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# Physical and Chemical Properties of 70% Cocoa Dark Chocolate Mixed with Freeze-Dried Arazá (*Eugenia stipitata*) Pulp

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**ABSTRACT:** This work aimed to determine the chemical and physical properties of 70% dark cocoa chocolate, including freeze-dried Arazá (*Eugenia stipitata*) pulp (FDAP). We studied chocolates incorporating three FDAP concentrations (1.0, 1.5, and 2.0%). No statistical differences were found in total polyphenol content, antioxidant capacity, and total catechin and epicatechin content. The dark chocolates' moisture and texture were unaffected by the FDAP. The Casson yield stress increased to 19.67±1.35 Pa, while the Casson plastic viscosity reduced to 1.68±0.03 Pa·s, Also, the particle size increased. The dark chocolates' flow behavior corresponded to a non-Newtonian fluid. Finally, the dark chocolate's properties were unaffected by a 2% FDAP concentration.

**Keywords:** antioxidants, catechin, chocolate, polyphenols, viscosity

## INTRODUCTION

Chocolate is a delicacy produced by combining cocoa derivatives, such as liquor, powder, or butter, with other components (Urbańska and Kowalska, 2019; Deus et al., 2021), with a final solid content between 65% and 75% (Glicerina et al., 2016; Ostrowska-Ligęza et al., 2019). These components are refined, combined, and subjected to conching and tempering, influencing the chocolate's final appearance, texture, taste, and aroma (Calva-Estrada et al., 2020). Moreover, the formulation and processing steps confer the rheological and textural characteristics, viscosity, mouthfeel, consistency, appearance, sensation, and acceptance of chocolates by the consumer (Glicerina et al., 2015; Gunaratne et al., 2019; Saputro et al., 2019). Additionally, the flow behavior is a critical quality attribute of chocolates.

High cocoa content chocolates offer phenolic compounds, including catechin and epicatechin (Nascimento et al., 2020), as well as oligomeric and polymeric procyanidins (Di Mattia et al., 2014; Gültekin-Özgüven et al., 2016), which are all known as polyphenols (Frank et al., 2020). Polyphenols have antioxidant properties that influence sensorial qualities, such as taste and color (Urbańska and Kowalska, 2019), impacting chocolate quality (de Rezende Mudenuti et al., 2018). The manufacturing of chocolate affects its total antioxidant capacity (AC) and total polyphenol content (TPC) (Gültekin-Özgüven et al., 2016; Martini et al., 2018). Currently, efforts are being made to preserve chocolate polyphenolic compounds by developing adequate treatments and minimal processing (Żyżelewicz et al., 2018).

The new flavored-chocolates development involves replacing traditional ingredients or adding new and nutritious ones, always trying to preserve their final texture and functionality (Wohlmuth, 2009; Zhao et al., 2018) since they can influence their quality. For example, adding fruit would be suitable to give the chocolate an attractive aroma and the possibility of masking the bitter taste of a high cocoa content chocolate (Komes et al., 2013). An underexplored alternative to improve chocolates' flavor and polyphenolic profile is to add natural polyphenols from plant sources (Todorovic et al., 2015; Sim et al., 2016). Hence, industries incorporate various types of fruits with bioactive compounds and antioxidant properties into their chocolates, generating the necessity to have

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a more profound knowledge of their polyphenols' content and their AC (Pedan et al., 2017). The work of Lončarević et al. (2018) in chocolates demonstrated that 10% of blackberry juice increased the Casson viscosity and polyphenol content, conferring an attractive color and a blackberry taste. Belščak-Cvitanović et al. (2012) showed that 1% of red raspberry's lyophilized leaf extract did not alter the chocolate's attributes. Todorovic et al. (2015) also found that incorporating raspberry into dark chocolates insignificantly influenced its TPC.

*Eugenia stipitata* (Arazá) is a highly aromatic and acidic fruit (de Araújo et al., 2019). The results of de Araújo et al. (2021) suggest that Brazilian Arazá has excellent functional properties and a high nutritional value. Furthermore, the edible fraction had minerals, sucrose, fructose, maltotetraose, organic and phenolic acids, flavonoids, carotenoids, terpenes, and a high AC (de Araújo et al., 2021). Its presence in the industry represents a new attractive polyphenol source with good antioxidant quality (de Araújo et al., 2019).

Although new approaches for producing polyphenols-enriched products have been reported, dark chocolates enriched with natural and readily available ingredients, including fruits, are lacking (Komes et al., 2013). Therefore, we investigated the results of adding three levels of freeze-dried Arazá (*E. stipitata*) pulp in 70% dark cocoa chocolates on their physical and chemical properties.

