H_2O/HCOOH$ in ratio of 70: 29.7: 0.3 v/v/v. Total phenolic content of BFMS extract was estimated by using the Folin's phenol reagent and standard curve was prepared by gallic acid. Results were expressed in mg/100g.

Aluminum chloride solution was used to evaluate TFC in which rutin trihydrate (RT) was used in preparation of standard curve and result mg/100g expressed in µg RT equivalent /ml of extract.

Beta carotene

â-Carotene was estimated by the method suggested by Srivastava and Kumar (2003). 5g of a sample of butter fruit milkshake powder (BFMS) was grinded with few crystals of Na2SO4 and homogenized with 10 ml $(CH_3)_2CO$. It was decanted, and then the supernatant was collected and transferred supernatant to a separatory flask. 10 ml of petroleum ether was added in separating funnel and mixed. Two layers were found, the lower layer discarded and the upper layer collected and optical density (OD) was recorded at 452 nm. The result was expressed in µg/ 100mg.

Table 1 Proximate composition of freeze dried butter fruit pulp (BFP) and freeze dried butter fruit milkshake powder (BFMS)

Parameter	BFP	BFMS
Moisture (%)	2.5±0.21	2.9±0.37
Protein (%)	6.12±0.11	17.2±0.14
Fat (%)	39.20±0.28	42.2±0.22
Ash (%)	2.2±0.04	2.78±0.09
Total carbohydrate(%)	49.17±0.68	33.16±0.51
Crude fibre(%)	0.81±0.08	0.84±0.1

Values are Mean ± Standard deviation (SD) of triplicates

Assessment of avocado milk powder by powder rheometer

Powder flowability is the powder's ability to flow test environment. Flowability is a one-dimensional character of powder. Powder rheology parameters help to know that whether powder is free-flowing or non-flowing.

An FT4 Powder Rheometer (Freeman Technology, Bouiters Farm Centre, India) shown Figure 2(a) was used to evaluate the rheological behavior of the powders that measuring the basic flow energy(BFE), conditioned bulk density(CBD), compressibility, aeration ratio, and shear test BFMS powder. The sample is placed in a Split Vessels as shown in Figure 2(b), which allows an exact volume to be managed, and density of the sample can be measured with accuracy. Precision blade or impeller shown in Figure2(c) is needed for test. Conditioning process performs before each test cycle to prepare the sample for measurement and eliminate any effects present in the sample by rotating and moving the impeller upwards and downwards through the powder 3 times.

Statistical analysis

Data of physicochemical analysis and antioxidant potential were taken in triplicate and expressed as mean with standard deviation (Mean±SD).MS excel software was used in calculation of mean and standard deviation.

Results and Discussion

Nutritional composition of BFMS

The results of the nutritional composition of butter fruit milkshake powder are presented in Table 1. BFMS contains good quantity of protein 17.2%, fat 42.2%, crude fiber 0.84% along with ash content 2.78 %. Ash content signifies the level of mineral present in the sample. The obtained value was higher than avocado fruit (butter fruit) pulp because the addition of milk and maltodextrin in avocado pulp significantly increased the protein, ash, fat in BFMS. Dantas et al. (2017) also found that maltodextrin successfully maintains high contents of protein because bioprotective osmolytes like maltodextrin stabilize the native conformation of globular proteins relative to the unfolded state under external stress like drying. The stabilization is due to unfavorable interactions between protein and cosolute leading to a preferential hydration of the protein. As water is being removed the folding equilibrium is shifted towards the native state and therefore stabilization occurs. The Freeze-dried BFMS

