

Nutritional/Chemical Composition and *In vitro* Antioxidant Properties of the Aqueous Extract of *Launaea taraxacifolia* Leaf Grown in Kwara State of Nigeria

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ABSTRACT

The nutritional/chemical composition and *in vitro* antioxidant potentials of *Launaea taraxacifolia* aqueous leaf extract was elucidated in our present study. Phytochemical screening, proximate analysis, vitamin content and antioxidant capacity of the aqueous leaf extract of the plant were assayed using standard procedures. Result of phytochemical screening shows the plant contains several secondary metabolites, notable among which are: Flavonoids (237.2 ± 0.26), Phenolics (16.83 ± 0.07), Triterpenoids (271.4 ± 0.62) and coumarins (13.97 ± 0.09), while the proximate composition indicates that the extract has high crude protein (24.28 ± 0.10), Crude Ash (15.85 ± 0.08), Crude Lipid (10.84 ± 0.55) and a reduced moisture content (8.97 ± 0.14). Findings from the analysis of the vitamins shows it contains several of the vitamins analyzed with vitamin B₆ (151.00 ± 0.30) having the highest value. The result of the antioxidant capability of *L. taraxacifolia* aqueous leaf extract indicates that it exhibited a concentration dependent potential in terminating the activities of free radicals in all the radical scavenging assay (1, 1, diphenyl 2-picryl hydrazyl (DPPH), Ferric Reducing Antioxidant Power (FRAP), Nitric Oxide, and Hydrogen peroxide) and compared favorably well with the Ascorbic acids used as reference. Hence the findings from this study imply that the aqueous leaf extract of *L. taraxacifolia* possess nutritional and health promoting metabolites that can be utilized both in the food and pharmaceutical industries.

Key words: Proximate composition; Phytochemical Screening; Antioxidant; Free radicals, Vitamins

INTRODUCTION

Medicinal plants have been an important therapeutic aid for alleviating ailments of mankind over the last 2500 years, which is due to their rich huge reservoir of bioactive compounds that has been established by several studies (Tian *et al.*, 2014; Awuchi *et al.*, 2021). These bioactive compounds are synthesized naturally from plants primary metabolites such as amino acids, carbohydrates and lipids, that produced a specific physiological action depending on their chemical constituents in human body and it is been utilized for therapeutic purposes (Borokini and Labunmi, 2017). The high rate of side effects encountered from conventional drugs coupled with the cost involved as drawn attention towards the use of medicinal plants for primary health care and treatment of diseases by most people in recent times especially in developing countries (Jain *et al.*, 2015; Shittu and Akor, 2015; Trevizan *et al.*, 2020). *Launaea taraxacifolia* is a neglected greenish leafy vegetable common among the people in the Western part of Nigeria, also in some part of Senegal, Ghana, Dahomey and Sierra Lone (Adebisi, 2000). It is known as Africa Lettuce or wild lettuce and it is a major part of most Africa diet due to its high level of nutrients. Studies have shown that the leaves contain substantial amount of macro and micro nutrients that can compared well with other conventional edible vegetables (Arawande *et al.*, 2013). The leaves are used as feed to stimulate milk production in cow and also to increase multiple birth in sheep and goat (Sofowora and Evans, 2008). According to a study by Bello *et al.*, 2018 the plant is said to have radical scavenging capacity likewise the ability to inhibits inflammation and over the years it has been stated to house many ethnopharmacological substances that act on disease conditions such as water retention disorders, conjunctivitis, improper bone fixation in infants, and diabetes mellitus (Laleye *et al.*, 2015). However, due to variation in soil morphology and geographical parameters, the leave constituents of plants vary from one region to another. Therefore, the study was thus aimed at investigating the bioactive constituents, nutritional composition and antioxidant potentials of *Launaea taraxacifolia* leaf extract grown in our own geographical location.