## MATERIALS AND METHODS

### Materials

Ripe Arazá fresh fruit was supplied by a local farmer from the Tsuntsunsa native community located in Bagua province, Amazonas, Perú. The fruits were delivered to the Laboratory of Agroindustrial Biotechnology at the Universidad Nacional Toribio Rodriguez de Mendoza de Amazonas (UNTRM-A). The APROCAM cooperative provided the dried fermented Criollo cocoa beans.

### The dark chocolate process

***The preparation of freeze-dried Arazá pulp*:** The ripe Arazá fruits were peeled and pulped as follows; 1,700 g pulp was cut into 2 to 3 cm cubes, then 40 g pulp was placed in 50-mL falcon tubes. Next, the tubes were dried in a freeze dryer compartments (Labconco Corp., Kansas City, MO, USA) for four days, grounded to powder (MM200, Retsch GmbH, Haan, Germany) for 4 min, and maintained at room temperature until chemical analysis.

***Dark chocolate production*:** The dark chocolate was produced at the Agroindustrial Pilot Plant of the UNTRM-A following the recipe provided by APROCAM and the method suggested by Abdul Halim et al. (2019) and Medina-Mendoza et al. (2021) with a few minor amendments.

**Table 1.** Formula for the production of dark chocolates (unit: %)

| Ingredient | Dark chocolate | | | |
|---|---|---|---|---|
| | Control | FDAP1 | FDAP1.5 | FDAP2 |
| Criollo cocoa liquor | 70.00 | 70.00 | 70.00 | 70.00 |
| Sugar | 25.00 | 24.00 | 23.50 | 23.00 |
| Cocoa butter | 5.00 | 5.00 | 5.00 | 5.00 |
| FDAP | 0.00 | 1.00 | 1.50 | 2.00 |

FDAP1, FDAP1.5, and FDAP2 are dark chocolates with 1, 1.5, and 2% of freeze dry Arazá pulp, respectively.

Four groups of 70% dark cocoa chocolates were produced [without and with three freeze-dried Arazá pulp (FDAP) concentrations]. Chocolates without FDAP served as controls. Adding three different FDAP concentrations replaced the sugar content (1%, 1.5%, and 2% for chocolate FDAP1, FDAP1.5, and FDAP2, respectively; Table 1). A tray dryer (Fischer Agro, Surquillo, Perú) was used for the roasting process at 120°C for 30 min. Then, the roasted beans were husked and crushed (DC-C, IMSA, Chorrillos, Perú) to obtain the nibs. Next, the nibs were ground with a cocoa bean grinding machine (Tritur-50, Prosol Inversiones S.A.C., San Miguel, Perú) until cocoa liquor was obtained. All ingredients were integrated with a two-roller granite stone concher (PG508, Premier Kitchen, Chennai, India) for 20 h at 60°C.

Three different FDAP concentrations were added separately to the mixture one hour before finishing the process according to the Table 1. The mixture was tempered (Templa05, Prosol Inversiones S.A.C.) at 34°C; then, 45 g was transferred into a chocolate mold. For unmoulding, the chocolate was maintained at 4°C for 15 min, foiled with aluminum, and reserved at room temperature (18°C) until the functional, physical, and sensory analysis (Komes et al., 2013).

### Chemical properties of dark chocolates

***Extraction of polyphenols*:** The Soxhlet extraction method described by Zhou et al. (2015), with minor changes, was used to defat the dark chocolates. For deffating, 5-g dark chocolate were mixed with petroleum ether ≥90% and put into a fat extractor (Det-Gras N, JP-Selecta, Barcelona, Spain). One gram of degreased chocolate was combined with a 10-mL methanol solution (80%), vortexed (1 min), placed in an ultrasonic bath (10 min at 30°C), and centrifuged (3,000 rpm for 10 min) (Gültekin-Özgüven et al., 2016). The collected supernatant was put in vials and refrigerated. The cocoa liquor was processed in the same way. The extraction of FDAP polyphenols was direct because they have no fat.

***TPC*:** The Folin-Ciocalteu technique was used to calculate the TPC (Singleton et al., 1999; Pantelidis et al., 2007). In summary, diluted extract (0.05 mL), water (0.45 mL),

and Folin-Ciocalteu phenol reagent 10-fold diluted (2.5 mL) were mixed with a 2-mL sodium carbonate at 7.5% (w/v). At 760 nm, absorbance was measured after 5 min at 50°C (S2100, Unico, Los Angeles, CA, USA). TPC was calculated using a standard curve of gallic acid. Ranges of 0～2,500 ppm of gallic acid dilutions ($R^2$=0.995) were used to make the calibration curve (y=0.107+0.0009x). The results used a gallic acid equivalent (mg GAE)/g defatted sample in milligrams.

