

Phytochemical analysis, total phenolic, total flavonoid contents and ferric reducing power of extracts from roots and leaves of *Searsia burchellii*

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Abstract

Searsia burchellii has therapeutic applications in traditional medicine. The aim of the present study was to analyse the phytochemical profiles, total phenolic contents (TPC), total flavonoid contents (TFC) and ferric reducing power of various extracts from roots and leaves of *S. burchellii* collected from the kingdom of Lesotho. A combination of maceration, hot solvent extraction and solvent-solvent partition techniques were used to obtain methanolic extracts, fractions such as hexane, chloroform, ethyl acetate and methanol/water from methanolic extracts and water extracts separately from the roots and leaves of *S. burchellii*. The presence of alkaloids, steroids, terpenoids, phenolics, tannins, flavonoids, coumarins, saponins, glycosides, carbohydrates, proteins and phlobatannins were identified from these extracts/fractions. The TPC of these extracts/fractions was found to be in the range of 3.22±0.37 to 247.46±4.33 mg TAE/g DW, respectively. The TFC of these extracts/fractions was found to be in the range of 34.70±4.31 to 278.20±2.01 mg QE/g DW, respectively. Additionally, these extracts/fractions showed a moderate to strong ferric reducing power compared to ascorbic acid. The IC₅₀ values were found to be in the range of 5.35-143.12 and 65.64-348.28 µg/mL for extracts/fractions from the roots and leaves, respectively. Ascorbic acid showed an IC₅₀ value of 20.93 µg/mL. Therefore, the extracts/fractions from *S. burchellii* have the potential to replace the existing synthetic antioxidants.

1. Introduction

Known by vernacular names such as Karoo kuni-bush, Karookeniebos, Motshotlho and Mokhoamphiri, *Searsia burchellii* belongs to the Anacardiaceae family of the *Searsia* (*Rhus*) genus (Smith, 1966; Van Wyk and Gericke, 2000; Germishuizen and Meyer, 2003; Moffett 2007). *S. burchellii* is widely distributed in Namibia, Lesotho and South Africa (Palmer and Pitman, 1972; Moffett, 1994; Palgrave, 2002). *S. burchellii* has grey coloured stem bark, densely populated olive-green waxy leaves and cream-coloured flowers of 4-6 cm long and yields reddish-brown fleshy fruits (Compton, 1976; Van Wyk and Van Wyk, 1997; Van Wyk *et al.*, 1997; New Winger, 2000; Palgrave, 2002). *S. burchellii* is an evergreen shrub and grows up to 5 meters height. Several plants from the *Searsia* genus have previously been reported with antimalarial, antiviral, anticancer, antidiarrheal, anti-inflammatory, antimicrobial, hepatoprotective and antioxidant activities (Djakpo and

Yao, 2010; Moteetee and Van Wyk, 2011; El-Salam and Mohammed, 2015; Moteetee and Kose, 2017; Mtunzi *et al.*, 2017). However, the species *S. burchellii*, which belongs to the same *Searsia* genus, has not been explored well for its biological and pharmacological activities so far. *S. burchellii* has been used to treat respiratory and pulmonary diseases, tuberculosis, cold, fever and problems associated with childbirth (Nielsen *et al.*, 2012; Umberto, 2012). The present study aimed to analyse the phytochemical profiles, total phenolic contents (TPC), total flavonoid contents (TFC) and ferric reducing power of various extracts from roots and leaves of *S. burchellii* collected from the kingdom of Lesotho. This is the first report of this kind, particularly the species from the Kingdom of Lesotho.

2. Materials and methods

2.1 Plant materials

The roots and leaves of *S. burchellii* were collected

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in January and March 2020 on the hills of Maseru district and along the Senqu River at Ha Ramatseliso village in Thaba-Tseka district in the Kingdom of Lesotho, Southern Africa. The plant materials were identified by Dr Lerato Seleteng-Kose, Department of Biology, National University of Lesotho, Roma campus, The Kingdom of Lesotho.

2.2 Processing of plant materials

The plant materials were air-dried at room temperature (25-27°C) for three weeks at the Organic Chemistry Laboratory and then pulverized into powder using a grinding machine (HSIANGTAI, Mode SM-450L, AC220V, 50HZ, MRC Laboratory Equipment). A mass of 1160.83 and 997.17 g of powdered roots and leaves, respectively were obtained.

2.3 Chemicals used

Analytical grades of the following solvents and chemicals were used for this study. Hexane, chloroform, ethyl acetate and methanol (all from Sigma-Aldrich), potassium ferricyanide (Holpro Analytics Pty Ltd), ascorbic acid (Associated Chemical Enterprises), ferric chloride, sodium phosphate (both from Saarchem Pty Ltd), trichloroacetic acid (BDH Chemicals Ltd), tannic acid and Folin-Ciocalteu (both from Sigma Aldrich), quercetin (Acros Organics), sodium carbonate (Radchem Laboratory Supplies), sodium hydroxide (Minema Ltd) and sodium nitrite (Unilab).

