OPEN ACCESS JOURNALS
Indiana Journal of Agriculture and Life Sciences
Abbriviate Tittle- Ind J Agri Life Sci
ISSN *(Online)*- 2583-1623
Journal Homepage Link- https://indianapublications.com/journal/IJALS
DOI- https://doi.org/10.5281/zenodo.10154132

INDIANA PUBLICATIONS
PRODUCTIVE AND QUALITY RESEARCH

Research Article

Volume-03|Issue-06|2023

ASSESSMENT OF PROXIMATE COMPOSITION AND ANTIOXIDANT POTENTIALS OF DATE PALM SEED (*Phoenix dactylifera* L.).

Chukwudi, P.[1, 2*], Emeka-Nwabunnia, I.[1], Ezeji, E.U.[1], Nsofor, C.A.[1], Nduka, C. A.[1, 3], Korie, S.C.[1]

[1]Department of Biotechnology, Federal University of Technology, Owerri, Nigeria.
[2]Environment & Climate Change Unit, Livelihood Improvement Family Enterprises –Niger Delta (LIFE-ND), Asaba, Nigeria.
[3]National Inland Waterways Authority (NIWA), Owerri, Nigeria.

Article History
Received: 06.11.2023
Accepted: 11.11.2023
Published:16.11.2023

Citation
Chukwudi, P., Emeka-Nwabunnia, I., Ezeji, E. U., Nsofor, C. A., Nduka, C. A., Korie, S. C. (2023). Assessment of Proximate Composition and Antioxidant Potentials of Date Palm Seed (Phoenix Dactylifera L.). *Indiana Journal of Agriculture and Life Sciences*, *3*(5), 1-8.

Abstract: : *Phoenix dactylifera* (Date palm) tree is a plant with unexplored potentials. Its seed otherwise known as pit is considered waste can serve both as nutritional and medicinal purposes. This research evaluated the proximate composition and antioxidant potentials of date palm seed in Nigeria. The proximate analysis was done quantitatively for carbohydrate, fibre, crude protein, ash, moisture and energy contents. The antioxidant activity was determined by the DPPH, NO, H_2O_2 and ferric reducing power. Carbohydrates was significantly higher (43.53%) followed by fiber (30.96%), lipid (9.35%), crude (6.34%), ash (5.23%) and finally moisture (4.60%) while the energy composition was 4978.22 KJ/100g. The free radical scavenging potentials of the extract of *Phoenix dactylifera* seed scavenged in a concentration dependent manner with the highest percentage inhibition occurring at the concentration of 2000 µg/ml for the H_2O_2, DPPH, NO and reducing antioxidant power. The findings of this study have shown that date palm seed can serve nutritional and therapeutic purposes.
Keywords: *Phoenix dactylifera*, Antioxidant, Proximate, Therapeutic.

INTRODUCTION

Since the ancient times, fruits have been integral part of diet consumed by humans. Hence, the promotion of good health and wellbeing of man cannot be complete with fruits due the nutritional value. Date palm fruit (Phoenix *dactylifera* L.) is known to be one of the oldest fruit trees in the world (Marzouk and Kassem, 2011). They are important subsistence crop grown in hot arid regions but marketed across the globe due to its high nutritional and confectionery value (Sanusi *et al.*, 2016). Date tree can tolerate extremely high temperatures and does not wither when cultivated in direct sunlight, thus, it thrives even when grown in the Sahel (Dada *et al.*, 2012). Date fruits are rich in both macro and micronutrients, including vitamins, consequently, their production and consumption have increased all over the world as a major dietary fruit (Kuras *et al.*, 2020).

In Nigeria, date palm is grown mostly in the northern region because of suitable climatic conditions. (Sanusi *et al.*, 2016).

The date fruit is constituted by a fleshy pericarp and a seed (pit). Flesh (the edible part) contains mainly sugars (~81-88%, mostly fructose, glucose and sucrose), but also some dietary fibre (~5-8.5%) and smaller amounts of protein, fat, ash and polyphenols [1]. However, the seeds (byproduct), so called pits, are often used in animal nutrition and can serve as coffee substitute since it is caffeine free.

The use of date seed in fiber-based foods and dietary supplements are suggested due to the excellent content of dietary fiber in the seed. Numerous studies have been conducted to find the benefits of the date palm, either from its fruit or seed, and it has been found that the date palm possesses several highly beneficial properties such as antibacterial, antiviral, antifungal, antioxidant, anti- hyperlidimic activity and hepato-protective activity (Al-Farsi & Lee., 2018). These seeds are also shown to lower the risk of cancer and some cardiovascular conditions as well as to improve the functionality and integrity of the immune system.

The objective of this research was to determine the proximate composition and antioxidant potentials of date palm seeds in order to highlight its application in food and pharmaceutical industrials.

