SCIENTIA PLENA VOL. 22, NUM. 03 2026
www.scientiaplena.org.br doi: 10.14808/sci.plena.2026.031503

Proximal, bioactive and antioxidant characterization of defatted avocado flour obtained by supercritical extraction and application in the development of a cereal bar: Comparative evaluation of freeze-dried avocado pulp

Caracterização proximal, bioativa e antioxidante da farinha de abacate desengordurada obtida por extração supercrítica e aplicação na elaboração de uma barra de cereal: Avaliação comparativa com polpa de abacate liofilizada

K. R. P. Pantoja[1]*; J. P. F. de Carvalho[2]; M. A. R. Cardoso[3]; J. M. L. Pinheiro[4]; P. R. M. da Costa Junior[5]; M. V. S. Bezerra[4]; R. N. Carvalho Junior[1,2,4,5]

[1]*Programa de pós-graduação em Engenharia de Recursos Naturais da Amazônia/Universidade Federal do Pará, 66075-110, Belém-Pará, Brasil*
[2]*Instituto de Tecnologia/Universidade Federal do Pará, 66075-110, Belém-Pará, Brasil*
[3]*Instituto de Ciências Exatas e Naturais/Universidade Federal do Pará, 66075-110, Belém-Pará, Brasil*
[4]*Programa de pós-graduação em Ciências e Tecnologia de Alimentos/Universidade Federal do Pará, 66075-110, Belém-Pará, Brasil*
[5]*Faculdade de Biotecnologia/Universidade Federal do Pará, 66075-110, Belém-Pará, Brasil*

**kelly.sousa@icen.ufpa.br*
(Recebido em 09 de julho de 2025; aceito em 10 de fevereiro de 2026)

Defatted avocado flour (DAF) obtained after supercritical extraction presents high nutritional value and can be valorized and applied in the development of food products. This study aims to characterize DAF obtained from supercritical extraction of freeze-dried avocado pulp (FDAP), under conditions of 40°C and 60°C combined with pressures of 300 and 400 bar, and its application in the development of a cereal bar. The study analyzed the proximate, bioactive, and antioxidant composition of DAF in comparison to FDAP. DAF showed a maximum residual yield of 62.63 ± 0.40%, and was rich in carbohydrates (18.36 ± 0.26%), proteins (8.83 ± 0.40%), lutein (173.55 ± 0.01 µg/g), phenolic compounds (753.21 ± 0.09 µg GAE/g), and presented antioxidant activity by DPPH (0.516 ± 0.01) and ABTS (0.413 ± 0.02), with values higher than those of FDAP: carbohydrates (12.31 ± 0.37%), proteins (5.94 ± 0.25%), lutein (99.20 ± 0.02 µg/g), phenolic compounds (458.57 ± 0.01 µg GAE/g), DPPH activity (0.056 ± 0.02), and ABTS (0.056 ± 0.01). The cereal bar formulated with DAF (CBDAF) presented high moisture content (25.19 ± 0.99%) and was a source of protein (12.29 ± 0.46%), while the bar formulated with FDAP (CBFDA) showed moisture of 22.19 ± 0.98% and protein content of 10.33 ± 0.98%, indicating the need for formulation adjustments to improve technological quality and ensure compliance with regulatory standards.
Keywords: supercritical extraction, avocado, food technology.

A farinha de abacate desengordurada (DAF) obtida após extração supercrítica possui alto valor nutritivo podendo ser valorizada e aplicada no desenvolvimento de produtos alimentícios. O presente estudo tem como objetivo realizar a caracterização da DAF obtida após extração supercrítica da polpa de abacate liofilizada (FDAP), nas condições de 40 ºC e 60 ºC combinadas a pressões de 300 e 400 bar e sua aplicação no desenvolvimento de uma barra de cereal. No estudo foi analisado a composição proximal, bioativa e antioxidante da DAF em comparação a FDAP. A DAF possui rendimento residual máximo de 62.63 ± 0.40%, rica em carboidratos (18.36 ± 0.26%), proteínas (8.83± 0.40%), luteína (173.55 ± 0.01 µg/g), compostos fenólicos (753.21 ± 0.09 µg GAE/g) com potencial atividade antioxidante DPPH (0.516 ± 0.01) e ABTS (0.413± 0.02), valores superiores aos FDAP com carboidratos (12.31± 0.37%), proteínas (5.94 ± 0.25%), luteína (99.20 ± 0.02 µg/g), compostos fenólicos (458.57 ± 0.01 µg GAE/g) e atividade antioxidante DPPH (0.056 ± 0.02) e ABTS (0.056 ± 0.01). A composição da barra de cereal desenvolvida com DAF (CBDAF) apresentou alto teor de umidade (25.19 ± 0.99%) e fonte de proteínas (12.29 ± 0.46%) enquanto a barra de cereal desenvolvida FDAP (CBFDA) exibiu umidade de 22.19 ± 0.98% e proteínas de 10.33 ± 0.98% com indicando necessidade de ajustes na formulação para melhor qualidade tecnológica e concordância com a legislação.
Palavras-chave: extração supercrítica, abacate, tecnologia de alimentos.

1. INTRODUCTION

The generation of industrial waste has been a persistent issue, particularly in the agro-industrial sector. In most cases, such waste is not repurposed and is instead improperly discarded, resulting in the accumulation of tons of organic matter and significantly contributing to severe environmental impacts. In recent years, however, both industry and society have increasingly recognized the importance of adopting sustainable strategies not only in production processes but also in consumption habits. This shift has promoted the valorization of waste, and the development of products derived from clean technologies and the full utilization of raw materials. [1, 2].