2.4 Preparation of plant extracts and fractions

A mass of 854.31 g of powdered leaves was macerated with 3.5 L of methanol for 72 hrs. The resulting solution was filtered off through a Whatman No.1 filter paper and a methanolic crude leaf extract was obtained on concentration. The above procedure was repeated twice, and the extract was combined. Finally, the same material was extracted under reflux condition for 10 hrs and the solution was concentrated. A mass of 244.33 g of combined methanolic crude leaf extract was obtained. Water extract from the leaves was obtained by boiling 142.86 g of powder with 300 mL of distilled water at 80°C for 10 hrs. A mass of 20.23 g of waterleaf extract was obtained on concentration. Using a similar extraction procedure, mass of 84.05 and 7.28 g of methanolic and water extracts, respectively were obtained from 1075.37 and 85.46 g of root powder. A mass of 5.15 and 85.15 g of these methanolic crude extracts from roots and leaves, respectively were kept separately for their radical scavenging activity and other studies.

A mass of 159.18 g crude methanolic leaf extract

was suspended in methanol-water (1:1 v/v) and subjected to solvent-solvent partition successively with hexane, chloroform and ethyl acetate. A mass of 18.95, 102.28 and 9.41 g of hexane, chloroform and ethyl acetate fractions, respectively were obtained. A mass of 22.88 g of the remaining methanol-water fraction was also obtained on concentration. Similarly, 2.70, 5.16, 15.85 and 46.54 g of hexane, chloroform, ethyl acetate and methanol-water fraction, respectively were obtained from 78.89 g crude methanolic root extract.

2.5 Phytochemical analysis

The extracts and fractions were analysed for the presence of various phytochemicals such as alkaloids, steroids, phenols, flavonoids, saponins, glycosides, coumarins, carbohydrates, proteins, tannins, terpenoids, and phlobatannins as per methods previously described in the literature (Njoku and Obi, 2009; Usman *et al.*, 2009; Bargah, 2015; Banu and Cathrine, 2015; Yakop *et al.*, 2020).

2.5.1 Detection of alkaloids

A mass of 0.1 g of each extract or fraction was dissolved in 6mL of 2% sulphuric acid separately and filtered. Each one of the resulting solutions was divided into three equal volumes (2 mL each) and transferred into three test tubes labelled A, B, and C.

Dragendorff's test: To test tube A, a few drops of Drangedorff's reagent were added. A formation of orange-red precipitate indicated the presence of alkaloids.

Mayer's test: To test tube B, a few drops of Mayer's reagent were added. The formation of a white creamy precipitate indicated the presence of alkaloids.

Wagner's test: To test tube C, a few drops of Wagner's reagent were added. A formation of a reddish-brown precipitate confirmed a positive test for alkaloids.

2.5.2 Detection of steroids

A mass of 0.1 g of the test extract or fraction was dissolved in 5 mL of chloroform and filtered. The resulting filtrate was divided into two halves with 2.5 mL each.

Salkows'ki test: To the 2.5 mL of filtrate, a few drops of concentrated sulphuric acid were added and the mixture was shaken and allowed to stand at room temperature (25-27°C) for a few min. The formation of a yellow or red precipitate indicated the presence of steroids.

Liebermann-Buchard's test: To another 2.5 mL of

filtrate, 1 mL of glacial acetic acid was added and mixed and followed by the addition of a few drops of concentrated sulphuric acid along the walls of the test tube. The development of blue to the green colour indicated the presence of steroids.

2.5.3 Detection of terpenoids

A mass of 500 mg of each extract or fraction was dissolved in 5 mL of distilled water separately and then filtered. Approximately 2 mL of each of this filtrate was dissolved in an equal volume of chloroform separately and evaporated to dryness. After the test tube contents have all evaporated, 2 mL of the concentrated sulphuric acid was then added and heated for approximately 2 mins. The development of a greyish colour indicated the presence of terpenoids.

2.5.4 Detection of phenolics

Ferric chloride test: A mass of 500 mg of extract or fraction was dissolved in 5 mL of distilled water separately. To each of this solution, 3 to 5 drops of 10% aqueous ferric chloride were added. A bright yellow solution was formed, which then changed to a dark green solution showed the presence of phenols.

2.5.5 Detection of tannins

A mass of 0.1 g of each extract or fraction was dissolved in 5 mL of 99% ethanol separately and the resulting mixture was filtered. The clear alcoholic filtrate was treated with a few drops of 1% ferric chloride solution. A formation of blue-black, green, or blue-green colour indicated the presence of tannins.

2.5.6 Detection of flavonoids

A mass of 0.1 g of extract or fraction was dissolved in 10 mL of distilled water separately and the resulting mixture was filtered.