***AC*:** The 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical assay (Brand-Williams et al., 1995), with some modifications, was used to determine the AC of dark chocolates (Scherer and Godoy, 2009; Żyżelewicz et al., 2018). For solution B, the DPPH (0.005 g) was added to 80% methanol solution (100 mL). Similarly, another 80% methanol solution (solution C) was used for the extract's dilutions. Different concentrations (1:1, 1:2, 1:5, 1:10, and 1:20) of the extract dilutions were prepared separately (solution A). Seven test tubes comprising 0.1 mL (solution A) and 3.9 mL (solution B) each were prepared. For the control, 0.1 mL solution C was mixed with 3.9 mL solution B. After a reaction of 30 min at room temperature in the dark, each sample's absorbance was read using a spectrophotometer at 517 nm. The percentage of DPPH radical inhibition (I%) was assessed using Equation 1, where $Abs_1$ and $Abs_0$ are the sample and control absorbance, respectively. The inhibition percentage versus extract concentration graph was used to obtain the extract concentration that produced 50% inhibition ($IC_{50}$) of the DPPH radical (Oke et al., 2009).

$$I\ (\%) = \left( \frac{Abs_0 - Abs_1}{Abs_0} \right) \times 100 \quad (1)$$

***Quantification of bioactive compounds by ultra-high performance liquid chromatographic-diode array detection (UHPLC-DAD)*:** With modest adjustments, the Demir et al. (2014) and Coklar and Akbulut (2017) methods were used to measure the content of epicatechin and catechin in the chocolate samples as a reference. G7167-B multisampler, G7116-B column oven, G7104-A flexible pump, and G7117-B diode array detector were employed using the Agilent 1290 Infinity Series UHPLC system (Agilent Technologies, Santa Clara, CA, USA). Before injection, a syringe filter of 0.45-μm pore size ×33 mm (Millex, Merck, Darmstadt, Germany) was used to filter the extracts. Samples of 10 μL were injected into the system using a multisample injector. The separation was conducted using a reversed-phase C18 column (5 μm, 250 mm×4.6 mm internal diameter). For the mobile phase, water/acetic acid (98:2) (a) along with water/acetonitrile/acetic acid (78:20:2) (b), were considered. The flow rate was 0.75 mL/min, and the used gradient went like this: 10～14% b (5 min), 14～23% b (11 min), 23～35% b (5 min), 35～40% b (14 min), 40～100% b (3 min), 100% b isocratic (3 min), 100～10% b (3 min), and 10% b isocratic (4 min). The set temperature value of the column was 40°C; for the detector, it was 280 nm, and the column temperature was set to 40°C. Comparison peak regions per standard were used to quantify the epicatechin and catechin content. For the analysis of the data, the Agilent ChemStation Software (Agilent Technologies) was used.

**Physical analysis**

***Texture*:** According to Biswas et al. (2017), a texture analyzer (CT3 Texture Analyzer, AMETEK Brookfield, Middleborough, MA, USA) fitted with a 25-kg load cell, and a stainless steel probe of TA2/1000 was utilized in measuring the texture. The samples were maintained at room temperature (20°C) for 1 h before texture measurement. The parameters used for pretest speed, test speed, product volume, penetration depth, time set, test speed, cell load, and trigger force were set at 0.5 mm/s, 2.0 mm/s, 0.7 mm×55 mm×50 mm, 3 mm, 1～2 min, 50 mm/s, 25 kg, and 20 g, respectively. The hardness (kg) was determined.

***Flow properties of dark chocolates*:** According to Medina-Mendoza et al. (2021) and Abdul Halim et al. (2019), rheological analysis was conducted to establish the flow behavior of FDAP chocolate. The Casson yield stress and plastic viscosity were surveyed using a Rheometer (Model MCR 92, Anton Paar, Graz, Austria) concentric cylinder equipped. This equipment quantified the rheological parameters based on the International Office of Cocoa, Chocolate and Sugar Confectionery (IOCCC). All samples were melted for 60 min at 50°C and put into a cup, and the measurements were achieved at 40°C. The cycle begins with a 60 s preconditioning phase at 40°C. Finally, the data were analyzed using the software (Rheo-Compass, Anton Paar) of the equipment, based on the Casson Model (Equation 2), [σ=yield stress (Pa); $\sigma_0$=Casson yield stress (Pa); $K_1$=Casson plastic viscosity (Pa·s), and γ=shear rate ($s^{-1}$)] (Abdul Halim et al., 2019).

$$\sigma^{0.5} = (\sigma_0)^{0.5} + K_1(\gamma)^{0.5} \quad (2)$$

***Particle size distribution*:** According to Zhao et al. (2018), Bek et al. (2020), and Lim et al. (2021), the particle size distribution of dark chocolates was quantified using a laser diffraction particle size analyzer (PSA1190, Anton Paar) in liquid mode and the Kalliope software (Anton Paar). A small amount of defatted chocolate fine particles was added to the circulating water in the analyzer. Next, the particle-water combination was run through the analyzer to obtain an average size distribution until an obscuration of ≥12.9% or more was attained before taking measurements. The dark chocolate samples in this investigation had a refractive index of 1.33.

