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Improved extraction of phenolic compounds from fruits, leaves, and stems of *Ugni candollei* B. and *Ugni molinae* T. using pressurized aqueous glycerol

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ABSTRACT

This study explores the polyphenolic composition and antioxidant potential of fruits, leaves, and stems from two Chilean murta species: *Ugni molinae* Turcz (red murta) and *Ugni candollei* Barm (white murta). While the fruits have been previously studied, limited information exists on the phytochemical content of leaves and, in particular, stems, representing a significant knowledge gap. To address this, polyphenols were extracted using pressurized liquid extraction (PLE) with an aqueous glycerol mixture and compared to conventional maceration with aqueous acetone. Spectrophotometric analysis showed that leaves yielded the highest extractable polyphenols (EPP; up to 131 mg GAE/g dried extract) *via* PLE, followed by stems and fruits. Antioxidant capacity, evaluated by DPPH and ORAC assays, was strongly correlated with EPP (Spearman coefficients: 0.870 and 0.818), with red murta leaf extracts exhibiting 90 % and 65 % higher antioxidant capacity than stems and fruits, respectively. UHPLC-ESI-MS-QTOF identified 37 phenolic compounds with organ-specific distribution: flavonols and anthocyanins in fruits, myricetin and tannins (corilagin) in leaves, and catechin and dihydroflavonols in stems. Notably, murta stems previously uncharacterized proved to be valuable sources of unique polyphenols that complement those found in fruits and leaves. PLE significantly improved the extraction of certain bioactive compounds from leaves and stems (*e.g.*, quinic acid 71 %, gallic acid 33 %, catechin 13 %), while conventional maceration was more effective for selected fruit compounds (e.g., quercetin-3-*O*-glucuronide 37 %). These findings expand the phytochemical knowledge of *Ugni* spp. and support the valorization of underutilized plant parts as functional ingredients for food and nutraceutical applications.

1. Introduction

Polyphenols are secondary plant metabolites with a chemical structure based on aromatic rings and hydroxyl groups. These compounds accumulate in the outer layers of plant material due to herbivore exposure and on seed surfaces to protect their reproductive processes (Guo et al., 2024). But, in a human health context, polyphenols are relevant because they have shown the ability to decrease the risk and modulate various chronic disorders, such as cardiovascular diseases or type 2 diabetes, through a combination of mechanisms of action, such as the ability to stimulate endogenous antioxidant enzymes or beneficial effects on gut microbiota (Di Lorenzo et al., 2021). They are classified into flavonoids (with several sub-categories, such as flavanols and flavonols) and non-flavonoids (phenolic acids, stilbenes, lignans, and others).

There is increasing interest in sustainable technologies for obtaining functional food ingredients, with polyphenols playing a key role due to their health benefits. Among these techniques, pressurized liquid extraction (PLE) stands out as an environmentally friendly, sustainable, and efficient alternative to conventional methods. PLE operates at high temperatures (323–473 K) and pressures (10–15 MPa), enhancing solvent diffusion and polyphenol solubility by reducing viscosity and

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surface tension. These conditions improve mass transfer and extraction yields. PLE's advantages—greater efficiency, selectivity, and lower solvent use—support circular economy principles by valorizing food waste. It has been successfully applied to recover anthocyanins from blackberries and *Morus nigra* L. leaves, flavonols and phenolic acids from goji berries (*Lycium barbarum L.*), and several phenolic compounds from blackberry (*Rubus* spp.) and blueberry (*Vaccinium myrtillus L.*) bagasse (Fraguela-Meissimilly et al., 2023; Li et al., 2022; Višnjevec et al., 2024).

Murta, *Ugni molinae* Turcz*., and Ugni candollei* Barm are Chilean plants from the *Myrtaceae* family. They are native to the edges of the forests of the coastal mountain range of The Andes. The fruits are edible and have a pleasant aroma; hence, they are consumed fresh or in derived products. The fruits and leaves of murta have traditionally been valued in ethnobotanical medicine for different properties. Studies developed during the last decades have shown the *in vitro* ability of murta extracts to inhibit several digestive enzymes (Escobar-Beiza et al., 2023; Rubilar et al., 2011). Extracts of murta leaves have been incorporated into cosmetic products due to their antioxidant capacity, which helps prevent premature skin aging. This set of properties highlights the potential of murta berries and leaves in food, health, and personal care.

The biological properties of murta are derived from their high content of phenolic compounds and antioxidant capacity (Castro et al., 2021; Pirce et al., 2021). Thus, several studies on different murta berries and leaf ecotypes have shown that phenolic acids, anthocyanins, and flavonoid glycosides, predominantly flavonols, are prevalent in these species. The principal polyphenols identified in murta berries include glucosides of myricetin, quercetin, cyanidin, and peonidin, as well as gallic acid, catechin, rutin, and kaempferol. Meanwhile, murta leaves are rich in gallic acid, catechin, *p*-coumaric acid, rutin, and ellagic acid (Junqueira-Gonçalves et al., 2015; Peña-Cerda et al., 2017; Vega-Galvez et al., 2020). In these studies, murta extracts were obtained through conventional solid-liquid extraction techniques, such as maceration with organic solvents, and only one previous publication explored using PLE to recover polyphenols from white murta (*Ugni candollei*) berries (Fuentes-Jorquera et al., 2024). Also, previous investigations have characterized the polyphenolic profiles of white and red murta fruit extracts (Ah-Hen et al., 2018; Avello et al., 2016), but a comprehensive comparative analysis of polyphenolic constituents across different anatomical structures of both white and red murta plant varieties remains absent from scientific literature.

Notably, the polyphenolic composition of white murta (*Ugni molinae* B.) stems has not been previously reported, highlighting a significant gap in the current understanding of the species' complete phytochemical profile. This lack of data underscores the importance of conducting comprehensive studies.