MATERIALS AND METHODS

Plant Authentication and preparation

Fresh leaves of *Launaea taraxacifolia* (Wild lettuce) was collected from Ojutaye Ilorin, Kwara State, Nigeria and was authenticated at the Department of Plant Biology, University of Ilorin, Kwara State where a voucher specimen was deposited. The fresh leaves were thoroughly washed, dried at 60^oc and pulverized into powder using an electric blender

Preparation of the aqueous extract of *Launaea taraxacifolia* leaves

A portion (100 g) of the powder was extracted in 1000 ml of distilled water for 48hrs with constant shaking on an orbital shaker. This was then filtered through a Whatmann No. 1 filter paper to obtain the filtrate. The filtrate was concentrated on an electro-thermal water bath to give

a residue of 29.30g which corresponds to a yield of 2.93%. The residue was reconstituted in distilled water to give the required concentrations used in this study.

Determination of the secondary plant metabolites of *Launeae taraxacifolia* leaves

Qualitative analysis of the plant was carried out using the method depicted by Odebiyi and Sofowora (1978) for the detection of saponins, tannins, phenolics, alkaloids, steroids, triterpenes, glycosides and flavonoids.

Quantitative screening of secondary plant metabolite of *Launeae taraxacifolia*

The secondary metabolites present in the plant extract were quantitatively determined using the methods of (Harborne *et al.*, 1998; Obadoni & Ochuko, 2001; Ramamoorthy & Bono, 2007; Jagadish *et al.*, 2009; Olajire *et al.*, 2011; Sazzad *et al.*, 2013).

Proximate composition and vitamin content of *Launeaea traxacifolia* leaves

Proximate composition of the *Launeae taraxacifolia* leaves was analyzed in triplicate in line with the methods stated by the Association of official analytical Chemist (AOAC, 2005). The following parameters were determined: Moisture content, crude lipid, crude protein crude fiber and crude ash. The vitamin composition of the plant extract was analyzed in accordance to the method described by Ayekyaw, (1978).

Determination of In *vitro* antioxidant parameters

Ferric Reducing Antioxidant Power (FRAP)

The evaluation of antioxidant activity is characterized by the calculation of the low pH mixture of iron (III) and 2,3,5-triphenyl-1,3,4-triaza-2-azoniacyclopenta-1,4-dienechloride (TPTZ) reduction to its ferrous form (II) which change the solution into violet-blue color (Benzie and Strain, 1996). FRAP solution (3.6 mL) add to distilled water (0.4 mL) and incubated at 37°C for 5 min. Then the solution was mixed with certain concentration of the plant extract (80 mL) and incubated at 37°C for 10 min. The absorbance of the reaction mixture was measured at 593 nm. For construction of the calibration curve, five concentrations of FeSO₄, 7H₂O (0.1, 0.4, 0.8, 1, 1.12, 1.5 mM) were used and the absorbance values were measured as for sample solutions (Benzie and Strain, 1996; Soto *et al.*, 2019).

In *vitro* DPPH (1, 1, diphenyl 2-picryl hydrazyl) assay

The method depicts by Molyneux, (2004) was adopted in this assay; first and foremost the antioxidant activity was assessed using the stable free radical DPPH. 1ml of different concentration of the extract was added to 1 ml of DPPH, 0.1mM in a test tube. Vitamin C (Ascorbic acid) was used as the standard. 30 min after incubation in the dark at room temperature, absorbance was recorded at 517 nm. The percent DPPH radical scavenging was evaluated using the equation stated below:

$$\% \text{ DPPH radical scavenging} = \frac{(\text{control absorbance} - \text{sample absorbance}) \times 100}{\text{Control absorbance.}}$$

Hydrogen peroxide (H₂O₂) scavenging activity

The hydrogen peroxide scavenging capacity was measured by the method of Jin *et al.* (1996). The hydroxyl radical was produced using a mixture of 1.0 mL of 0.75 mM 1,10-phenanthroline, 2.0 mL of 0.2 M sodium phosphate buffer (pH 7.4), 1.0 mL of 0.75 mM FeSO₄ and 1.0 mL of H₂O₂ (0.01%, v/v). The mixture was incubated at 37°C for 30 min immediately after addition of 1.0 ml sample solution and the absorbance was measured at 536 nm. Deionized water and Ascorbic acids were used as the blank and positive control respectively. The scavenging activity on -OH was calculated using the following equation:

$$\text{Scavenging activity (\%)} = \frac{(\text{Abs}_{\text{sample}} - \text{Abs}_{\text{blank}})}{(\text{Abs}_0 - \text{Abs}_{\text{blank}})} \times 100$$

Where Abs is the absorbance of the deionized water instead of H₂O₂ and sample in the assay system.