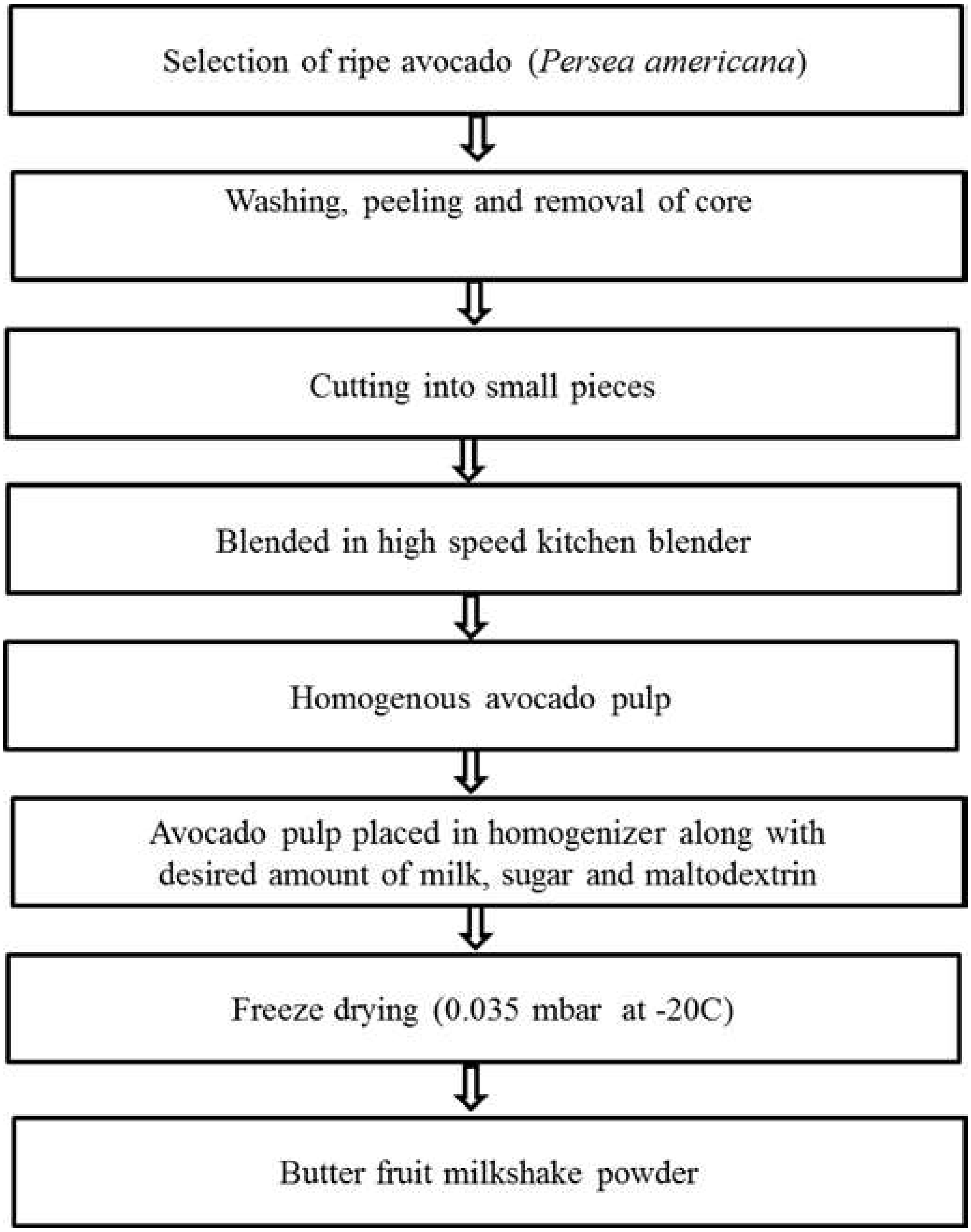


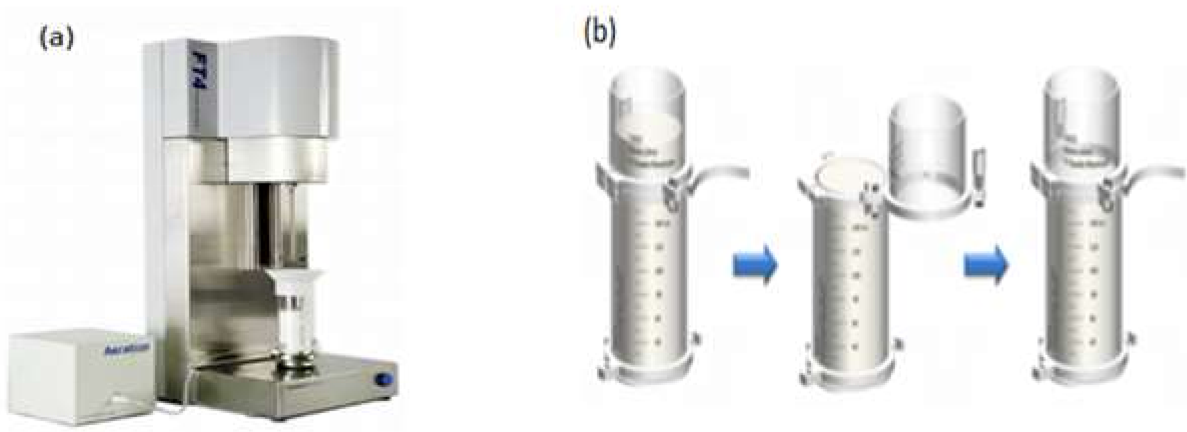
Fig. 1 Flow diagram of procedure of butter fruit milkshake powder (BFMS)

powder had a moisture content of 2.9%. Hence the product can be stable at room temperature for longer periods.

Physico-chemical analysis of BFMS

TSS, pH and titratable acidity

The result of TSS, pH, and titratable acidity was shown in Table 2. The content of total soluble solids of avocado pulp without the addition of milk was around 8.68°Brix while after the addition of milk and maltodextrin TSS of BFMS was 16°Brix. As expected, the presence of milk and maltodextrin increased the total soluble solid of BFMS significantly. The acidity of avocado pulp was 1.05%, which was reduced by the addition of milk 0.88%. A similar trend was also noticed in addition to milk in passion fruit (do Espírito Santo et al. 2012). The pH of avocado pulp was between 6.71-6.74 and 6.94 after addition milk in the avocado pulp. The result conforms to Ceballos et al. (2012), who reported that pH and total soluble solid content was increase while titratable acidity



decrease when freeze-dried soursop fruit pulp treated with maltodextrin.

Color

The results of the color measurements of BFMS were shown in Table 2. Avocado is naturally green in color, so the value of a* is negative. However, the addition of milk and maltodextrin in avocado pulp leads to, brighter, less green and less yellow (L*=77.62, a*=-2.22, b*=19.63) butter fruit milkshake powder is obtained. Ceballos et al. 2012 also noticed that freeze-dried maltodextrin soursop fruit pulp was lighter in color than the control soursop fruit pulp.