In the avocado (*Persea americana* Mill.) oil extraction chain, up to 30% of solid residues (peels and seeds) can be generated, in addition to the defatted pulp remaining after processing. Traditional oil extraction by cold pressing, although classified as a green technology, produces a residual flour with high lipid content, which limits its reuse in food and functional formulations. Moreover, most of these residues are discarded without proper treatment, resulting in the loss of valuable nutrients and increasing environmental impacts [3, 4].

A promising alternative for utilizing avocado peel and seed lies in obtaining extracts rich in bioactive compounds through green extraction techniques, such as microwave-assisted extraction, ultrasound-assisted extraction, pressurized liquid extraction, eutectic solvents, and supercritical fluid extraction, among others. However, most studies treat the extract as the final product, with little exploration of the solid residues post-extraction. Furthermore, the extraction process often significantly reduces the remaining bioactive and functional content of these residues, limiting their applicability in food, nutraceutical, or cosmetic formulations [1, 3, 5].

Clean technologies such as SFE-CO_2 (Supercritical Fluid Extraction with Carbon Dioxide) enable the production of defatted avocado flour (DAF) with high added value, enriched with micro and macronutrients and low in lipid content, thus facilitating their recovery and valorization [6, 7]. However, despite the great potential that avocado by-products present, there are no studies that characterize the flour obtained after supercritical extraction and explore it as an ingredient in the formulation of new products, such as cereal bars (CB), which are composed of fruit pulps [8], cereals, chocolates, and nuts aiming to attract consumers attention and meet their caloric needs. Additionally, they have functional value being practical and healthy.

Thus, this study aims to characterize the defatted avocado flour (DAF) obtained after supercritical extraction of freeze-dried avocado pulp (FDAP) and apply it as an ingredient in the formulation of a cereal bar, evaluating its proximate composition, phenolic compounds, lutein, and antioxidant activity in comparison to FDAP.

2. MATERIAL AND METHODS

2.1 Raw material

DAF is a byproduct obtained after supercritical extraction of the FDAP from the Margarida variety, in order to obtain a supercritical oil rich in antioxidants, as carried out in a previous study by Pantoja et al. (2025) [9], the process carried out in this present study can be seen in Figure 1. For the study, ripe avocados were depulped, homogenized, and lyophilized for 24 hours. The FDPA was extracted using supercritical CO_2 to obtain avocado oil, the following operating conditions were adopted: CO_2 output flow rate of 5.93 kg/min, pressures of 300 and 400 bar combined with temperatures of 40 and 60 ºC. A static period of 30 minutes and a dynamic period of 60 minutes. After the process, two products are obtained: a supercritical oil and DAF.

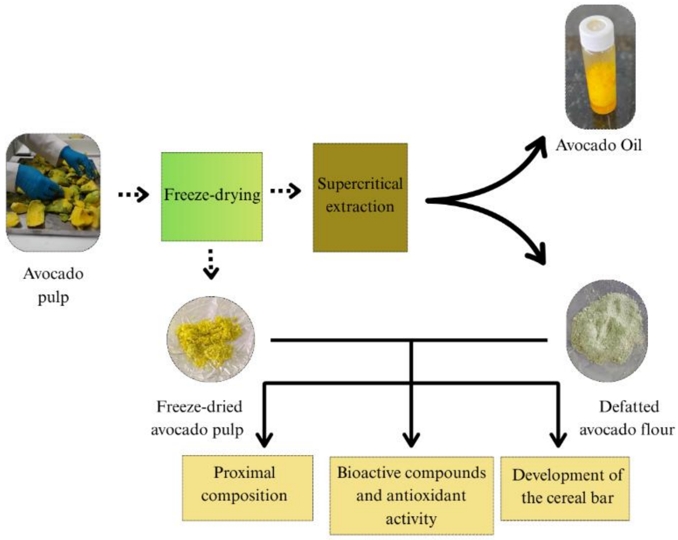

Figure 1: Process of obtaining and studying defatted avocado flour.

2.2 Proximate composition

The FDAP, DAF (best extraction condition) and the formulations of CBFDA (Cereal bar with freeze-dried avocado) and CBDA (Cereal bar with defatted avocado) were evaluated for ash, lipid, carbohydrate, protein, and moisture contents, according to the Official Methods of Analysis of AOAC International (2023) [10]. All analyses were performed in triplicate. The best condition was evaluated as the one that showed the best results regarding the content of Lutein and Total Phenolic Compounds (TPC).

2.3 TPC

The DAF obtained under all conditions and the FDAP, were evaluated TPC content using the Folin–Ciocalteu method [11]. Approximately 100 mg of each sample were weighed and homogenized in an extracting solution (70% ethanol / 29.5% water / 0.5% acetic acid) for 15 minutes. The samples were then centrifuged in a centrifuge (Model CT15RE, Hitachi, Japan) for 20 minutes. About 0.5 mL of each supernatant was transferred into cuvettes and reacted with 1.25 mL of 7.5% sodium carbonate solution and 0.25 mL of 1 N Folin–Ciocalteu reagent. Distilled water was used as a blank. The reactions were allowed to proceed for 30 minutes in the absence of light. All measurements were performed in triplicate, and the results were expressed as milligrams of gallic acid equivalent per gram of dry sample (mg GAE/g d.b.). The calibration curve was constructed using gallic acid in six concentrations (15–50 mg/L), according to the equation: $Y = 0.0151x + 0.0332$, with $R^2 = 0.9954$. Absorbance readings were taken using a UV/VIS spectrophotometer (model UV-M90, Bel Engineering, Italy) at 750 nm.