Alkaline test: A 2 mL of the filtrate and a 5 mL of 10% sodium hydroxide were mixed. The presence of flavonoids was detected by the formation of an intense yellow solution, which decolourized when a few drops of dilute sulphuric acid were added.

Lead acetate test: A 2 mL of the filtrate and 2 mL of 10% lead acetate was mixed. The formation of a yellow precipitate indicated the presence of flavonoids.

2.5.7 Detection of coumarins

A mass of 100 mg of each extract or fraction was mixed with 5 mL of distilled water. The resulting solution was filtered. 1 mL of 10% sodium hydroxide was added to the filtrate followed by the addition of 1 mL of chloroform. The formation of a yellow colour

indicated the presence of coumarins.

2.5.8 Detection of saponins

A mass of 0.1 g of each extract or fraction was dissolved in 5 mL of distilled water separately in centrifuge tubes. The resulting solution was shaken separately. The formation of 1 cm foam that persists for 15 min indicated the presence of saponins.

2.5.9 Detection of glycosides

Keller-Killiani's test: A mass of 500 mg of each extract or fraction was dissolved in 5 mL of distilled water. 2 mL of glacial acetic acid containing few drops of ferric chloride was added to the filtrate. 1 mL of concentrated sulphuric acid was further added along the side of the test tube. The formation of a brown ring at the interface or violet colour below the ring indicated the presence of glycosides.

2.5.10 Detection of carbohydrates

A mass of 0.1 g of each extract or fraction was dissolved in 5 mL of distilled water and the resulting mixture was filtered. The filtrate was used for the following tests.

Benedict's test: To the 2 mL of the filtrate, 2 mL of Benedict's reagent was added. The mixture was then heated on a boiling water bath for 2 min. A brick-red precipitate indicated the presence of reducing sugars.

Fehling's test: A volume of 2 mL of the filtrate was treated with a few drops of dilute hydrochloric acid and the resulting solution was heated with Fehling's A and B solutions. The formation of a red precipitate indicated the presence of reducing sugars.

2.5.11 Detection of proteins

Biuret test: A mass of 500 mg of each extract or fraction was dissolved in 10 mL of distilled water separately and filtered. A volume of 2 mL of each filtrate was treated with a few drops of 2% copper sulfate solution separately, followed by the addition of 1 mL of 95% ethanol. Excess potassium hydroxide pellets were added to the resulting solution. A formation of pink colour in the ethanolic layer indicated the presence of proteins.

2.5.12 Detection of phlobatannins

A mass of 500 mg of each extract or fraction was dissolved in 5 mL of distilled water separately and then filtered. To 1 mL of each of this filtrate, 2 mL of 1% hydrochloric acid was added and the mixture was boiled. The formation of the red precipitate indicated the presence of phlobatannins.

2.6 Determination of total phenolic contents (TPC)

The total phenolic contents of various extracts or fractions of *S. burchellii* were evaluated using the modified calorimetric Folin-Ciocalteu method as described previously (Kosar et al., 2007; Itidel et al., 2013; Khan et al., 2018). Tannic acid served as a positive control. Briefly, 1000 µg/mL of stock solution of positive control was prepared by dissolving 50.0 mg of tannic acid in 50 mL of 50% methanol (v/v). The stock solution was serially diluted to the following concentrations: 0, 25, 100, 150, 250, 500, 750 and 1000 µg/mL. A solution of 10 mL of each extract or fraction in 50% methanol at a concentration of 1000 µg/mL was prepared separately. An aliquot of 0.3 mL of extract or fraction or positive control was mixed separately with 2.5 mL of Folin-Ciocalteu (10%, v/v. in water) solution, followed by the addition of 5 mL of 7.5% sodium carbonate (w/v) solution. The contents were vortexed and allowed to stand in the dark at room temperature (25-27°C) for 2 hrs. The absorbance of the reaction mixtures was measured against a blank solution (50% methanol) at 760 nm. All analysis was conducted in triplicates. The TPC of each extract or fraction was estimated using the calibration curve from the tannic acid solution. The results were expressed as tannic acid equivalents (TAE) per gram of dry weight of extract or fraction.

2.7 Determination of total flavonoid contents (TFC)

The total flavonoid contents of each extract or fraction were determined by using the calorimetric aluminium chloride as per the method described by Bouaziz et al. (2009) with slight modification. Quercetin served as a positive control. A stock solution of quercetin was prepared by dissolving 5mg of quercetin in 5 mL of 50% of methanol. Serial dilutions of 0, 5, 10, 15, 20, 25, and 30 µg/mL were made from this stock solution. A 10 mL of 1000 µg/mL solution of each extract or fraction was prepared separately. A volume of 0.3 mL of standard or sample was mixed with 0.3 mL of 5% sodium nitrite. A volume of 0.3 mL of 10% aluminium chloride was added after 5 mins. The solution was allowed to stand for a minute and 2 mL of 1.0 M sodium hydroxide was added. The mixture was diluted with 6 mL of distilled water and thoroughly mixed. The absorbance was measured at 510 nm. The experiment was conducted in triplicates. The total flavonoid contents of each of these extracts or fractions were estimated from the quercetin calibration curve and are expressed as milligrams of quercetin equivalent (QE) per dry weight of extract or fraction.