Water activity

Water activity (a_w) plays an important role in growth of microorganism cause spoilage of food and loss functional properties of food due to mobility of molecule (Correia et al. 2017). Safety and mechanical point of view a_w value is also important. From mechanical point of view if water activity in the range of 0.35-0.50 unacceptable organoleptic changes could be produced. From a safety viewpoint, it has been reported that values below 0.6 can be considered microbiologically or chemically stable because the amount of free water available for biochemical reactions is low (Dantas et al. 2018). From Table 2 water activity (a_W) of BFMS powders is 0.518, which can be considered stable from microbial spoilage.

Wettability

Wettability time of lyophilized BFMS powders is presented in Table 2. Wettability is described as a circumstance of a solid food surface that determines how fast a liquid will wet and spread on the surface. According to Vega et al. (2005) wettability depends on density, surface charge, porosity, particle size, surface roughness, surface area, surface composition and surface activity of the food particles. Wettability of BFMS

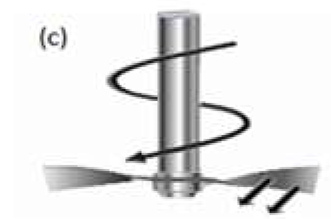


Fig. 2 Powder rheometer (a) FT4 powder rheometer (b) Split Vessel for FT4 test (c) Impeller for FT4 test