2.4 Lutein

Lutein analysis was performed using a method adapted from Rodrigues-Amaya (2001) [12] Approximately 100 mg of DAF under conditions, as well as FDAP were used. Ethanol was employed as the extracting solvent. Absorbance readings were taken using a UV/VIS spectrophotometer (model UV-M90, Bel Engineering, Italy) at 445 nm. Lutein content was calculated based on its specific absorption coefficient in ethanol (2550). All measurements were performed in triplicate, and the results were expressed as micrograms per gram of dry sample (µg/g d.b.).

2.5 Antioxidant Activity

2.5.1 DPPH Assay

The DAF obtained under all conditions and FDAP, were evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl hydrate) spectrophotometric method, according to Brand-Williams et al. (1995) [13]. Approximately 100 mg of each sample were weighed and homogenized in an extracting solution (Ethyl alcohol) for 15 minutes. The supernatant was separated by centrifugation (Model CT15RE, Hitachi, Japan) for 20 minutes. An aliquot of 0.5 mL from each extract was transferred into cuvettes and reacted with 1.95 mL of a 60 µM DPPH solution. The analysis was performed in triplicate, and the results were expressed as micromoles of Trolox equivalent per gram of dry sample (µmol TE/g d.b.). A Trolox calibration curve was constructed using six concentration points (25 to 1000 µmol/L), according to the linear equation: $Y = 0.0005x + 0.0396$, with $R^2 = 0.9901$.

2.5.2 ABTS Assay

The DAF obtained under all conditions and FDAP, was evaluated using the ABTS (2,2′-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid)) spectrophotometric method, according to Re et al. (1999) [14]. Approximately 100 mg of each sample was extracted using an ethanolic solution (70% ethanol, 29.5% water and 0.5% acetic acid) for 15 minutes. The ethanolic extract was then centrifuged, and the supernatant was separated using a centrifuge (Model CT15RE, Hitachi, Japan) for 20 minutes. An aliquot of 20 µL from each sample was added to cuvettes and reacted with 2 mL of ABTS radical solution for 6 minutes. The analysis was performed in triplicate, and the results were expressed as micromoles of Trolox equivalent per gram of dry sample (µmol TE/g d.b.). A Trolox calibration curve was constructed using six concentration points (0.625 to 25 µmol/L), following the linear equation: $Y = 0.0217x + 0.0294$, with $R^2 = 0.9911$.

2.6 Elaboration of CB

Different tests were performed, varying the types of ingredients and their concentrations, aiming at the best formulation in terms of visual aspect, texture and interaction between the ingredients. In the preliminary tests, the proportions of dry ingredients varied between 7 and 25% (oats, maltodextrin, Brazil nuts), while the liquids varied between 10 and 15% (glucose syrup, honey, water). Changes were allowed, as these tests produced a bar that was unstable after cooling and heating, and which fell apart. For the new formulation (Table 1), between 0.8 and 27.8% of dry ingredients and between 0.6 and 27.8% of liquids were added for 100 g of product.

Table 1: Concentration of the new formulation ingredients

Ingredients	(%)
FDAP or DFA	11.1
Thick oat flakes	27.8
Fine oat flakes	16.7
Honey	27.8
Glycerin	8.3
Maltodextrin	4.2
Cocoa powder	0.8
Chia seeds	1.9
Vanilla extract	0.6
Water	0.8

In this formulation, fine and coarse oat flakes, maltodextrin, cocoa powder, DFA or FDAP, and chia seeds were mixed in a stainless steel container. In another container, honey and glycerin were heated in a bain-marie at 40°C. The ingredients were mixed together using silicone protection until a malleable dough was formed, and small amounts of water were added when necessary. The CB were then shaped into a rectangular shape, individually wrapped in plastic wrap and cooled at 10°C for two hours. After cooling, the bars were placed in aluminum molds and baked in an oven at 105°C.

3. RESULTS AND DISCUSSION

3.1 DAF obtained after extraction

The quantification of the DAF generated after the supercritical extraction process is presented in Table 2. Large amounts of DAF were observed at 40 °C/300 bar (62.63 ± 0.40%) and 60 °C/300 bar (54.31 ± 0.01%). In the previous research [9], the operational variables temperature and pressure significantly influenced oil extraction and, consequently, the residual yield (RY) of the DAF highlighting the impact of process conditions on extraction efficiency. Under isobaric conditions (300 bar), increasing the temperature from 40 to 60 °C led to an approximate 8% decrease the DAF.

Table 2: DAF generated after extraction

Experimental Conditions	RY (%)	Density (kg/m^3)
40ºC/300 bar	62.63 ± 0.40	926.78
40ºC/400 bar	49.71 ± 0.01	990.90
60ºC/300 bar	54.31 ± 0.01	829.01
60ºC/400 bar	49.12 ± 0.03	911.03

At the isothermal condition of 40 °C, increasing the pressure from 300 to 400 bar resulted in a RY reduction of over 10%. A similar pattern was observed at 60 °C, confirming that higher pressure negatively affected DAF recovery at both temperatures. These results can be explained by the direct influence of pressure and temperature on the density of supercritical CO_2. In general, increasing pressure raises the solvent density, enhances the solubilization of compounds and, consequently, improving extraction oil but decreasing of the RY [9, 15].

