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Phytochemicals Screening, Proximate composition and Antioxidants Analysis of Italian *Citrus paradisi* Fruits

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Abstract

Citrus paradise is a largely consumed fruit for nutrition and medicinal importance. This study was conducted to evaluate the phytochemicals, proximate, and antioxidants contents of Italian *Citrus paradisi* fruits. The phytochemicals screening and proximate analysis were performed using the methods of AOAC. Antioxidants contents and capacities of the extract were determined by DPPH and FRAP Assay. The grapefruits aqueous extract revealed the presence of various phytochemicals and contains significant ($p < 0.05$) amount of carbohydrates (89.36 %), moisture (74.80 %), lipids (4.56 %), protein (3.36 %), crude fiber (2.36 %), and ash content (0.83 %). The extract exhibited significant ($p < 0.05$) increase in DPPH percentage inhibition of free radical scavenging activity and FRAP properties comparable to that of the standard, ascorbic acid. The grapefruits extract contains various phytochemicals, proximate nutrients, and exhibited free radical scavenging activity. The grapefruits have nutritional status, antioxidant properties, and medicinal importance.

Keywords: antioxidants, *Citrus paradise*, nutrients, phytochemicals, proximate composition

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1 Introduction

Varieties of plant materials including fruits have nutritional and medicinal importance. Globally, people rely on the plant materials for nutritional benefits and medicinal uses. Nutritional status of foods and plant materials is determined by their nutrients contents. Phytoconstituents have important roles in biological processes and are responsible for the medicinal and pharmacological properties of plant materials [1]. Phytochemicals demonstrated significant pharmacological activities, hence used in drug discovery and development [1]. Plants contain abundant natural antioxidant compounds that play significant roles in protection against production or synthesis of reactive oxygen species. Natural antioxidants prevent and fight against many diseases including chronic and degenerative disorders [2]. Low antioxidants contents in plant-based foods leads to high production of endogenous reactive oxygen species (ROS) resulting to severity of many chronic diseases [3]. Researchers have been concentrated on plants due their phytoconstituents contents, proximate composition, and antioxidants properties.

Citrus is an evergreen, small tree or shrub plant belonging to the family Rutaceae found abundantly in the world. Citrus is characterized by flavour, attractive evergreen foliage and flower as well as the extraordinary fragrance [4]. The Rutaceae family consists of about 150 genera and 1,500 species. The world most cultivated and consumed members of the Rutaceae family include Citrus sinensis (sweet oranges), Citrus reticulata (tangerines), Citrus paradisi(grapefruits), Citrus limon (lemon) and Citrus aurantifolia (limes) [5]. Citrus fruits are the most widely produce fruits with a significant economic value [6, 7]. About 115 million tons of citrus fruits are produced per year worldwide [5]. In Africa, about 3,741,000 tonnes of varieties of citrus fruits are produced yearly [8]. However, approximately 3% (3,240,000) of the world's citrus fruits are produced in Nigeria [8]. In Nigeria, citrus fruits are produced in large quantity and have been contributed immensely to the economic development of the country. Due to inadequate and small capacity processing industries, much quantity of citrus fruits produced in Nigeria are directly consumed [8].

Citrus fruits have been used in food industry, cosmetics and folk medicine [9, 10]. Wastes including peels, seeds and pulp from the citrus fruits processing industry are used as a source of many important products. Citrus fruits contain many bioactive compounds including carotenoids and phenolics [11, 12]. The phytoconstituents of citrus fruits include plant antioxidants that possess many medicinal and pharmacological activities [13]. Ctrus fruits demonstrated many pharmacological activities including anti-cardiovascular and anti-cancer activity [11, 14]. Citrus fruits extracted juice exhibited antiviral and anticancer activities [15, 16]. Citrus fruits peels have demonstrated antioxidant and antimicrobial properties [17]. Grapefruits (Citrus paradisi) are one of the dominant members of the Citrus fruits family. They are important citrus fruits characterized by a number of secondary metabolites. *Citrus paradisi* contain many phytoconstituents including vital nutrients, phytochemicals, and antioxidants [18]. Grapefruits have many medicinal uses due to the presence of flavonoid compounds which are potent antioxidants [19, 20, 21]. The medicinal important of Citrus paradise include lowering cholesterol level [22], used in treatment of cancer [23], fungal infections [24].