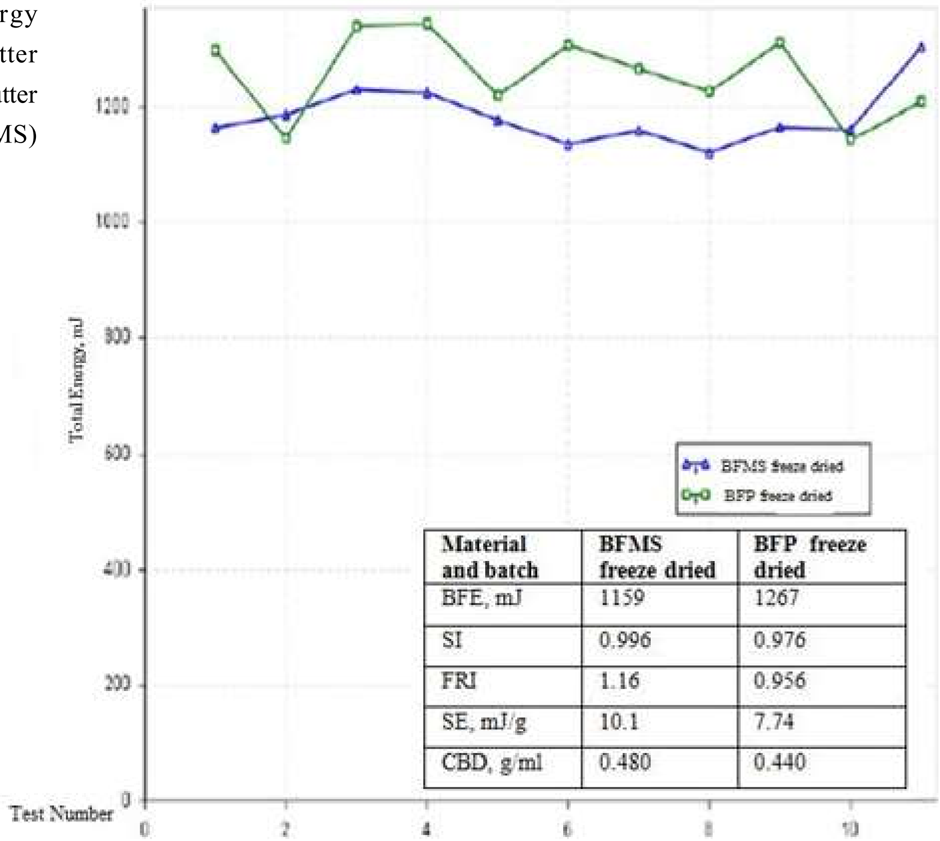
powder was 30 sec, which was lower than the result reported by Calýskan et al. (2015) in freeze-dried kiwi puree powder with MD (186 s).

Bulk density and tapped density

Tapped and untapped density is significant parameter for the powder because it indicates the area cover by the powder, which is vital for packing industry. Tapped and untapped density results are presented in Table 2, where the untapped density known as bulk density and tapped density were found to be 0.492 and 0.99 gcm^{-3} respectively. The researchers reported that the remaining solids after removal of moisture have higher densities as compared to water, and the overall solid density increase as moisture is removed. The addition of maltodextrin decreased cohesiveness and increased bulk and tapped densities. Calýskan et al. (2015) dried the kiwi puree with 10% MD in freeze dryer, bulk and tapped density increase in freeze dried kiwi puree with maltodextrin compared to freeze dried kiwi puree without MD.

Nutraceutical characteristics of BFMS

Table 3 shows total phenolic, flavonoid and β carotene of BFMS. Phenols are an essential constituent of fruit and vegetables.



Material and batch	BFMS freeze dried	BFP freeze dried
BFE, mJ	1159	1267
SI	0.996	0.976
FRI	1.16	0.956
SE, mJ/g	10.1	7.74
CBD, g/ml	0.480	0.440

Fig. 3 Flow energy measurements of butter fruit pulp (BFP) and butter fruit milkshake (BFMS) powder

Table 2 Quality parameters of freeze dried butter fruit pulp (BFP) and freeze dried butter fruit milk shake powder (BFMS)