***Moisture content*:** The dark chocolate moisture content was obtained using a halogen moisture analyzer (Excellence plus HX204, Mettler-Toledo GmbH, Greifensee, Switzerland) following the Elbl et al. (2020) method, with some variations. For this, a 1-g sample each was accurately weighed in a crucible, and the thermogravimetric principle was used for measurement. Then, all samples were heated to 105°C until a constant weight was reached. The weight difference provides the amount of moisture presented.

### Statistical analysis

Minitab Software version 19 (Minitab LLC., State College, PA, USA) was used for the statistical data analysis. From each chocolate batch, three independent samples (n=3) were extracted for their respective analysis. One-way ANOVA and Tukey's range test were used to evaluate the significant differences among each treatment. The spider chart for sensory analysis was made using Excel 2013 for Windows (Microsoft Corp., Redmond, WA, USA).

## RESULTS AND DISCUSSION

### TPC, AC, and bioactive compounds of dark chocolates

Table 2 shows that the FDAP (7.01±1.45 mg GAE/g) used in this study has higher TPC than the Brazilian Arazá (1.84±0.83 mg GAE/g) (Neri-Numa et al., 2013), mainly due to the cultivation environment (Di Mattia et al., 2014; Jiang et al., 2021). However, the FDAP TPC was lower than that obtained for Criollo cocoa liquor (9.52± 1.34 mg GAE/g). de Rezende Mudenuti et al. (2018) produced 70% Brazilian forastero cocoa chocolates, reporting a TPC of 15.8 mg GAE/g. Moreover, Vertuani et al. (2014) reported 29.17±1.21 mg GAE/g in 70% dark cocoa chocolate from Madagascar, and Urbańska and Kowalska (2019) found a TPC of 40.55 mg GAE/g in chocolate products made of Colombian roasted beans. These values are superior to those obtained in this study (10.96±0.92~11.93±1.03 mg GAE/g), possibly because they were made from Criollo cocoa. Urbańska and Kowalska (2019) reported values of 9.10 mg GAE/g in chocolates made with Criollo cocoa from Venezuela, similar to our findings. Chocolate TPC depends on the formulation and the technological parameters during production (Belščak-Cvitanović et al., 2012; Di Mattia et al., 2014), particularly the temperature (Urbańska and Kowalska, 2019). The term "enrichment" is applied when nutrients are added to a food that already contains them (Sik et al., 2021). In this study, we expected an increased TPC due to FDAP addition to 70% Criollo cocoa dark chocolates. However, this was not the case, perhaps because of the interaction of other components (for example, sugar) with the polyphenols (or their oxidation) by the action of the conching time and temperature (Sulistyowati and Misnawi, 2008; Ziegleder, 2009).

Our experimental results are similar to those reported by Belščak-Cvitanović et al. (2012) and Todorovic et al. (2015). They added 1% red raspberry lyophilized leaf extract to dark chocolates without increasing the chocolates' TPC, although TPC increased at 3%. According to Sim et al. (2016), incorporating 3% of mangosteen pericarp into dark chocolates increased its TPC. These differences in the results found by the above authors and ours suggest that increasing the TPC of chocolates would depend on the fruit's nature and the used concentration. Conversely, the temperature can degrade the procyanidin complex to monomers, thus affecting the overall product TPC (Urbańska and Kowalska, 2019). Polyphenols (both in their oxidized and nonoxidized forms) can form alkaloids, protein, and polysaccharides complexes, causing the Folin-Ciocalteu method to underestimate them when quantifying (Belščak-Cvitanović et al., 2015; Urbańska and Kowalska, 2019).