NO₂ (Nitric Oxide) Scavenging Activity

The nitric oxide radical scavenging capacity of the fractions was measured using Griess reaction (Sangameswaran *et al.*, 2009). Sodium nitroprusside (2.5 mL, 10 mM) in phosphate buffered saline (PBS) was added to 0.5 mL at different concentrations of the extract. The reaction mixture was incubated at 25°C for 150 min. After incubation, 0.5 mL aliquot was removed and 0.5 mL of Griess reagent: (1% (w/v) sulfanilamide, 2% (v/v) H₃PO₄ and 0.1% (w/v) naphthylethylene diamine hydrochloride) was added. The absorbance was measured at 546 nm. Ascorbic acid was used as reference and similar procedure was used as that of the fractions. Sodium nitroprusside in PBS (2 mL) serve as control. The nitric oxide radicals scavenging activity of the extracts and ascorbic acid was calculated in line with the equation below:

$$\text{Percentage of inhibition} = \frac{(\text{Ao} - \text{A1}) \times 100}{\text{Ao}}$$

where Ao is the absorbance of sodium nitroprusside in PBS (without extracts and ascorbic) and A1 is the absorbance in the presence of the fractions and ascorbic acid.

Statistical Analysis

The results represent the mean of three replicates $\pm$ SEM.

RESULTS

Phytochemical constituents of *Launaea taraxacifolia*

Qualitative and quantitative phytochemical screening of the crude leaf extract of the plant shows the presence of Saponin, Tannins, Phenolics, Flavonoids, Steroids, Triterpenoids and glycosides. Alkaloids were not detected; Triterpenoid and flavonoid were the most abundant in the leaves while Saponin was in trace amount (Table 1).

Table 1. Phytochemical Constituents of *Launaea taraxacifolia* leaf extract

S/N	Secondary plant metabolites	Concentration(mg/100g)
1	Saponins	2.86 ± 0.03
2	Tannins	7.38 ± 0.01
3	Steroids	4.65 ± 0.35
4	Triterpenoid	271.4 ± 0.62
5	Phenolics	16.83 ± 0.07
6	Flavonoids	237.2 ± 0.26
7	Glycosides	7.74 ± 0.02
8	Coumarins	13.97 ± 0.09

Values are means of three determinations $\pm$ SEM

Proximate composition of *Launaea taraxacifolia* leaves

Analysis of the proximate composition of *Launaea taraxacifolia* leaf shows it contains tangible amount of crude protein (24.28 ± 0.10), Crude ash (15.85 ± 0.08) and Crude fiber (12.9 ± 0.10) while the Moisture content (8.97 ± 0.14) and Crude lipid (10.84 ± 0.55) were found to be minimal (Table 2).

Table 2: Proximate composition of *Launaea taraxacifolia* leaves

S/N	Parameter	Composition (%)
1	Moisture content	8.97 ± 0.14
2	Crude lipid	10.84 ± 0.55
3	Crude protein	24.28 ± 0.10
4	Crude fibre	12.98 ± 1.65
5	Crude ash	15.85 ± 0.08

Values are means of three determinations $\pm$ SEM

Vitamin Constituents of *Launaea taraxacifolia* leaf

The vitamin content of the leaf of *Launaea taraxacifolia* shows it contains Vitamin B6 (Pyridoxine), Vitamin E, Vitamin D, Vitamin K, Vitamin B₁₂, Vitamin A and Vitamin C, although Vitamin A and C were found to be in trace amount (Table 3).

Table 3: Vitamin content of *Launaea taraxacifolia* dried leaves

S/N	Vitamins	Concentrations (mg/100g)
1	A	0.04± 0.01
2	C	4.08± 0.02
3	D	30.25± 0.05
4	E	44.5± 3.18
5	K	6.25± 0.25
6	B₆	151.00 ± 0.30
7	B₉	11.30 ± 0.02
8	B₁₂	8.50 ± 0.00

Values are means of three determinations ± SEM

In vitro Antioxidants parameters of *launaea taraxacifolia* leaves

The free radical scavenging activity of *launaea taraxacifolia* is presented in Figures 1 to 4. The graphs show the %inhibition of the plant and standard (BHT) at different concentrations. The graphs also revealed that the leaf extract possess a concentration dependant scavenging activity and compared favorably well with that of the standard.