2.8 Evaluation of ferric reducing power

The ferric reducing power of various extracts or

fractions was determined according to the methods previously described in the literature with some modifications (Sim et al., 2010; Kossah et al., 2011; Tlili et al., 2014). Briefly, a stock solution of 10 mg of each extract or fraction was dissolved in 10 mL of 50% methanol (v/v) separately. Further dilutions were made from the stock solution such as 0, 5, 10, 20, 40, 60, 80 and 100 µg/mL. A volume of 2.5 mL of each test solution was mixed separately with 2.5 mL of 0.2 M sodium phosphate (pH 6.6), and 2.5 mL of 1% potassium ferricyanide. The mixture was then incubated in a water bath at 50°C for 20 mins. After the incubation, 2.5 mL of 10% trichloroacetic acid was added and the mixture was centrifuged at 1000 rpm for 10 min. A volume of 2.5 mL aliquot of the upper layer was mixed with an equal volume of distilled water and 0.5 mL of a 0.1% ferric chloride solution. The absorbance of the reaction mixture was measured at 700 nm against a blank solution (50% v/v methanol in distilled water). The IC₅₀ values of various extracts or fractions were calculated from the graph, which was obtained by plotting the absorbance versus various concentrations of various extracts or fractions. The higher absorbance of the reaction mixture indicates the stronger reducing power of extracts or fractions and *vice versa*. Ascorbic acid served as a positive control. All experiments were conducted in triplicates ($n = 3$) and the results are expressed as mean \pm standard deviation.

2.9 Statistical analysis

Data analysis was performed using the SPSS 17.0 statistics program by means of a two-way analysis of variance. The differences were considered statistically significant when $p \leq 0.05$.

3. Results and discussion

The methanolic crude extract (**1**) was prepared from the roots of *S. burchellii* and four fractions were obtained from this methanolic crude extract viz. hexane fraction (**2**), chloroform fraction (**3**), ethyl acetate fraction (**4**) and methanol-water fraction (**5**). Additionally, water extract (**6**) was also prepared separately from the root powder. Similarly, methanolic crude extract (**7**) was prepared from the leaves of *S. burchellii* and four fractions were obtained from this methanolic crude extract viz. hexane fraction (**8**), chloroform fraction (**9**), ethyl acetate fraction (**10**) and methanol-water fraction (**11**). Additionally, water extract (**12**) was also prepared separately from the powdered leaves. All these extracts and fractions (**1-12**) were subjected to phytochemical analysis and the results are summarized in Table 1.

The TPC of **1-12** is summarised in Table 2. Tannic acid served as the standard for determining TPC and its

Table 1. Phytochemical screening of various extracts/fractions from roots and leaves of *S. burchellii*

Phytoconstituents	Extracts/Fractions											
	1	2	3	4	5	6	7	8	9	10	11	12
Alkaloids	-	-	+	-	-	+	+	+	+	-	-	-
Steroids	+	+	+	+	+	+	+	+	+	-	-	-
Terpenoids	+	-	-	+	-	+	-	-	-	-	-	-
Phenolics	+	+	+	+	+	+	-	-	-	+	+	+
Tannins	+	+	+	+	+	+	+	+	+	+	+	+
Flavonoids	+	-	+	-	+	-	-	-	-	+	+	+
Coumarins	-	-	-	-	-	-	-	-	-	+	+	+
Saponins	+	-	-	-	+	+	-	-	+	+	-	+
Glycosides	+	-	+	+	+	+	-	-	+	-	-	-
Carbohydrates	+		+	+	+	+	+	+	+	-	+	+
Proteins	+	-	-	+	+	+	-	-	+	-	+	-
Phlobatannins	+	-	-	+	+	+	+	-	-	+	+	+

1 = methanolic crude extract from root, 2 = hexane fraction from root, 3 = chloroform fraction from root, 4 = ethyl acetate fraction from root, 5 = methanol-water fraction from root, 6 = water extract from root, 7 = methanolic crude extract from leaves, 8 = hexane fraction from leaves, 9 = chloroform fraction from leaves, 10 = ethyl acetate fraction from leaves, 11 = methanol-water fraction from leaves, 12 = water extract from leaves. (+) and (-) signs indicate the presence and absence of phytoconstituent, respectively and these signs remained the same for a particular extract for which more than one phytoconstituent test was conducted.