The present study hypothesizes that PLE using an aqueous glycerol mixture is more efficient than conventional acetone extraction in recovering phenolic compounds from the fruits, leaves, and stems of two varieties of Chilean murta (*Ugni molinae* T. and *Ugni candollei* B.). The main objective is to obtain polyphenol-rich extracts from different anatomical parts of this native species. Specifically, the study aims to: (i) compare the efficiency of PLE and conventional extraction methods in terms of extractable polyphenol yield; (ii) evaluate the correlation between polyphenol content and antioxidant capacity in the extracts of each plant part; and (iii) identify the predominant polyphenolic compounds of fruits, leaves, and stems of two varieties of Chilean murta.

2. Materials and methods

2.1. Chemicals and reagents

The following chemicals were used in this study and purchased from Merck, Germany: analytical grade, acetone (C_3H_6O), glycerol ($C_3H_8O_3$), methanol (CH_4O), dibasic potassium phosphate (K_2HPO_4), monobasic potassium phosphate (KH_2PO_4), and Folin-Ciocalteu phenol reagent. While the next materials were purchased from Sigma-Aldrich, USA: analytical grade, sodium carbonate (Na_2CO_3), 2,2-diphenyl-1-picrylhydrazyl (DPPH) reagent, (±)-6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) reagent ($C_{14}H_{18}O_4$), fluorescein, 2,2′-Azobis(2-methylpropionamidine) dihydrochloride (AAPH). For HPLC grade: water, formic acid (HCOOH), and acetonitrile (CH_3CN).

2.2. Berries, leaves, and stems of white and red murta

Berries, leaves, and stems of wild white and red murta (*Ugni candollei* B., *Ugni molinae* T.) were collected in the forest of the Nahuelbuta Cordillera in April 2019 from Mahuilque (38°13′13.3″ S 73°15′19.5″ W), Biobio region of southern Chile. The collected material was dry-cleaned, and the fruits were separated and freeze-dried at 193 K (FDT-8620, Operon Co., Ltd., Gimpo, Republic of Korea). Finally, the dried samples were ground to a particle size of 0.5 mm and stored at 253 K until extraction. Table 1 lists the samples studied in this work and their abbreviations.

2.3. Solid-Liquid extraction of phenolic compounds

We compared the efficiency and reliability of the PLE process with those of a standard acetone maceration process. Both types of extraction were conducted using a solid-liquid ratio of 1/10 (*w/v*). All extractions were conducted in triplicate. In the PLE extraction, 10 g of a ground murta sample (from fruits, leaves, or stems) from each variety, white or red, was mixed with 55 g of quartz sand in a 100 mL steel extraction cell that had been previously filled with an additional 50 g of quartz sand. The extraction cell was then introduced into an accelerated solvent extractor (Dionex ASE 150, Thermo Fisher Scientific, Waltham, MA, USA) at 393 K and 10 MPa, using a 30 % *v/v* glycerol-water solution as the solvent. The extraction was conducted in a static cycle of 10 min, followed by rinsing with 100 mL of solvent and purging with pressurized nitrogen, resulting in a total extraction time of 20 min. In maceration extraction, 5 g of a murta sample was extracted in an Erlenmeyer flask previously covered with aluminum foil paper and mixed with 50 mL of a 60 % *v/v* acetone-water solution at 301 K for one hour on a hotplate magnetic stirrer (L32, Labinco BV, Breda, The Netherlands) at 500 rpm. The resulting extract was filtered using filter paper and stored at 253 K until analysis (Erpel et al., 2021; Huamán-Castilla et al., 2021).

2.4. Characterization and analysis of murta extracts

2.4.1. Extractable polyphenols (EPP)

The extractable polyphenols (EPP) were determined following the methodology proposed by Singleton and Rossi (1965). In summary, 3.75 mL of distilled water was combined with 0.5 mL of the murta extract and 0.25 mL of the diluted Folin-Ciocalteu (FC) reagent in a vial (diluted 1/1 with distilled water). Subsequently, 0.5 mL of 10 % *w/v* Na_2CO_3 was added to the mixture. The reaction was conducted in the dark for one hour. Then, the absorbance was measured at 765 nm using a

Table 1
Description and codes for samples included in the study.

	PLE-Glycerol		Maceration-Acetone	
Sample	White murta	Red murta	White murta	Red murta
Fruit	WM-F-G	RM-F-G	WM-F-A	RM-F-A
Leaf	WM-L-G	RM-L-G	WM-L-A	RM-L-A
Stem	WM-S-G	RM-S-G	WM-S-A	RM-S-A

PLE: pressurized liquid extraction. WM-F-G: White murta fruit glycerol extract. WM-L-G: White murta leaf glycerol extract. WM-S-G: White murta stem glycerol extract. RM-F-G: red murta fruit glycerol extract. RM-L-G: red murta leaf glycerol extract. RM-S-G: red murta stem glycerol extract. WM-F-A: White murta fruit acetone extract. WM-L-A: White murta leaf acetone extract. WM-S-A: White murta stem acetone extract. RM-F-A: red murta fruit acetone extract. RM-L-A: red murta leaf acetone extract. RM-S-A: red murta stem acetone extract.

Genesys 150 UV–Vis spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA). All analyses were carried out in triplicate. The results were compared with a gallic acid calibration curve (10–90 mg/L; r^2: 0.9974) and expressed as mg GAE/g dried extract.

2.4.2. Antioxidant capacity

2.4.2.1. DPPH radical method. The antioxidant capacity (AC) was determined using the DPPH method described by Brand-Williams et al. (1995). In summary, a 0.039 mg/mL methanolic DPPH solution was prepared for each analysis conducted. For the analysis, 0.1 mL of murta extract was added and mixed with 3.9 mL of methanolic DPPH solution, and the mixture was homogenized. Blank and control samples were prepared from methanol and the methanolic DPPH solution, respectively. The mixture was then kept in the dark for 30 min to eliminate colour interferences. Subsequently, the absorbance was measured at 520 nm (Genesys 150 UV–Vis, Thermo Fisher Scientific, Waltham, MA, USA) and compared with a Trolox curve (2–9 mg/L). All analyses were performed in triplicate. The results were expressed as mg Trolox equivalents (TE)/g dried extract.

