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ORIGINAL ARTICLE

Lipophilic and polar extracts in the cork-rich bark of the Miombo species *Parinari curatellifolia*

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Abstract

The Miombo species *Parinari curatellifolia* has a bark with a cork-rich rhytidome that was studied here for the first time regarding extract yields and composition, using the complete rhytidome and also the separated cork fraction. The objective was to analyse their potential as sources of extracts with antioxidant activities aiming at increasing the species' valorization and our knowledge on cork-rich barks.The content in total extractives was 6.1% and 8.4% in rhytidome and cork, respectively, with the lipophilic extracts soluble in dichloromethane corresponding to 23% and 38% of the total extracts, respectively. In cork, triterpenes and sterols were the main families of lipophilic components (58.9% and 20.8% of all compounds, respectively) while the rhytidome lipophilic extracts had a much higher content of long-chain lipids, including alkanoic acids and α,ω-diacids (28.8% and 7.0%, respectively), with terpenes representing 27.7% and sterols 8.4% of all compounds. Aromatics were also present in significant amounts of 12.1%. Triterpenes included compounds known for various therapeutic properties, e.g. betulinic acid, corosolic acid, maslinic acid and ursolic acid, thereby explaining the use of *P. curatellifolia* in traditional medicine. Ethanol-water extracts contained phenolics, flavonoids, and tannins, and showed low (cork) or good (rhytidome) antioxidant properties. The outerbark of *P. curatellifolia* is therefore a potential source of bioactive compounds.

1 Introduction

Barks are complex tree components showing significant structural diversity and chemical richness. Barks are constituted by different tissues that develop radially: the phloem tissues, generated by the cambium, are located at an interior position, while one periderm, or various periderms, generated by the phellogen, are located externally. The outerbark, or rhytidome, comprises all the tissues that extend radially from the functioning periderm near the phloem to the outside. It is the frontier between the interior tissues of the tree and the outside, and therefore it is at the forefront of biotic and abiotic protection. Some rhytidomes have substantial layers of cork in successive periderms, i.e., cork-rich barks, which may contribute to increase tree protection against heat, drought or fires (Leite and Pereira 2017).

The African miombo, an extensive tropical seasonal woodland and dry forest covering about 10% of the continent, is characterized by a pronounced dry season, and a semi-arid climate (Chiteculo and Surovy 2018; Frost 1996; Montfort et al. 2021) with fire critically impacting tree cover (Malmer 2007; Ryan and Williams 2011). *Parinari curatellifolia* Planch ex. Benth (Chrysobalanaceae) is a miombo tree species that grows in tropical Africa from Senegal to Kenya with the highest concentration in Zimbabwe and the lowveld region of South Africa (Benhura et al. 2013). It is an evergreen tree growing up to 20 m with a distinct mushroom shape and a rough bark that develops a cork-rich rhytidome containing layers of cork between layers of phloem, as shown in Fig. 1. Little scientific information exists on the species, and its bark was not studied previous to our research on the cellular and chemical characteristics of its cork fraction, which confirmed the protective features towards environmental conditions (Malengue et al. 2023). *P. curatellifolia* is used in traditional medicine for treatment in many diseases and health disorders, such as cancer, pneumonia, fever, malaria, typhoid, hypertension, microbial infections,

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Fig. 1 Cross-section of part of a stem disc of *Parinari curatellifolia* showing the wood, phloem and the rhytidome with successive periderms separated by phloem layers. Each periderm contains a substantial cork layer that is singled out by its light brown colour

pain, anti-inflammation, and toothaches (Kundishora et al. 2020; Mawire et al. 2021; Omoniwa et al. 2021; Peni et al. 2010). A recent ethnopharmacological survey on medicinal plants used by traditional healers in Togo reports *P. curatellifolia* among the top species to treat cancer and old wounds (Kola et al. 2020). The bioactive properties of *P. curatellifolia* extracts have been attributed to the presence of phytochemicals such as triterpenoids, saponins, alkaloids, flavonoids, tannins and phenols (Halilu et al. 2008, 2019; Edeoga et al. 2005).