calibration curve is shown in Figure 1. The TPC of 1-6 was found to be 205 ± 3.83 , 5.03 ± 0.53 , 140.46 ± 6.93 , 247.46 ± 4.33 , 246.10 ± 3.93 and 185.71 ± 4.20 mg TAE/g of dry extract weight, respectively. This result showed that 4 and 2 showed the highest and lowest TPC, respectively among the extracts/fractions from roots. Similarly, the TPC of 7-12 was found to be 1.77 ± 0.16 , 3.22 ± 0.37 , 46.95 ± 3.54 , 186.25 ± 4.56 , 69.53 ± 4.96 and 48.41 ± 6.33 mg TAE/g of dry extract weight, respectively. This result revealed that 1 and 4 showed the lowest and highest TPC among the leaf extracts/fractions. Additionally, the phytochemical analysis showed the absence of phenolics in 7, 8 and 9 (Table 1). Therefore, it is expected that 7, 8 and 9 will not be shown any TPC. Nevertheless, they showed much lower or appreciable TPC (Table 2). It might be due to the availability of hydrolyzable tannins in 7, 8 and 9 (Table 1), which could release free phenolics on dilution with 50% methanol (Refer to 2.6).

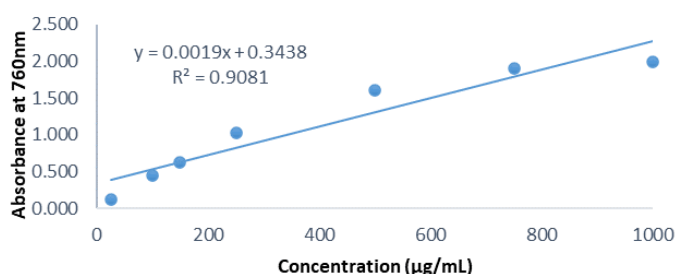


Figure 1. The calibration curve of tannic acid used to estimate TPC of various extracts/fractions from *S. burchellii*

The TFC of 1-12 is summarised in Table 2. Quercetin served as the standard for determining TFC and its calibration curve is shown in Figure 2. The TFC of 1-6 was found to be 90.07 ± 1.90 , 149.67 ± 5.65 ,

34.70 ± 4.31 , 126.43 ± 4.90 , 40.63 ± 1.46 and 54.53 ± 4.42 mg TAE/g of dry extract weight, respectively. This result indicated that fraction 2 and 3 showed the highest and lowest TFC among the root extracts/fractions. The TFC of 7-12 was found to be 163.03 ± 4.03 , 179.47 ± 2.40 , 123.83 ± 3.66 , 278.20 ± 2.01 , 231.67 ± 3.41 and 114.10 ± 3.90 mg TAE/g of extract weight. Amongst the leaf extracts/fractions, 10 and 12 exhibited the highest and lowest TFC, respectively. In general, the extracts/fractions from *S. burchellii* have an abundance of total flavonoid contents. However, the phytochemical analysis showed the absence of flavonoids in 2, 4 and 6-9 but these fractions showed reasonably higher TFC values. This might be due to the presence of hydrolysable condensed tannins which consist of flavan-3-ol subunits that give free flavonoid moieties (Naumann *et al.*, 2017).

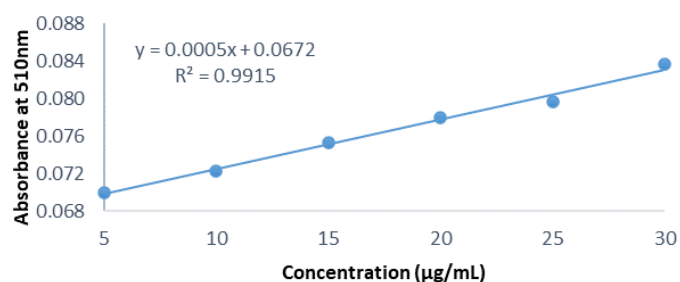


Figure 2. The calibration curve of quercetin used to estimate TFC of various extracts/fractions from *S. burchellii*

The ferric reducing agent power of 1-12 and ascorbic acid are summarized in Table 2. The ferric reducing power of 1-6 and ascorbic acid was found to be in the range of 0.265 ± 0.001 - 1.706 ± 0.013 , 0.147 ± 0.002 - 0.404 ± 0.005 , 0.200 ± 0.010 - 0.592 ± 0.019 , 0.370 ± 0.005 - 2.137 ± 0.003 , 0.284 ± 0.002 - 1.195 ± 0.042 , 0.207 ± 0.002 -

Table 2. Total phenolic contents (TPC), total flavonoid contents (TFC) and ferric reducing agent power (FRAP) of various extracts/fractions from *S. burchellii*