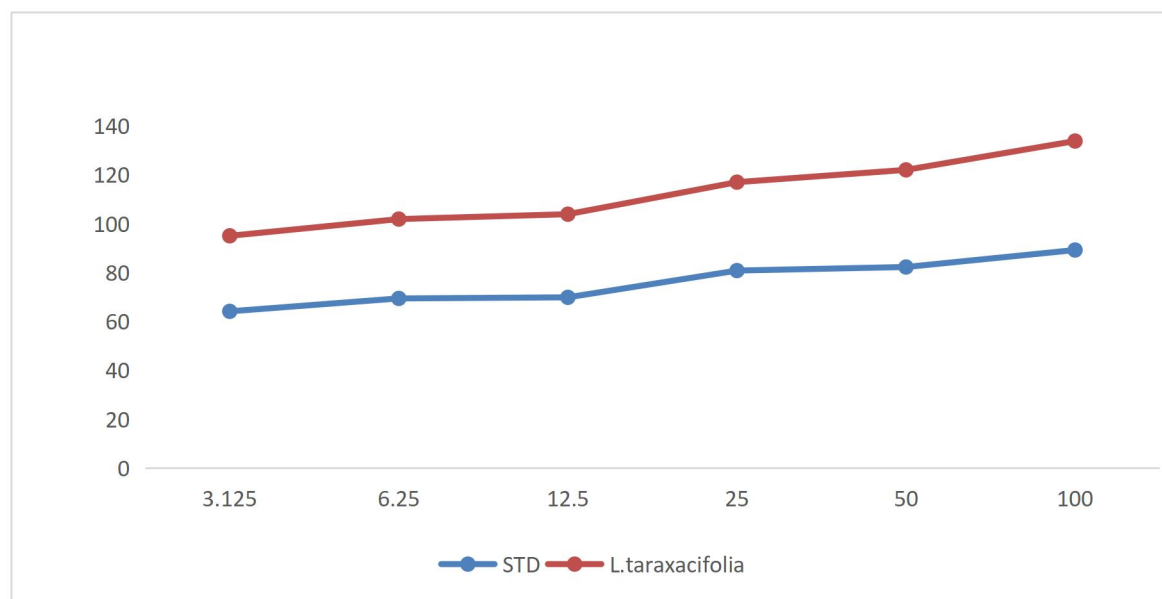


Figure 1: 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging activity of *launaea taraxacifolia*.