Studies have been reported on the phytochemicals and nutritional contents analysis of different species of *Citrus paradisi* fruits produced and/or consumed in different areas in the world. However, study has been reported on the phytochemical composition and

nutritive values of Egyptian *Citrus paradisi* fruits consumed in Wamakko Local Government Area of Sokoto State. Despite the limitation of the study in one of the twenty three local government areas in the state, the study has not been reported anything about the antioxidant capacity of the plant and certain important phytoconstituents and proximate parameters. Furthermore, Italian *Citrus paradisi* fruits have been consumed more in different areas in the state than the Egyptian *Citrus paradisi* fruits. To ascertain the nutritional status, phytoconstituents, and antioxidants capacity of the grapefruits consumed in this state, this study aims at evaluating the phytochemical, proximate and antioxidants contents and capacity of Italian *Citrus paradisi* fruits.

2 Experimental section

2.1 Materials

All the chemicals and reagents used in this study were of analytical grade. The chemicals were manufactured by Sigma-Aldrich (St. Louis, MO, USA; Irvine, UK; Chemie, Steinheim, Germany) and Guangdong Chemical Reagent Engineering, (Guangdong, China).

2.2 Collection and Authentication of the Plant Sample

Fresh *Citrus paradisi* fruits were purchased from different areas in all the three zones in Sokoto state. The areas are Ramen Kura, Kara Market in the state metropolis, Illela Market, Sabon Birni market, Bodinga market, and Dange Shuni Market, Sokoto, Nigeria. The sample was identified and authenticated at Botany Unit, Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto and the voucher material (UDUBSH/ANS/0336) was deposited.

2.3 Preparation of Plant Extract

The preparation of plant extract was carried out using the method of [1] with some modifications. The *Citrus paradisi* fruits were washed thoroughly with tap water and then rinsed in distilled water. Outer layer of the fruits was peeled and the seeds were removed. The remaining contents were cut into pieces, air dried at room temperature for fourteen days, and then pulverized into fine powder using an electric grinding machine. The grapefruits powder (500g) was soaked in 2L of distilled water for 72 hours with constant stirring at interval of 60 minutes. The grapefruits aqueous extract was filtered using Whatman filter paper and concentrated using rotary evaporator at 40°C under reduced pressure for 180 minutes. The extract obtained weighed 200.5g with percentage yield of 40.1% w/w approximately. The extract was then stored in desiccators until further analysis.

2.4 Qualitative Phytochemical Screening

2.4.1 Test for Alkaloids

Alkaloids in the grapefruits aqueous extract were identified using Wagner's test as described by Mosa et al. [25], Abubakar et al. [1, 26] and Trease and Evans [27]. Three (3) ml of the extract was added to 3 ml of 1% HCl, heated for 20 min. and then cool. One mile of Wagner's reagent was added drop by drop into a test tube containing the filtrate. The formation of a reddish-brown precipitate indicated the presence of alkaloids.

2.4.2 Test for Flavonoids

Sodium Hydroxide Test was used for the qualitative determination of flavonoids in the grapefruits aqueous extract as described by Mosa et al. [25] and Abubakar et al. [1, 26]. The extract (3 ml) was treated with 1 ml of 10% NaOH solution. The formation of an intense yellow colour indicated the presence of flavonoids which became colourless after the addition of dilute hydrochloric acid.

2.4.3 Test for Tannins

Tannins present in the grapefruits aqueous extract were analyzed by ferric chloride test using method of Trease and Evans [27]. Two milliliters of 5% solution of $FeCl_3$ was added to 1 ml of the crude extract. Presence of a black or blue-green colour indicated the presence of tannins.

2.4.4 Tests for Glycosides

The qualitative determination of glycosides in the grapefruits aqueous extract was done using Salkowski's test in accordance to the method of Abubakar et al. [1, 26] and Trease and Evans [27]. Twenty five miles of 1% sulphuric acid was added to 5 ml of the extract

in a test tube and boiled for 15minutes. The mixture was cool, neutralized with 10% sodium hydroxide, and then 5ml of Fehling's solution A and B was added. A brick red precipitate of reducing sugars indicated the presence of glycosides.