It is well known that barks, and especially the outerbarks, are chemically rich with a large diversity of compounds that may offer a large bioactivity potential. The cork component differs in chemical composition from the lignocellulosic phloemic fraction, namely regarding the secondary metabolites that are present and can be extracted by solubilization with adequate solvents (Leite and Pereira 2017). For instance, cork usually contains a higher proportion of lipophilic soluble compounds in comparison to phloem, e.g. the content in lipophilic extractives in cork and in phloem was respectively 10.9% and 1.7% in the rhytidome of *Quercus cerris* (Şen et al. 2020), 5.1% and 0.2% in *Quercus suber* (Costa et al. 2019) and 5.6% and 1.1% in *Pseudotsuga menziesii* (Ferreira et al. 2015). Therefore, knowledge on the specific composition of cork within the complete cork-rich rhytidome is required when assessing a potential source of bioactive compounds and the valorization of the outerbark.

This is the driver for the present study that focus on the content and composition of lipophilic and polar extracts in the outer bark of *P. curatellifolia* from trees sampled in the miombo woodland in Angola. The complete rhytidome was analysed, as well as its cork fraction after a careful separation from the associated phloem layers. The results will be a first report on the bark rhytidome and cork extracts for *P. curatellifolia*. The objective is to analyse the potential of the cork and the outerbark of *P. curatellifolia* as sources of extracts though phytochemical screening and antioxidant activities evaluation in order to contribute to the species' valorization potential. The results will also enhance our knowledge on cork-rich barks and specifically on the cork.

2 Materials and methods

2.1 Samples

Bark samples were collected from young *Parinari curatellifolia* trees in the community of Cachindongo, Province of Huambo, Angola (13°15'33'61"S, 15°36'96"E, altitude 1582 m) pertaining to the miombo ecosystem. The trees were on average 13 years old, with 12.8 cm stem diameter at breast height (1.30 m above ground).

The bark was separated manually from the wood in stem discs taken at approximately 1.30 m of tree height, and the rhytidome was cut out from the phloem. The rhytidome contained successive periderms with a substantial amount of cork, and pure cork samples were also prepared. For that, rhytidome pieces were granulated with a knife mill, and the granules of cork and phloem separated by density and visual observation with the help of a magnified glass to sort out the cork granules.

2.2 Extraction

The rhytidome and the pure cork samples were ground in a Retsch SK hammer mill, sieved and the 40–60 mesh fractions were kept for analysis. The soluble extractible compounds were determined using a Soxhlet apparatus with a

solvent sequence of dichloromethane, ethanol and water during 6 h, 16 h and 16 h, respectively.

2.3 Composition of lipophilic extractives

Aliquots of the dichloromethane extracts were evaporated under N_2 flow and dried under vacuum at room temperature overnight.

The samples were derivatized by trimethysilylation to trimethylsilylate all the hydroxyl and carboxyl groups into trimethylsilyl (TMS) ethers and esters, respectively, and immediately analyzed by GC-MS. The equipment used was an Agilent 5973 MSD that was operated under the following conditions: Zebron 7HG-G015-02 column (Phenomenex, Torrance, CA, USA) (30 m, 0.25 mm; ID, 0.1 µm film thickness), flow 1 mL/min, injector 380 °C, oven temperature program: 50 °C (held 1 min), rate 10 °C/min up to 150 °C, rate of 5 °C/min up to 200 °C, 4 °C/min up to 300 °C, and rate of 10 °C/min up to 380 °C (held 5 min). The MS source was kept at 220 °C and the electron impact mass spectra (EIMS) taken at 70 eV of energy. The area of peaks in the total ion chromatograms of the GC–MS analysis was integrated and their relative proportions expressed as percentage for semi-quantitative analysis. Compounds were identified as TMS derivatives by comparing their mass spectra with a GC–MS spectral library (Wiley, NIST), and by comparing their fragmentation profiles with published data, reference compounds, ion fragmentation patterns, and retention times.

Each aliquot was injected in triplicate and the results are presented by their mean.

2.4 Composition of polar extractives

Polar extracts were obtained by direct contact of the cork and rhytidome powders with a solution of ethanol/water (80/20, v/v) with a 1:10 (m/v) solid-liquid ratio for 60 min at 40 °C in an ultrasonic bath. The sample was filtrated, and the supernatant extract was used to determine the contents in total phenolics, flavonoids and condensed tannins, as well as the antioxidant activity using DPPH and FRAP methods. The yields of extracts were calculated and expressed as the percentage of mass of extract per gram of raw material.