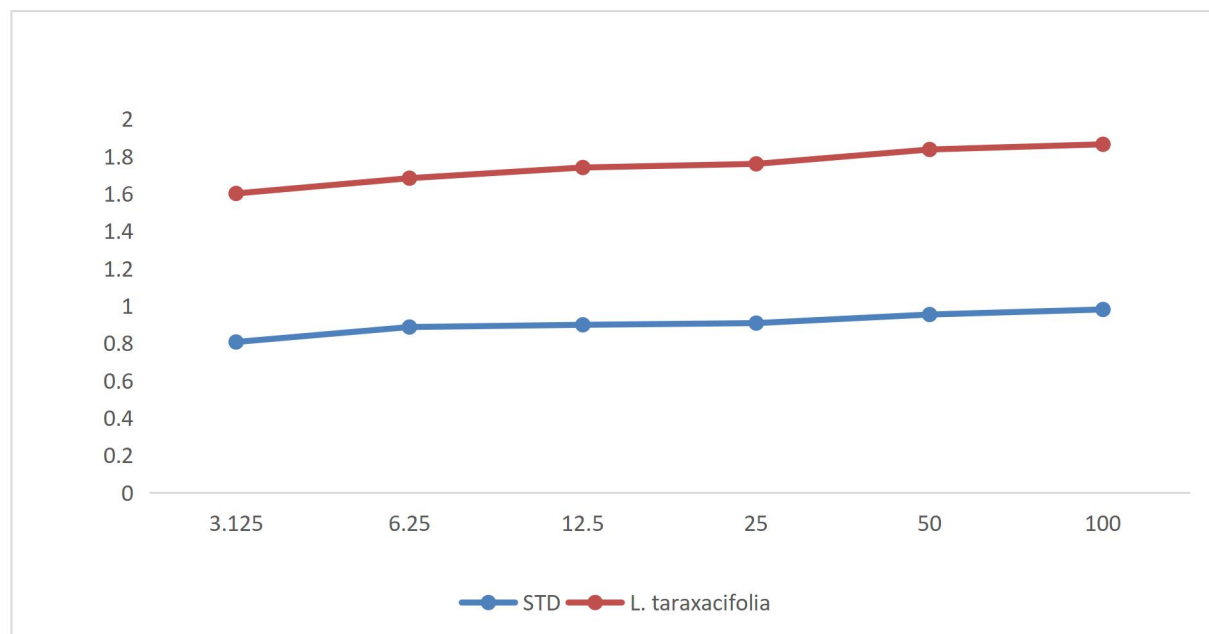


Figure 2: Ferric reducing antioxidant power (FRAP) free radical scavenging activity of *Launaea taraxacifolia* leaves

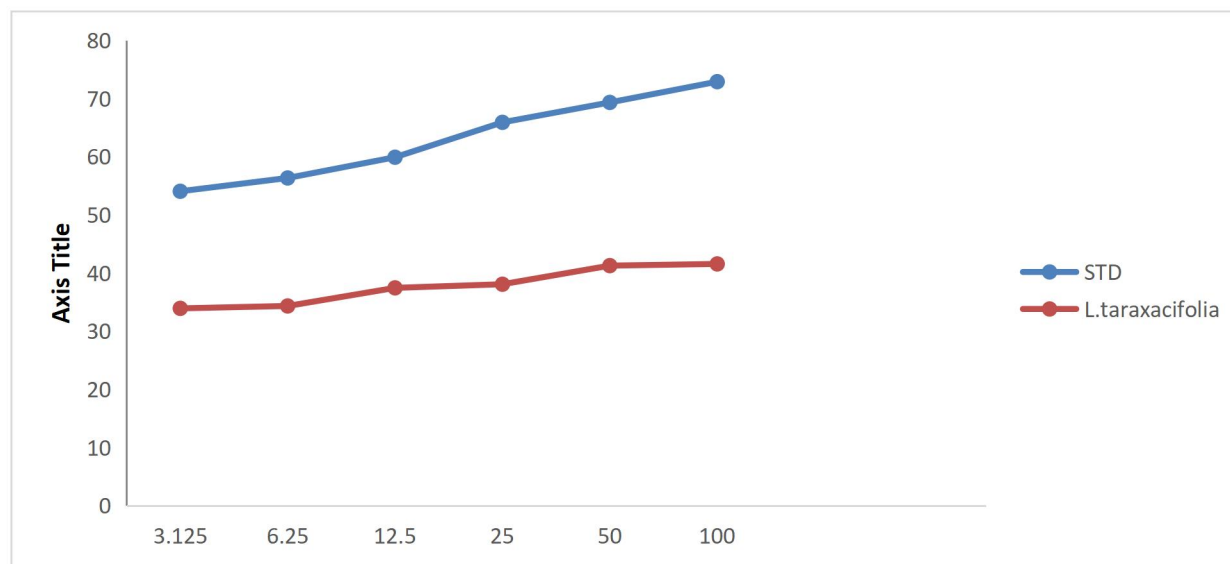


Figure 3: Hydrogen peroxide (H₂O₂) free radical scavenging activity of *launaea taraxacifolia* leaves.