Extracts/ Fractions	TPC (mg TAE/g)	TFC (mg QE/g)	Ferric reducing power/concentration of extracts or fractions in µg/mL							IC ₅₀ (µg/mL)
			5	10	20	40	60	80	100	
1	205.26±3.83	90.07±1.90	0.265±0.001 ^a	0.359±0.007 ^c	0.520±0.002 ^a	0.853±0.012 ^b	1.335±0.003 ^a	1.647±0.003 ^a	1.706±0.013 ^b	17.39
2	5.03±0.53	149.67±5.65	0.147±0.002 ^a	0.165±0.007 ^a	0.207±0.004 ^a	0.251±0.005 ^b	0.277±0.002 ^a	0.329±0.003 ^a	0.404±0.005 ^a	143.12
3	140.46±6.93	34.70±4.31	0.200±0.010 ^c	0.210±0.006 ^a	0.257±0.002 ^a	0.351±0.002 ^a	0.431±0.003 ^a	0.502±0.006 ^c	0.592±0.019 ^c	77.12
4	247.46±4.33	126.43±4.90	0.370±0.005 ^d	0.511±0.100 ^f	0.786±0.008 ^b	1.282±0.005 ^b	1.932±0.008 ^c	2.049±0.015 ^a	2.137±0.003 ^a	5.35
5	246.10±3.93	40.63±1.46	0.284±0.002 ^a	0.363±0.010 ^b	0.471±0.017 ^b	0.675±0.097 ^c	0.953±0.020 ^b	1.127±0.133 ^a	1.195±0.042 ^d	22.79
6	185.71±4.20	54.53±4.42	0.207±0.002 ^a	0.248±0.003 ^a	0.314±0.003 ^a	0.466±0.011 ^d	0.548±0.028 ^c	0.733±0.018 ^d	0.750±0.013 ^b	50.83
7	1.77±0.16	163.03±4.03	0.118±0.002 ^a	0.120±0.002 ^a	0.134±0.002 ^a	0.162±0.002 ^a	0.194±0.005 ^a	0.251±0.002 ^a	0.281±0.001 ^a	222
8	3.22±0.37	179.47±2.40	0.126±0.001 ^a	0.133±0.001 ^a	0.173±0.006 ^c	0.234±0.004 ^a	0.239±0.003 ^a	0.273±0.002 ^a	0.567±0.008 ^c	112.32
9	46.95±3.54	123.83±3.66	0.132±0.002 ^a	0.137±0.004 ^a	0.152±0.001 ^a	0.180±0.001 ^a	0.205±0.001 ^a	0.228±0.002 ^a	0.265±0.002 ^a	348.28
10	186.25±4.56	278.20±2.01	0.158±0.001 ^a	0.169±0.001 ^a	0.198±0.001 ^a	0.294±0.002 ^a	0.327±0.001 ^a	0.408±0.001 ^a	0.467±0.001 ^a	109
11	69.53±4.96	231.67±3.41	0.157±0.001 ^a	0.189±0.007 ^a	0.250±0.002 ^a	0.360±0.003 ^a	0.457±0.011 ^c	0.569±0.007 ^a	0.699±0.042 ^c	65.64
12	48.41±6.33	114.10±3.90	0.139±0.001 ^a	0.148±0.002 ^a	0.193±0.006 ^a	0.250±0.002 ^a	0.311±0.002 ^a	0.395±0.002 ^a	0.411±0.005 ^a	118.58
Asc. acid	N/A	N/A	0.140±0.001 ^a	0.148±0.001 ^a	0.289±0.003 ^a	1.027±0.011 ^c	1.790±0.021 ^b	2.286±0.004 ^a	2.427±0.006 ^a	20.93

1-12 refer to footnote of Table 1. TPC = Total phenolic contents. TFC = Total flavonoid contents; Asc. acid = ascorbic acid; N/A = Not applicable; The experiment was conducted in triplicate (*n* = 3). Values with different superscript letters are significantly different within column.

0.750±0.013, and 0.140±0.002-2.427±0.006, respectively (refer to Table 2). Fraction 4 showed the highest ferric reducing power of 2.137±0.003 at a concentration of 100 µg/mL followed by 1, 5, 6, 3 and 2, respectively. The ferric reducing power of 7-12 was found in the ranges of 0.118±0.002-0.281±0.001, 0.126±0.001-0.567±0.008, 132±0.002-0.265±0.002, 0.158±0.001-0.467±0.001, 0.157±0.001-0.699±0.042 and 0.139±0.001-0.411±0.005, respectively. Fraction 11 showed the highest ferric reducing power at the concentration of 100µg/mL followed by 8, 10, 12, 7 and 9, respectively.