Table 1 Extraction yields (% of dry mass) of cork and rhytidome of *Parinari curatellifolia* trees, with dichloromethane (DCM), ethanol (EtOH) and water (H2O), as mean and standard deviation of three trees

% of dry mass	Cork	Rhytidome
Total extractives	8.41 ± 0.90	6.09 ± 0.55
DCM extractives	3.19 ± 1.00	1.38 ± 0.32
EtOH extractives	1.26 ± 0.05	1.95 ± 0.31
H_2O extractives	3.95 ± 0.06	2.76 ± 0,41

2.4.1 Determination of total phenolic, flavonoid and condensed tannins contents

The total phenolic content was evaluated using the Folin-Ciocalteu reagent (Singleton et al., 1965) and determined through a standard curve of gallic acid (0.014 to 0.762 g/L) and expressed as mg of gallic acid equivalents (GAE) per g of extract.

Total flavonoids content was determined by an aluminium chloride colorimetric assay (Zhishen et al. 1999) and condensed tannins by the vanillin-H_2SO_4 method (Abdalla et al. 2014) using (+)-catechin as standard. The results were expressed as mg of catechin equivalents (CE) per g of extract.

2.5 Antioxidant activity of ethanol-water extracts

The antioxidant activity of the ethanol-water extracts was determined by two methods to cover the various mechanisms of antioxidant action: 2,2-diphenyl-1-picryhydrazyl (DPPH), which measures the free radical scavenging capacity, and ferric reducing/antioxidant power (FRAP), which measures the sample's ferric reducing power.

The DPPH assay was performed using 2,2-diphenyl-1-picrylhydrazyl hydrate, and expressed in terms of: (i) the amount of extract required to reduce by 50% the DPPH concentration (IC_{50}); and (ii) Trolox equivalents (TEAC) on a dry extract (Sharma and Bhat 2009).

The FRAP assay was performed following the methodology described by Benzie and Strain (1996), and the results were expressed as mg of Trolox equivalents (TE) or gallic acid equivalents (GAE) per g of extract.

3 Results and discussion

The bark of *Parinari curatellifolia* contains a substantial rhytidome, clearly distinguished in the cross-section from the internal layer of phloem. The rhytidome is formed by multiple periderms showing layers of cork with a light brown color that alternate with dark colored phloem layers (Fig. 1). The analysis of extractives content and composition was made on the complete rhytidome (i.e. including the cork and phloem layers) and on the pure cork that was separated from the associated phloem layers.

3.1 Extraction yields

The yields obtained by the solvent extraction of the cork and rhytidome samples of *P. curatellifolia* bark are given in Table 1. Extractives amounted to 8.4% of the cork, of which 38% are lipophilic compounds soluble in dichloromethane,

and 62% are polar compounds solubilised by ethanol and water. The rhytidome contained a total of 6.1% extractives with 23% of lipophilic and 77.0% of polar extracts.

The total extractives content found in *P. curatellifolia* cork is similar to the 9.6% reported for *Quercus variabilis* (Miranda et al. 2013), but lower than values determined for corks of other species, e.g. *Quercus suber* (8.6–32.9%; Pereira 2013), *Quercus cerris* (16.7%; Şen et al. 2010), *Kielmeyera coriacea* (15–20%; Rios et al. 2014) and *Pseudotsuga menziesii* (29.2%; Ferreira et al. 2015). The proportion of lipophilic extractives in the total extractives is usually high in corks (Leite and Pereira 2017), and that also happened in *P. curatellifolia* with the DCM solubles representing 38% of the total extractives.

The rhytidome contained less extractives than cork (Table 1) with a lower proportion of lipophilic extractives, that represented 23% of the total extractives. This is the result from the lower content in DCM solubles of the phloem fraction contained in the rhytidome. This agrees with studies that compared the content in lipophilic extractives of cork and phloem: for instance, the lipophilic fraction of extractives in cork and phloem corresponded, respectively, to 25.6% and 12.4% of the total extractives in *Quercus cerris* (Şen et al. 2020), 43.6% and 4.4% in *Quercus suber* (Costa et al. 2019) and 19.2% and 3.9% in *Pseudotsuga menziesiii* (Ferreira et al. 2015).

A previous report of a mild maceration extraction of the whole bark of *P. curatellifolia* with methanol revealed a 4.5% yield (Halilu et al. 2023) while extraction with petroleum ether yielded 0.3% and a subsequent ethyl acetate extraction 0.8% (Halilu et al. 2013).