2.4.2.2. Oxygen radical absorbance capacity (ORAC) method. The ORAC assay followed the previously described protocol with some modifications (Huang et al., 2002). In a 96-well microplate, 250 μL of fluorescein at 55 nM, prepared in a 75 mM phosphate buffer (pH 7.4), was added to 25 μL of extract or standard diluted in the same buffer. The microplate was incubated for 30 min at 310 K, and the reaction was initiated by adding 25 μL of a freshly prepared solution of AAPH at 153 mM in phosphate buffer. Fluorescence was measured every minute for 1 h at 310 K using a Synergy HTX multi-mode microplate reader (Biotek Instruments, Winooski, VT, USA), with an excitation wavelength of 485 nm and an emission wavelength of 528 nm. All assays were performed in triplicate. Data analysis, area under the curve (AUC) calculations, standards, and buffer blanks were performed using Gen5 software (Biotek Instruments, Winooski, VT, USA). The net AUC values of samples and standards were calculated as follows:

$$Net\ AUC = AUC\ sample \vee Std. - AUC\ blank \quad (1)$$

The ORAC values of the samples were interpolated from the standard Net AUC Std. vs. Trolox concentration curve (1–10 mg/L) and expressed as mg Trolox equivalents (TE)/ g dried extract.

2.5. Polyphenols profile by UHPLC-ESI-MS-QTOF analysis

The following samples were analyzed: white murta extracts in aqueous acetone (fruit, WM-F-A; stem, WM-S-A), white murta extract in aqueous glycerol (stem, WM-S-G), red murta extract in aqueous acetone (stem, RM-S-A), and red murta extracts in aqueous glycerol (fruit, RM-F-G; leaf, RM-L-G), ensuring a proper representation of the three analyzed parts, the two murta cultivars, and the two extraction procedures. One milliliter of the sample was extracted and transferred to HPLC vials. Chromatographic separation was conducted using a 2.1 mm x100 mm Kinetex C18 column with a particle size of 1.7 μm. The following solvents were employed for metabolomic analysis: Solvent A: Water +0.1 % Formic Acid; Solvent B: 90 % Acetonitrile +0.1 % Formic Acid. The temperature of the column was maintained at 313 K. The samples were maintained at a temperature of 279 K. A 10-μL sample was injected. Mass spectrometry analysis was conducted in positive and negative modes. Data analysis was conducted using Metaboscape 4.0 software (Bruker). Identification was based on a) exact mass (with a maximum error of 10 ppm); b) MS/MS fragmentation pattern, for which the software suggested an MS/MS score based on the spectral libraries of the MassBank of North America (MoNa), in conjunction with experimental spectra. The area was obtained for the peaks with assigned identities.

2.6. Statistical analysis

The Student's *t*-test was employed to compare the two groups to assess significant differences (*p*-value <0.05) between both murta varieties and between extraction methods. The nonparametric Mann-Whitney *U* test was utilized when the data did not exhibit a normal distribution. The nonparametric Spearman coefficient evaluated the correlation between the phenolic content and the antioxidant capacity of the murta extracts. These analyses were conducted using SPSS software version 29.

3. Results and discussion

3.1. Extractable polyphenols and associated antioxidant capacity

3.1.1. Comparative analysis of extractable polyphenolic content across fruits, leaves, and stems

The EPP analysis of extracts revealed substantial differences in the distribution of phenolic compounds among the various anatomical structures of *Ugni molinae* T. (red murta) and *Ugni candollei* B. (white murta) (Fig. 1). In both cultivars, leaves consistently demonstrated higher phenolic accumulation than fruits and stems, yielding 77 % and 67 % more, respectively. Red murta leaf extracts exhibited the highest EPP value (130 mg GAE/g dried extract with the glycerol-PLE method). This preferential accumulation in leaf tissues likely reflects their physiological role in plant defense mechanisms against UV radiation and pathogens, consistent with previous findings (Martemucci et al., 2024).

Red murta consistently demonstrated higher EPP content than white murta in the three parts of the plant: 32 % more in fruits, 34 % more in leaves, and 36 % more in stems. Nevertheless, these differences were statistically significant (*p-value* < 0.05) only for fruits and leaves. The visibly darker pigmentation of red murta may explain these differences, suggesting potential genotypic influences on secondary metabolite biosynthesis pathways. The EPP content of our acetone maceration extract of red murta fruit (28 mg GAE/g dried extract) aligns with those reported by López de Dicastillo et al. (2017), who documented EPP values ranging from 23 to 34 mg GAE/g in ethanolic extracts, obtained at 313 K and atmospheric pressure. However, our results differ from

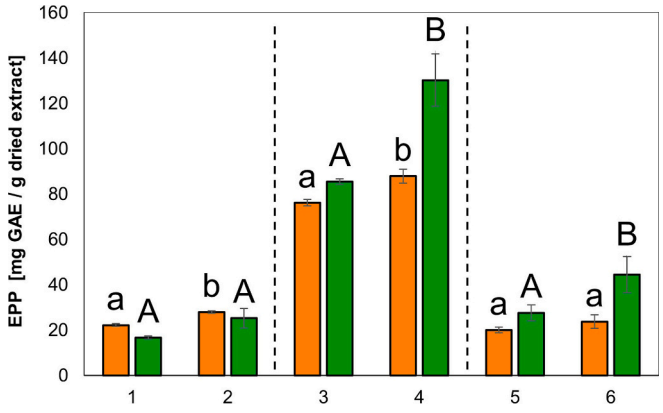

Fig. 1. Extractable polyphenols (EPP) of murta extracts obtained by PLE-glycerol (green) and maceration-acetone (orange) of both murta varieties. WM-F: white murta fruits, RM-F: red murta fruits, WM-L: white murta leaves, RM-L: red murta leaves, WM-S: white murta stems, RM-S: red murta stems. The lowercase letters represent statistically significant differences (*p-value* <0.05) between both murta varieties for the type of sample (fruit, leaf, or stem). The uppercase letters represent statistically significant differences (*p-value* <0.05) between the two types of extraction (maceration or PLE). A Student's *t*-test was used to compare the two groups. When the data did not have a normal distribution, the nonparametric Mann-Whitney *U* test was used. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