MATERIALS AND METHODS

Sample Preparation

***Corresponding Author:** Chukwudi, P

Date palm fruits (*Phoenix dactylifera*) used for this study was sourced from Asaba market, Delta State, Nigeria. The seeds were isolated from date fruits collected at the full ripen stage. The seeds were washed with running water and air-dried. The date seeds were ground in a sterile milling machine to fine powder and stored in an air-tight container for further use.

Chemicals / Reagents

The chemicals and reagents were of analytical grade.

Determination of Moisture Content

The weight of empty crucible was weighed and recorded (W_1), 5 grams of date seed powder was measured into the crucible and weight of the empty crucible with content (W_3) was recorded. The crucible with the content was placed in an air circulating oven set at 105^0C for 2 hours. At the end of drying, the container and its content was placed in a desiccator to cool at room temperature (A.O.A.C, 2000). With minimum exposure to air, the container and its content was weighed again as (W_2).

Calculation: $$Moisture\ Content\ (\%) = \frac{W3 - W2}{W3 - W1}\ X\ 100$$ Equation 3

Where,
W_3 = weight of container and sample before drying
W_2 = weight of container and sample after drying
W_1 = weight of container alone
W_3-W_2 = weight of moisture
W_3 -W_1 = weight of sample

Determination of Ash Content

The weight of crucible was weighed and recorded; 5 grams of date seed powder was measured into the crucible and recorded the weight and its content (W_2) (A.O.A.C, 2000). The crucible and its content was placed in a muffle furnace, set 500-600^{o}C, till the sample was ashed. Allowed to cool for a while, and placed in a desiccator to cool to room temperature, and weighed and recorded again as (W_3).

Calculation: $$Ash\ Content\ (\%) = \frac{W3 - W2}{W3 - W1}\ X\ 100$$ Equation 4

Where,
W_3 = weight of crucible and ash
W_2 = weight of crucible and sample
W_1 = weight of crucible alone
100 = scaling factor for conversion to percentage

Determination of Fibre Content

A 2.0g of date seed extract was weighed into a conical flask, 100ml of H_2SO_4 was added into the flask and heated for 1 hour, then, the solution will be sieved. The residue was added into the conical flask again mixed with 100ml of 2.5 NaOH, heated for an hour and sieved. Rinse the residue on the sieve with ethanol to stop the reaction between the NaOH and fiber. Rinse again with distilled water (A.O.A.C, 2000). The residue was transferred into an empty crucible and dried in the oven at 105^{o}C, weighed the crucible as (W_1), ashed and weighed again as (W_2).

$$Fibre\ (\%) = \frac{W3 - W2}{W1}\ X\ 100$$ Equation 5

Where,
W_3 = weight of residue after acid/ base agitation
W_2 = weight of ash
W_1 = weight of sample

Determination of Total Lipid Content

The thimble and cotton wool was placed on a tarred analytical weighing balance, then the weight recorded (W_1). 5g of powdered date seed was placed into the thimble and cotton wool and weighed again (W_2). The thimble and cotton and its contents was placed in an extractor and connected the extractor into a heating mantle and fitted the water condenser in place. The extractor process was run for about 6-12 hours. At the end of the extraction, the thimble, cotton wool and its contents was transferred to the oven, set at the evaporation temperature of the extraction solvent (n-Hexane) for about 1-2 hours (A.O.A.C, 2000). Then cooled the thimble, cotton wool and contents in a desiccator to room temperature and quickly weighed again (W_3).

Calculation: $$Total\ Lipid\ Content\ (\%) = \frac{W3 - W2}{W1}\ X\ 100$$ Equation 6

Where,
X% = percentage lipid content
W_3 = weight of thimble, cotton wool and sample after extraction
W_2 = weight of thimble, cotton wool and sample before extraction
W_1 = weight of thimble and cotton wool alone
100 = scaling factor for conversion to percentage

***Corresponding Author:** Chukwudi, P

Determination of Protein Content

A 2g of date seed extract was added into a conical flask, added 20ml of concentrated H_2SO_4. Added a pinch of selenium metal powder, mixed all together and a blue coloration was formed. Measured 7 grams of selenium catalyst into the conical flask and agitated, placed on a heating mantle in a fume cupboard and allowed to heat for 1 hour, a presence of discoloration from black to green or blue was noticed. Allowed to cool and transferred to the kjeldahl distillation using HCl to titrate (A.O.A.C, 2000).

Stage 2: kjeldahl distillation

A 30ml of 4% boric acid was added to a flask and 3 drops modified methyl red indicator to the boric acid. The presence of black to blue to black coloration appeared. Titrated using 0.1ml of HCl until a blue to yellow coloration was observed.

Sample Preparation for In-Vitro Scavenging Activity

The ground date pits was sieved to uniform particle size using 2mm mesh sieve. Soxhlet extraction technique was employed in order to obtain the ethanolic extract using absolute ethanol.

