

Phytochemical and antioxidant properties of methanolic extracts of pulp, seed, leaf and stem bark of velvet tamarind (*Dialium guineense*) plant

Oluwole-Banjo AK

Department of Biochemistry,
Faculty of Basic Medical Sciences,
College of Medicine of the University of Lagos,
Idi Araba, Lagos, Nigeria

E-mail: drkolabanjo@yahoo.co.uk. aoluwole-banjo@unilag.edu.ng

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Abstract

Velvet tamarind (*Dialium guineense*) is a tall tropical fruit bearing tree belonging to the genus: Leguminosae. It has small edible fruit with brown hard inedible shell. It grows in the savanna region of West Africa and is widely spread in Nigeria. This uncultivated and underutilized plant has been identified to possess several ethnomedicinal and pharmacological properties. However, scientific evidences to prove its effectiveness for most of the claims are unavailable. The present study was carried out to determine the phytochemical and antioxidant properties of the various parts (pulp, stem bark, seed and leaf) of the plant using standard laboratory procedures. The tree sample for this experiment was obtained from Ota, in Ogun State, Nigeria. From the results, the phytochemical composition includes: 25.82 – 48.10% saponin, 7.59 – 23.06% tannin, 12.73 – 17.33% phenol, 9.78 - 25.41% steroid, 12.04 – 47.64% flavonoids, 19.06 – 25.66% terpenoids, 22.35 – 45.63% cardiac glycoside, and 85.36 – 106.21% vitamin C. The four extracts showed a concentration dependent increase in 1, 1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging and nitric oxide (NO) scavenging activities with maxima at 100µg/ml. The values ranged from 71.96 – 77.80% for DPPH and 75.70 – 80.52% for NO relative to ascorbic acid at 83.80% and 85.95% respectively. Velvet tamarind exhibited good phytochemical and antioxidant properties for its acclaimed ethnomedicinal properties. However, further research is necessary on the pulp and seed of this underutilized medicinal plant.

Keywords: Velvet tamarind, Leguminosae, ethnomedicinal plant, antioxidants, phytochemical.

Introduction

There is a rich biodiversity of plants of economic and medicinal importance in the African continent. These plants are good candidates for drug development that can reduce global health expenditure and also meet human health needs. Most global health issues are due to metabolic diseases such as diabetes, cancer, stroke, cardiovascular diseases, etc, which are mainly due to activities of free radicals or reactive oxygen species (ROS). Thus, current research emphasis on healthy living is based on the intake of antioxidants to reduce/combat the effects of free radicals and ROS. Antioxidants are both endogenous (e.g. catalase, glutathione peroxidase) and exogenous (e.g. vitamins and phenolics). Exogenous antioxidants are either synthetic or natural from plants and microbes. Synthetic antioxidants e.g. Butylated hydroxytoluene BHT, Butylated hydroxyanisole BHA, are

expensive and highly toxic compare to the natural antioxidants which are cheaper, readily available and less toxic (Jayaprakash *et al*, 2000; Patel *et al*, 2010).