those of Alfaro et al. (2013), who observed substantial EPP variations (283–2152 mg GAE/100 g dw) depending on genotype and harvest timing. These extractions were performed using methanol at 303 K and atmospheric pressure. The EPP content of our red murta leaf extracts (130 mg GAE/g dried extract) fell within the range reported by López de Dicastillo et al. (2017) (92–136 mg GAE/g). However, Peña-Cerda et al. (2017) obtained much higher values (260 mg GAE/g) using organic solvents such as hexane, dichloromethane, ethyl acetate, and ethanol at room temperature and atmospheric pressure. These discrepancies likely reflect variations in extraction methodologies, solvent systems, and environmental growth conditions, influencing polyphenolic yields. Notably, our study provides the first comprehensive EPP characterization of white murta (*Ugni candollei* B.) extracts across all plant structures, addressing a significant gap in scientific literature. Additionally, the phenolic content of murta stem extracts has been minimally documented, with only Rubilar et al. (2011) reporting EPP contents of 15.8 and 11.9 mg GAE/g for red murta stems crude and aqueous fractionated extracts, respectively. The crude extracts were obtained by maceration with ethanol (50 % *v/v* in water) at room temperature and atmospheric pressure. The aqueous fractionated extract was obtained by defatting the crude extract with petroleum ether, suspending it in distilled water, adding acetic acid, and mixing the monomeric and oligomeric components. These EPP levels are substantially lower than ours (45 mg GAE/g dried extract), probably due to differences in extraction methodologies and experimental conditions.

3.1.2. Extraction method efficiency: PLE versus conventional maceration

Comparing extraction methodologies revealed that PLE with an aqueous glycerol mixture significantly enhanced polyphenolic recovery by 32 % and 45 % from red murta leaves and red murta stems, compared to conventional maceration with aqueous acetone. The most pronounced difference was observed in red murta leaves, where PLE achieved 32 % higher extraction efficiency than maceration (130 *vs* 88 mg GAE/g dried extract). This substantial improvement can be attributed to PLE's elevated operational temperature (393 K) and pressure (10 MPa), conditions that reduce solvent viscosity and surface tension while increasing the solubility and diffusivity of the target compound (Plaza & Turner, 2015). This extraction efficiency pattern was reversed for fruit samples, where conventional maceration yielded marginally higher EPP values than PLE, though the differences were not statistically significant ($p\text{-value} > 0.05$). This exception may reflect the distinct matrix characteristics of fruit tissues, which may contain more accessible phenolic compounds that require less intensive extraction than those in lignified stem tissues or structurally complex leaf matrices. Our findings regarding PLE's superior extraction efficiency align with previous research by Huamán-Castilla et al. (2021), who reported that PLE with 50 % aqueous glycerol enhanced polyphenolic recovery from grape pomace by 17 % compared to extraction with eutectic solvents. Similarly, Bebek Markovinović et al. (2023) demonstrated that PLE doubled the EPP yield from strawberry fruit compared to conventional extraction methodologies. Despite requiring specialized equipment, the enhanced extraction efficiency of PLE can be attributed to several factors: 1) increased solubility and diffusion at elevated temperatures, 2) high pressure maintaining solvent liquidity and enhancing matrix penetration, and 3) the optimal polarity range of the glycerol-water mixture for diverse phenolic compounds. Additionally, PLE's compatibility with environmentally benign solvents makes it a superior technology for obtaining bioactive compounds from plant matrices (Boateng, 2024; Hernández-Corroto et al., 2020). It is worth considering, though, that the best extraction method depends on the plant tissue and polyphenol family to be recovered. For example, conventional maceration is more effective for recovering anthocyanins and quercetin glycosides from fruit berries, highlighting the need for tailored optimization.

3.1.3. Antioxidant capacity and correlation with extractable polyphenols

The AC assessment using DPPH and ORAC assays revealed patterns that closely mirrored the EPP distribution (Fig. 2), with leaf extracts demonstrating substantially higher antioxidant potential than fruit and stem extracts across both murta varieties. Statistical analysis confirmed a strong positive correlation between EPP and antioxidant capacity, with Spearman correlation coefficients of 0.870 and 0.818 for DPPH and ORAC assays, respectively ($p\text{-value} < 0.001$, the correlation is significant at the 0.01 level, 2-tailed). Moreover, the red murta leaf extract obtained *via* PLE showed the highest AC with the DPPH assay (188 mg TE/g dried extract) and, at the same time, the highest EPP content (130 mg GAE/g dried extract). The highest AC in fruit extracts measured by the ORAC method (69 mg TE/g dried extract) was observed in red murta samples obtained through maceration. Red murta PLE extracts exhibited the highest AC measured using the ORAC method (67 mg TE/g dried extract) in stems. López de Dicastillo et al. (2017) obtained substantially higher AC values, using the DPPH assay, for red murta fruit and leaf extracts, 110 and 361 mg TE/g extract, respectively. This discrepancy may reflect differences in extraction methodologies, plant growth conditions, or harvest timing. Rubilar et al. (2011) observed that stem extracts exhibited approximately half the antioxidant capacity of leaf extracts, a finding consistent with our results, which showed that the AC of stem extracts was 2.6 and 2.9 times lower than that of leaf extracts in the ORAC and DPPH assays, respectively.

It is essential to recognize that while DPPH and ORAC assays offer valuable insights into antioxidant potential, they utilize artificial radical systems that may not accurately simulate physiological conditions (Schaich et al., 2015). Nevertheless, the strong correlation between EPP and AC across both assays reinforces the conclusion that phenolic compounds are primary contributors to the observed antioxidant properties, with potential implications for the nutraceutical and functional food applications of murta extracts. At the same time, it is worth noting that our analysis focused exclusively on extractable polyphenols, and a fraction of non-extractable polyphenols (NEPP) likely remained in the residual plant material. Only a few studies have explored NEPP in murta extracts (Fuentes-Jorquera et al., 2025; López et al., 2017); hence, future studies might benefit from characterizing these non-extractable fractions to provide a more comprehensive understanding of the total phenolic potential of murta biomass.