DPPH SCAVENGING ACTIVITY ASSAY

The free radical scavenging activity of the extract was analyzed by DPPH assay using spectrophotometer (Bruce, 2015). Each of the test extract (2ml) at different concentration was mixed with 0.5µm DPPH in 1ml of ethanol in a cuvette, incubated for 30 minutes in a dark room temperature cork before incubation. 1ml of DPPH and 1ml of methanol was added to the negative control. It was read in a spectrophotometer at the absorbance at 517nm. The concentration was performed in triplets and the percentage of antioxidant activity calculated as follows:

$$DPPH\ scavenging\ activity = \frac{Absorbance\ of\ control - Absorbance\ of\ sample}{Absorbance\ of\ control} \quad \text{Equation 7}$$

ASCROBIC ACID

The same concentration of the ascorbic acid was prepared to serve as the control, 2ml of each of the ascorbic acid taken and added to the labeled tube according to their concentration

NITRIC OXIDE NO_2 SCANVENGING ACTIVITY

A 2ml of 10 milimolar sodium nitroprusside in 0.5M of phosphate buffer solution (pH 7.4) was mixed with 0.5ml 0f the extract at various concentrations and the mixture was incubated at 25℃ for 150minutes, from the incubated mixture 2ml was measured out of the extract and added into 2ml sulphallic acid reagent (33% in 20% glacial acetic acid) and incubated at room temperature for 5minutes. 1.0ml of napthyethylenediamedihydrocholride (0.1%w/v) was mixed and incubated at room temperature for 30minutes before measuring the absorbance at 540nm (Amessis-Ouchemoukh *et al.*, 2019).

FOR CONTROL

A 2ml phosphate buffer saline (pH 7.4) and 2ml of sulphallic acid reagent was mixed.

HYDROGEN PEROXIDE SCANVENGING ACTIVITY

This assay was based on the benzoic acid hydroxylation method (Chung, 2017). A solution of hydrogen peroxide (40M) was prepared in phosphate buffer (50M) pH7.4 the concentration of hydrogen peroxide was determined using a spectrophotometer, the extract in distilled water was added to 2ml of hydrogen peroxide in phosphate buffer solution and absorbance at 230nm was determined after 10minutes against a blank solution containing phosphate buffer without hydrogen peroxide. Then the percentage of hydrogen peroxide scavenging was calculated.

FOR CONTROL

A 1ml ascorbic acid was added to 2ml of hydrogen peroxide

REDUCING POWER

Reducing power assay was carried out following the method described by Habibur *et al.* (2013). Different concentration of the extract in 1ml water will be added into the labeled test tube according to their concentration, 2.5ml of phosphate buffer pH 6.6 and potassium ferricyanide (2.5ml, 1%w/v). The mixture will be incubated at 50^0C for 20mins after incubation. 2.5ml of 10% trichloroacetic acid solution was added to each tube and the mixture centrifuge at 3000rpm for 10minutes. Subsequently 5ml of the upper layer solution was mixed with 5ml of distilled water and 1ml of ferric chloride solution (0.1%w/v) and the absorbance was measured at 700nm using a spectrophotometer. Ascorbic acid was used as standard, 2.5ml of phosphate buffer (pH 6.6 was used as a blank solution. Percentage inhibition will be calculated using the formula below.

$$\%\ \boldsymbol{inhibition} = \frac{\boldsymbol{Absorbance\ of\ control - Absorbance\ of\ sample}}{\boldsymbol{Absorbance\ of\ control}} \quad \textbf{Equation 8}$$

Data and Statistical Analysis

The data obtained was analyzed with one-way analysis of variance (ANOVA) using the statistical package for the social science (SPSS) program, version 11 followed by least significant difference (LSD) to compare significance between groups

***Corresponding Author:** Chukwudi, P

RESULTS

The result of the proximate analysis (fig 3.1) of *Phoenix dactylifera* shows a significance difference in yield of proximate analysis (Nutritional and non-nutritional) content of phoenix dactylifera. Carbohydrates was significantly higher (43.53%) followed by fiber (30.96%), lipid (9.35%), crude (6.34%), ash (5.23%) and finally moisture (4.60%). Fig. 3.2. shows the energy composition value of *Phoenix dactylifera* seed to be 4978.22 KJ/100g. The free radical scavenging potentials of the extract of *Phoenix dactylifera* seed scavenged in a concentration dependent manner with the highest percentage inhibition occurring at the concentration of 2000 µg/ml for the H_2O_2, DPPH, NO and reducing antioxidant power.