3.2 Composition of lipophilic extractives

Table 2 shows the different families and individual compounds identified in the dichloromethane extract of cork and of the complete rhytidome of *P. curatellifolia* bark. In cork, triterpenes and sterols are the main families of lipophilic components, representing 58.9% and 20.8% of all compounds, respectively. Among triterpenes, betulinic acid (24.6%), corosolic acid (8.0%) and ursonic acid (7.3%) are the main compounds. The major sterol is β-sitosterol (13.3% of all compounds), with ϒ-sitostenone and stigmasterol also identified. Saturated, unsaturated and substituted alkanoic acids, and α,ω-diacids were also found in the lipophilic extracts in small amounts (3.2%, 1.7%, 0.6% and 2.2%, respectively), with hexadecanoic and tetracosanoic acids as the most represented saturated alkanoic acids (0.8%, and 0.6%, respectively) and hexadecanedioic acid (1.9% of all compounds identified) as the most represented α,ω-diacid. Alkanols comprised only 0.6% of all compounds. Aromatics were present in small amounts (0.7% of all compounds), including vanillin and isovanillic acid.

Overall, all the identified lipophilic compounds in the cork of *P. curatellifolia* bark have been previously reported in cork extractives from other species although with differences in compositional proportions. The triterpene betulinic acid, detected here in high quantity in the cork lipophilic extracts of *P. curatellifolia* bark, is also the main compound found in the dichloromethane extract of *Q. suber* cork (Sousa et al. 2006). However, in the lipophilic extracts of cork from *Q. cerris* friedelin was the major compound identified, accompanied by β-sitosterol, betulin and squalene (Şen et al. 2015), while the lipophilic extracts of cork from *Pseudotsuga menziesii* did not show the presence of triterpenes (Ferreira et al. 2015). This clearly shows that the lipophilic cork extracts differ in amount and composition between different species.

The rhytidome lipophilic extracts (Table 2) differ from the cork extracts by a much higher content of long-chain lipids: saturated and unsaturated alkanoic acids represented 20.7% and 5.9% of all compounds, respectively, α,ω-diacids 7.0%, alkanols 2.4%, and glycerol 2.6%. Aromatics were also present in significant amounts (12.1%). Terpenes were a major chemical class in the extracts, with 27.7%, and sterols corresponded to 8.4%, both much under their content in cork extracts.

A work comparing the composition of dichloromethane extracts of cork and phloem tissues of *P. menziesii* bark (Ferreira et al. 2015) reported the presence of fatty acids as the main components of phloem samples (26.4% vs. 19.0%), and the presence of higher amount of sterols (β-sitosterol 18.1%). Comparable relative quantities of fatty acids were found also in dichlorometane extracts of phloem tissues of *Quercus cerris* bark rhytidome (Şen et al. 2010).

The presence in the cork and rhytidome of *P. curatellifolia* of compounds that show bioactivity and have been reported as having important therapeutical quality for various disorders and diseases, as discussed for the main compounds in the last section of the paper, explains the use of this bark in traditional medicine. This is the case for several of the pentacyclic triterpenoids that are present in *P. curatellifolia* cork and rhytidome: betulinic acid is reported to have several bioactive properties namely antiinflamatory and antiviral, and with promising anticancer activity (Jiang et al. 2021; Oliveira-Costa et al. 2022), corosolic acid has a large array of bioactivities and great attention has been given to its anti-diabetic properties as well as the anticancer effect while being innocuous to normal cells (Sharma et al. 2018; Sivakumar et al. 2009; Zhao et al. 2021), and ursolic acid also shows similar bioactivities (Liu et al. 2023; Woźniak et al. 2015). Oleanolic acid extracted from *P. curatellifolia*

Table 2 Composition of the dichloromethane extracts of cork and rhytidome in the bark of *Parinari curatellifolia* trees, as % of total peak area in the chromatograms. Mean and standard deviation of three trees