Additionally, the IC₅₀ values of 1-12 were determined and were found to be 17.39, 143.12, 77.12, 5.35, 22.79, 50.83, 222.00, 112.12, 348.28, 109.00, 65.64 and 118.58 µg/mL, respectively. This result revealed that 1 and 4 were the most potent with their IC₅₀ values less than the positive control. The positive control ascorbic acid showed an IC₅₀ value of 20.93 µg/mL. Fraction 5 showed a slightly higher IC₅₀ value than the positive control. All other extracts showed much higher IC₅₀ values than the positive control (Figure 1). As stated previously that *S. burchellii* has not been studied previously. However, the antioxidant activity of other species from the *Searsia* genus has previously been reported. For example, various extracts from leaves and stem-bark of *Searsia leptodictya* and *Searsia tripartita* have been reported to have promising radical scavenging activity (El-Salam and Mohammed, 2015; Moteetee and Kose, 2017; Mtunzi et al., 2017). Phytochemicals such as phenolics, tannins, saponins and flavonoids have been identified as active ingredients for this radical scavenging activity (El-Salam and Mohammed, 2015; Moteetee and Kose, 2017; Mtunzi et al., 2017).

4. Conclusion

The phytochemical screening, TPC, TFC, and ferric reducing power of various extracts/fractions from roots and leaves of *S. burchellii* were studied. The presence of phytochemicals such as alkaloids, steroids, terpenoids, phenolics, tannins, flavonoids, coumarins, saponins, glycosides, carbohydrates, proteins, and phlobatannins were identified from these extracts/fractions. The TPC of these extracts/fractions was found to be in the range of 1.77±0.16 to 247.46±4.33 mg TAE/g DW. The TFC of these extracts/fractions was found to be in the range of 34.70±4.31 to 278.20±2.01 mg QE/g DW. The root and leaf extracts/fractions showed ferric reducing power ranging from 0.147±0.002 to 2.137±0.003, and 0.118±0.002 to 0.699±0.042, respectively. The IC₅₀ values of root and leaf extract/fractions were found to be in a range of 5.35 to 143.12 and 65.64 to 348.28 µg/mL, respectively. From this study, we conclude that *S.*

burchellii is a phytochemical-rich plant with antioxidant potential. Therefore, further study can be made on this plant to replace the existing synthetic antioxidants and for other therapeutic applications.

Conflicts of interests

The authors declare no conflicts of interests.

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References

- Banu, K.S. and Cathrine, L. (2015). General techniques involved in phytochemical analysis. *International Journal of Advanced Research in Chemical Science*, 2(1), 25-32.
- Bargah, R.K. (2015). A preliminary test of phytochemical screening of crude ethanolic and aqueous extracts of *Moringa pterygosperma* Gaertn. *Journal of Pharmacognosy and Phytochemistry*, 4 (1), 7-9.
- Bouaziz, M., Dhouib, A., Loukil, S., Boukhri, M. and Sayadi, S. (2009). Polyphenol content, antioxidant, and antimicrobial activities of extracts of some wild plants collected from the South of Tunisia. *African Journal of Biotechnology*, 8, 7017-7027.
- Compton, R.H. (1976). The Flora of Swaziland. *Journal of South African Botany (Supplementary)*. Vol. 11, p. 1-484. Kirstenbosch, South Africa: National Botanic Gardens of South Africa.
- Djakpo, O. and Yao, W. (2010). *Rhus chinensis* and *Galla Chinensis*-Folklore to modern evidence: Review. *Phytotherapy Research*, 24(12), 1739-1747. <https://doi.org/10.1002/ptr.3215>
- El-Salam, A. and Mohammed, I. (2015). Phytoconstituents and the study of antioxidant, antimalarial and antimicrobial activities of *Rhus tripartita* growing in Egypt. *Journal of Pharmacognosy and Phytochemistry*, 4(2), 276-281.
- Germishuizen, G. and Meyer, N.L. (2003). Plants of Southern Africa: An annotated checklist. *Steltizia, Naturalis*, Leiden.
- Itidel, C., Khokri, M., Mohamed, B. and Yosi, Z. (2013). Antioxidant activity, total phenolic, and flavonoid content variation among the Tunisian natural