Parameter	BFP	BFMS
pH (reconstituted)	6.71±0.77	6.94±0.88
Titratable acidity (%)	1.05±0.12	0.88±0.05
TSS (reconstituted)	8.68±0.38	16±0.26
Color : L*	65.83±0.58	77.62±0.69
a*	-4.5 ±0.59	-2.22±0.57
b*	22.83±0.10	19.63±0.17
Water activity	0.61±0.01	0.52±0.01
Wettability (sec)	36	30
Free fats (%)	11.87±0.18	8.625±0.38
Tapped density(g/cm^3)	0.81±0.01	0.99 ±0.06
Bulk density (g/cm^3)	0.389±0.11	0.492 ±0.14

According to Kaur et al. (2016), there is a link between antioxidant activity and phenols because compounds of phenol are active H+ donors, making food product rich in antioxidant. The avocado pulp contains total phenolic, and flavonoid content was 410.41 mg/100g and 21.9 mg/100g, respectively. Butter fruit milk powder has a slightly higher content of total phenolic content (416.2 mg/100g) and total flavonoid content (24 mg/100g) than avocado pulp. These changes are due to variety of avocado and addition of maltodextrin because maltodextrin has an additional stabilizing effect and due to this ability it reduces reactant mobility and preserves TPC and TFC.The BFMS powder is also a good source of â-carotene (502µg/100g). Silva-Espinoza et al. (2020) reported that freeze-drying better preserves bioactive compounds in an orange puree.

Rheology of BFP and BFMS

BFP,SI,SE,FRI and CBD of freeze dried butter fruit milk shake powder (BFMS) and butter fruit pulp (BFP) was shown Table 4 and Figure 3.

Basic flow energy (BFE)

Basic flow energy estimation is the most illuminating parameter as it can detect minimal differences in the physical properties of samples such as particle shape, size, distribution, surface irregularity, electrostatic potential, and moisture content. Better flowability contributes to minimum agglomeration, easy processing and handling, and convenient in manufacture and transport of powder (Vega et al. 2005). The agglomerated larger-sized particles of BFP (1267 mJ) needed more energy to move as compared to BFMS powder (1159mJ) in the forced flow regime. Similarly, Bian et al. (2015) observed the BFE value of 680 mJ for