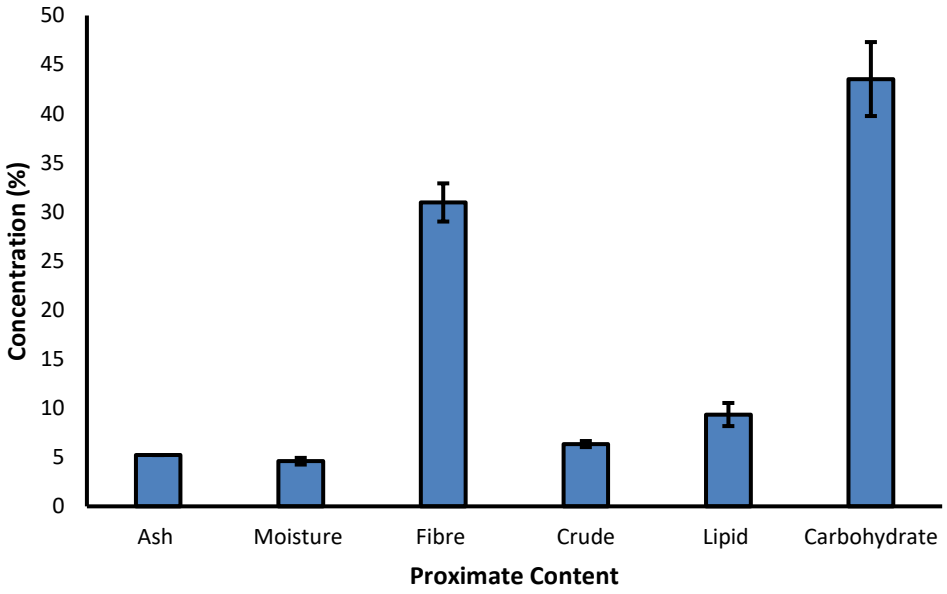

Fig. 3.1: Proximate analysis of *Phoenix dactylifera* seed.

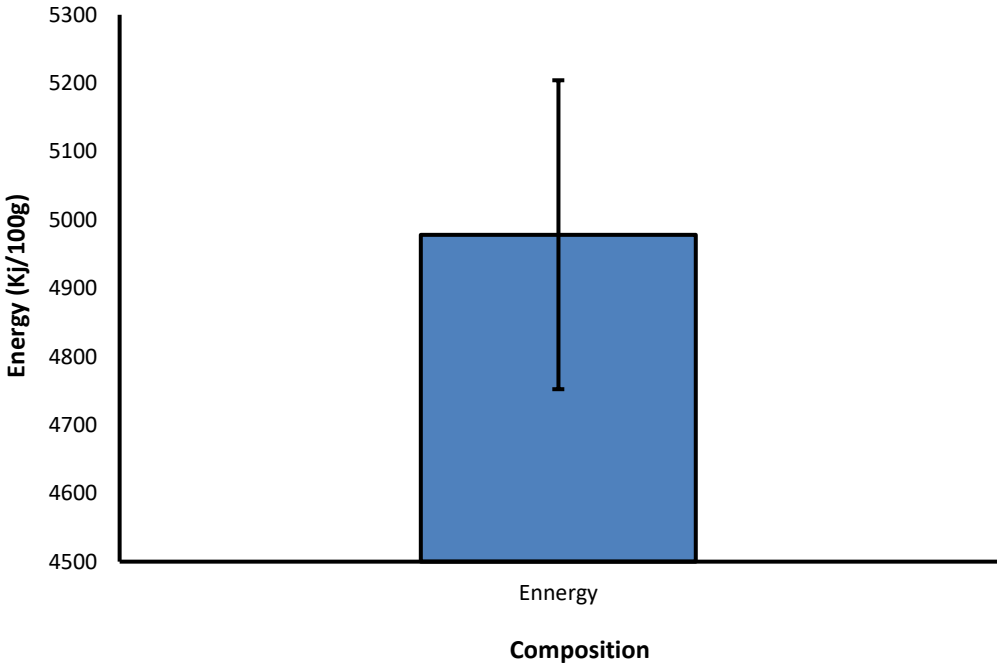

Fig 3.2: Energy composition of *Phoenix dactylifera* seed.