2.4.5 Test for Cardiac Glycosides

Cardiac glycosides in the grapefruits aqueous extract were analyzed by Keller-Killani test as described by Mosa et al. [25] and Trease and Evans [27]. Five milliliters (5ml) of the extract was treated with 2ml of glacial acetic acid containing one drop of ferric chloride solution. This was under lay with 1ml of concentrated sulphuric acid. A brown ring at the interface was formed which indicated the presences of deoxysugar, a characteristic of cardenolides. The violet colour appeared below the brown ring, while in the acetic acid layer, the greenish colour formed gradually throughout the layer.

2.4.6 Test for Steroids

Steroids present in the grapefruits aqueous extract were determined by using the method of Trease and Evans [27]. Five milliliters of chloroform and 5 ml of sulphuri acid were added to 500 µl of the prepared plant extract. Violet colour changed to blue-green which indicated the presence of steroids.

2.4.7 Test for Saponins

The Froth test as described by Mosa et al. [25], Abubakar et al. [1, 26] and Trease and Evans [27] was used for the qualitative determination of saponins in the grapefruits aqueous extract About 3 ml of the plant extract was added to 3 ml of distilled water in a test tube. The test tube was shaken vigorously for about 30 seconds and allowed to stand for 30 minutes. The stable persistent froth was formed which indicated the presence saponins.

2.4.8 Test for Phenols

Phenols present in the grapefruits aqueous extract were determined by using the method of Trease and Evans [27]. The extract (5 ml) was put in a test tube and then 5 ml of ethanol and 5 ml of the ferric chloride was added into the test tube. The formation of an ink blue coloration indicated the presence of phenols.

2.4.9 Test for Terpenoids

Terpenoids in the grapefruits aqueous extract were analyzed using the method described by Trease and Evans [27]. Portion of the plant extract was shaken with ethanol and acetic anhydride (1ml) 10 ml of concentrated sulphuric acid was added. A pink color formed which indicated the presence of terpenoids.

2.5 Quantitative Determination of Phytochemicals

2.5.1 Determination of Alkaloids Content

Total alkaloids in the grapefruits aqueous extract were analyzed in accordance to the method of Trease and Evans [27] with some modifications. The dried extract (5 g) was dissolved in 100ml of methanol and then the solvent was evaporated. Twenty milliliters (20 ml) of 2mM H_2SO_4 was added into the residue, mixed thoroughly, and then partitioned with ether. Strong NH_3 solution was used to basify aqueous portion. The alkaloidal fraction was obtained by extracting the basified aqueous fraction with excess chloroform for many times and then the extract was concentrated to dryness. The final alkaloid residue was weighed and the alkaloids content was obtained using the following formula in equation 1.

$$\text{Alkaloids Content (\%)} = \frac{\text{Weight of alkaloids residue}}{\text{Weight of extract}} \times 100 \quad \text{(Equation 1)}$$

2.5.2 Determination of Flavonoids Content

The grapefruits aqueous extract was quantitatively analyzed for flavonoids content using the method described by Harborne [28] with some modifications. The dried extract (5 mg) was heated in 50 ml of 2M HCl solution at 100°C for 25 minutes under reflux. The mixture was allowed to cool and then filtered using Whatman filter paper. The ethyl acetate was added into the mixture in drops and then to the mark 50 ml. The mixture was filtered using filter paper to recover the flavonoid precipitate which was dried in rotary evaporator. The dried flavonoids residue was weighed and the

flavonoids content was calculated using the formula in equation 2.

$$\text{Flavonoids Content (\%)} = \frac{\text{Weight of flavonoids residue}}{\text{Weight of extract}} \times 100 \quad \text{(Equation 2)}$$

2.5.3 Determination of Saponins Content

Saponins presence in the grapefruits aqueous extract was evaluated as described by El-Olemyl *et al*. [29] method with some modifications. The dried extract (5 g) and 150 ml of 50% alcohol were mixed together in 250 ml flask, boiled for half hour and then filtered using Whatman filter paper. The charcoal (1 g) was added to the filtrate and the content was boiled for 30 minutes. The hot mixture was filtered and then allowed to cool at room temperature. For total saponins precipitation, 150 ml of acetone was added into the filtrate and the mixture was filtered using Whatman filter paper. The filter paper was quicklytaken into the desiccator containing anhydrous $CaCl_2$ solution. The saponins residue obtained was concentrated to dryness. The dried saponins residue was weighed and the percentage of saponins in the extract was obtained by the following formula in equation 3.