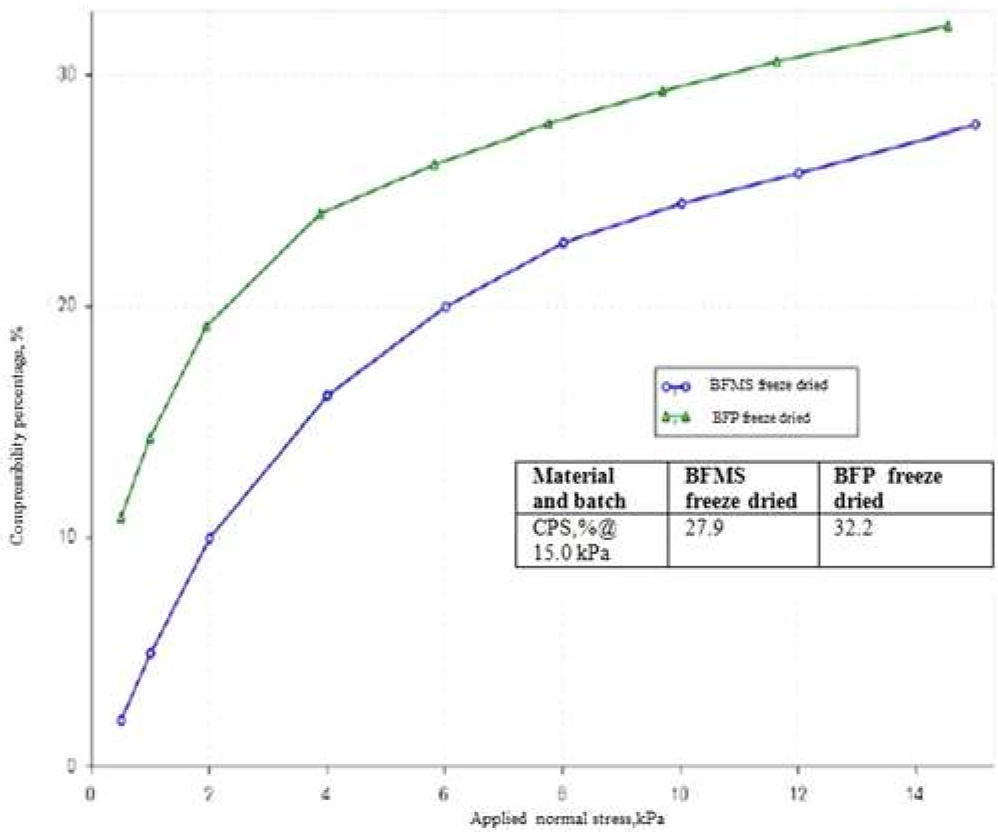


Fig. 4 Compressibility graph of butter fruit pulp (BFP) and butter fruit milkshake (BFMS) powder

Table 3 Total phenolic contents, total flavonoid contents and beta carotene of freeze dried butter fruit pulp (BFP) and freeze dried butter fruit milk shake powder (BFMS)

Parameter	BFP	BFMS
TPC (mg/100g)	410.41±4.19	416.2±5.04
TFC (mg/100g)	21.9±1.22	24±1.54
β-carotene (μg/100g)	512±3.63	502 ±7.09

Values are Mean ± Standard deviation (SD) of triplicates

hard wheat and 713 mJ for soft wheat flour at 11.4% moisture content.

Stability index

To know the stability of powder during flow testing environment SI value is measured. BFMS and BFP have almost the same stability index (SI) that is 0.996 and 0.976, respectively, and both the samples are stable within the test environment. Mitra et al. (2017) also found that the Basundi mix had the same SI (0.9-1.1). Above or below this range, food powders are prone to attrition and would be categorized as unstable. Instability may occur when powders are very cohesive and compressible, may become caked or agglomerated during flow, thereby changing the flow properties significantly.

Table 4 Flowability parameters of freeze dried butter fruit pulp (BFP) and freeze dried butter fruit milk shake powder (BFMS) and butter fruit pulp (BFP)

Measurements	BFMS	BFP
BFE(mJ)	1159	1267
SI	0.996	0.976
FRI	1.16	0.956
SE (mJ/g)	10.1	7.74
CBD (g/ml)	0.480	0.440
Aeration Test		
AE_10 (mJ)	702	1043
AR_10	1.71	1.89
Compressibility Test		
CPS(% @ 15.0kPa)	27.9	32.2
Shear Test		
Cohesion (kPa)	4.13	1.82
UYS (kPa)	17.30	9.37
MPS (kPa)	20.12	24.7
FF	1.16	2.57
AIF	38.98	47.5