Free radical and ROS are generated by living cells during respiration and other cellular activities such as defence against microbes and foreign substances in biological systems. They are also generated on exposure to pollutants such as automobile and industrial exhaust, radiations, pesticides, industrial waste, ozone, drugs etc (Masoko and Eloff, 2007). Free radicals have lone unpaired electrons in the outer shell which makes them highly reactive and unstable. Thus, endogenous antioxidants scavenge/quench these free radicals/ROS to prevent them from attacking biomolecules and causing damaging effects in the cell /body (Masoko and Eloff, 2007). Oxidative damage to biological systems occurs when free radicals and ROS produced by biological systems exceeds the scavenging/quenching capacity of the cell's endogenous antioxidants. The excess free radical electrons pair with nucleic acids, amino acid and polyunsaturated fatty acids in DNA, proteins and lipids (in cell membranes and intracellular organelles) respectively. These may result in DNA damage, damage or degradation of protein and induction of lipid peroxidation. This oxidative stress may eventually lead to metabolic disorder such as diabetes mellitus, cancer, cardiovascular diseases, ageing, inflammatory diseases, neurodegenerative diseases etc (Masoko and Eloff, 2007; Hodzie *et al*, 2009; Nagmoti *et al*, 2011; Agarwal *et al*, 2012). Naturally occurring antioxidants in plants are due to the phytochemical constituents (e.g. phenolics, flavonoids) of the plants. These phytochemicals have redox properties and are thus reducing agents, hydrogen donors, singlet oxygen quenchers, free radical scavengers etc (Agarwal *et al*, 2012). Current worldwide research on drug development focuses on plants which possess natural antioxidant capacity in the treatment of metabolic disorders which is rising globally. Various *in vitro* methods of evaluating plant extract antioxidant capacity includes nitric oxide (NO) radical scavenging assay, ferric reducing potential assay, total antioxidant capacity, DPPH radical scavenging capacity etc.

Velvet tamarind (*Dialium guineense*) is a tall tropical fruit bearing leguminous tree belonging to the genus *Dialium*, family Fabaceae, sub family Caesapinioideae. It has small edible fruit with brown hard inedible shell. It grows in the savanna region of West Africa and is widely spread in Nigeria. It is known in western Nigeria (Yoruba) as 'awin', in northern Nigeria (Hausa) as 'tsamiyar or kurm' and in eastern Nigeria Igbo as 'icheku or nkwa' (Akinpelu *et al*, 2011). It is an uncultivated and underutilized plant which has been identified as a nutritional and medicinal plant that could cure a number of illnesses such as cancer, fever, cough, headache, bronchitis, toothache, stomach disorder, liver and gall bladder problems (Odugbemi, 2008). The medicinal and non-medicinal uses of velvet tamarind were extensively reviewed by Besong *et al*, (2016). Though several studies have reported on the ethnomedicinal values of the various morphological parts of the plant, especially the leaf and stem bark, little or no data exist on the phytochemical and antioxidant properties of the plant especially the pulp and seed. The aim of this study was to evaluate the phytochemical composition and antioxidant properties of the various morphological parts of the plant (especially the pulp and seed) and thus provide possible scientific evidence for the reported ethnomedicinal properties of the plant.

Materials and Methods

Sample collection

The samples: ripe fruit, stem bark and leaves were collected from *Dalium guineense* plant in a compound in Ota, Ogun state, Nigeria with coordinates 6°31'26"N 3°12'27"E. The samples were identified at the Herbarium at Department of Botany, University of Lagos. Spoilt and bruised fruits were removed. The whole fruit was washed thoroughly with tap water to remove extraneous materials. The fruits were then air dried. The pulp was then separated from the seed and shell. The seeds were thoroughly rinsed with tap water to remove any trace of pulp. Seed, pulp, stem bark and leaves were all shade dried in open air for three weeks. Air dried samples were then grounded to coarse powder using a manual milling machine. Powdered samples were stored in air tight containers under cool and dry conditions until required for further analysis.

Sample extraction

100gm of air-dried samples were weighed into a container, with 200ml of methanol as extracting solvent using the maceration method. The mixture was filtered after 48 hours using Whatman filter paper. The methanol extract obtained was evaporated to dryness in a rotary evaporator and designated as crude methanol extract.

Phytochemical Screening

The methanolic extracts of the samples were qualitatively and quantitatively screened for the presence of phytochemical constituents. Qualitative screening of the methanolic extracts for the presence of alkaloids, terpenoids, saponins, tannins, flavonoids, cardiac glycosides and phenols were carried out using the procedures of Sofowora (1993), Trease and Evans, (1989) and Harbnone (1973). Quantitative estimation of phytochemical constituents was done using the procedures of Chang *et al*, (2002) for Flavonoids, Obadoni and Ochuko (2001) for Saponin, Van Buren and Robinson (1981) for tannin, Singleton *et al* (1999) for total phenol, Ferguson (1956) for terpenoids, El-Olemy (1994), for cardiac glycoside and Barakat *et al* (1973) for ascorbic acid.