$$\text{Alkaloids Content (\%)} = \frac{\text{Weight of saponins residue}}{\text{Weight of extract}} \times 100 \quad \text{(Equation 3)}$$

2.5.4 Determination of Steroids Content

The quantitative determination of steroids content in the grapefruits aqueous extract was carried out according to the method described by Evans [30] with some modifications**.** Two miles of sulphuric acid and iron chloride were added into the 10 ml volumetric flask containing 1 ml of the extract. Two miles of potassium hexacynoferrate (III) solution was added into the mixture and then incubated at 70 °C for half hour with constant shaking. The absorbance against the blank was measured at 780 nm wavelength. The steroids content was obtained using the following formula in equation 4.

$$\text{Steroids Content (\%)} = \text{Absorbance of sample} \times 100 \quad \text{(Equation 4)}$$

2.5.5 Determination of Tannin Content

Total tannin content in the extract was analyzed using the AOAC [31] method with certain modifications. Ten milligrams of tannic acid as dissolved in 100 ml water to obtain the tannic acid standard solution. Tannic acid standards (0 – 2.5 ml aliquots) were prepared in 25 ml volumetric flasks. The Folin-Denis reagent (2.5 ml) and sodium carbonate solution (1.25 ml) were added into the flask and then the mixture was made up to the volume. The absorbance was read after 30 minutes spectrophotometrically at 760 nm wavelength. The dried powder extract (1 g) was boiled in 80 ml of water for 30 minutes. The content was allowed to cool and then taken into the 100 ml volumetric flask and diluted to the volume. The mixture was filtered using Whatman filter paper and the filtrate was then tested for tannin content. Total tannin in the extract was obtained using tannic acid standard curve and expressed as TAE/100 g of the extract.

2.6 Determination of Proximate Composition

Proximate contents (moisture, carbohydrate, crude protein, ash and fiber) of the grapefruits aqueous extract were evaluated using the method of AOAC [32]. The tests were conducted in triplicate and data were analyzed and expressed as the mean and standard error in percentage.

2.7 Determination of Antioxidant Content

2.7.1 Analysis of Total Phenolic Content

The total phenolic content of the grapefruits aqueous extract was determined using folic-ciocalteau method as described by Singleton et al. [33] and Abubakar et al. [34], with little modification. The grapefruits aqueous extract (0.5ml) and 2.5ml of 1N folin-ciocalteu reagent were mixed and incubated for 5 minutes at 25 °C. Sodium carbonate (Na_2CO_3,

7.5%, w/v) (2 ml) was added into the content and then diluted with 2ml of distilled water. The contents were incubated at room temperature for two hours. The absorbance was measured spectrophotometrically at 760nm against the blank. The standard curve (2, 4, 6, 8, 10 µg/ml) was constructed using gallic acid as standard and the total phenolic content in the extract was expressed mg GAE/g of the extract.

2.7.2 Estimation of Total Flavonoids Content

The total flavonoids content of the grapefruits aqueous extract was determined using the method of Lamaison and Carret [35] and Abubakar et al. [34]. The experiment was conducted using Quercetin as standard and for constructing the calibration curve. The quercetin (10 mg) was dissolved in 80% ethanol and then diluted to 12.5, 25, 50, 100 µg/ml. The standard solutions (0.5 ml) were mixed with 95% ethanol (1.5 ml), 10% aluminium chloride (0.1 ml), 1M potassium acetate (0.1ml) and distilled water (2.8ml). The grapefruits aqueous extract (0.5 ml) was added into 0.1ml of 10% $AlCl_3$ solution and 0.1ml of distilled water in the blank. The contents were incubated at room temperature for 30 minutes and the absorbance was read at 415nm wavelength against the blank. The data were expressed as mg QE/g of the extract.