However, in this study, the highest RY of DAF was associated with conditions of lower solvent density. In this context, the average porosity of the lyophilized sample was 0.7 [9], indicating a greater volume of empty spaces within the matrix, which likely facilitated solvent flow through the extraction bed. On the other hand, increased pressure may have caused bed compaction, limiting compound diffusion and solubilization, thereby reducing the overall efficiency of the process [6, 16].

The resulting DAF, which corresponds to 50 to 60% of the pulp used for extraction, can be valorized as a functional flour. When this percentage is combined with the peel and seed together accounting for approximately 30% of the fruit's weight and typically discarded, a significant volume of organic waste is observed. If not properly managed, such waste could lead to serious environmental impacts, especially in large-scale industrial processes [4, 7].

3.2 Proximal composition

The results of the proximal composition of FDAP and DAF are organized in Table 3. The moisture content of the FDAP was 3.40%, and DAF was 4.30%. This increase may be associated with sample handling, during which the material absorbed moisture from the environment. According to the Codex Alimentarius [17], the moisture content in food products should be below 14%. In the study conducted by Mooz et al. (2012) [18], the fruit exhibited a moisture content of

79.37%. Daiuto et al. (2014) [19] reported that the fresh pulp of the Hass variety contained 69.85% moisture, while the lyophilized pulp presented 13.99%. It is important to note that high moisture levels can affect macronutrient concentration and hinder the extraction process. Moreover, elevated moisture can increase microbial activity and negatively impact sensory characteristics such as flavor and texture.

Table 3: Proximal composition of FDAP and DFA

Analysis (%)	FDAP	DAF
Moisture	3.40 ± 0.26	4.30 ± 0.20
Ashes	0.79 ± 0.02	10.00 ± 0.24
Carbohydrates	12.31 ± 0.37	18.36 ± 0.26
Proteins	5.94 ± 0.25	8.83 ± 0.40
Lipids	45.33 ± 0.90	10.00 ± 0.35

The DAF exhibited a high ash content (10%) when compared to the FDAP (0.79%). Studies indicate that such variations may occur depending on the part of the fruit analyzed and are associated with the concentration of minerals present in the matrix. In a study conducted by Eggbound et al. (2018) [20], avocado seeds 3.82% ash. Similarly, Daiuto et al. (2014) [19], upon evaluating different freeze-dried parts of the Hass avocado, observed low mineral contents in the peel (2.15%) and seed (2.81%), in contrast to significantly higher values in the pulp (11.56%). These findings suggest that the supercritical extraction process may have contributed to the concentration of minerals in the residual flour. In this context, Peccin et al. (2022) [21] emphasize that avocado is a relevant source of macro and microminerals, such as potassium, nitrogen, calcium, magnesium, and sodium, which play essential roles in human metabolism, thereby conferring notable nutritional value to fruit.

The carbohydrate content was higher in the DAF (18.36%) compared to the values reported by Daiuto et al. (2014) [19] for the pulp of the Hass variety, which exhibited a low carbohydrate content (0.26%) in contrast to the seed (1.64%). Studies indicate that the highest concentrations of carbohydrates are generally found in fruit seeds. For instance, Cordeiro et al. (2025) [22] reported 83.94% carbohydrates in avocado seeds, while Peccin et al. (2022) [21] observed an even higher content of 83.25%. According to Ford et al. (2023) [23], avocado pulp contains an average of 8.64% carbohydrates. These findings suggest that flour obtained through supercritical extraction may serve as a viable alternative for the partial replacement of wheat or rice flour in food formulations.

The protein content of the DAF was comparable to the values reported in the literature. Silva et al. (2019) [24] developed flour from avocado seeds for use in biscuit formulations, which contained 5.17% protein. Daiuto et al. (2014) [19] observed a lower protein content of 1.27% in the pulp. Cordeiro et al. (2025) [22] identified a value of 4.50% in flour seed, and Peccin et al. (2022) [21] reported 3.63% protein in flour derived from the seed. These authors emphasize that avocado is a source of essential amino acids, which are associated with a reduced risk of chronic diseases such as type II diabetes mellitus and cardiovascular disorders. Furthermore, the defatted flour obtained after the extraction process qualifies as a source of protein according to the criteria established by legislation [25].

DAF considerable residual lipid content, consistent with values reported by other authors, which range from 2 to 8% [19, 21, 24]. This lipid content contributes to the functional quality of the flour, as avocado is rich in monounsaturated fatty acids, such as oleic acid (C18:1), linoleic acid (C18:2, polyunsaturated), and palmitic acid (C16:0, saturated) [9, 23]. These fatty acids are associated with reductions in LDL cholesterol levels and improvements in cardiovascular health. Therefore, avocado flour represents an excellent alternative as an ingredient in dietary supplements, particularly due to its low total fat content, making it suitable for lipid-restricted diets.

3.3 Bioactive compounds

The results are presented in Table 4. Phenolic compounds are natural antioxidants known for their anti-inflammatory activity and their ability to neutralize free radicals, contributing to the prevention of chronic diseases and enhancing the oxidative stability of foods. When present in high concentrations, they add significant functional value to agro-industrial by-products [3, 5, 26].