Chemical compounds, % of total area	Cork	Rhytidome
1-Alkanols	**0.61 ± 0.20**	**2.41 ± 0.42**
1-Dodecanol		0.51 ± 0.30
1,12-Dodecanediol		0.90 ± 0.08
1-Hexadecanol	0.04 ± 0.00	
1-Octadecanol	0.05 ± 0.00	
1-Docosanol	0.09 ± 0.02	0.68 ± 0.29
1-Tetracosanol	0.22 ± 0.04	0.08 ± 0.07
1-Hexacosanol	0.21 ± 0.14	0.23 ± 0.22
Saturated alkanoic acids	**3.22 ± 0.56**	**20.70 ± 2.10**
Hexanoic acid		0.42 ± 0.24
Heptanoic acid		1.30 ± 0.52
Octanoic acid		2.62 ± 1.50
Nonanoic acid	0.04 ± 0.01	4.01 ± 0.68
Decanoic acid	0.03 ± 0.02	1.70 ± 0.57
Dodecanoic acid		1.19 ± 0.44
Tetradecanoic acid	0.07 ± 0.01	1.15 ± 0.35
Pentadecanoic acid	0.04 ± 0.00	
Hexadecanoic acid	0.79 ± 0.05	5.24 ± 0.68
Heptadecanoic acid	0.03 ± 0.00	
Octadecanoic acid	0.42 ± 0.03	1.55 ± 0.32
Heneicosanoic acid	0.05 ± 0.00	
Docosanoic acid	0.38 ± 0.01	0.74 ± 0.36
Tricosanoic acid	0.26 ± 0.09	
Tetracosanoic acid	0.59 ± 0.06	0.68 ± 0.36
Pentacosanoic acid	0.19 ± 0.16	
Hexacosanoic cid	0.34 ± 0.24	
Unsaturated alkanoic acids	**0.74 ± 0.14**	**5.88 ± 1.24**
9-Hexadecenoic acid	0.02 ± 0.02	0.25 ± 0.17
9,12-Octadecadienoic acid	0.54 ± 0.35	
9-Octadecenoic acid	0.09 ± 0.09	5.63 ± 1.19
11-Octadecenoic acid	0.13 ± 0.13	
13-Docosenoic acid	0.10 ± 0.01	
Substituted alkanoic acids		**2.18 ± 2.02**
Pentanoic acid, 4-oxo		2.18 ± 2.02
Saturated α,w - alkanoic diacids	**2.23 ± 0.73**	**7.00 ± 4.40**
Octanedioic acid	0.07 ± 0.04	0.58 ± 0.22
Nonanedioic acid	0.13 ± 0.07	1.08 ± 0.33
Hexadecanedioic acid	1.92 ± 0.54	2.86 ± 2.38
Octadecanedioic acid	0.11 ± 0.08	0.74 ± 0.51
Eicosanedioic acid	1.13 ± 1.02	1.59 ± 1.24
Docosanedioic acid		0.15 ± 0.14
Terpenes/Triterpenoids	**58.91 ± 0.78**	**27.71 ± 6.52**
Pimaric acid	0.16 ± 0.16	
Dehydroabietic acid	1.04 ± 1.04	
Abietic acid	0.06 ± 0.01	
Gibberellin	3.48 ± 2.80	1.97 ± 1.12
Gibberellic acid	0.61 ± 0.48	
Betulinol	1.30 ± 0.38	0.27 ± 0.28
Oleanolic acid	4.42 ± 0.08	1.65 ± 0.43
Betulinic acid	24.60 ± 0.95	8.67 ± 2.36
Ursolic acid	7.34 ± 0.28	2.59 ± 0.80
Hederagenin acid	2.44 ± 0.37	2.21 ± 0.53

Table 2 (continued)