3.2. Polyphenol profile analysis

3.2.1. Comprehensive characterization of phenolic compounds in Ugni species

UHPLC-ESI-MS-QTOF analysis was performed on a selected subset of samples due to limitations in the amount of extract obtained under certain extraction conditions. To ensure reliable chemical characterization while minimizing additional sources of variability, only samples with sufficient extract volume were included in this analysis. Conducting new extractions would have required additional sample collection, potentially from different seasons and locations, which could have introduced further variability and compromised the comparability of the results. The chromatographic analysis by UHPLC-ESI-MS-QTOF enabled the identification of 37 distinct phenolic compounds across the six selected murta extracts (WM-F-A, RM-F-G, RM-L-G, WM-S-G, WM-S-A, and RM-S-A), revealing significant variations in the polyphenolic fingerprint according to plant structure, murta variety, and extraction methodology (Table 2, Fig. 3). The identified compounds encompassed multiple phenolic classes: phenolic acids (6 compounds, including hydroxybenzoic and hydroxycinnamic acid derivatives), flavonoids (30 compounds distributed among flavanols, flavonols, dihydroflavonols, and anthocyanins), and tannins (1 compound). This diverse phenolic profile suggests a complex biosynthetic pathway in the *Ugni* species that warrants further exploration for potential metabolomic differentiation between varieties. The phenolic acids identified included gallic acid, which was ubiquitous across all analyzed extracts, along with more complex derivatives such as 5-(beta-*D*-glucopyranosyloxy)-2-hydroxybenzoic acid, glucosyringic acid, and 3,4-di-*O*-galloylquinic

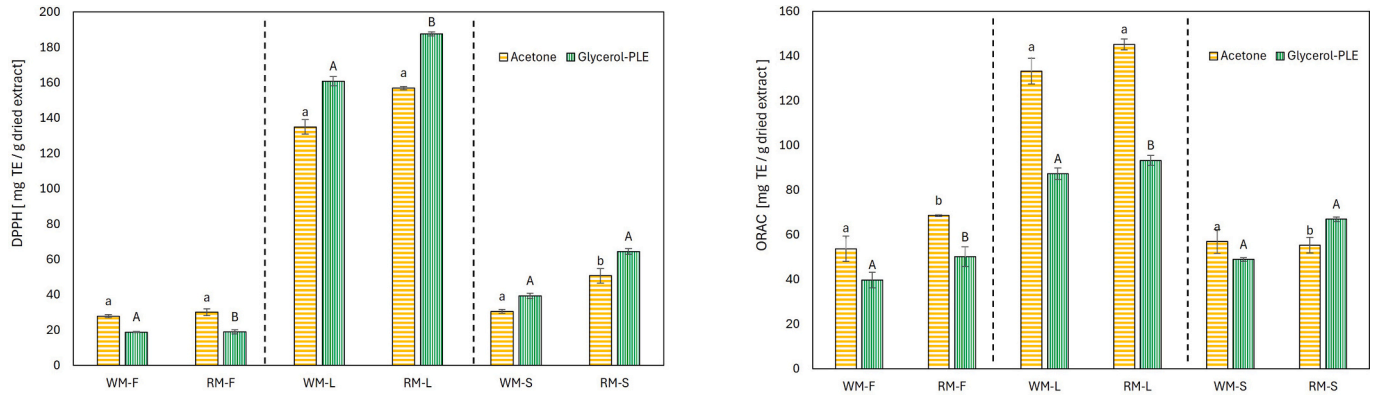

Fig. 2. Antioxidant capacity (AC) of white and red murta extracts obtained by PLE-glycerol (green) and maceration-acetone (orange) for fruits, leaves, and stems. **A**: DPPH assay. **B**: ORAC assay. WM-F: white murta fruits, RM-F: red murta fruits, WM-L: white murta leaves, RM-L: red murta leaves, WM-S: white murta stems, RM-S: red murta stems extracts. The lowercase letters represent statistically significant differences (*p-value* <0.05) between both murta varieties for the type of sample (fruit, leaf, or stem). The uppercase letters represent statistically significant differences (*p-value* <0.05) between the two extraction methods. A student's t-test was used to compare the two groups. When the data did not have a normal distribution, the nonparametric Mann-Whitney U test was used. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

acid. The hydroxycinnamic acid derivatives included quinic acid and chlorogenic acid hemihydrate, which are present in all extracts, consistent with their widespread distribution in the plant kingdom (Narra et al., 2024). The presence of these compounds across all anatomical structures suggests a fundamental role in the plant's physiological functions rather than a structure-specific specialization. Flavonoids constituted the most abundant and diverse group of identified phenolic compounds, with prominence of flavonols and anthocyanins. Among flavonols, quercetin and myricetin glycosides predominate, with quercetin-3-*O*-rhamnoside, quercetin-3-*O*-glucuronide, and myricetin-3-*O*-galactoside detected across all extracts. More complex flavonol derivatives, including 2”-*O*-galloylquercitrin and gallomyricitrin, were exclusively detected in fruit and leaf extracts, suggesting tissue-specific biosynthetic pathways or accumulation patterns. Notably, the anthocyanin profile exhibited marked structure-specificity, with cyanidin 3-*O*-hexoside, peonidin 3-*O*-arabinoside, and petunidin 3-*O*-arabinoside detected in fruit and leaf extracts but absent in stem extracts. Conversely, more complex anthocyanins, including malvidin 3-*O*-(6″-acetyl-hexoside), cyanidin 3-*O*-rutinoside, and peonidin 3-*O*-rutinoside, were identified exclusively in stem extracts obtained by acetone maceration. This distinct anthocyanin distribution pattern represents a novel finding that may reflect the differential regulation of the anthocyanin biosynthetic pathway across plant tissues, suggesting potential evolutionary adaptations for tissue-specific functions (Shene et al., 2009). Finally, the identification of corilagin, an ellagitannin with documented anti-inflammatory and antioxidant properties (Mateș et al., 2024), in red murta leaf extracts (RM-L-G) and white murta stem extracts (WM-S-G and WM-S-A) constitutes a significant novel finding, as this compound had not been previously reported in *Ugni* species. The presence of corilagin in these extracts may enhance their bioactive potential beyond general AC.