Table 4: Phenolic compounds e Lutein content in samples

Experimental conditions	TPC (µg GAE/g)	Lutein (µg/g)
40ºC/300 bar	753.21± 0.09	173.55 ± 0.01
40ºC/400 bar	630.00 ± 0.03	136.38 ± 0.01
60ºC/300 bar	646.61± 0.01	143.69 ± 0.02
60ºC/400 bar	579.11± 0.03	119.41 ± 0.01
FDAP	458.57± 0.01	99.20 ± 0.02

The highest levels of TPC were observed under the conditions of 40 °C/300 bar and 60 °C/300 bar, suggesting that higher pressures (400 bar) reduced the recovery of these antioxidants. In other studies, the concentration of TPC has been shown to vary widely depending on the part of the fruit analyzed and the type of solvent used. Peccin et al. (2022) [21] reported values ranging from 52,790 to 86,030 µg GAE/g in flour obtained from avocado seeds, using hexane and ethanol as extraction solvents, respectively. Similarly, in another study [19] was found TPC ranging from 3,300 to 63,500 µg GAE/g across pulp, peel, and seed, with the pulp being the least concentrated fraction in phenolic compounds.

In avocado pulp, TPC can range from 220 to 400 µg GAE/g, depending on the fruit variety. The Margarida cultivar, for example, presents high levels in the seed (88,380 µg GAE/g) and lower concentrations in the pulp (400 µg GAE/g) [27]. According to Amado et al. (2019) [28], the pulp of the Margarida variety contains 1,910 µg GAE/g, while the peel exhibits 5,450 µg GAE/g. The Hass and Quintal varieties show phenolic contents of 16,150 and 38,820 µg GAE/g in the pulp, respectively. As observed, the TPC in the DAF obtained by supercritical extraction were higher than those in the FDAP, indicating that the defatting process with CO_2 promoted a relative concentration of these antioxidants in the solid residue. These findings reinforce the functional potential of DAF as a value-added by-product in the extraction chain.

Lutein, in particular, is the most abundant carotenoid in avocado pulp. Studies indicate that its concentration tends to be higher in dark green pulp varieties and is associated with eye health. Reported concentrations in the pulp range from 0.001 to 0.004 g/kg, and in some varieties, values as high as 271 µg/100 g of lutein have been observed [18, 29]. The highest value was observed under the condition of 40 °C/300 bar, with a concentration of 173.55 ± 0.01 µg/g. The scientific literature still lacks studies specifically evaluating lutein content in DAF, highlighting the relevance of the data obtained in this study. Carotenoids are compounds known for their antioxidant and anti-inflammatory potential, as well as for their ability to inhibit enzymes associated with the onset of chronic diseases [3, 9].

According to literature data, lutein content varies considerably depending on the part of the fruit analyzed and the extraction method employed. Studies have reported concentrations ranging from 0.322 to 0.324 µg/g in the peel and seed [30], and between 0.14 and 0.842 µg/g in the pulp [23]. Ashton et al. (2006) [31] reported a content of 20.5 µg/g using hexane maceration extraction. Therefore, it is evident that the lutein levels obtained in the flours extracted with supercritical CO_2 in this study were significantly higher than those reported using other methods.

In present study, the use of supercritical CO_2 as a solvent enabled simultaneous defatting of the pulp and concentration of lutein in the solid residue. Supercritical CO_2 has a high solvating capacity for lipophilic compounds, such as carotenoids, particularly those located on the surface of the cell wall, which are often extracted along with the oil. However, intracellular lipid emulsions, which are less accessible, may offer greater resistance to extraction, thereby limiting the complete removal of these compounds [3, 9].

3.4 Antioxidant activity

The antioxidant activity evaluated using two distinct methods DPPH and ABTS is presented in Table 5. The DAF showed higher antioxidant activity under the conditions of 40 °C/400 bar and 40 °C/300 bar, as evaluated by the DPPH and ABTS methods. The results demonstrated variations among the operational conditions, indicating a direct influence of pressure and temperature on the preservation of antioxidant compounds.

Table 5: Antioxidant Activity of DAF and FDAP

Experimental conditions	DPPH (µmol Trolox eq./g)	ABTS (µmol Trolox eq./g)
40ºC/300 bar	0.376 ± 0.02	0.413 ± 0.02
40ºC/400 bar	0.516 ± 0.01	0.316 ± 0.02
60ºC/300 bar	0.416 ± 0.05	0.132 ± 0.02
60ºC/400 bar	0.376 ± 0.03	0.190 ± 0.02
FDAP	0.056 ± 0.02	0.056 ± 0.01

In the literature, antioxidant activity values vary widely depending on the part of the fruit, the variety, and the extraction method used. Daiuto et al. (2014) [19], in a study on the pulp of the Hass variety, reported values of 8.1 µmol Trolox eq./g (DPPH) and 15.5 µmol Trolox eq./g (ABTS). In another study, Bezerra et al. (2022) [32] reported antioxidant activity in the peel ranging from 0.710 to 1.29 µmol Trolox eq./g (DPPH) and from 132 to 321 µmol Trolox eq./g (ABTS), values similar to those found by Amado et al. (2019) [28], who reported 2.34 µmol Trolox eq./g.