2.8 Assessment of Antioxidant Capacities

2.8.1 DPPH Free Radical Scavenging Activity Analysis

Free radical scavenging activity of the grapefruits aqueous extract was determined using 1,1-diphenyl-2-picryl-hydrazyl (DPPH) method as described by Gyamfi et al. [36] and Abubakar et al. [34] with some modification. One mile (1 ml) of 0.3 mM DPPH solution in 100% ethanol was added to 2 ml of the grapefruits extract (50, 100, 200, 400, and 500 µg/ml) in the test tubes. However, the same amount of of 0.3 mM DPPH solution was added to 2 ml of distilled water in the blank tube and 2 ml of the ascorbic acid in the standard tube. The contents were mixed thoroughly, and incubated at room temperature for 30 minutes. The absorbance of the tests and standard against the blank was measured using spectrophotometer at 520nm wavelength. All the experiments were conducted in triplicate, and the results were expressed as the mean of the values. The percentage inhibition activity of the extract was calculated using the following formula in equation 5.

$$\text{Percentage inhibition} = \frac{A0 - A1}{A0} \times 100 \quad \text{(Equation 5)}$$

Where :
A_0 is the absorbance of the control and
A_1 is absorbance of the EAF/standard.

2.8.2 Ferric Reducing Antioxidant Power (FRAP) Assay

The ferric reducing antioxidant properties of the grapefruits aqueous extract was determined using the method of Benzie and Strain [37] with certain modifications. The FRAP solution was prepared by mixing 25 ml of 300 mM acetate buffer pH 3.6, 2.5 ml of 10 mM 2,4,6tripyridyls- triazine (TPTZ) in 40 mM HCl, and 2.5 ml of 20 mM ferric chloride $(FeCl{\cdot}6H_2O)_3$ solution. The freshly prepared FRAP reagent (2 ml) was added into the flask containing 1 ml of the grapefruits aqueous extract at concentrations of 100, 200, 300, 400, and 500 µmol/L. The contents were mixed thoroughly, and then incubated at 37◦ C for 30 minutes. Ascorbic acid was used as standard and for constructing the standard curve. The absorbance of the test was measured using spectrophotometer at 595 nm wavelength. The tests were performed in triplicate and the results were expressed as µmol Fe^{2+} per gram of the extract (µmol Fe^{2+}/g of the extract).

2.9 Statistical Analysis

The tests were conducted in triplicate and the data were expressed as mean ± SEM. The values were analyzed using Statistical Package for Social Sciences (SPSS) Statistics version 22 software (IBM Corp., Armonk, NY, USA). One-way analysis of variance (ANOVA) was used to compute the significant differences between the mean values at confidence level (95%) by Tukey-Kramer multiple comparisons test. The two-tailed (p<0.05) were considered significant.

3 Results and Discussion

3.1 Phytochemicals Composition of the Grapefruits Aqueous Extract

The phytochemicals constituent presence in the aqueous extract of the *Citrus paradisi* fruits is shown in Table 1. The grapefruits aqueous extract revealed the high presence of alkaloids, flavonoids, saponins, steroids, and tannins. The extract also showed the presence of moderate amount of cardiac glycosides, glycosides and phenols. Trace amount of terpenoids was also found in the grapefruits aqueous extract (Table 1).

Table 1 Qualitative Phytochemicals Screening of the Grapefruits Aqueous Extract

Phytochemical	Grapefruits Aqueous Extract
Alkaloids	+++
Cardiac glycosides	++
Glycosides	++
Flavonoids	+++
Phenols	++
Saponins	+++
Steroids	+++
Tannins	+++
Terpenoids	+

+++ = Highly present, ++ = Moderately present, + = Fairly present

Figure 1 shows the percentage composition of phytochemicals in the aqueous extract of the *Citrus paradisi* fruits. The *Citrus paradisi* fruits aqueous extract contains highest significant ($p < 0.05$) amount of flavonoids (29.52 %), alkaloids (26.03 %), tannins (23.43 %) and steroids (15.93 %). The extract also contains 8.26 % proportion of the saponins (Figure 1).

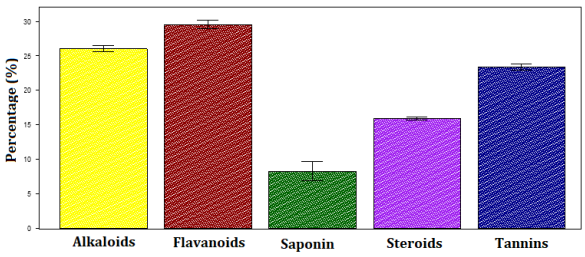

Figure 1 Quantitative Phytochemicals Composition of the Grapefruits Aqueous Extract. Values are mean ± SEM (n = 3)