3.2.2. Characteristic distribution patterns and the influence of the extraction methods on phenolic profile and compound recovery

Analysis of the identified polyphenols revealed a distinctive accumulation of phenolic compounds in the three studied plant structures, with significant implications for their potential bioactivity and applications (Fig. 3). Relative abundances (RA) were calculated based on the total absorbance (areas) of the 12 polyphenols identified in each extract, shown in Table 3. Table S1, in the supplementary material, shows the areas of all phenolic compounds identified in murta extracts. The predominant compounds in the fruit extracts varied significantly between extraction methodologies and varieties. Following analysis by compound family and extraction method, it was determined that the PLE method was superior to maceration for acids. In general terms, the red murta leaves exhibited a higher level of quinic acid (71 % RA in RM-L-G extracts) than the fruits and stems. The highest concentration of gallic acid was found in the white murta stems (33 % RA in WM-S-G extracts), which is consistent with what has been reported before on PLE of these compounds (Huamán-Castilla et al., 2021). Regarding flavonols, the quercetin glycosides, quercetin 3-*O*-galactoside and quercetin-3-*O*-glucuronide, were found in white murta fruit extracts (maceration with acetone), with 27 % and 37 % of RA, respectively. In contrast, the flavonol, myricetin 3-*O*-rhamnoside, was found mainly in the red murta leaves (10 % RA in RM-L-G). In addition, the dihydroflavonol, dihydroquercetin 3-*O*-rhamnoside, was present in significant quantities in red murta stem extracts obtained by maceration (47 % RA). This preferential accumulation of dihydroflavonols in stem tissues represents a novel finding that may reflect their role in lignification processes or defensive functions in woody tissues. White murta stem extracts obtained by glycerol-PLE (WM-S-G) exhibited higher flavanols (+)-catechin and (−)-epicatechin content than fruits and leaves, with 12.8 % and 9.7 % of RA, respectively. Regarding the anthocyanin group, a wide variety of these compounds were found in both fruit extracts (WM-F-A and RM-F-G), with PLE yielding higher amounts than the conventional method. As previously mentioned, the high levels of flavonols, particularly quercetin glycosides (quercetin 3-*O*-galactoside and quercetin 3-*O*-glucuronide) and anthocyanins likely contribute to the documented high antioxidant capacity of murta fruits, and align with their traditional ethnopharmacological applications (Avello et al., 2013). Finally, it is worth noting the presence, albeit in low amounts, of the ellagitannin corilagin in red murta leaf extracts by glycerol-PLE (1 % RA). The phenolic compounds observed across anatomical structures also suggest accumulation and biosynthetic regulation patterns specific to each structure, which may reflect their distinct physiological functions within the plant.

4. Conclusions

This study demonstrated phenolic compounds' differential distribution and extraction efficiency across distinct anatomical structures of *Ugni molinae* T. and *Ugni candollei* B. Red murta leaves exhibited the highest polyphenolic concentration when extracted using PLE with aqueous glycerol, following a consistent pattern of extractable phenolic content (leaves > stems > fruits) across both varieties. PLE consistently outperformed conventional maceration, increasing the recovery of bioactive compounds from leaves and stems by up to 32 %. Antioxidant capacity, assessed via DPPH and ORAC assays, confirmed the

Table 2
Identification of phenolic compounds by UHPLC-ESI-MS-QTOF present in white and red murta extracts in fruit, leaves, and stems.