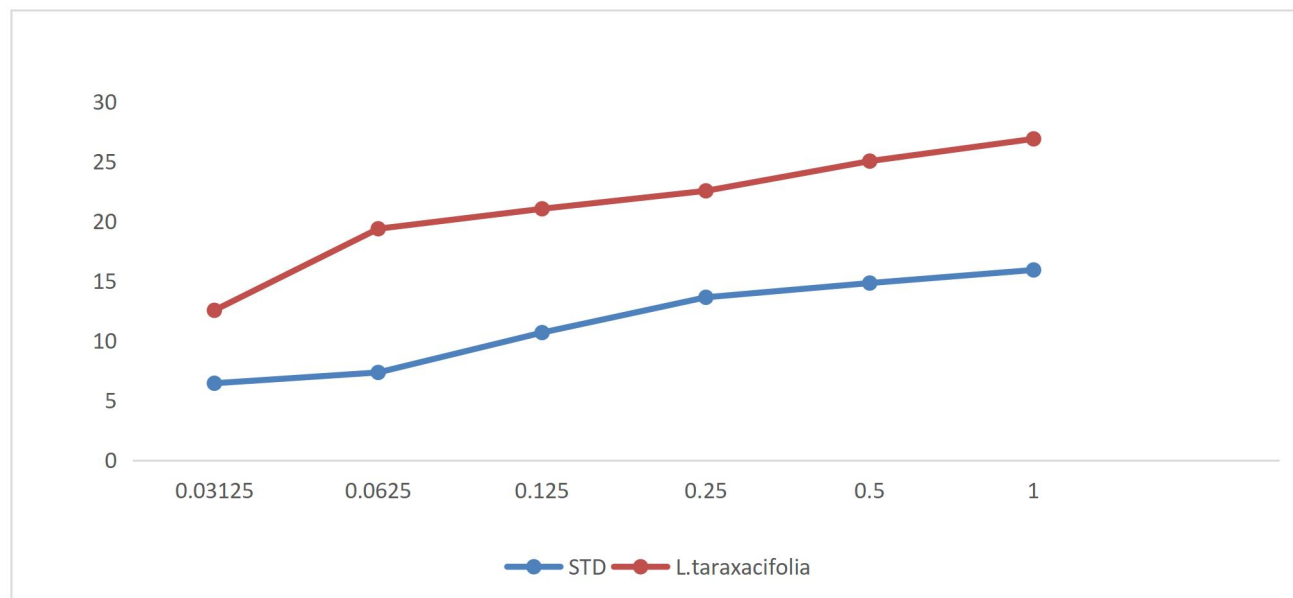


Figure 4: Nitric Oxide (NO₂) Free radicals scavenging activity of *Launaea taraxacifolia* leaves

DISCUSSION

The phytochemical, proximate composition, vitamin contents and antioxidant potential of *L. taraxacifolia* leaves were elucidated in this study. The use of this plant by human as alternative to most frontline drugs in the treatment of diseases has been on from time past, however there is paucity of information on the chemical constituent and biological properties imbedded in this plant. The findings from the phytochemical analysis of the crude plant extract of *L. taraxacifolia* shows it contains several secondary metabolites that are of medicinal values notable among which are: Flavonoids, Phenolics, glycosides, Triterpenoids, and Tannins. This finding is in agreement with that of Arawande *et al.*, 2013 as similar phytochemical were also found in their study on *L. taraxacifolia* leaf extract. The antioxidant and some medicinal properties exhibited by the plant could be ascribed to the presence of these metabolites present in it. For instance, phenolic compound has been proven to possess the ability to terminate free radicals and also possess a strong chain breaking antioxidant (Flora, 2009). Likewise, flavonoids are known to counter the risk of degenerative disease associated with oxidative damage caused by free radicals generated in the course of metabolic processes (Dai and Mumper, 2010; Ghasemzadeh and Ghasemzadeh, 2011; Sangeetha *et al.*, 2016).

The proximate composition of *L. taraxacifolia* (table 2) revealed it contains lesser amount of water although it is higher than those of some common leafy vegetables such as *Xanthosem sagittifolium* (7.7%), *Adansonia digitata* (8.5%), but lower when compared to *Gnetum buchholsonianum* (33.8%), *Vernonia amygdaline* (27.4%) (Ladan *et al.*, 2016) and 76.53% in

Moringa oleifera leaves (Oduro *et al.*, 2008). The value indicates the plant would be stored and preserved for a period of time without any interference with microbial and biochemical spoilage organism or reactions. Moisture content of any food can be used as an indicator of the ability of the plant to stay longer during storage. Water is a suitable medium for most biochemical reactions. Food samples with water content between 12% or more are prone to high biochemical activities and usually have short life (Joslyn, 1970).