Antioxidant Properties

Nitric Oxide scavenging activity

This assay is based on the principle that sodium nitroprusside in aqueous solution at physiological pH (7.2) generates NO, which reacts with oxygen to produce nitrate and nitrite that is estimated using Griess reagent. Scavengers of NO compete with oxygen leading to reduced production of nitrite ions. In this study, 2ml of sodium nitroprusside in 0.5ml phosphate buffered saline solution was mixed with different concentrations (20-100µg/ml) of methanolic extract of each sample dissolved in methanol and incubated at room temperature for 150 minutes. After incubation period, 0.5ml of Griess reagent was added. The absorbance of the chromophore formed was immediately read at 546nm. Ascorbic acid was used as positive control, (Nabavi *et al*, 2008). Inhibition of nitrate formation by the plant extract and the standard antioxidant ascorbic acid were calculated relative to control. Percentage inhibitions were linearised against concentration of each extract and standard antioxidant. IC₅₀ - Inhibition concentration of each extract required to reduce 50% of the NO formed was determined

DPPH radical scavenging assay

The free radical scavenging activity of Velvet tamarind plant was assessed using the scavenging activity of 1, 1-diphenyl-2-picrylhydrazyl (DPPH) according to method of Mensor *et al*, 2001. Different concentrations of the plant extract (20 – 100µg/ml) were prepared. 0.1mM of DPPH solution was also prepared in methanol solvent. Then 1ml of extract from the serial dilution was added to 2ml of DPPH solution. The DPPH solution was the control. Ascorbic acid was the standard. The mixture was shaken and left to stand in the dark for 30minutes, and the absorbance of the resulting solution was read spectrophotometrically at 517nm. Lower absorbance value for the mixture indicates a higher free radical scavenging potential. The radical scavenging activity of the extract was expressed as percentage inhibition and was calculated using the formula:

$$\% \text{ Inhibition} = [(A_{\text{DPPH}} - A_{\text{extract}}) / A_{\text{DPPH}}] \times 100$$

A_{DPPH} = Absorbance of DPPH at 517nm, A_{extract} = Absorbance of plant extract at 517nm. All tests were performed in triplicates and expressed as mean ± Standard Deviation (SD)

Total Antioxidant Capacity TAC

The total antioxidant capacity of the plant extracts was evaluated spectrophotometrically by the Phosphomolybdenum method (Prieto *et al*, 1999). This assay is based on the reduction of Mo (VI) to Mo (V) by antioxidant compounds and subsequent formation of a green phosphate / Mo (V) complex at acidic pH. 1ml of sample extract was combined with 3ml of TAC reagent solution (0.6M H₂SO₄, 28mM of sodium phosphate and 4mM of ammonium molybdate). The tubes containing the reaction solution were incubated at 95°C for 90minutes. After cooling to room temperature, the absorbance was read at 695nm with a spectrophotometer

against methanol as blank Ascorbic acid was used as the standard. The experiment was performed in duplicate and the mean value was used to express the TAC.

$$\text{TAC} = [\text{Absorbance}/1.224] \times 100$$

IC₅₀ calculations

IC₅₀ defined as the antioxidant concentration required to achieve 50% inhibition. A scatter graph using Excel was plotted, where X axis is the concentration and Y axis is % activity, to obtain the slope of the graph $Y = mX + C$. The value of $X = IC_{50}$ when $Y = 50$.

Statistical analysis

All experiments were performed twice and the results averaged. Data were expressed as mean \pm SD. Levels of significance were determined using ANOVA. Tukey post-hoc test was used to compare column treatment means. $P \leq 0.05$ values were considered to indicate statistical significant difference. IC₅₀ was obtained by linear regression.