The chocolate AC comes from the TPC, but the heat affects this property during conching (Sulistyowati and Misnawi, 2008). The results will depend on the time and

**Table 2.** Content of polyphenols, antioxidant capacity, and bioactive compounds of dark chocolates and their ingredients

| Property | Ingredient | | Dark chocolate[2)] | | | |
|---|---|---|---|---|---|---|
| | FDAP | Criollo cocoa liquor[1)] | Control | FDAP1 | FDAP1.5 | FDAP2 |
| TPC (mg GAE/g) | 7.01±1.45$^{b}$ | 9.52±1.34$^{ab}$ | 10.96±0.92$^{a}$ | 11.23±1.10$^{a}$ | 11.41±1.79$^{a}$ | 11.93±1.03$^{a}$ |
| DPPH $IC_{50}$ (mg/mL) | 21.27±4.46$^{a}$ | 5.98±3.17$^{b}$ | 5.44±3.59$^{b}$ | 5.19±1.73$^{b}$ | 9.97±2.56$^{b}$ | 5.93±2.10$^{b}$ |
| Epicatechin (mg/g) | 0.04±0.01$^{c}$ | 0.48±0.07$^{a}$ | 0.34±0.02$^{b}$ | 0.33±0.04$^{b}$ | 0.31±0.06$^{b}$ | 0.23±0.02$^{b}$ |
| Catechin (mg/g) | ND | 0.08±0.01$^{a}$ | 0.06±0.02$^{ab}$ | 0.06±0.01$^{ab}$ | 0.05±0.01$^{ab}$ | 0.04±0.01$^{b}$ |

Values are presented as mean±SEM.
Different letters (a-c) in a row indicate a significant difference at $P<0.05$.
[1)]The properties of the cocoa liquor were measured after the grinding process.
[2)]The properties of the dark chocolates were measured after the tempering process.
FDAP1, FDAP1.5, and FDAP2 are dark chocolates with 1, 1.5, and 2% of freeze dry Arazá pulp, respectively.
TPC, total polyphenols content; GAE, gallic acid equivalent; DPPH $IC_{50}$, 50% of 2,2-diphenyl-1-picrylhydrazyl radical inhibition; ND, not determined.

temperature parameters (Di Mattia et al., 2014). Bolenz and Glöde (2021) used 3.5% grape pomace and a 70∼75°C conching process. They reported that it significantly increased the TPC of milk chocolates. Gültekin-Özgüven et al. (2016) used conching temperatures and time between 60∼80°C and 11∼13 h without increasing the TPC content, suggesting that when using high conching temperatures, it is necessary to use short exposure times. Nevertheless, the information on this topic is limited. Moreover, our study utilized a 20-h conching process at 60°C, and the general total chocolate polyphenols content and the product's AC can increase proportionally (Vertuani et al., 2014; Bolenz and Glöde, 2021). The $IC_{50}$ value indicates the sample size that can inhibit 50% of the DPPH radical. Thus, the analyzed samples denote that FDAP has the lowest AC (21.27±4.46 mg/mL DPPH $IC_{50}$) due to its low TPC. These AC values are also lower than those of cocoa liquor and chocolate FDAP1, FDAP 1.5, and FDAP2 (5.19±1.73, 9.97±2.56, and 5.93±2.10 mg/mL DPPH $IC_{50}$, respectively). No significant difference was observed in the AC of the FDAP1, FDAP1.5, FDAP2, and the control. However, they all have lower AC than the 70% Madagascar cocoa chocolates (0.36±0.02 mg/mL of DPPH $IC_{50}$) (Vertuani et al., 2014).

Catechin and epicatechin are the main bioactive compounds in dark chocolate (Gültekin-Özgüven et al., 2016) and correlate with the AC (Wan et al., 2001; Todorovic et al., 2015). Regarding the bioactive compounds, cocoa liquor has epicatechin and catechin contents (0.48±0.07 and 0.08±0.01 mg/g, respectively), higher than all chocolate samples, meaning that it could be degraded, possibly due to the excessive conching time (20 h), which could not be recovered by incorporating FDAP in the chocolates (Table 2). The loss of these essential molecules can be compensated for by incorporating high-flavonoid ingredients, as Martini et al. (2018) demonstrated. In their study, increased epicatechin in dark chocolates was related to Sakura green tea leaves extract incorporation. Their results are contrary to ours, perhaps because the FDAP does not contain the necessary amounts of epicatechin and catechin to enrich the chocolates. In our study, it seems that conching reduced the chocolate's functional properties, demonstrated by reducing TPC, AC, epicatechin, and catechin. These results agree with those reported by Di Mattia et al. (2014), Bordiga et al. (2015), and Pedan et al. (2017). The conching parameters (Sulistyowati and Misnawi, 2008) may be responsible for reducing the evaluated parameters, as evidenced by the discrepancies in the TPC and AC values of the cocoa liquor and the control chocolate. Thus, it is crucial to deeply understand the cocoa beans' TPC and their transformations during the manufacturing process since its AC depends on it (Pedan et al., 2017). The TPC of chocolates may be improved by incorporating a higher FDAP percentage (≥3%), but at the risk that the consumer may not like it due to the FDAP's high acidity.