Chemical compounds, % of total area	Cork	Rhytidome
Maslinic acid	3.32 ± 2.23	1.80 ± 0.56
Corosolic acid	8.04 ± 2.89	5.12 ± 1.30
Euscaphic acid		3.43 ± 1.10
Tomentic acid	2.54 ± 0.18	
Steroids	**20.79 ± 5.27**	**8.39 ± 3.93**
Campesterol	0.16 ± 0.01	0.02 ± 0.03
Stigmasterol	1.20 ± 0.81	0.65 ± 0.26
β-sitosterol	13.29 ± 5.09	6.30 ± 3.26
Stigmast-4-en-3-one		1.43 ± 0.56
Stigmastanol	0.17 ± 0.05	
4, 22-Stigmtadiene-3-one	0.46 ± 0.10	
ϒ-Sitostenone	5.51 ± 0.66	
Aromatics	**0.69 ± 0.23**	**12.08 ± 2.78**
Benzoic acid	0.02 ± 0.00	2.51 ± 0.77
Vanillin	0.23 ± 0.12	3.83 ± 1.10
Cinnamic acid	0.06 ± 0.06	
3-Hydroxybenzoic acid		2.19 ± 0.79
Sinapaldehyde	0.06 ± 0.01	0.90 ± 0.08
Isovanillic acid	0.18 ± 0.00	1.49 ± 0.19
Vanillic acid		
Isoferulic acid	0.01 ± 0.01	0.39 ± 0.10
p-coumaric alcohol	0.01 ± 0.01	
Ferulic acid	0.11 ± 0.02	0.67 ± 0.37
Glycerol and derivatives	**0.96 ± 0.68**	**2.58 ± 1.55**
Glycerol	0.28 ± 0.17	2.58 ± 1.55
2,3-Dihydroxypropyl hexadecanoate	0.46 ± 0.34	
2,3-Dihydroxypropyl octadecanoate	0.22 ± 0.17	
Sugars	**0.04 ± 0.00**	
Sitosteryl-3β-D-glucopiranoside	0.04 ± 0.00	
Identified	**89.46 ± 0.42**	**88.93 ± 2.67**

bark was reported to have anticonvulsant and sedative properties (Halilu et al. 2019).

3.3 Composition and antioxidant properties of polar extractives

The phenolic composition of the ethanol-water extracts obtained under mild extractions conditions (40ºC and 80:20 v/v ethanol-water) is shown in Table 3 regarding content of total phenolics, flavonoids and condensed tannins. The antioxidant activity of the extract is also included, as measured by the DPPH and FRAP methods.

The extraction yield under these mild conditions was much lower than the total polar extractives determined by the successive extraction with ethanol and water (1.7% vs. 5.2% in cork, 1.6% vs. 4.7% in rhytidome). There is therefore scope to improve yields by optimizing extraction conditions.

The ethanol-water extract composition of *P. curatellifolia* cork and rhytidome is here reported for the first time (Table 3) and compared to that of other corks. In general, the total phenolic content was low, with 290.0 mg GAE/g extract. This compares with values reported for *Q. suber* cork extracts (Ferreira et al. 2020; Santos et al. 2010, 2013), although lower than for *Q. cerris* cork extracts (Şen et al. 2020), but higher than for *Beaucarnea recurvata* cork (Şen et al. 2018).

The content of flavonoids and proanthocyanidins in *P. curatellifolia* cork ethanolic extract is above values previously reported for *Q. suber* cork ethanol-water extracts (82.4 mg and 90.0 mg CE/g extract; Ferreira et al. 2020) and methanol-water extracts (49.9 mg quercetin eq. and 29.5 mg CE/g extract for flavonoids and condensed tannins, respectively; Touati et al. 2015), as well as for *Beaucarnea recurvata* cork ethanolic extract (61.6 mg and 19.5 mg CE/g extract for flavonoids and tannins content, respectively; Şen et al. 2018). Similar flavonoids and tannins content was observed in 50% ethanol extract of *Q. cerris* cork (tannins 220.8 CE/g extract and flavonoids 318.8 mg CE/g extract; Şen et al. 2020).

Table 3 Composition and antioxidant properties of the ethanol-water extracts of cork and rhytidome in the bark of *Parinari curatellifolia* trees. Mean of three trees and standard deviation

	Cork	Rhytidome
Extraction yield (%)	1.73 ± 0.26	1.55 ± 0.11
Total phenolic compounds (mg GAE g^{-1} extract)	289.95 ± 147.46	263.87 ± 43.96
Flavonoids (mg CE g^{-1} extract)	198.74 ± 119.00	236.86 ± 88.32
Condensed tannins (mg CE g^{-1} extract)	202.54 ± 131.15	275.10 ± 48.06
Antioxidant capacity TEAC (mg TE g^{-1} extract)	351.69 ± 237.4	609.70 ± 161.41
IC_{50} (µg mL^{-1})	13.30 ± 7.66	7.76 ± 2.24
FRAP (nM TEAC g^{-1} extract)	2.19 ± 1.0	3.08 ± 0.70
FRAP (mg/g extract)	548.24 ± 272.26	770.04 ± 161.85