Silva-Reis et al. (2024) [33] evaluated the effect of solvent type on the antioxidant activity of avocado pulp and observed that more polar solvents, such as methanol, promote greater solubilization of antioxidants, resulting in higher antioxidant activity values. In that study, DPPH-based activity reached 35.54 µmol Trolox eq./g. Moreno et al. (2014) [34], in turn, compared six avocado varieties and reported a maximum value of 1.61 µmol Trolox eq./g for the pulp. In the study conducted by Nascimento et al. (2021) [35], analyzing the Hass variety, a wide range was identified: from 41 to 98 µmol Trolox eq./g using the DPPH method and from 141 to 402 µmol Trolox eq./g using the ABTS method.

In the present study, the antioxidant activity values obtained for DAF were considered low when compared to those reported in the literature. This reduced activity may be associated with the limited solubility of the antioxidants after oil extraction with supercritical CO_2. A significant portion of these compounds is carried away with the oil during extraction, while the remaining antioxidants in the solid matrix may be strongly bound to structural components such as fibers and carbohydrates, making their extraction with common solvents more difficult. Furthermore, it is widely recognized that the peel and seed generally exhibit higher antioxidant content compared to the pulp [9, 19].

3.5 Cereal bar composition

The results of the proximate composition analysis in CBFDA and CBDA are presented in Table 6. The product developed using freeze-dried pulp showed a high carbohydrate content (19.46%) due to the concentration of fruit solids caused by lyophilization. An increase in protein content (10.33%) and a low lipid percentage (4.31%) was also observed, indicating its potential as an excellent protein source. In a study using Baru seeds [36], the cereal bars contained 50% carbohydrates, 11.67% moisture, and 12.24% protein. The authors emphasized the importance of varying seed proportions to better evaluate the physical attributes of the bars. Another important factor is the addition of sweeteners such as honey and glucose, which influence the carbohydrate content of the bars and may limit their consumption by individuals following low-carbohydrate diets.

Table 6: Proximate composition of CBFDA and CBDA

Analysis (%)	CBFDA	CBDA
Moisture	22.19 ± 0.98	25.19 ± 0.99
Ashes	1.66 ± 0.09	2.17 ± 0.12
Carbohydrates	19.46 ± 0.91	6.41 ± 0.26
Proteins	10.33 ± 0.98	12.29 ± 0.46
Lipids	4.31 ± 0.13	3.22 ± 0.06

CBDA was formulated with the aim of obtaining a protein-rich product, as the defatted avocado flour (DAF) used in the supercritical extraction process showed significantly reduced moisture, and high concentrations of carbohydrates and proteins. However, it was observed that even with standardized amounts of ingredients, CBDA exhibited a low carbohydrate content (6.41%) and a protein level (12.29%) like that of CBFDA. In both cases, the bars exceeded the maximum moisture limit accepted, indicating the need for formulation adjustments to ensure that the composition of CBDA and CBFDA complies with regulatory standards [17, 25]. Regarding visual aspects, CBFDA presented a darker coloration due to the addition of chocolate and a firmer texture for handling. CBDA, even after cooling and oven heating, remained sticky, which made handling difficult (Figure 2).

CBFDA CBDA

Figure 2: Formulated Cereal Bars.

In a study conducted by Melati et al. (2021) [37], the authors evaluated different binding agents in the formulation of cereal bars, such as guar gum, chia, and psyllium. The results indicated that the formulation containing psyllium showed better sensory characteristics in terms of flavor, texture, and color. The texture of cereal bars can also be adjusted through the use of dry ingredients such as rice flour, oats, and granola, which contribute to product stability and crunchiness, while also enhancing its nutritional value. In another study [38] cereal bars with avocado seeds were elaborated and showed texture and crunchiness due to the low protein content, which according to the authors reduces the capacity to absorb water. In the present study, oats were used to provide texture and crunchiness to the bar, while honey, chia seeds, and maltodextrin served as binding and thickening agents. Cocoa was added to enhance the nutritional value, and small amounts of water were incorporated to aid in bar cohesion and improve ingredient mixing. However, adjustments are needed to optimize the amounts of FAD to be incorporated, aiming for maximum utilization of these inputs and improvements in attributes such as texture and consistency.

4. CONCLUSION

The application of defatted avocado flour (DAF) as an ingredient in the development of food products represents a sustainable alternative aimed at reducing and valorizing agro-industrial waste, as well as minimizing the environmental impacts associated with its disposal. This approach is further strengthened by the use of green and eco-friendly technologies. This study demonstrated that supercritical fluid extraction of freeze-dried avocado pulp (FDAP) enabled the production of a nutritious DAF, rich in carbohydrates, proteins, carotenoids, phenolic compounds, and with higher antioxidant potential compared to FDAP. These findings indicate that the extraction process concentrated macro- and micronutrients in DAF, enhancing its functional and bioactive value. The incorporation of DAF into formulations such as cereal bars contributes to the development of a product with nutritional and functional value, particularly in terms of carbohydrate and protein content. However, adjustments to the physical attributes of the product such as moisture, texture and consistency are necessary to improve bar quality both with the incorporation of DAF and FDAP. In this context, further studies on the functional and technological properties of DAF are needed to maximize its use in the development of new food products and to identify potential limitations. Additionally, conducting sensory analyses is essential to assess consumer perception and guide formulation improvements.