- population of *Rhus tripartite* (Ucria) Grande and *Rhus pentaphylla* Desf. *Industrial Crops and Products*, 51, 171-177. <https://doi.org/10.1016/j.indcrop.2013.09.002>
- Khan, M.S., Yusufzai, K.S., Rafatullah, M., Sarjadi, M.S. and Razlan, M. (2018). Determination of total phenolic content, total flavonoid content, and antioxidant activity of various organic crude extracts of *Licuala spinose* leaves from Sabah, Malaysia. *American Society for Microbiology*, 11(3), 53-58.
- Kosar, M., Bozan, B., Timellii, F. and Baser, K.H.C. (2007). Antioxidant activity and phenolic composition of sumac (*Rhus coriaria* L.) extracts. *Food Chemistry*, 103(3), 952-959. <https://doi.org/10.1016/j.foodchem.2006.09.049>
- Kossah, R., Zhang, H. and Chen, W. (2011). Antimicrobial and antioxidant activities of Chinese sumac (*Rhus typhina* L.) fruit extracts. *Food Chemistry*, 22(1), 128-132. <https://doi.org/10.1016/j.foodcont.2010.06.002>
- Moffett, R.O. (1994). Names of the Southern African species of *Rhus* (Anacardiaceae) and their etymology, *Bothalia*, 24(1), 67-76. <https://doi.org/10.4102/abc.v24i1.753>
- Moffett, R.O. (2007). Name changes in the old world *Rhus* and recognition of *Searsia* (Anacardiaceae). *Bothalia*, 37(2), 165-175. <https://doi.org/10.4102/abc.v37i2.311>
- Moteetee, A. and Kose, L.S. (2017). A review of medicinal plants used by the Basotho for treatment of skin Disorders: Their phytochemical, antimicrobial, and anti-inflammatory potential. *African Journal of Traditional, Complementary and Alternative medicine*, 14(5), 121-137. <https://doi.org/10.21010/ajtcam.v14i5.16>
- Moteetee, A. and Van Wyk, B.E. (2011). The medicinal ethnobotany of Lesotho: a review. *Bothalia*, 41(1), 209-228. <https://doi.org/10.4102/abc.v41i1.52>
- Mtunzi, F.M., Ejidike, P.I., Matamela, T., Dikio, E. and Klink, J.M. (2017). Phytochemical profiling, antioxidant and antibacterial activities of leaf extracts from *Rhus leptodictya*. *International Journal of Pharmacognosy and Phytochemical Research*, 9(8), 1090-1099. <https://doi.org/10.25258/phyto.v9i08.9616>
- Naumann, D.H., Tedeschi, L.O., Zeller, W.E. and Huntley, N.F. (2017). The role of condensed tannins in ruminant animal production: advances, limitations, and future directions. *Journal of Animal Science*, 46(12), 929-949. <https://doi.org/10.1590/s1806-92902017001200009>
- New Winger, H.D. (2000). African traditional medicine: A dictionary of the plant used and applications. p. 589. Stuttgart, Germany: Medpharm Scientific.
- Nielsen, T.R.H., Kuete, V., Jager, A.K., Meyer, J.J.M. and Lall, N. (2012). Antimicrobial activity of selected South African medicinal plants. *BMC Complementary and Alternative Medicine*, 12, 74. <https://doi.org/10.1186/1472-6882-12-74>
- Njoku, V.O. and Obi, C. (2009). Phytochemical constituents of some selected medicinal plants. *African Journal of Pure and Applied Chemistry*, 3 (11), 228-233.
- Palgrave, M.C. (2002). Palgrave's Trees of Southern Africa. 3rd ed. Capetown: Struik Publishers.
- Palmer, E. and Pitman, N. (1972). Trees of Southern Africa. Capetown and Amsterdam: Balkema.
- Sim, S.K., Sri Nurestri, A.M. and Norhanom, A.W. (2010). Phenolic content and antioxidant activity of *Pereskia grandifolia* Haw. (Cactaceae) extracts. *Pharmacology magazine*, 6(23), 248-254. <https://doi.org/10.4103/0973-1296.66945>
- Smith, C.A. (1966). Common names of South African plants. Memoirs of Botanical Survey of South Africa No. 35, Government Printer, Pretoria.
- Tlili, N., Mejri, H., Yahia, Y., Saadaoui, E., Rejeb, S., Khaldi, A. and Nasri, N. (2014). Phytochemicals and antioxidant activities of *Rhus tripartitum* (Ucria) fruits depending on locality and different stages of maturity. *Food Chemistry*, 160, 98-103. <https://doi.org/10.1016/j.foodchem.2014.03.030>
- Umberto, Q.F.L.S. (2012). Medicinal and Poisonous Plants. *Nature*. New York: CRC Press.
- Usman, H., Abdulrahman, F.I. and Usman, A. (2009). Qualitative phytochemical screening and in vitro antimicrobial effects of methanol stem bark extract of *Ficus thonningi* (Moraceae). *African Journal of Medicine, Complementary and Alternative Medicines*, 6(3), 289-295. <https://doi.org/10.4314/ajtcam.v6i3.57178>
- Van Wyk, B.E. and Cricke, N. (2000). People's Plants. Pretoria: Briza Publications.
- Van Wyk, B.E. and Van Wyk, P. (1997). Field guide to trees of Southern Africa. Capetown: Struik Publishers.
- Van Wyk, B.E., Van Outshoorn, B. and Gericke, N. (1997). Medicinal plants of South Africa. Pretoria: Biza Publications.
- Yakop, F., Hamid, M.H.S.A., Ahmad, N., Majid, M.A., Pillai, K.M. and Taha, H. (2020). Phytochemical screening, antioxidant and antimicrobial activities of extracts and fractions of *Dillenia suffruticosa*. *Malaysia Applied Biology*, 49(1), 121-130.