Results and Discussion

Table 1 shows the result of preliminary phytochemical screening. Alkaloid was not detected in all the samples analyzed. Saponin and tannin were not detected in the pulp and leaf, but there were high contents of flavonoids and phenols in all the samples analyzed. The seeds however contained all the phytochemicals analyzed. Table 2 shows the result of quantitative evaluation of the phytochemical constituents. The seed had the highest ($p \leq 0.05$) concentration of saponins ($48.10 \pm 0.14 \text{ mg}/100\text{g}$), Tannins ($23.06 \pm 0.12 \text{ mg}/100\text{g}$) and cardiac glycosides ($46.48 \pm 0.22 \text{ mg}/100\text{g}$), while the stem bark had the highest ($p \leq 0.05$) concentration of phenol and flavonoids at $17.34 \pm 0.09 \text{ mg}/100\text{g}$ and $47.64 \pm 0.15 \text{ mg}/100\text{g}$ respectively. The pulp and seed contained the highest ($p \leq 0.05$) concentration of antioxidant vitamin C (ascorbic acid) at $107.12 \pm 1.89 \text{ mg}/100\text{g}$ and $85.45 \pm 2.99 \text{ mg}/100\text{g}$ respectively.

Table 3 shows the NO scavenging activity of the samples. The sample extracts exhibited a concentration dependent increase in NO scavenging activity with maximum values at $100 \mu\text{g}/\text{ml}$. The pulp, stem bark, seed and leaf showed a maximum NO scavenging activity of $75.70 \pm 0.26\%$, $78.17 \pm 0.15\%$, $77.52 \pm 0.25\%$ and $79.35 \pm 0.25\%$ respectively, comparatively lesser ($p \leq 0.05$) than $85.95 \pm 0.40\%$ for ascorbic acid (standard). Regression analysis (fig 1) resulted in IC₅₀ values of $61.55 \mu\text{g}/\text{ml}$, $58.75 \mu\text{g}/\text{ml}$, $38.33 \mu\text{g}/\text{ml}$, $45.16 \mu\text{g}/\text{ml}$ and $34.00 \mu\text{g}/\text{ml}$ for pulp, stem bark, seed, leaf and ascorbic acid respectively.

Table 4 shows the result of the DPPH radical scavenging activity of the sample extracts. The samples showed a concentration dependent increase in percentage inhibition of DPPH scavenging activity with maximum values at $100 \mu\text{g}/\text{ml}$. The pulp, stem bark, seed and leaf showed maximum percentage inhibition of DPPH radical scavenging activity of $71.20 \pm 0.40\%$, $71.96 \pm 0.13\%$, $73.00 \pm 0.39\%$ and $77.80 \pm 0.25\%$ respectively, comparatively lower ($p \leq 0.05$) than $83.25 \pm 0.25\%$ for ascorbic acid (standard). Regression analysis (fig 2) resulted in IC₅₀ values of $50.23 \mu\text{g}/\text{ml}$, $44.02 \mu\text{g}/\text{ml}$, $45.63 \mu\text{g}/\text{ml}$, $33.95 \mu\text{g}/\text{ml}$ and $20.35 \mu\text{g}/\text{ml}$ for pulp, stem bark, seed, leaf and ascorbic acid respectively.

Table 5 shows the result of TAC for the various parts of plant extract studied. The pulp has the highest ($p \leq 0.05$) TAC of $36.85 \pm 0.85 \text{ mgAAE}/100\text{g}$ extract, followed by the seed at $33.15 \pm 0.57 \text{ mgAAE}/100\text{g}$ extract, and leaf at $27.96 \pm 0.22 \text{ mgAAE}/100\text{g}$. The least value of $26.84 \pm 0.27 \text{ mgAAE}/100\text{g}$ extract was for the stem bark which was not different ($p \geq 0.05$)