Many phytochemicals including alkaloids, flavonoids, saponins, steroids, tannins, cardiac glycosides, glycosides, phenol, and terpenoids are found in the grapefruits aqueous extract. Result of the presence research indicated that the grapefruits aqueous extract demonstrated high content of flavonoids, alkaloids, tannins, steroids and saponins. It has been reported that phytochemicals demonstrated pharmacological activities and medicinal properties [38]. Studies showed that flavonoids demonstrated antioxidant properties and pharmacological activities including anti-inflammatory and anticancer activities. Alkaloids have been reported to demonstrate many pharmacological activities. Study by [39] showed that alkaloids isolated from plants extracts exhibited analgesic activity. It has been reported that tannins (polyphenolic compounds) exhibited antioxidant properties and anti-inflammatory and anti-carcinogenic activities [40]. Steroids from the plants serve as precursors for the synthesis of sex hormones and many steroidal drugs including corticosteroid [41]. Saponins are vital source of steroidal hormones and demonstrated hypocholesterolmiae properties [42].

3.2 Proximate Composition of the Grapefruits Aqueous Extract

The proximate composition of aqueous extract of the *Citrus paradisi* fruits is shown in figure 2. The grapefruits extract contains the highest of carbohydrate (89.36 %) and the moisture (74.80 %) content. The extract revealed the moderate amount of lipid (4.56 %), protein (3.36 %) and crude fiber (2.36 %). The extract also contains least amount (0.83 %) of ash content (Figure 2).

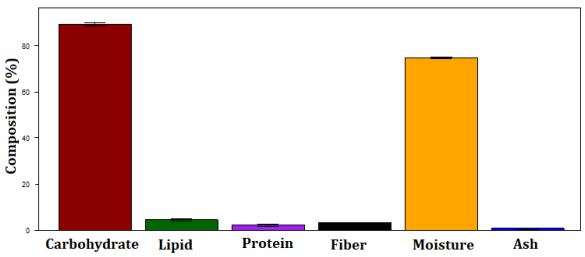

Figure 2 Proximate composition of the Grapefruits Aqueous Extract. Values are expressed as mean ± SEM (n = 3)

Result of the proximate analysis in this study revealed that the grapefruits aqueous extract contains high significant amount of carbohydrate and moisture content. The high carbohydrate and moisture content of the grapefruits extract could be attributed to its high caloric value and short shelf life, respectively. Also, the grapefruits aqueous extract is rich in proteins, crude fiber, lipids, and ash content. Proteins serve vital functions in many biological processes including breast feeding, enzymes synthesis and activities, hormones production and activities [43]. Dietary fiber served as important agent for decreasing high risk of many disorders including coronary heart diseases, obesity, type 2 diabetes and cancer [44]. Ash content is an important factor determining the mineral content of food stuffs [45]. The result of this study showed that the grapefruits aqueous extract might be an important source of minerals. The low lipid content the grapefruits aqueous extract signified that the grapefruits are important in reducing high risk of hypertension and coronary heart disease.

3.3 Total Flavonoids and Phenolic Content of the Grapefruits Aqueous Extract

Table 2 shows the total flavonoids and phenolic content of aqueous extract of the *Citrus paradisi* fruits. The extract contains the highest amount of total flavonoids and phenolics (225.56 and 355.06 mg/g) content, respectively (Table 2).

Table 2 Total Flavonoids and Phenolic Content of the Grapefruits Aqueous Extract

Antioxidant Content	Amount (mg/g)
Total Flavonoids	225.56 ± 0.53
Total Phenolics	355.06 ± 0.44

Data are expressed as mean ± SEM (n = 3)

Flavonoids, the potent antioxidants prevent oxidative cell damage by scavenging free radicals [46]. They have been reported to demonstrate pharmacological activities including anticancer, anti-ulcer activities [46]. Phenolics demonstrated many biological and pharmacological activities including antioxidant, antiulcer, anti-inflammatory, anti-tumour and antidepressant activities [40]. They promote good health due their capacity to scavenge free radicals including reactive oxygen and nitrogen species. The result of the current study indicated that the grapefruits aqueous extract contains significant amount of total flavonoids and phenolics content.