5. ACKNOWLEDGMENTS

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001

6. REFERENCE

1. Kumar RS, Sasikumar R, Dhilipkumar T. Exploiting agro-waste for cleaner production: A review focusing on biofuel generation, bio-composite production, and environmental considerations. J Cleaner Prod. 2024;435:140536. doi: 10.1016/j.jclepro.2023.140536
2. Ali EA, Mohammed DM, Abd El Gawad F, Orabi MA, Gupta RK, Srivastav PP. Valorization of food processing waste byproducts for essential oil production and their application in food system. Waste Manag Bull. 2025;3(3):100200. doi: 10.1016/j.wmb.2025.100200
3. Pantoja KRP, Melo GCA, Ferreira CP, Lima MC, Menezes EGO, Carvalho Junior RN. Supercritical technology as an efficient alternative to cold pressing for avocado oil: a comparative approach. Foods. 2024;13(15):2424. doi: 10.3390/foods13152424
4. Tesfaye T, Ayele M, Gibril M, Ferede E, Limeneh DY, Kong F. Beneficiation of avocado processing industry by-product: a review on future prospect. Curr Res Green Sustain. Chem. 2022;5:100253. doi: 10.1016/j.crgsc.2021.100253
5. Abaide ER, Zabot GL, Tres MV, Martins RF, Fagundez JL, Nunes LF, et al. Yield, composition, and antioxidant activity of avocado pulp oil extracted by pressurized fluids. Food Bioprod Process. 2017;102:289-98. doi: 10.1016/j.fbp.2017.01.008
6. Knez Ž, Pantića M, Cöra D, Novaka Z, Knez Hrnčič M. Are supercritical fluids solvents for the future? Chem Eng Process Intensif. 2019 Jul;141:107532. doi: 10.1016/j.cep.2019.107532
7. Uwineza PA, Waskiewicz A. Recent advances in supercritical fluid extraction of natural bioactive compounds from natural plant materials. Molecules. 2020;25(17):3847. doi: 10.3390/molecules25173847
8. Jabeen S, Huma N, Sameen A, Zia MA. Formulation and characterization of protein-energy bars prepared by using dates, apricots, cheese and whey protein isolate. Food Sci Technol. Campinas. 2021;41(Suppl 1):197-207. doi: 10.1590/fst.12220
9. Pantoja KRP, Aires GCM, Oppata SA, Carvalho JPF, Cordeiro RM, Carvalho Junior RN. High-quality Margarida avocado oil via supercritical CO_2 compared to commercial Hass avocado oil obtained by cold pressing: a comparative lipid profile analysis by UPC^2 and GC–MS. The Journal of Supercritical Fluids. 2026;229:106819. doi: 10.1016/j.supflu.2025.106819
10. Latimer Junior, GW. Official Methods of Analysis of AOAC International. 22. ed. Oxford (UK): Oxford University Press; 2023.
11. Zieliński H, Kozłowska H. Antioxidant activity and total phenolics in selected cereal grains and their different morphological fractions. J Agric Food Chem. 2000;48(6):2008-16. doi: 10.1021/jf990619o

