

## Comparative Analysis of Antioxidants and Phytochemical Properties of Selected Varieties of Mango (*Mangifera indica* L.) Kernel Seeds

<sup>1</sup>Joshua Ndukwe; <sup>2</sup>Dr. Reuben N. Okechi; <sup>3</sup>Okechukwu C. Josephat;  
<sup>4</sup>Emmanuel C. Ebem

Corresponding Authors: [Joshua Ndukwe](#)

**Abstracts:** In Nigeria, the kernel of a ripe mango fruit which is approximately 35-60% of the absolute mango fruit weight still remains a waste and contaminant after the pulp consumption. This could be as a result of lack of adequate information available on the different predominant varieties. Current research was therefore carried out to determine the anti-oxidant and phytochemical properties of four selected varieties of mango kernel seeds (German, Fazli, Cherry and Safeda) using standard techniques. The radicals scavenging activity of the extracts were performed using 1,1-diphenyl-2-picrylhydrazyl (DPPH)-radical scavenging assay, and 2,2'-Azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) (ABTS)-radical scavenging assay, Lipid peroxidation inhibition assay, Super-Oxide Dismutase assay, Glutathione Reductase assay and Catalase assay. Determination of Vitamin C was done using titrimetric method and all compared using standard methods. Phytochemical and biochemical screenings showed the presence of Saponin, Flavonoids, Alkaloids, Phytate, Oxalate, Hemagglutinin, Phenol, Cardiac glycoside, Terpenoids and Tannin. The quantitative analysis result indicated that tannin (3.078%), phenol (8.091mg/kg), and oxalate (0.108mg/100) were present in higher concentration in the seed kernel of Cherry compared to others, while terpenoids (16.00%), cardiac glycoside (8.845%) and hemagglutinin (3.558mg/kg) were present in higher concentration in the seed kernel of German compared to the others. Phytate (0.927mg/100) and flavonoids (5.594%) were higher in safeda whereas saponin (4.00%) and alkaloids (4.781%) were higher in fazli. Ferric Reducing Antioxidant property (FRAP) results showed that ripe mango kernel of Fazli exhibited highest Ferric Reducing power of 61mg/ml concentration and absorbance of 0.510 at wavelength of 593nm using UV- Spectrophotometer. This study therefore showed that different varieties of mango seed kernel contain varying amount of pharmacologically dynamic substances that are valuable and available as antioxidants, potential additives and other therapeutic purposes.

**Keywords:** Mango kernel, phytochemical properties, anti-oxidants, composition, mango seed processing, varietal differences

## 1.0. Introduction

Annually, it's alarming how tones of mango kernels generated from mango fruits after pulp consumption turns to be waste which constitutes environmental nuisance and source of environmental contaminant in Nigeria and the world at large. Mango (*Mangifera indica* L.) Is an invaluable tropical fruit with vast production, worldwide acceptance, wide marketing tendency, extensive distribution for industrial purposes and dynamic relevance to human health. Mango wastes, such as the seed kernel and peel, have high functional and nutritional potential (Torres-Leon *et al.*, 2016). It is one of the most vital edible tropical fruits in the world, because of its pleasant taste and aroma, eye catching colors and refreshing nutritional values (Ibarra *et al.*, 2015). Mango is rich in carbohydrates, water, vitamins, fibre, minerals, and antioxidants (Tharanathan *et al.*, 2006). On the basis of its chemical composition, demand and consumption it is regarded as the king of fruits, a distinction that credits it the second most traded edible tropical fruit worldwide and fifth in total production (FAOSTAD, 2015). The estimated world production of mango per year is 42 million tons; with India emerging the largest producer of mango, having 1,525,000 tons per year, seconded by China, Kenya, Thailand, Indonesia, Pakistan, and Mexico sequentially. Mexico is recorded the largest exporter of ripe mango with estimation of 287,771 tons per year (FAOSTAD, 2015). It's supposed that processing of ripe mango yields approximately 150,000 - 400,000 tons of kernel waste worldwide. Nigeria ranked the world 10<sup>th</sup> largest producer, contributing about 3% to the world total production and it's estimated that after processing, 35%-60% of the total fruit is wasted annually (Torres-León *et al.*, 2016).