*GAE: Gallic acid equivalents, CE: Catechin equivalents, TE:,Trolox equivalents, TEAC: Trolox equivalents antioxidant capacity, IC50: Half maximal inhibitory concentration to reduce DPPH radical formation, DPPH: 2,2-diphenyl-1-picrylhydrazyl hydrate (DPPH), FRAP: Ferric-reducing/antioxidant power

The ethanol–water extracts of *P. curatellifolia* cork (Table 3) revealed very low antioxidant activity with IC_{50} values of 13.3 µg extract/ mL, as compared to IC_{50} values of known antioxidant standards e.g. Trolox (3.3 µg/mL) and (+) catechin (2.2 µg/mL). Corks from other species were reported to have polar extracts with better antioxidant properties e.g. *Quercus suber* with IC_{50} of 3.5–5.8 µg extract/ mL (Ferreira et al. 2020; Santos et al. 2010, 2013; Touati et al. 2015), *Beaucarnea recurvata* cork with 4.4 µg extract/ mL (Şen et al. 2018), and *Quercus cerris* with IC_{50} values between 2.5 and 7.3 µg extract/mL (Şen et al. 2020).

The rhytidome ethanol–water extracts (Table 3) had a content of total phenolics and flavonoids similar to cork, while content of condensed tannins was higher. The most important difference was related to the antioxidant activity of the ethanol-water extracts (IC_{50} of 7.8 µg extract/mL) which was much better than that of cork although below the value for reference antioxidants. Şen et al. (2020) characterized waste cork and phloem fractions of *Q. cerris* bark obtained after an industrial scale fractionation of bark, and the phloem extracts showed antioxidant activity, with IC_{50} values between 5.8 and 11.7 µg extract/mL.

3.4 Potential applications

Extractives are currently under high scrutiny as possible targets for biomass valorization, and several designs for biorefineries propose extraction as a first step in processing, namely when barks are considered as feedstocks because of their high content in extractives (Şen et al. 2023). Extractives are chemically very diversified between species, and many possess bioactive properties or are chemically valued molecules.

In the case of the outer bark of *P. curatellifolia*, the content (Table 1) and the composition of lipophilic (Table 2) and polar (Table 3) extractives allow to consider them as a valorisation target. Of special importance are the potential medicinal applications of several triterpenes and sterols that exist in appreciable amounts in the *P. curatellifolia* outer bark, and which are reported to have therapeutical properties against, e.g. cancer, diabetes, obesity among many other diseases or ailments. This is the case, for instance, of the triterpenes corosolic acid (Miura et al. 2012; Zhao et al. 2021), ursolic acid (Erdmann et al. 2021; Seo et al. 2018;), maslinic acid (Choudhary et al. 2021; He et al. 2022; Lozano-Mena et al. 2014;), betulinic acid (Cai et al. 2018; Oliveira-Costa et al. 2022), oleanolic acid (Castellano et al. 2022; Li et al. 2021), hederagenin acid (Wang et al. 2020; Zeng et al. 2018), tormentic acid (Olech et al. 2021), or euscaphic acid (Jeong et al. 2022). In another field, gibberellin is a known plant regulator (Gupta et al. 2013).

Certainly this chemical richness of *P. curatellifolia* outer bark explains its use in traditional medicine with a long list of beneficial properties for various problems (Kola et al. 2020; Kundishora et al. 2020; Omoniwa et al. 2021).

P. curatellifolia outer bark, and especially its cork component, appear as potential source for the pentacyclic triterpenoids betulinic acid, corosolic acid and ursolic acid because they are present in considerable proportions in a lipophilic extract that is obtained with a high yield (Tables 1 and 2).

4 Conclusion

The content and chemical composition of lipophilic and polar extractives of *P. curatellifolia* outerbark and of its cork fraction were here reported for the first time. The solvent extraction of *P. curatellifolia* outerbark may be included in a valorisation framework for the species, supporting its sustainable exploitation and contributing to an economic valorisation of the *miombo* region. The outerbark and its cork fraction contain several triterpenes reported to be therapeutically active for several important diseases, e.g. cancer or diabetes, and explain their use in traditional medicine. *P.*

curatellifolia outerbark, and especially its cork fraction, are potential sources of betulinic, corosolic and ursolic acids.

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Data availability All data supporting the findings of this study are available from the corresponding author upon request.

Declarations

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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