12. Rodriguez-Amaya, DB. A guide to carotenoid analysis in foods. 1. ed. Washington (DC): ILSI Press; 2001.
13. Brand-Williams W, Cuvelier ME, Berset C. Use of a free-radical method to evaluate antioxidant activity. LWT Food Sci Technol. 1995;28(1):25-30. doi: 10.1016/S0023-6438(95)80008-5
14. Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic Biol Med. 1999;26(9-10):1231-7. doi: 10.1016/s0891-5849(98)00315-3
15. Ahmad T, Danish M. A review of avocado waste-derived adsorbents: Characterizations, adsorption characteristics, and surface mechanism. Chemosphere. 2022;296:134036. doi: 10.1016/j.chemosphere.2022.134036
16. Majid A, Phul AR, Khaskheli AH, Abbasi S, Sirohi MH, Ahmed I, et al. Applications and opportunities of supercritical fluid extraction in food processing technologies: A review. Int J Adv Appl Sci. 2019;6(7):99-103. doi: 10.21833/ijaas.2019.07.013
17. Codex Alimentarius Commission. Codex standard for vegetable protein products (CODEX STAN 152-1985). Rome (IT): FAO/WHO; 1985. Available from: https://www.fao.org/fao-who-codexalimentarius/sh-proxy/ru/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXS%2B152-1985%252FCXS_152e.pdf.
18. Mooz ED, Gaiano NM, Shimano MYH, Amancio RD, Spoto MHF. Physical and chemical characterization of the pulp of different varieties of avocado targeting oil extraction potential. Food Sci Technol. 2012;32(2):274-80. doi: 10.1590/S0101-20612012005000055
19. Daiuto ER, Tremocoldi MA, de Alencar SM, Vieites RL, Minarelli PH. Composição química e atividade antioxidante da polpa e resíduos de abacate *Hass*. Rev Bras Frutic. 2014;36(2):417-24. doi: 10.1590/0100-2945-102/13
20. Egbuonu AC, Opara IC, Onyeabo C, Uchenna NO. Proximate, functional, antinutrient and antimicrobial properties of avocado pear (*Persea americana*) seeds. J Nutr Health Food Eng. 2018;8(1):260. doi: 10.15406/jnhfe.2018.08.00260
21. Peccin MM, Bernardi JL, Nascimento LH, Ambrósio N, Cansian RL, Paroul N, et al. Caracterização físico-química da farinha e dos extratos das sementes de abacate (*Persea americana* (Mill.) *Lauraceae*). Rev Colomb Cienc Quím Farm. 2022;51(1):369-86. doi: 10.15446/rcciquifa.v51n1.95598
22. Cordeiro JFS, Shinohara NKS, Alves JCA, Santos TG, Pinto Neto WT, Lacerda JPC, et al. Caracterização e utilização de farinha de semente de abacate "Hass" (*Persea americana*) e resíduo de malte de cerveja (*Hordeum vulgare*) para elaboração de biscoitos. Rev Observ Econ Lat. 2025;23(5):1-21. doi: 10.55905/oelv23n5-019
23. Ford NA, Spagnuolo P, Kraft J, Bauer E. Nutritional composition of *Hass* avocado pulp. Foods 2023;12:2516. doi: 10.3390/foods12132516
24. Silva IG, Andrade APC, Silva LMR, Gomes DS. Elaboração e análise sensorial de biscoito tipo cookie feito a partir da farinha do caroço de abacate. Braz J Food Technol. 2019;22:e2018209. doi: 10.1590/1981-6723.20918
25. Brasil. Agência Nacional de Vigilância Sanitária (Anvisa). Resolução da Diretoria Colegiada – RDC nº 429, de 8 de outubro de 2020. Dispõe sobre a rotulagem nutricional de alimentos embalados. Brasília (DF): Diário Oficial da União; 2020 Oct 09;106(Seção 1). Available from: http://www.in.gov.br/en/web/dou/-/resolucao-de-diretoria-colegiada-rdc-n-429-de-8-de-outubro-de-2020-282070599.
26. Nardini M. Phenolic compounds in food: Characterization and health benefits. Molecules. 2022;27(3):783. doi: 10.3390/molecules27030783
27. Oliveira CS, Andrade JKS, Rajan M, Narain N. Influence of the phytochemical profile on the peel, seed and pulp of Margarida, Breda and Geada varieties of avocado (*Persea Americana* Mill) associated with their antioxidant potential. Food Sci Technol. 2022;42: e25822. doi: 10.1590/fst.25822
28. Amado DAV, Detoni AM, Carvalho LSC, Torquato AS, Martin TS, Tiuman TS, et al. Tocopherol and fatty acids content and proximal composition of four avocado cultivars (*Persea americana* Mill). Acta Alimentaria. 2019;48(1):47-55. doi: 10.1556/066.2019.48.1.6
29. Ramos-Aguilar AL, Ornelas-Paz J, Tapia-Vargas LM, Ruiz-Cruz S, Gardea-Béjar AA, Yahia EM, et al. The importance of the bioactive compounds of avocado fruit (*Persea americana* Mill) on human health. Biotecnia. 2019;21(3):154-62. doi: 10.18633/biotecnia.v21i3.1047
30. Marra A, Manousakis V, Zervas GP, Koutis N, Finos MA, Adamantidi T, et al. Avocado and its by-products as natural sources of valuable anti-inflammatory and antioxidant bioactives for functional foods and cosmetics with health-promoting properties. Appl Sci. 2024;14(14):5978. doi: 10.3390/app14145978

31. Ashton OBO, Wong M, McGhie TK, Vather R, Wang Y, Requejo-Jackman C, et al. Pigments in avocado tissue and oil. J Agric Food Chem. 2006;54:10151-8. doi: 10.1021/jf061809j
32. Bezerra RAD, Silva NM, Tuzzi BF, Marchi FE, Feihrmann AC, Santos GT. Extração de compostos fenólicos e atividade antioxidante da casca de abacate (*Persea americana* Mill) através de diferentes solventes. Res Soc Dev. 2022;11(13):e480111335602. doi: 10.33448/rsd-v11i13.35602
33. Silva-Reis CM, Faria CMDR, Gil ACD, Castro JC, Mardigan LP, Zanutto CA, et al. Influência do solvente e do método de extração na atividade antioxidante e no conteúdo total de polifenóis de extratos de polpas de abacate, pitaia e uva. Contrib. Cienc Soc. 2024;17(4):e6277. doi: 10.55905/revconv.17n.4-236
34. Moreno E, Ortiz BL, Restrepo LP. Contenido total de fenoles y actividad antioxidante de pulpa de seis frutas tropicales. Rev Colomb Quim. 2014;43(3):41-8.
35. Nascimento FCG, Sousa MM, Bermúdez VMS, Silva EF, Nascimento VLV. Avaliação de carotenoides e atividade antioxidante da polpa madura do abacate (*Persea americana* Mill). Res Soc Dev. 2021;10(8):e45010817500. doi: 10.33448/rsd-v10i8.17500
36. Arruda LC, Faria WCS, Siqueira NFMP, Campos MFS, Barros WM. Development of cereal bar with baru almonds and pineaplle peels. Res Soc Dev. 2021;10(5):e21610514684. doi: 10.33448/rsd-v10i5.14684
37. Melati J, Luccheta L, Prado NV, Oliveira DF, Tonial IB. Características físicas e sensoriais de barras de cereais salgados com diferentes ligantes. Food Sci Technol. 2021;41(Sup 1):150-4. doi: 10.1590/fst.07820
38. Koeswanto A, Kusuma K. Formulation and evaluation of a novel functional cereal containing avocado seed flour. Braz J Dev. 2023;9(11):30308-21. doi: 10.34117/bjdv9n11-065