Studies conducted in Brazilian Arazá report considerable concentrations of quercetin (5.1 mg/100 g), myricetin (17.0 mg/100 g), and kaempferol (3.7 mg/100 g) (Neri-Numa et al., 2013). Although the FDAP bioactive compounds were unidentified, we assume that the peaks labeled ×1 and ×2 (Fig. 1C) correspond to some of these compounds. Likewise, the peaks ×3 and ×4 (Fig. 1D) in chocolates were unidentified; further studies are necessary. Fig. 1 shows the chromatograms obtained using UHPLC-DAD for the analyzed dark chocolates; only epicatechin and catechin contents were found in the samples. The standard chromatogram peaks showed a good separation at 280 nm (Fig. 1A). The identification using UHPLC-DAD of ×1 and ×2 labeled peaks in the FDAP (Fig. 1B) and ×3 and ×4 peaks in the cocoa liquor (Fig. 1C) and chocolate A (Fig. 1D) was not found.

**Physical analysis of dark chocolates**

One of the main quality parameters of chocolate texture is hardness (Bahari and Akoh, 2018; Toker et al., 2018). Incorporating FDAP did not affect the hardness values (5.33±2.05∼6.07±2.67 kg) (Table 3). No statistical differences in dark chocolate hardness were observed, differing from those found by Biswas et al. (2017) and Limbardo et al. (2017). It is possible that conching favored coating chocolate particles significantly with cocoa fat making the chocolate more compact. Furthermore, the FDAP percentages and 20-h conching did not affect the hardness (Augusto et al., 2019). Casson is the standard model approved (by the IOCCC) to analyze chocolate rheology (Afoakwa, 2010; Fernandes et al., 2013).

The initiation of the chocolate flow is denoted through the Casson yield stress value (Afoakwa et al., 2007; Ahmed, 2017), while the Casson plastic viscosity represents the required energy for maintaining this flow (Afoakwa et al., 2007; Biswas et al., 2017; Bahari and Akoh, 2018). In this work, the FDAP increased the Casson yield stress in chocolate FDAP1, FDAP1.5, and FDAP2 compared with the control chocolate (16.16±0.57 Pa), and significant effects were found between them. Significant differences in the viscosity parameter were found, the values were lower in the chocolate FDAP1, FDAP1.5, and FDAP2 compared with the control (2.15±0.02 Pa·s). Due to the FDAP proportions incorporated into the chocolate, FDAP1, FDAP1.5, and FDAP2 did not affect their flow behavior. According to Beckett (2008), our results are within the established range for chocolates and coincide with the values reported by Glicerina et al. (2016), Bahari and Akoh (2018), and Lončarević et al. (2018) for blackberry-enriched white chocolates. According to Bolenz and Glöde (2021), we could establish the hypothesis that the behavior of these rheological parameters in choco-