In recent years, scientists have developed significant interest in studying the medicinal and nutraceutical properties of plants due to their antiradical scavenging and antioxidant properties. There are also claims of non-

toxic property of natural antioxidants compared to synthetic antioxidants, (Maswada 2013). Several works reported on the phytochemical constituents of the stem bark and leaves of *Dialium guineense*: David *et al* (2011), Ogu and Amiebenemo (2012), David and Raphael (2013), and the antioxidant capacity of the methanolic leaf extract (Gideon *et al*, 2013). These reports are in agreement with our result. This study further reports the phytochemical constituents and antioxidant properties of the pulp and seed, which have not been reported. The presence of various phytochemical compounds in the sample extracts may adduce evidence for the reported ethnomedicinal uses of *Dialium guineense* plant, Besong *et al* 2016. The seed has high concentrations of all the phytochemicals under study and may be a good potential for ethnomedicinal use and antioxidant property. These phytochemicals have good curative properties on disease pathogens. The plant stem bark and leaves have been reported to exhibit properties such as antibacterial (Orji *et al*, 2012), molluscicidal (Odukoya *et al*, 1996), antiplasmodial (Bero *et al* 2009; Adumaya *et al* 2013), anti-diarrheal (Gideon *et al*, 2012), anti vibrio (Akinpelu *et al*, 2011), analgesic (Ezeja *et al* 2011), anti-hepatotoxic (Abdulwasiu *et al*, 2014), anti-ulcer (Balogun *et al*, 2013), antimicrobial (Gideon *et al*, 2013), anti-hemorrhoid (Odukoya *et al*, 2009), and oral care (Okwu and Okeke, 2003).

The quantitative analysis of the phytochemicals showed high concentration of saponin (25.82 and 48.10mg/100g) in stem bark and seed respectively. Saponins form foams in aqueous solution (Habu and Ibeh, 2015) and prevent carries and plaque (Okwu and Okeke, 2003), hence the use of the stem bark as chewing stick for oral care. Saponins also have tendency to bind sterols of cell membrane, reduce cholesterol and thus exhibit hypocholesterolemic effects and have tendency to ward off microbes. This may also contribute to the free radical scavenging activity of the seed. There was higher concentration of tannin in the seed than stem bark of velvet tamarind. Tannins are poorly absorbed and have astringent property. They bind protein forming cross-linking that tightens and thickens tissues, and could help to reduce inflammation, internal bleeding and heal wounds. In the digestive canal, tannin alter the permeability of the mucous membrane, reduces the influx of water and decreases water stool, hence it is anti- diarrheal, neutralizing exotoxins produced by diarrhea causing microbes e.g. *E.coli* and *cholera*, (Verhelst *et al*, 2008, Cherubin *et al*, 2016). They also have antioxidant properties. Phenols, flavonoids and terpenoids are present in the various parts of the plant. Flavonoids protect against allergies, inflammation, free radicals, microbes, ulcers, hepatoxins, and tumors etc, (Miller, 1996, Besong *et al*, 2016). Cardiac glycoside has anticancer effect and helps to increase the tone, excitability and contractility of the cardiac muscle, and also helps to exert diuretic effect due to increased renal circulation.