***Corresponding Author:** Chukwudi, P

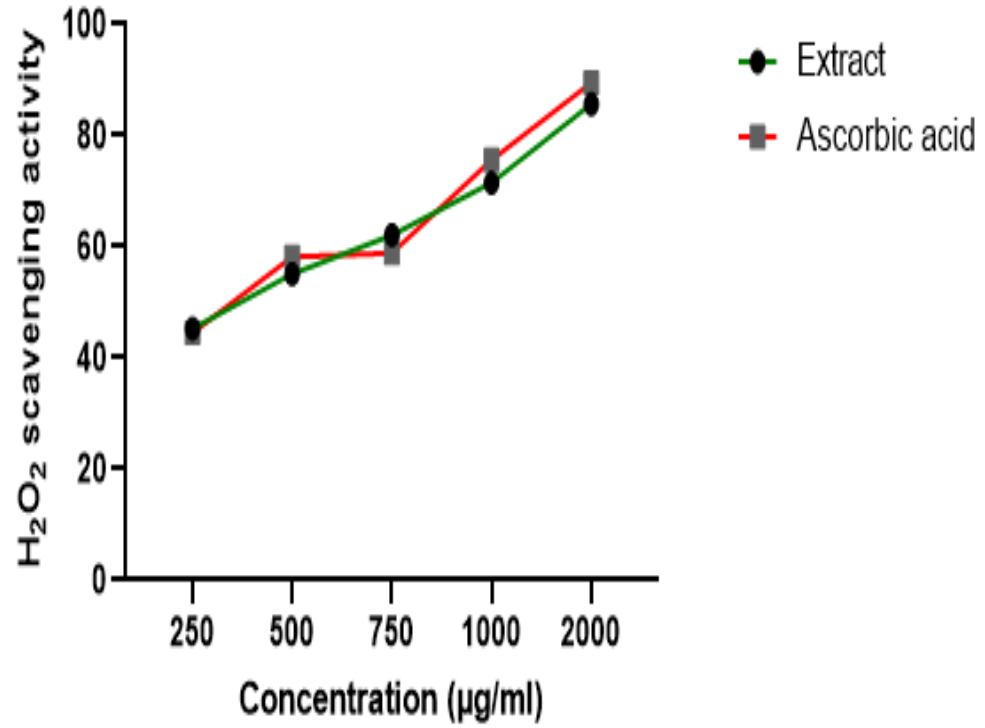

Fig. 3.3: Percentage inhibition of H_2O_2 radical by *Phoenix dactylifera* seed extract