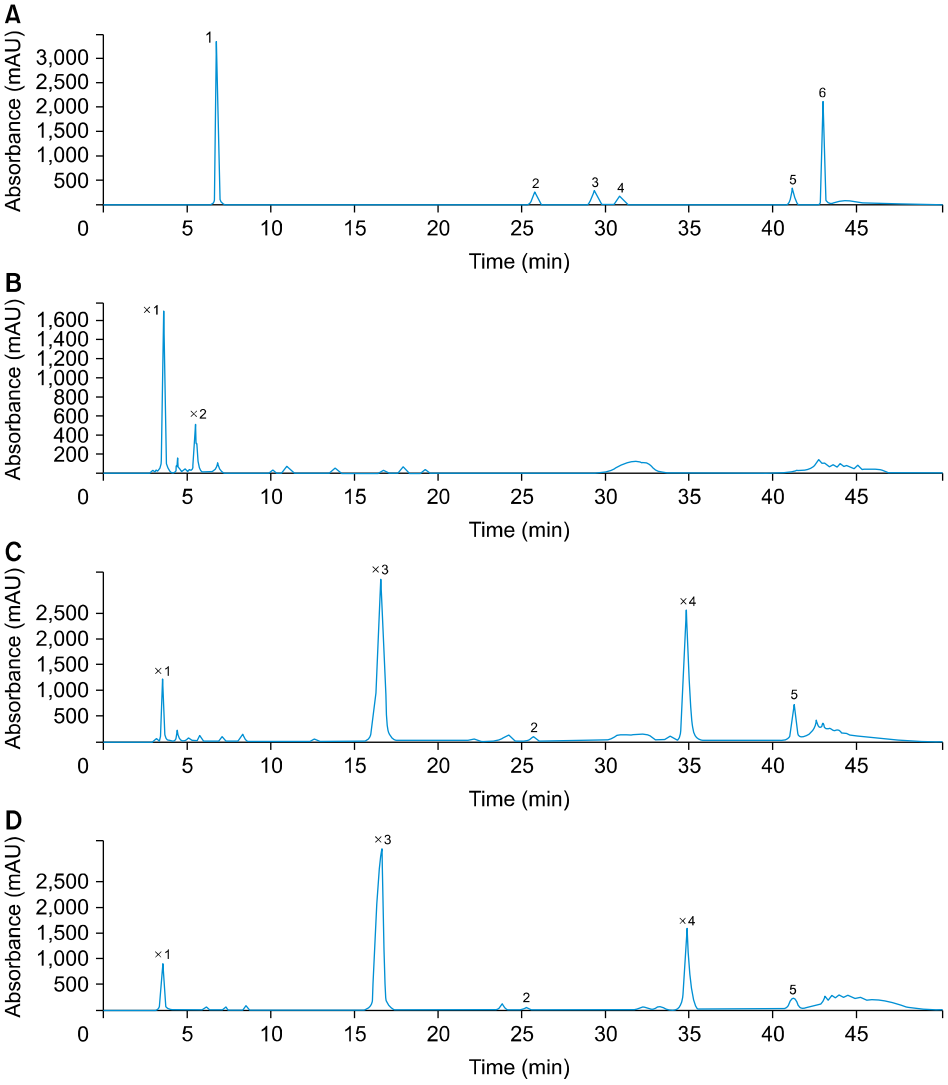


**Fig. 1.** Chromatograms of standards of phenolic compounds (A) and those found in freeze-dried Arazá pulp (FDAP) (B), cocoa liquor (C), and chocolate FDAP1 (D). The peaks belong to the phenolic compounds that were searched for in the samples: gallic acid (1), catechin (2), chlorogenic acid (3), caffeic acid (4), epicatechin (5), and *p*-coumaric acid (6). Peaks labeled with × were not identified with ultra-high-performance liquid chromatographic-diode array. FDAP1, dark chocolate with 1% of freeze dry Arazá pulp.

**Table 3.** Physical properties of dark chocolates

| Physical property | Dark chocolate | | | |
|---|---|---|---|---|
| | Control | FDAP1 | FDAP1.5 | FDAP2 |
| Texture (kg) | 5.67±1.75$^{a}$ | 5.33±2.05$^{a}$ | 6.07±2.67$^{a}$ | 5.50±2.96$^{a}$ |
| Casson yield stress (Pa) | 16.16±0.57$^{b}$ | 18.04±0.97$^{ab}$ | 19.67±1.35$^{a}$ | 19.63±1.16$^{a}$ |
| Casson plastic viscosity (Pa·s) | 2.15±0.02$^{a}$ | 1.80±0.02$^{ab}$ | 1.68±0.03$^{b}$ | 2.11±0.34$^{ab}$ |
| Particle size, D90 (μm) | 15.71±0.56$^{c}$ | 17.71±1.26$^{c}$ | 29.26±5.14$^{b}$ | 67.84±3.64$^{a}$ |
| Moisture (%) | 0.48±0.14$^{a}$ | 0.33±0.03$^{a}$ | 0.31±0.08$^{a}$ | 0.41±0.04$^{a}$ |

Values are presented as mean±SEM.
Different letters (a-c) in a row indicate a significant difference at $P<0.05$.
FDAP1, FDAP1.5, and FDAP2 are dark chocolates with 1, 1.5, and 2% of freeze dry Arazá pulp, respectively.

late FDAP1, FDAP1.5, and FDAP2 (Table 3) was because the cocoa butter did not sufficiently cover the FDAP particles or maybe even agglomerated during the process.

The D90 parameter is commonly used in the chocolate industry to indicate the large particle proportion found in chocolates ( Beckett, 2008; Afoakwa, 2010). The PS of the chocolates depends on the refining/conching time (Saputro et al., 2019) and the refiner type (Rohm et al., 2018). Zhao et al. (2018) employed water as a dispersion medium to measure the particle size of the previously defatted chocolates, obtaining D90 values of 18.33±0.19 μm for chocolates with 10 h of conching. Lim et al. (2021) used isopropanol as a dispersion medium for the chocolate particles made with different sweeteners, obtaining D90 values between 35.23±1.45 and 44.90±3.50 μm. Table 3 shows the PS of all chocolates, measured as a D90 value (90% of the particles size are lesser than this rate). The PS increased when adding a higher FDAP percent-