Antioxidant properties of *Dialium guineense* plant was monitored using *in vitro* methods: DPPH radical scavenging activity, Total antioxidant capacity (TAC) and inhibition of NO formation. Nitric oxide is a pleiotropic mediator of physiological process. It is involved in neuronal signaling, inhibition of platelet aggregation, smooth muscle relaxation, regulation of cell mediated toxicity, antimicrobial and antitumor activities. It is also involved in host defense activities (Hazeena and Muthukumaran, 2014). However, overproduction of nitric oxide alongside with superoxide radical contributes to pathogenesis of certain inflammatory diseases (Guo *et al* 1999). Hence NO inhibitors will help to reduce the inflammation and tissue damage seen in inflammatory diseases (Moncada *et al*, 1991). The dose dependent increase in NO inhibition by extracts of *Dialium guineense* plant shows that it contains phytochemicals responsible for inhibiting NO, hence its ethnomedicinal property. The very high NO inhibition values and corresponding low IC₅₀ shown by the seed and stem bark may be due to the high concentrations of saponins and flavonoids in these parts. Boora *et al* (2014), reported that saponin followed by flavonoids were more potent in quenching NO radical. Flavonoids have been reported to exhibit a wide range of biological and chemical activities (Zheng and Wang 2001). A high concentration dependent increase in DPPH radical scavenging activities of the methanolic extract of *Dialium guineense* plant shows that the plant is capable of scavenging free radicals. This may be due to the presence of flavonoids and phenolics in the plant extracts. All the plant part extracts showed highest DPPH radical scavenging at 100ug/ml and associated with low IC₅₀ values. The TAC assay detects antioxidants such as ascorbic acids, tocopherols, carotenoids and phenolics, (Prieto *et al* 1999). The very high TAC value of the pulp and seed extracts may be due to the high value of these constituents

in these extracts compared to the stem bark and leaf extracts. These constituents help to neutralise and absorb free radicals, quench singlet and triplet oxygen (Osawa, 1994). Though several authors have reported linear relationship between total phenolics and flavonoids contents of plant extracts with antioxidant/antiradical capacities, (Katalinic *et al.*, 2006; Wojdylo *et al.*, 2007; Moussa *et al.*, 2011, Maswada 2013), others have reported otherwise (Capecka *et al.*, 2005, Wong *et al.*, 2006)

Conclusion

The data obtained from this study have clearly indicated that methanolic extract of *Dialium guineense* pulp, stem bark, seed and leaves are rich sources of phytochemical constituents that are responsible for the high antioxidant activities, and hence their therapeutic activities in preventing and slowing down oxidative stress related degenerative diseases. This work also reported the phytochemical constituent and antioxidant properties of the *Dialium guineense* seed for the first time, and thus suggests further work on this less utilized seed to harness its high antioxidant potential for ethnomedicinal and nutraceutical purposes.

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Conflict of Interests

The authors declare that there was no conflict of interest.

Tables, Figures and Charts

Table 1: Qualitative Phytochemical Screening of Velvet Tamarind plant.

	Pulp	Stem bark	Seed	Leaf
Saponin	-	+	++	-
Tannin	-	+	++	-
Phenol	+	+	+	+
Flavonoids	++	++	++	+
Terpenoids	+	-	+	+
Cardiac Glycoside	+	+	++	-
Alkaloid	-	-	-	-

+ present, - absent

Table 2: Phytochemical composition of Velvet Tamarind plant

	Pulp	Stem bark	Seed	Leaf
			(mg/100g ±SD)	
Saponin	0.00±0.00 ^a	25.82±0.20 ^d	48.10±0.14 ^c	0.00±0.00 ^a
Tannin	0.00±0.00 ^a	7.59±0.09 ^b	23.06±0.12 ^c	0.00±0.00 ^a
Phenol	13.43±0.08 ^a	17.34±0.09 ^d	12.75±0.09 ^a	15.77±0.26 ^{ab}
Flavonoids	35.44±0.09 ^a	47.64±0.15 ^b	36.60±0.20 ^a	12.44±0.16 ^c
Terpenoids	19.07±0.13 ^a	0.00±0.00 ^d	21.79±0.33 ^c	25.68±0.25 ^d
Cardiac glycoside	22.35±0.16 ^a	24.02±0.39 ^a	46.48±0.22 ^d	0.00±0.00 ^c
Ascorbic acid	107.12±1.89 ^a	23.84±3.97 ^d	85.45±2.99 ^c	21.48±1.99 ^d

a, b, c, d = horizontal means with different superscripts differ (p<0.05) significantly.