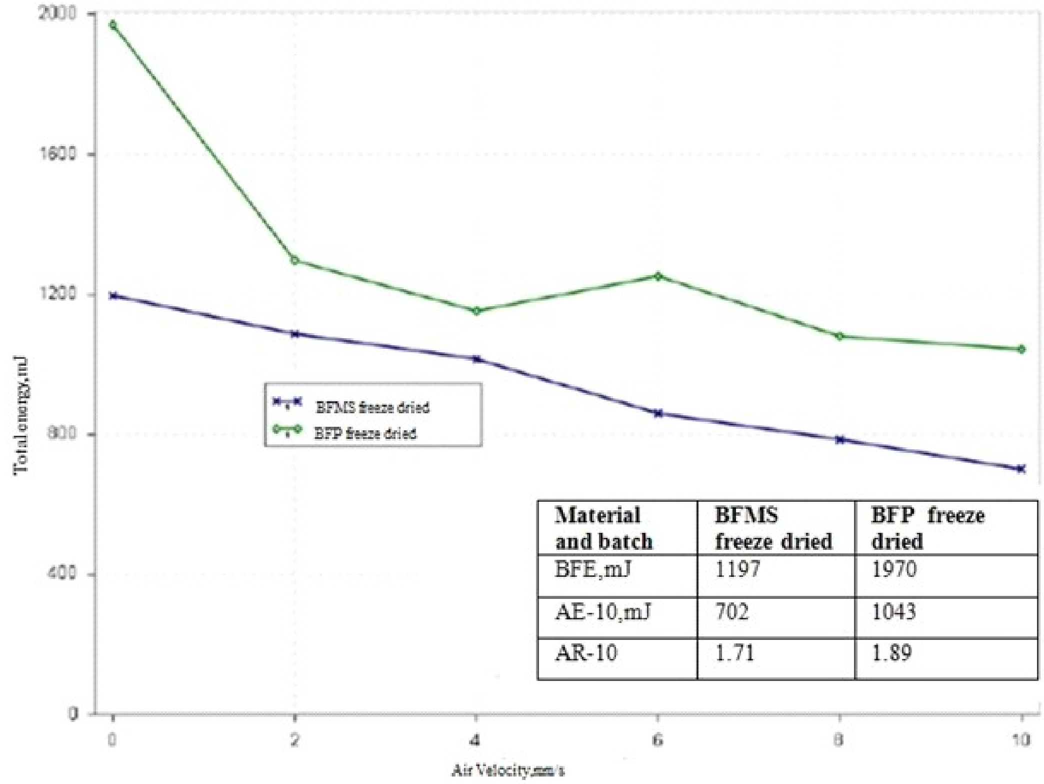


Fig. 5 Flow Energy at an airflow of freeze-dried butter fruit pulp (BFP) and butter fruit milkshake (BFMS) powder

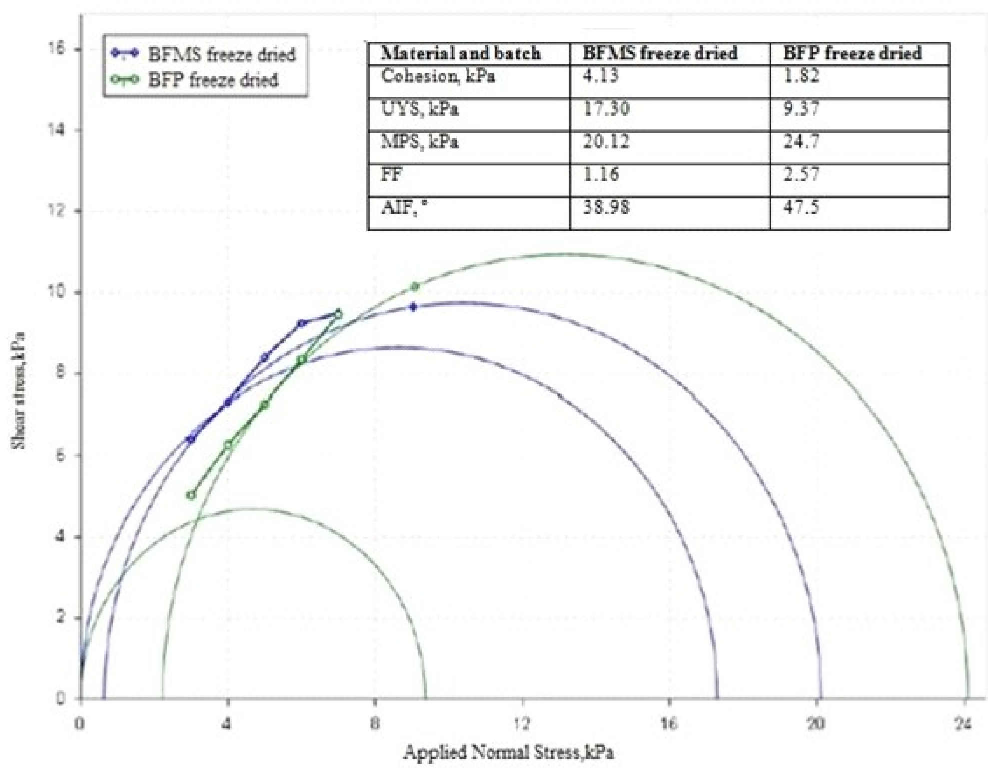


Fig. 6 Mohr cycle of studied freeze-dried butter fruit pulp (BFP) and butter fruit milkshake (BFMS) powder

Flow rate index (FRI)

Flow rate index is measured to know the powder sensitivity to blade tip speed, and the powders which are cohesive in nature are commonly more sensitive to FRI. The FRI values of BFMS and BFP was 1.16 and 0.959 respectively, representing the powder was insensitive to fluctuating flow rate. The cohesive powders required greater flow energy at lower flow rates because the entrained air was able to escape, leaving a stiffer material which was more resistant to flow. The non-cohesive powders were less sensitive to flow rate because of the presence of entrained air. A similar result was found in the basundi mix, which FRI was closer to 1.2, indicating the powder was insensitive to varying flow rate (Mitra et al. 2017).