ID	Family/Class	Identification	Molecular Formula	MS/MS score (%)	Molecular ion (*m/z* theo)	Molecular ion (*m/z* obs)	Error (ppm)					
							WM-F-A	RM-F-G	RM-L-G	WM-S-G	WM-S-A	RM-S-A
	Phenolic acids											
1	*Hydroxybenzoic acids*	Gallic acid	$C_7H_6O_5$	100	169.0143	169.0140	1.54	1.54	1.54	1.54	1.54	1.54
2		5-(beta-D-glucopyranosyloxy)-2-hydroxybenzoic acid	$C_{13}H_{16}O_9$	83	315.0722	315.0719	n.d.	0.76	0.76	0.76	0.76	0.76
3		Glucosyringic acid	$C_{15}H_{20}O_{10}$	85	359.0984	359.1010	n.d.	n.d.	−7.32	−7.32	−7.32	−7.32
4		3,4-di-O-galloylquinic acid	$C_{21}H_{20}O_{14}$	76	495.0780	495.0819	n.d.	−7.72	−7.72	n.d.	n.d.	n.d.
5	*Hydroxycinnamic acids*	Quinic acid	$C_7H_{12}O_6$	96	191.0561	191.0568	−3.82	−3.82	−3.82	−3.82	−3.82	−3.82
6		Chlorogenic acid hemihydrate	$C_{16}H_{20}O_{10}$	98	371.0984	371.0985	−0.40	−0.40	−0.40	−0.40	−0.40	−0.40
	Flavonoids											
7	*Flavanols*	(+)-Catechin	$C_{15}H_{14}O_6$	82	289.0718	289.0721	−1.04	−1.73	−1.73	−1.04	−1.04	−1.73
8		(−)-Epicatechin	$C_{15}H_{14}O_6$	99	289.0718	289.0721	−1.73	n.d.	−1.04	−1.04	−1.04	−1.04
9	*Flavone*	Scutellarein-7-O-glucoside/ Plantaginin	$C_{21}H_{20}O_{11}$	100	447.0933	447.0904	6.46	6.46	n.d.	n.d.	n.d.	n.d.
10	*Flavonols*	Quercetin-3-O-arabinoside	$C_{20}H_{18}O_{11}$	99	433.0776	433.0780	−0.95	−0.95	−0.95	n.d.	n.d.	n.d.
11		Quercetin-3-O-rhamnoside / Quercitrin	$C_{21}H_{20}O_{11}$	97	447.0933	447.0904	6.46	6.46	6.46	6.46	n.d.	6.46
12		Myricetin-3-O-xyloside	$C_{20}H_{18}O_{12}$	97	449.0726	449.0759	−7.48	n.d.	−7.48	n.d.	−7.48	−7.48
13		Myricetin 3-O-rhamnoside/ Myricitrin	$C_{21}H_{20}O_{12}$	99	463.0882	463.0880	0.35	0.35	0.35	n.d.	0.35	0.35
14		Quercetin 3-galactoside/ Hyperin	$C_{21}H_{20}O_{12}$	99	463.0882	463.0882	0.11	0.11	n.d.	0.11	0.11	0.11
15		Quercetin-3-O-glucuronide	$C_{21}H_{18}O_{13}$	99	477.0675	477.0678	−0.75	−0.75	−0.75	−0.75	−0.75	−0.75
16		Myricetin-3-galactoside	$C_{21}H_{20}O_{13}$	99	479.0831	479.0848	−3.55	−3.55	−3.55	−3.55	−3.55	−3.55
17		2''-O-Galloylquercitrin/ quercitrin	$C_{28}H_{24}O_{15}$	89	599.1042	599.1063	−3.42	−3.42	−3.42	n.d.	n.d.	n.d.
18		Gallomyricitrin / 2''-O-galloylmyricitrin	$C_{28}H_{24}O_{16}$	96	615.0992	615.1045	−8.65	n.d.	−8.65	n.d.	n.d.	n.d.
19		6-O-p-Coumaroyl-1,2-di-O-galloyl-b-D-glucopyranose	$C_{29}H_{26}O_{16}$	80	629.1148	629.1162	−2.24	−2.24	−2.24	n.d.	n.d.	n.d.
20		[(2*R*,3*R*,4*S*,5*R*,6*S*)-6-[5,7-dihydroxy-4-oxo-2-(3,4,5-trihydroxyphenyl)chromen-3-yl]oxy-3,4,5-trihydroxyoxan-2-yl]methyl 3,4,5-trihydroxybenzoate	$C_{28}H_{24}O_{17}$	86	631.0941	631.0987	−7.34	n.d.	−7.34	n.d.	−7.34	−7.34
21	*Dihydroflavonols*	Dihydroquercetin 3-O-rhamnoside/ Taxifolin 3-O-rhamnoside	$C_{21}H_{22}O_{11}$	93	449.1089	449.1090	−0.04	−0.04	n.d.	−0.04	−0.04	−0.04
22		Taxifolin-3-glucoside /Dihydroquercetin-3-O-glucoside	$C_{21}H_{22}O_{12}$	96	465.1039	465.1043	−1.05	−1.05	−1.05	n.d.	n.d.	n.d.
23	*Anthocyanins*	Cyanidin	$C_{15}H_{11}O_6$	99	287.0550	287.0548	0.73	n.d.	n.d.	n.d.	n.d.	n.d.
24		Pelargonidin 3-O-hexoside	$C_{21}H_{21}O_{10}$	99	433.1129	433.1129	n.d.	n.d.	n.d.	n.d.	0.05	n.d.
25		Delphinidin 3-O-pentoside	$C_{20}H_{19}O_{11}$	99	435.0922	435.0917	n.d.	n.d.	1.13	n.d.	n.d.	n.d.
26		Cyanidin 3-O-hexoside	$C_{21}H_{21}O_{11}$	99	449.1078	449.107	1.87	1.87	1.87	n.d.	n.d.	n.d.
27		Peonidin 3-O-arabinoside	$C_{21}H_{21}O_{11}$	99	449.1078	449.1077	0.31	0.31	0.31	n.d.	n.d.	n.d.
28		Petunidin 3-O-arabinoside	$C_{21}H_{21}O_{11}$	99	449.1078	449.1118	−8.82	−8.82	−8.82	n.d.	n.d.	n.d.
29		Peonidin 3-O-hexoside/ Malvidin 3-O-arabinoside	$C_{22}H_{23}O_{11}$	99	463.1235	463.1238	n.d.	−0.67	n.d.	n.d.	n.d.	n.d.
30		Delphinidin 3-O-(6''-acetyl-hexoside)	$C_{23}H_{23}O_{13}$	99	507.1133	507.1164	n.d.	n.d.	n.d.	n.d.	−6.07	n.d.
31		Malvidin 3-O-(6''-acetyl-hexoside)	$C_{25}H_{27}O_{13}$	99	535.1446	535.1453	n.d.	n.d.	n.d.	n.d.	−1.27	−1.27
32		Pelargonidin 3-O-rutinoside	$C_{27}H_{31}O_{14}$	99	579.1708	579.1738	n.d.	n.d.	−5.13	n.d.	n.d.	n.d.
33		Cyanidin 3-O-(6''-p-coumaroyl-glucoside)	$C_{30}H_{27}O_{13}$	99	595.1446	595.1477	n.d.	n.d.	n.d.	n.d.	n.d.	−5.18
34		Cyanidin 3-O-rutinoside	$C_{27}H_{31}O_{15}$	99	595.1658	595.1614	n.d.	n.d.	n.d.	n.d.	7.31	7.31
35		Peonidin 3-O-rutinoside	$C_{28}H_{33}O_{15}$	99	609.1814	609.1783	n.d.	n.d.	n.d.	n.d.	5.09	7.31
36		Cyanidin 3-O-(6''-caffeoyl-glucoside)/Delphinidin 3-O-(6''-p-coumaroyl-glucoside)	$C_{30}H_{27}O_{14}$	99	611.1395	611.1389	1.03	n.d.	n.d.	n.d.	n.d.	n.d.
	Tannins											
37	*Ellagitannins*	Corilagin	$C_{27}H_{22}O_{18}$	92	633.0806	633.0776	n.d.	n.d.	4.79	4.79	4.79	n.d.