Table 3: Scavenging of nitric oxide by the methanolic extract of Velvet tamarind plant

Concentration (µg/ml)	Pulp	Stem bark	Seed	Leaf	Ascorbic acid
20	23.77±0.26 ^a	31.66±0.40 ^b	39.28±0.25 ^c	36.20±0.25 ^d	43.47±1.85 ^e
40	29.20±0.22 ^a	44.96±0.26 ^b	49.80±0.15 ^c	47.48±0.15 ^{bc}	54.08±2.81 ^d
60	47.44±0.40 ^a	60.34±0.39 ^b	65.75±0.40 ^c	58.15±0.26 ^b	60.15±0.35 ^b
80	68.35±0.39 ^a	67.32±0.39 ^a	70.55±0.26 ^a	69.51±0.26 ^a	74.35±1.15 ^b
100	75.70±0.26 ^a	78.17±0.15 ^{ab}	80.52±0.25 ^b	77.35±0.25 ^{ab}	85.95±0.40 ^c
IC ₅₀	23.77±0.26 ^a	31.66±0.40 ^b	39.28±0.25 ^c	36.20±0.25 ^d	43.47±1.85 ^e

a, b, c, d = horizontal means with different superscripts differ (p<0.05) significantly.

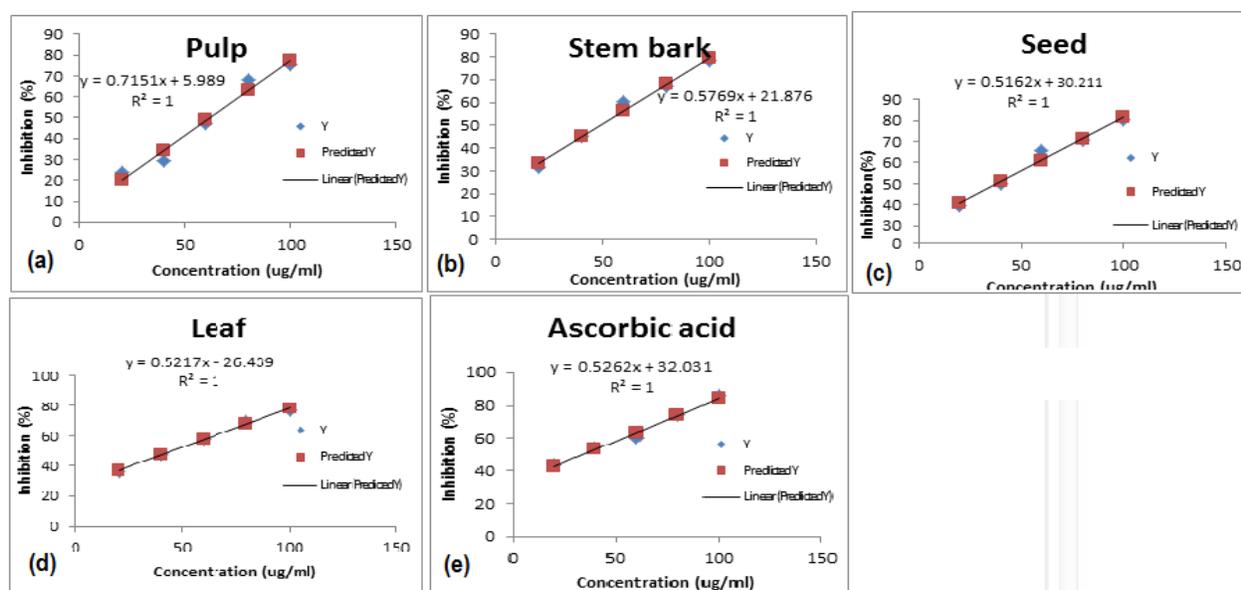
Table 4: DPPH Inhibition by the methanolic extract of Velvet tamarind plant