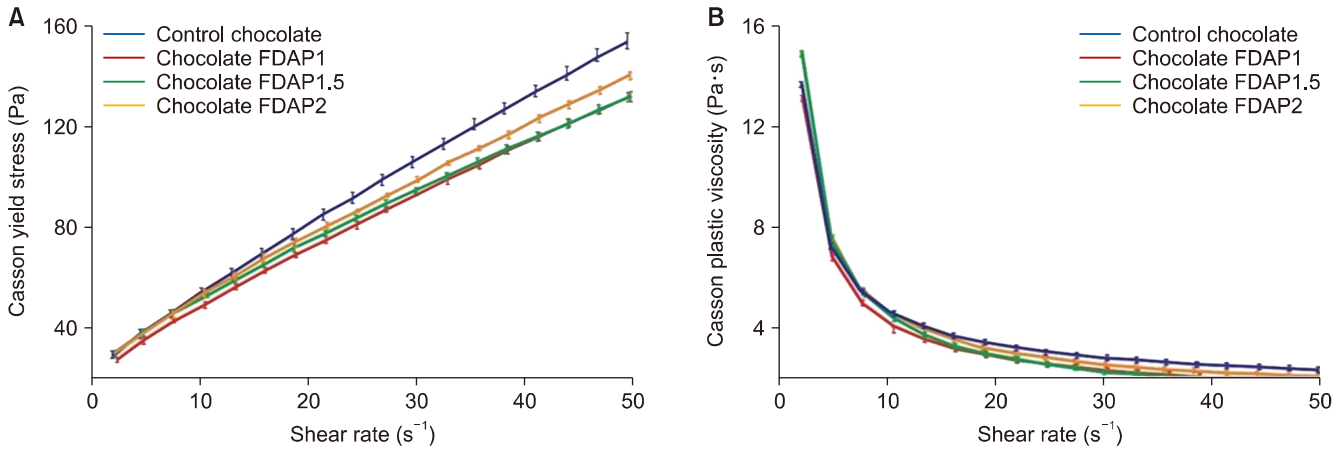


**Fig. 2.** Casson yield stress (A) and Casson plastic viscosity (B) of dark chocolates. FDAP1, FDAP1.5, and FDAP2 are dark chocolates with 1, 1.5, and 2% of freeze dry Arazá pulp, respectively.

age. PS was according to Belščak-Cvitanović et al. (2012), Rohm et al. (2018), and Bolenz and Glöde (2021). Beckett (2008) established a range of 18 μm for fine particles and 50 μm for coarse ones. Thus, the PS of the chocolate control, FDAP1, and FDAP1.5 were below this range. Table 3 also shows that the moisture of the chocolates were not significantly different. Knowing that excessive moisture in the chocolates can cause agglomeration of particles, affecting their thickness and flow behavior (Rodriguez Furlán et al., 2017; Saputro et al., 2017; Ibrahim et al., 2020), we can affirm that the chocolate moisture was adequate because the rheological parameters were unaffected.

The chocolate's quality and stability are linked to the flow behavior (Toker et al., 2019). The flow curve in Fig. 2 shows that the chocolates have a non-Newtonian fluid behavior, where the Casson plastic viscosity decreases with an increasing shear rate. However, the opposite occurs with the Casson yield stress. These results coincide with those reported by Afoakwa (2010), Glicerina et al. (2016), Biswas et al. (2017), and Toker et al. (2018). In our study, like the results by Acan et al. (2021) in the chocolate flow behavior formulated with grape pomace, the Casson yield stress values increased with FDAP incorporation (Fig. 2A). The Casson plastic viscosity's behavior in the chocolates' flow curve shows no changes due to FDAP incorporation (Fig. 2B).

We can conclude that the chocolates produced in this study complied with the rheological parameters and texture required by commercial chocolate. Changes were only seen in the Casson plastic viscosity, Casson yield stress, and particle size. Furthermore, incorporating FDAP in concentrations <2% did not improve the functional properties of 70% Criollo cocoa dark chocolates. Therefore, we suggest that it is feasible to increase the functional characteristics of dark chocolates by increasing FDAP concentrations. We also believe that further studies are needed to identify and quantify the ×1, ×2, ×3, and ×4 peaks since they appear in considerable proportions.

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## AUTHOR DISCLOSURE STATEMENT

The authors declare no conflict of interest.

## AUTHOR CONTRIBUTIONS

Conceptualization, methodology, formal analysis, writing-review & editing: EMCA, ISCC. Resources, supervision, reviewing and editing: CRBZ, LDMA. Data curation, visualization, methodology, software: RJRP, ERO, MMM, LTV.

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