Ionization was performed in negative for all classes, but anthocyanins were detected with positive ionization. WM-F-A: White murta fruit acetone extract. RM-F-G: red murta fruit glycerol extract. RM-L-G: red murta leaf glycerol extract. WM-S-G: White murta stem glycerol extract. WM-S-A: White murta stem acetone extract. RM-S-A: red murta stem acetone extract. n.d.: non-detected.

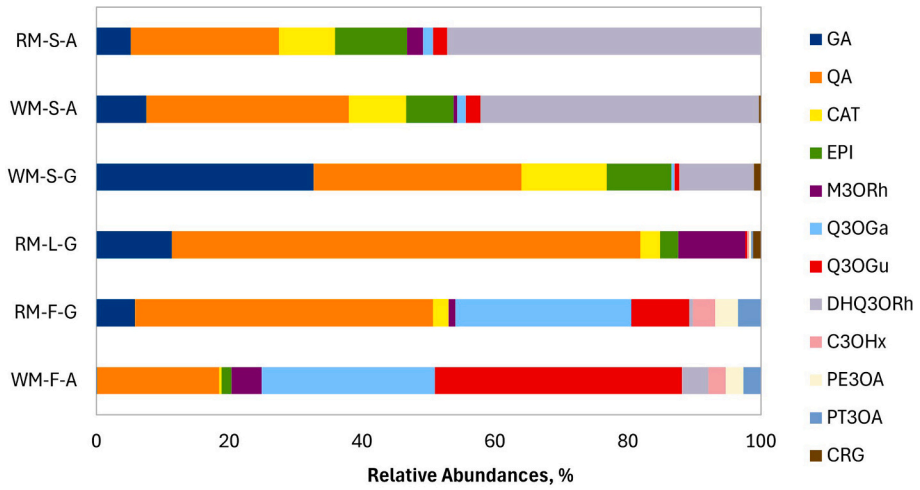

Fig. 3. Relative abundances (RA) of polyphenols in murta extracts. WM-F-A: White murta fruit acetone extract. RM-F-G: red murta fruit glycerol extract. RM-L-G: red murta leaf glycerol extract. WM-S-G: White murta stem glycerol extract. WM-S-A: White murta stem acetone extract. RM-S-A: red murta stem acetone extract. GA: gallic acid. QA: quinic acid. CAT: (+)-catechin. EPI: (−)- epicatechin. M3ORh: myricetin 3-*O*-rhamnoside. Q3OGa: quercetin 3-*O*-galactoside. Q3OGu: quercetin 3-*O*-glucuronide. DHQ3ORh: dihydroquercetin 3-*O*-rhamnoside. C3OHx: cyanidin 3-*O*-hexoside. PE3OA: peonidin 3-*O*-arabinoside. PT3OA: petunidin 3-*O*-arabinoside. CRG: corilagin. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 3
Relative abundances of the major phenolic compounds present in the murta extracts.

	RELATIVE ABUNDANCES, %											
Sample	GA	QA	CAT	EPI	M3ORh	Q3OGa	Q3OGu	DHQ3ORh	C3OHx	PE3OA	PT3OA	CRG
WM-F-A	0.1	18.4	0.3	1.5	4.5	26.1	37.2	3.9	2.6	2.6	2.6	0
RM-F-G	5.8	44.8	2.4	0	1.0	26.5	8.7	0.5	3.4	3.4	3.4	0
RM-L-G	11.3	70.5	3.0	2.7	10.1	0	0.2	0	0.3	0.3	0.3	1.2
WM-S-G	32.7	31.3	12.8	9.7	0	0.5	0.7	11.3	0	0	0	1.0
WM-S-A	7.5	30.5	8.6	7.1	0.5	1.3	2.2	41.9	0	0	0	0.3
RM-S-A	5.2	22.2	8.5	10.8	2.4	1.5	2.1	47.2	0	0	0	0

WM-F-A: White murta fruit acetone extract. RM-F-G: red murta fruit glycerol extract. RM-L-G: red murta leaf glycerol extract. WM-S-G: White murta stem glycerol extract. WM-S-A: White murta stem acetone extract. RM-S-A: red murta stem acetone extract GA: Gallic acid. QA: Quinic acid. CAT: (+)-Catechin. EPI: (−)- Epicatechin. M3ORh: Myricetin 3-*O*-rhamnoside. Q3OGa: Quercetin 3-*O*-galactoside. Q3OGu: Quercetin-3-*O*-glucuronide. DHQ3ORh: Dihydroquercetin 3-*O*-rhamnoside. C3OHx: Cyanidin 3-*O*-hexoside. PE3OA: Peonidin 3-*O*-arabinoside. PT3OA: Petunidin 3-*O*-arabinoside. CRG: Corilagin.

remarkable bioactive potential of murta extracts, with red murta leaf extracts demonstrating superior antioxidant capacity compared to fruit and stem extracts. This activity was strongly correlated with extractable phenolic content. Chromatographic analysis identified 37 phenolic compounds, highlighting structure-specific accumulation patterns: flavonols and anthocyanins in fruits, tannins and myricetin in leaves, and catechins and dihydroflavonols in stems. Notably, this study provided the first data on murta stems as valuable sources of unique polyphenolic compounds, complementary to those found in fruits and leaves. Our findings highlight the potential of murta biomass, particularly its leaves and stems, as a valuable source of bioactive compounds with applications in the food, nutraceutical, and cosmetic industries, aligning with circular bioeconomy principles and promoting sustainable extraction methods.

CRediT authorship contribution statement

Natalia Fuentes-Jorquera: Writing – original draft, Visualization, Methodology, Investigation, Formal analysis. **Marisol Villalva:** Writing – review & editing, Validation, Methodology, Data curation. **Jara Pérez-Jiménez:** Writing – review & editing, Visualization, Validation, Supervision, Methodology, Data curation, Conceptualization. **María Salomé Mariotti-Celis:** Writing – review & editing, Validation, Methodology, Data curation. **Roberto I. Canales:** Writing – review & editing, Supervision, Funding acquisition, Conceptualization. **José Ricardo Pérez-Correa:** Writing – review & editing, Supervision, Resources, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ifset.2026.104489.

Data availability

Data will be made available on request.

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