Concentration (µg/ml)	Pulp	Stem bark	Seed	Leaf	Ascorbic acid
20	35.40±0.26 ^a	39.29±0.25 ^b	36.31±0.38 ^a	43.95±0.25 ^c	46.50±0.25 ^d
40	40.10±0.10 ^a	43.25±0.26 ^b	41.47±0.15 ^a	49.36±0.26 ^c	61.40±0.10 ^d
60	59.70±0.26 ^a	62.80±0.26 ^b	65.15±0.26 ^c	65.65±0.26 ^c	70.20±0.30 ^d
80	67.85±0.13 ^a	69.12±0.39 ^a	71.06±0.26 ^b	72.25±0.13 ^b	74.50±0.15 ^c
100	71.20±0.40 ^a	71.96±0.13 ^{ab}	73.00±0.39 ^b	77.80±0.25 ^c	83.25±0.25 ^d
IC ₅₀	50.23	44.02	45.63	33.95	20.35

Table 5: Total Antioxidant Capacity (Mean ± SD)

Plant part	TAC mg/100g Extract
Pulp	36.85±0.85 ^a
Stem bark	26.84±0.27 ^b
Seed	33.15±0.57 ^c
Leaf	27.96±0.22 ^b

a, b, c = vertical means with different superscripts differ (p<0.05) significantly

Fig 1(a-e). Regression analysis of percentage NO inhibition by *Dialium guineense* plant and ascorbic acid

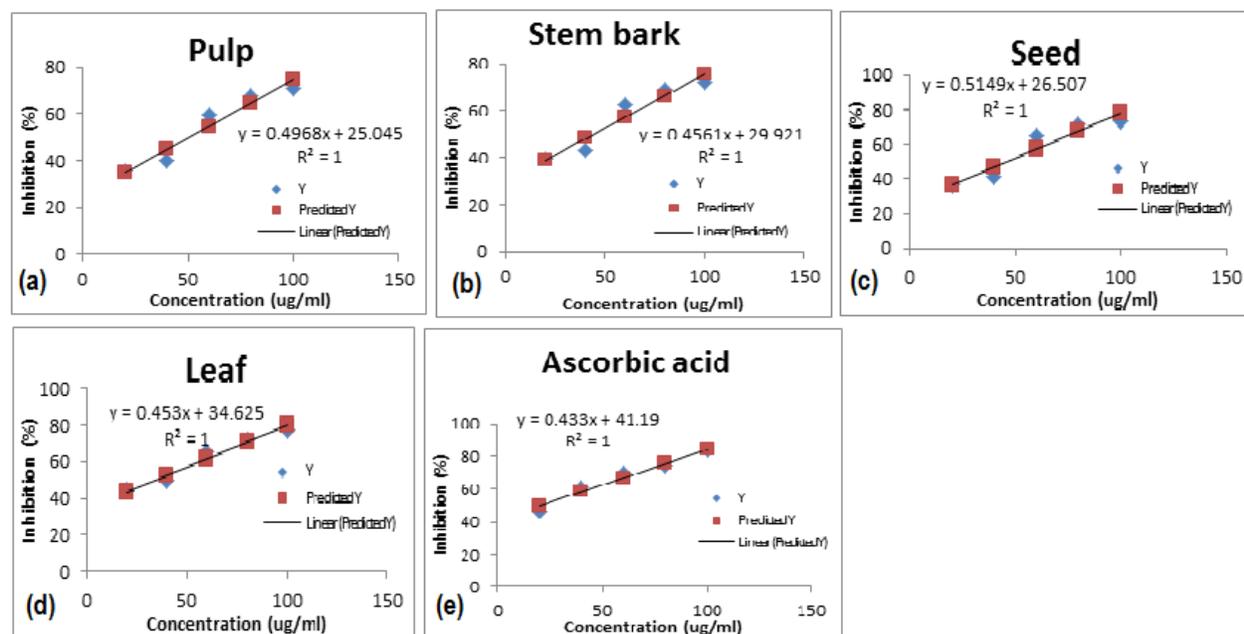


Fig 2(a-e). Regression analysis of percentage DPPH inhibition by methanolic extract of *Dialium guineense* plant and ascorbic acid

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