Specific energy

Specific energy is measured to identify powder flowability in free and low-stress area, and providing a helpful sign that powder has cohesion or not. Han et al. (2011) stated the parameter of cohesion and their specific energy: A powder has low cohesion when SE value is less than 5; moderate cohesion when SE value in the range of 5 -10; and powder has high cohesion for SE > 10. From Table 4 and figure 3, specific energy increased significantly with the addition of milk and maltodextrin to an avocado pulp that indicates BFMS has high cohesion (10.13mJ/g) as compared to BFP (7.74mJ/g). SE of BFMS is more than BFP because interlocking forces develops between different shapes and sizes of particles.

Conditioned bulk density (CBD)

CBD is just one physical property of a powder that influences the flowability powder. It depends upon powder properties such as particle shape, size, texture, etc. Both BFMS and BFP samples are having similar condensed bulk density values that are 0.480 and 0.440 respectively.

Compressibility of BFMS and BFP

Compressibility values give the idea that how to handle and transportation of powder. The compressibility test is a bulk property measurement that reveals how density changed as a function of applied normal stress. As summarized in Table 4 and figure 4, the compressibility value of avocado pulp powder in 15kPa is 32.2% which decreased after the addition of milk and maltodextrin (27.86%) because the particle size is in different shapes so there is bond formed between the particles and low space for air. Lower compressibility indicates there is efficient packing among the particles and a minimal amount of excess air in the bulk, which is a desirable property for pharmaceutical powders intended for solid dosage forms (Han et al. 2011).

Aeration energy

Aeration energy indicated that the relationship between total energy and air velocity of BFMS and BFP. High AE represents a *cohesive powder*, while *low* AE represents a free-flowing *powder (*Leturia et al. 2014).The data obtained from Figure 5 showed that less energy was required to aerate BFMS compared to BFP. Mitra et al. (2017) also found that basundi mix at 3% moisture content had the lowest aeration energy as compared to 6% and 9%.

Mohr cycle

The value of flow function (FF), obtained from divide major principle stress (MPS) with uncofined yield strength (UYS). Han et al. (2011) reported that FF value could directly reveal the flowability of sample, if flow function value is less than 1 means not flowing; for very cohesive FF value between 1-2;cohesive powder has FF value between 2-4 and easy flowing powder has FF in the range of 4-10. From above statement BFP (FF=2.57) powder can be considered as easy flowing powder as compared to BFMS (FF=1.16) as shown in Table 4 and figure 6. Cohesion is a point of crossing of the yield locus from y – axis. The cohesion of BFP was 1.82, which free flowing than BFMS 4.13.

Conclusions

It is concluded that developed avocado milkshake power is nutritiously superior, highly acceptable, and healthy at low cost. Incorporation of avocado pulp in milk increased in protein, fat, ash, and crude fibre content, whereas it has low moisture content and water activity which revealed that it has a longer shelf life with low wettability time and instantly soluble in water. BFMS is rich in phenolic, flavonoid, and beta-carotene, which is helpful in maintenance of the good health and treatment of many contagious diseases. Powder rheology with FT4 freeman technology study shows that avocado milkshake powder has good flowability. This type of BFMS powder may increase food product market with bioactive goodness, immunity booster, and be advantageous to people of all ages, especially for army men due to long shelf life, instant to make, and provide enough energy. Further research is needed for the fat encapsulation that can improve the sensory acceptability of this product, and to observe the effect of drying on avocado pulp, SEM analysis to be done. Butter fruit milkshake can be tried with different flavors and various value-added food products can be made by incorporating BFMS like in chocolate, milk bar.

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