The study also shows the plant contains an appreciable quantity of proteins (24.28%). Hence it thus entails that in every 100 g of *L. taraxacifolia* leaves would contain 24.28 g of protein. The crude protein content obtained in this study is higher when compared to most common vegetables such as: water Spinach (6.30%), *Momordica foetida* leaves (4.60%), Balsam apple leaves (11.29%) (Hassan and Umar, 2006), in sweet potatoes leaves (24.85%) (Oduro *et al.*, 2008). However, the crude protein of *Launaea taraxacifolia* leaves was found to be lower when compared to 27.51% in *Moringa oleifera* leaves (Oduro *et al.*, 2008) and 31.00% in *Talinum triangulare* (Akindahunsi and Salawu, 2005). Protein is one of the major classes of food required for the building of body tissue, it is the main source of essential amino acids in the body. Excess consumption of fatty foods has been attributed to various types of disease such as: cardiovascular disease, aging and cancer (Aruah *et al.*, 2011). The crude lipid (10.84%) contained in the plant implies a suitable calorific value and hence possible presence of fat-soluble vitamins that are beneficial to health. The Crude fiber (12.98%) contained in *L. taraxacifolia* leaves indicates it could be a good source of dietary fiber. Fibers are important constituents in diet as it causes distinction in faecal water content, faecal bulk, transit time and removal of bile acids and neural sterols, which lowers the body's cholesterol pool. The percentage ash content (15.85%) contained in the plant leaf implies that it contains some minerals and are possibly a good source of minerals in the diet, nutritionally ash serve as anchor in the metabolism of protein, carbohydrate and fat (Okaka, 2005).

Vitamins are essential component of normal metabolic process and cell regulation, and are required for growth and development; hence their benefits in human diet and animals in general cannot be overemphasized (Khayat *et al.*, 2017). Result of assay of various vitamin constituent of *L. taraxacifolia* shows it contains more of vitamin B₆ (Pyridoxine) with its active form as pyridoxal phosphate it functions as coenzymes for many reactions in amino acids metabolism, other vitamins of importance present in the plants include: Vitamin E, A and C although they are found in trace amount. It thus indicates that *L. taraxacifolia* can be a good source of vitamins in human diet.

Antioxidant are known to obstruct the activities of free radicals, these properties in-turn helps in preventing the progression of most oxidative stress mediated disorder which arise due to the presence of these radicals (Rangasamy and Numasivayam, 2014; Dose *et al.*, 2016). The ability of the leaf extract of *L. taraxacifolia* to scavenge free radicals was assayed using DPPH, FRAP,

H₂O₂, NO radical scavenging assayed. Findings from the study shows the extract of *L. taraxacifolia* exhibits a concentration-dependent potentials to terminate the activities of free radicals in all assayed which matched favorably well with that of Ascorbic acid (Standard) used as reference. The capacity of the extract to exhibit such action is often link to their potential to terminate the reaction process of stable or highly reactive free radicals (Krishnaradu *et al.*, 2009). These properties are attributed to the presence of some secondary metabolite imbedded in the plant extract such as phenolic and flavonoids, this agrees with the findings of Adinortey *et al.* 2012; Borokini and Labunmi, 2017 whose study also confirmed the antioxidant effect of *L. taraxacifolia* against nitric oxide (NO), 2,2-diphenyl-1-picrylhydrazyl (DPPH), and hydroxyl (OH) radicals. These activities were reported to be significant due to the high content of phenolic acids and flavonoids detected in the leaves. Thus *L. taraxacifolia* can be included among the list of plants with antioxidant potentials and highly nutritional constituents that can be utilized by the food and pharmaceutical industries.

Conclusion

The results of this study revealed that aqueous extract of *Launaea taraxacifolia* leave contain certain nutritional and phytochemical constituents that are required for normal metabolic functions of the body. The leaves of the plant also possessed high antioxidant activity which might be helpful in preventing or slowing the progress of various oxidative stress related disorders and therefore could serve as a suitable raw material for the food and pharmaceutical industries.

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