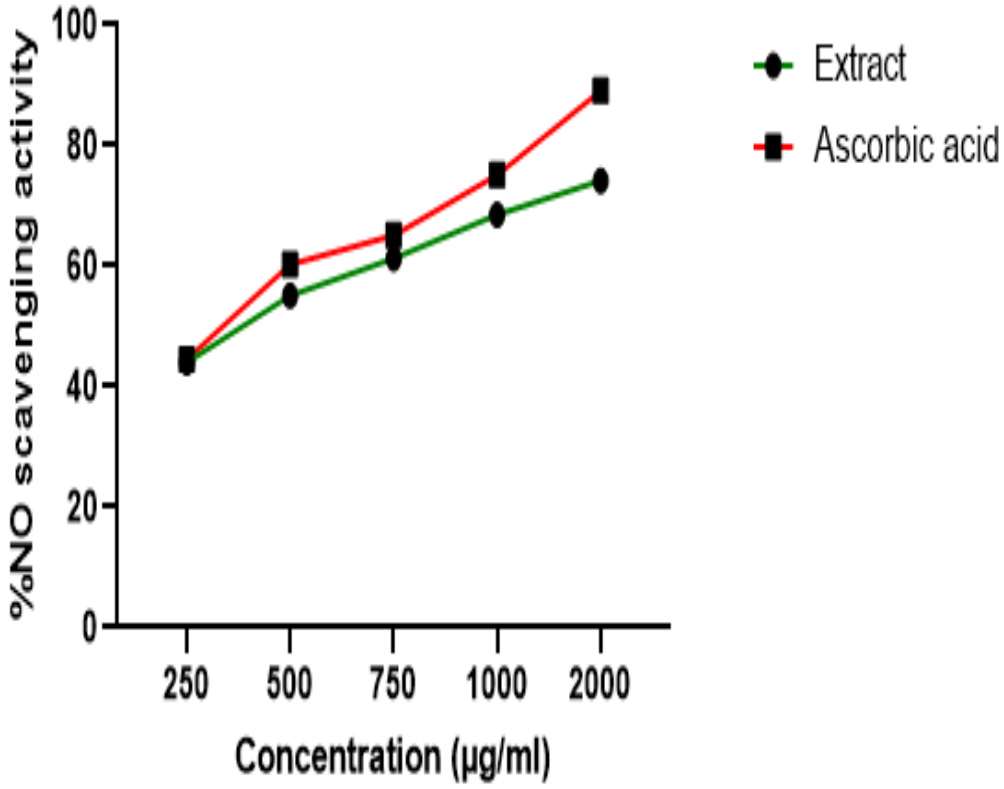

Fig 3.4: percentage inhibition of NO by *Phoenix dactylifera* seed extract

***Corresponding Author:** Chukwudi, P

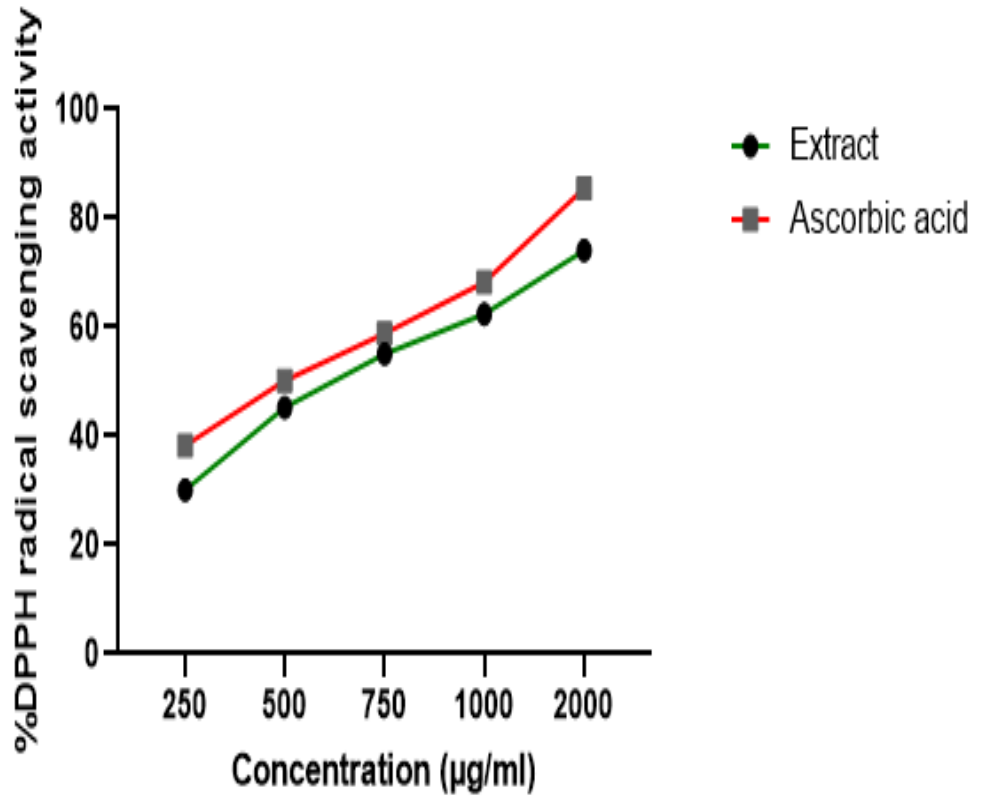

Fig. 3.5: Percentage inhibition of DPPH radical by *Phoenix dactylifera* seed extract

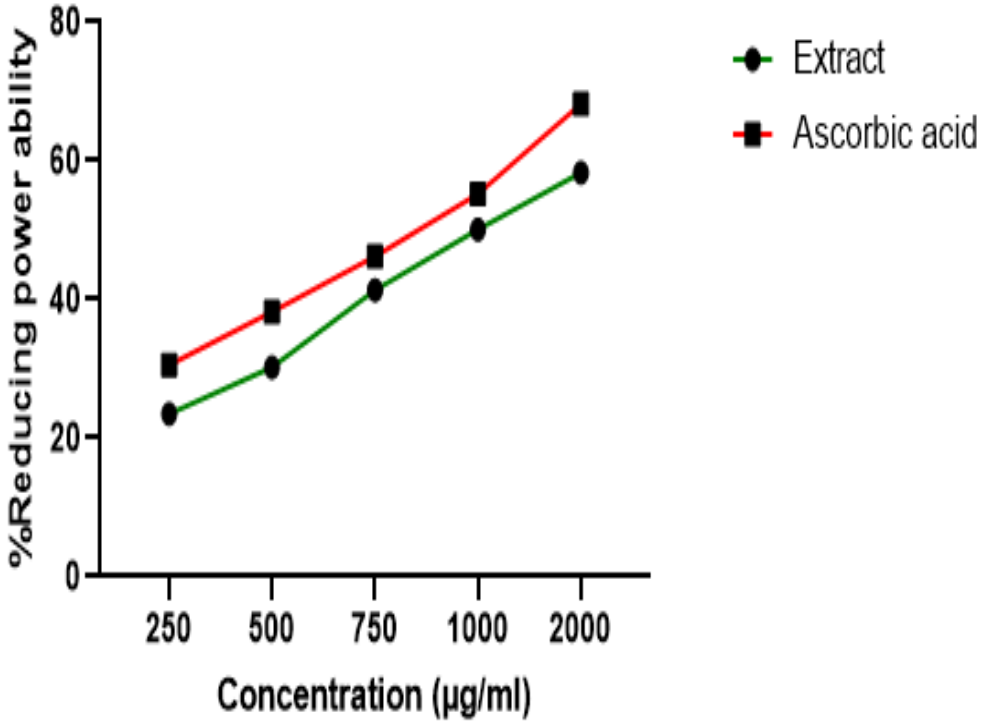

Fig 3.6: Percentage inhibition of reducing power radical by *Phoenix dactylifera* seed extract.