3.4 Antioxidant Capacities of the Grapefruits Aqueous Extract

3.4.1 DPPH free radical scavenging activity of the Grapefruits Aqueous Extract

Figure 3 shows the DPPH free radical scavenging activities of aqueous extract of the *Citrus paradisi* fruits. The DPPH percentage inhibition of radical scavenging activity of the extract and the standard, ascorbic acid increased significantly ($p < 0.05$) with increase in concentration of the solution (Figure 3). At all the concentration values, the extract demonstrated maximum DPPH free radical scavenging activity comparable to that of the standard, ascorbic acid (Figure 3).

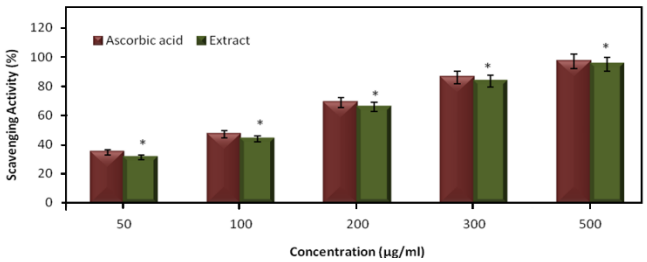

Figure 3 DPPH free radical scavenging activity of the Grapefruits Aqueous Extract. Values are expressed as mean ± SEM (n = 3). *p<0.05 statistically significant when compared with the standard (One-way ANOVA) followed by Tukey's multiple comparison test.

3.4.2 Ferric Reducing Antioxidant Power (FRAP) of the Grapefruits Aqueous Extract

The ferric reducing antioxidant power of aqueous extract of the *Citrus paradisi* fruits is shown in figure 4. The extract and ascorbic acid (standard) exhibited significant ($p < 0.05$) increase in ferric reducing antioxidant power in concentration dependant manner (Figure 4). The extract exhibited maximum ferric reducing antioxidant power comparable to that of the ascorbic acid (standard) at all the concentration values (Figure 4).

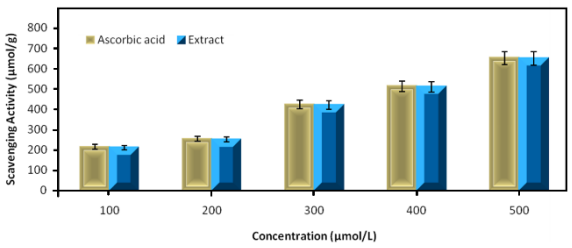

Figure 4 Ferric Reducing Antioxidant Power (FRAP) of the Grapefruits Aqueous Extract. Data are mean ± SEM (n = 3). *p<0.05 statistically significant when compared with the standard (One-way ANOVA) followed by Tukey's multiple comparison test.

The result of this study revealed that the grapefruits aqueous extract demonstrated maximum DPPH free radical scavenging and ferric reducing antioxidant properties comparable to that of the standard (ascorbic acid). This suggested that the grapefruits extract could possess potent free radical scavenging properties. Free radicals damage membrane lipids, proteins and DNA molecules resulting in oxidative stress [47]. Antioxidants protect biological systems against oxidative stress by scavenging free radicals. Natural antioxidants present in plants demonstrate free radical scavenging activities thereby inhibiting or neutralizing the effects of oxidative stress in biological systems [48].

4 Conclusions

The grapefruits extract contains several phytochemicals, proximate nutrients, and with significant amount of total flavonoids and phenolics content. The extract has free radical scavenging activity and ferric reducing antioxidant power. This implies that the grapefruits have nutritional value, antioxidant properties, and potential medicinal uses. Thus, the grapefruits could be consumed for nutritional status, biological functions, and medicinal importance.

5 Declarations

5.1 Acknowledgments

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5.2 Author Contributions

The experimental studies were designed by Ibrahim IklimahBandi and Abubakar Ibrahim. Phytochemical tests were carried out by Ibrahim IklimahBandi, Ibrahim Shehu and Adiya Zainab Sarkin Gobir, Abubakar Ibrahim, Buhari HafsatBature, and Shehu Samira Rara conducted the proximate and antioxidants tests. Data analysis was performed Abubakar Ibrahim.The original draft manuscript was wrote and reviewed by Abubakar Ibrahim. The final draft of the manuscript was read, revised and approved by all the author and co-authors.

5.3 Funding Statement

This research was not supported by any funding sources.

5.4 Conflicts of Interest

The authors declare that no conflict of interest.

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