DISCUSSIONS

The result of the proximate analysis of *Phoenix dactylifera* shows a significance difference in yield of proximate analysis (Nutritional and non-nutritional) content of phoenix dactylifera. Carbohydrates was significantly higher (43.53%) followed by fiber (30.96%), lipid (9.35%), crude (6.34%), ash (5.23%) and finally moisture (4.60%). While the energy composition value of *Phoenix dactylifera* seed was shown to be 4978.22 KJ/100g.

The date palm seed examined in this study showed high carbohydrate content, hence, can be considered a good source of energy. Carbohydrate contributes the greatest amount of energy needed by man animals. This probably explains why is considered a staple fruit in semi-arid regions of the globe. Generally, the carbohydrate content of date palm has been reported to largely contain fructose and glucose (Myhara, Karkalas & Taylor, 2009) which are easily metabolized in cell energy production pathways.

The low moisture content observed in this research supports the long shelf-life generally associated with date fruits. Vegetables or fruits with low moisture contents have low susceptibility to microbial degradation and consequently, high durability. This is in consonance with the works of Assirey, (2014); Agboola & Adejumo, (2013), who observed low moisture content in dates. The high fibre content of the seed depicts its importance in animal feed composition as it aids digestion and metabolism in the rumen (Mashadi & Al-Muhammad, 2015).

The lipid content might not be enough for industrial application but has essential oils which can prevent cardiovascular disease (Mashadi & Al-Muhammad, 2015).

The effect of antioxidants on DPPH is thought to be due to their hydrogen donating ability (Baumann, Wurn & Bruchlausen, 2013). Radical scavenging activities are very important to prevent the deleterious role of free radicals in different diseases, including cancer. 1,1,-diphenyl-2-picrylhydrazyl-radical scavenging activity (DPPH) is a stable free radical and accepts an electrons or hydrogen radicals to become a stable diamagnetic molecule the reduction capability of DPPH radicals is determined by the decrease in absorbance at 517nm induced by antioxidants. Data indicate that the palm date seed extract are able to reduce the stable radical DPPH to the yellow- colored diphenylpicrylhrazine. From this study, the date palm seed extract scavenged DPPH radicals in a concentration- dependent manner comparable with the standard ascorbic acid ranging between 250 and 2000µg/ml. palm date seed exhibited a robust antioxidant potential. At the concentration of 2000µg/ml, the palm date seed extract demonstrated a strong free radical scavenging activity having a percentage inhibition of 64.8%. The percentage inhibition of the extract was lower than the standard ascorbic acid. Data obtained from this research showed that the reductive capability of palm date seed compared to ascorbic acid. For the measurement of the reductive ability, the amount of Fe^{2+} complex can be monitored by measuring the formation of color complex at 700nm. it has been investigated from the Fe^{3+} —Fe^{2+} transformation in the presence of tested materials like the antioxidant activity, the reducing power of palm date seed extract increases with increasing concentration, the reducing properties are generally associated with the presence of reductones, which have been shown to exert antioxidant action by breaking free radical chain by donating a hydrogen atom. In this study, the palm date seed have revealed the ability to cause reduction to the ferric ions which is quite comparable to the standard ascorbic acid, at the highest concentration of 2000µg/ml, the standard ascorbic acid with the % inhibition of 70% demonstrated a potent antioxidant effect with the palm date seed extract almost closed to it. Nitric oxide (reactive nitrogen species), formed during their reaction with oxygen or with superoxide, are very reactive. These compounds are responsible for altering the structural and functional behavior of many cellular components. Nitric oxide is also implicated for inflammation, cancer and other pathological condition (Monacade, Palmer & Higgs, 2017). Incubation of sodium nitroprusside resulted in linear time-dependent nitrate production. The results revealed that palm date seed extract have the nitric oxide scavenging activity. In general there was a positive correlation between the palm date seed and the scavenging activity against nitric oxide, date palm seed have the ability to counteract the effect of NO formation and in turn may be of considerable interest in preventing the ill effects of excessive NO generation in the human body. Furthermore, the scavenging activity may also help to arrest the chain of reactions initiated by excess generation of NO that are detrimental to the human health. Free radicals and other reactive specie are constantly generated in vivo both by accidents of chemistry and for specific metabolic purposes. The most important reaction of free radicals in aerobic cells involves molecular oxygen and its radical's derivates, peroxide and transition metals. Reactive specie is thought to play an important role in aging and in the pathogenesis of numerous degenerative or chronic diseases, such as cancer, cardiovascular disease, diabetes and atherosclerosis (Kanayama & Miyamoto, 2007). The scavenging ability of ascorbic acid and palm date seed extract on hydroxyl radical inhibition were done as control. Hydroxyl radical scavenging activity was seen in palm date seed extract, the scavenging activity was increased with increasing the concentration of palm date seed. It was observed that at the concentration of 750µg/ml the palm date seed extract demonstrated a strong free radical scavenging activity, the percentage inhibition of the palm date extract was more pronounced than the standard ascorbic acid. Generally palm date seed extract demonstrated a high antioxidant effects against

***Corresponding Author:** Chukwudi, P

peroxidation of biomolecules to scavenge the hydroxyl radicals.

Oxidative stress occurs when there is an imbalance in the production and neutralization of free radicals which may result in significant damage to cell structure and function. Recently, there has been an increased interest on natural antioxidant of plant origin with strong antioxidant activities. And has been known that free radicals build up in the body and has been associated with many diseases states due to more free radical present than can be kept in balance, and hence the need for antioxidant. Antioxidants are known to repress the buildup of these free radicals or keep them in balance thereby improving health.

CONCLUSION

This study has revealed that *Phoenix dactylifera* (Date palm) seed has abundance of beneficial proximate properties. The high carbohydrate and energy composition can be harnessed by food processing industries to maximize their profit. The antioxidant potentials can as well be utilized in the pharmaceutical industries.

REFERENCE

1. A.O.A.C. (2000). Official methods of analysis. International. (17th Edition).
2. Agboola, O. S., & Adejumo, A. L. (2013). Nutritional composition of the fruit of the Nigerian wild date palm, Phoenix dactylifera. *World Journal of Dairy and Food Sciences*, *8*(2), 196-200.
3. Al-Farsi, M. A. & Lee, C.Y. (2018). Nutritional and functional properties of dates: a review". *Crit Rev Food Sci Nutr* 48: 877–887.
4. Amessis-Ouchemoukh, N., Abu-Reidah, I. M., Quirantes-Piné, R., Madani, K. & Segura-Carretero, A. (2019). Phytochemical Profiling, In vitro Evaluation of Total Phenolic Contents and Antioxidant Properties of Marrubium Vulgare (horehound) Leaves of Plants Growing in Algeria," *Industrial Crops and Products*, 61: 120-129
5. Assirey, E. A. R. (2015). Nutritional composition of fruit of 10 date palm (Phoenix dactylifera L.) cultivars grown in Saudi Arabia. *Journal of Taibah University for science*, *9*(1), 75-79.
6. Baumann, J., Wurn, G. & Bruchlausen, F.V. (2013). Prostaglandin synthetase inhibiting O2 radical scavenging properties of some flavonoids and related phenolic compounds. Deutsche Pharmakologies Gesellschaft abstracts of the 20th spring meeting. *Arc Pharmacol*. 307:1-77
7. Chung, S. K., Osawa, T. & Kawakishi, T. (2017). Hydroxyl radical scavenging effects of speices and scavengers from brown mustard (Brassica nigra). *Bioscience Biotechnolgy and Biochemistry* 61:118-123
8. Dada, M., Nwawe, C. N., Okere, R. A., & Uwubanmwen, I. O. (2012). Potentials of date palm tree to the Nigerian economy. *World Journal of Agricultural Sciences*, *8*(3), 309-315.
9. Kanayama, A., & Miyamoto, Y. (2007). Apoptosis triggered by phagocytosis-related oxidative stress through FLIPS down-regulation and JNK activation. *Journal of Leucocyte Biology*, *82*(5), 1344-1352.
10. Kuras, M. J., Zielińska-Pisklak, M., Duszyńska, J., & Jabłońska, J. (2020). Determination of the elemental composition and antioxidant properties of dates (Phoenix dactyliferia) originated from different regions. *Journal of Food Science and Technology*, *57*, 2828-2839.
11. Rahman, H., Manjula, K., Anoosha, T., Nagaveni, K., Eswaraiah, M. C., & Bardalai, D. (2013). In-vitro antioxidant activity of Citrullus lanatus seed extracts. *Asian J Pharm Clin Res*, *6*(3), 152-157.
12. Marzouk, H. A., & Kassem, H. A. (2011). Improving fruit quality, nutritional value and yield of Zaghloul dates by the application of organic and/or mineral fertilizers. *Scientia Horticulturae*, *127*(3), 249-254.
13. Mashadi, A.S. & Al-Muhammad, M.M. (2015). Physical and chemical characterisation of the major date varieties grown in Saudi Arabia ii. Sugars, tannins, vitamins a and c, *Journalof Arid Plants*, Pp 56-59
14. Moncada, S. R. M. J., Palmer, R. M. L., & Higgs, E. (1991). Nitric oxide: physiology, pathophysiology, and pharmacology. *Pharmacological reviews*, *43*(2), 109-142.
15. Myhara, R.M., Karkalas, J. & Taylor, M.S. (2009).The composition of maturing Omani dates. *Journal of the science of food and Agriculture*; 79:1345-1350.
16. Sanusi M. K, Omokhudu, C. A., Bello, O. G., and Yahaya S. A. (2016). An Assessment of Knowledge Level of Date Palm (Phoenix dactylifera L) Farmers in Dutse Local Government Area of Jigawa State, Nigeria. World Journal of Agricultural Research, 4(2): 36-42.

***Corresponding Author:** Chukwudi, P