Mango leaves are consumed by ruminants, the pulps are consumed by mammals including humans, the back and root is suggested to be medicinal. That leaves researchers with the wise thought that the kernel which accounts 35-60% of the total fruit weight could serve as a non-conventional source of pharmaceuticals, therapeutics and in nutrition industries, animal feed production not left alone. It has raised potential scientific interest because till date it is reported as bio-waste with considerable amount of bioactive compounds such as (phenolic compounds, flavonoids, carotenoids, vitamin C, and dietary fibre) that contributes to animal wellbeing and resuscitates human health (Jahurul *et al.*, 2015). As a result of its proven antioxidant activity, mango kernel is documented to have anticancer properties against breast and colon cancer, antimicrobial activity against Gram positive and Gram negative bacteria (Khammuang and Sarnthima, 2011).

Therefore, incorporating this unconventional amount of waste to animals feed could have an essential and significant space to fill in the scarcity and unavailability problems of animal health. However, mango kernels contain anti-nutrients and toxic components such as tannins, saponins, trypsin inhibitors, lectins, and cyanogenic glycosides which

pose digestibility issues and make them unsafe as carbohydrate and protein in poultry production. Soaking, boiling, leaching, drying and fermentation have been recorded to be effective means to detoxify and drastically reduce the effect of these anti-nutrients and toxic components in mango kernel (Beyene and Araya, 2015).

Aging is one of the inexorable problems related to the degeneration of cell throughout the human lifetime and skin aging become one of the most dermatologic burden since people want to be healthy and look younger despite all synthetic manipulation of the skin. Antioxidants especially natural products are one of the most famous solutions to this problem. Antioxidants are referred to radical scavenger which can either directly scavenge the free radicals or stop chain reaction of the oxidation process. Antioxidant compounds can be both synthetic and natural compounds. The natural compounds mostly from fruits and herbal extracts are currently of interest worldwide. The anti-wrinkle products containing antioxidant ingredients are used expansively because of the ease of application, lower price, and no injury compared to a medical device treatment such as iontophoresis, botox injection, etc.

The mango production in Nigeria is mainly but not limited to Benue, Jigawa, Plateau, Yobe, Kebbi, Niger, Kaduna, Kano, Bauchi, Sokoto, Adamawa and Taraba States (FAO, 2015). Among various Nigerian mango varieties, the most dominant are German, fazli, safeda and cherry. Due to large-scale production, consumption and utilization of mango, large quantity of its kernels is produced on a yearly basis and has the potential to be used in industries for value addition. Presently, no detailed study has been reported on the phytochemicals and antioxidant attributes of mango kernels from the preferred mango varieties in Nigeria. Looking at the functional food, nutritional, and nutra-pharmaceutical potentials of mango kernel seeds, this research therefore carried out a comprehensive analysis of the phytochemicals and antioxidant attributes of kernels of selected Nigerian mango varieties.

## **2.0. Materials and Methods**

### **2.1. Samples collection and processing into Mango kernel.**

Four mango varieties were sourced from Eke-Ukwu Owerri market, in Owerri, Imo State. These varieties were identified as Garman, Safeda, Falzi and Cherry cultivars, by a botanist, Dr. C.M. Duru, of the Department of Biology, Federal University of Technology, Owerri. The ripe mango fruits were washed in a clean running tap for 10 minutes. The peels and pulps were decorticated using stainless steel knife and the seeds dried in hot air oven at 60°C for 5 hours. The seeds were separated and cracked manually to remove the shells and hulls to get the mango kernels. The mango kernels were oven dried (Gallenkamp) at  $60 \pm 1$  °c for 4 hours to a constant weight after which it was ground, sieved and stored in air tight container. Each sample was emaciated in hexane for about 24

hours to eliminate unwanted lipid sand waxes. Whatman filter paper No.1 (pore size 11  $\mu\text{m}$ ), was used to carry out filtration and the solvent was evaporated using rotary evaporator to acquire four concentrated extracts: ripe German extract, Safeda extract, Fazli extract and cherry extract. The extracts were kept in a refrigerator at 4°C in the dark for further investigation. The extractions were performed in triplicate.

## 2.2. Chemicals and reagents Used

All the reagents used in this study, including 1,1-diphenyl-2-picrylhydrazyl (DPPH), 2,2'-Azobis (2-amidinopropane) dihydrochloride (AAPH), linoleic acid, 2,4,6-tripyridyltriazine (TPTZ) and 2,2'-Azino-bis (3-ethylbenzothiazoline-6- sulphonic acid) (ABTS), hydrogen peroxide, pyrogallol, glutathione, phosphate buffer, ammonium hydroxide, dinitro salicylic acid and acetic acid were sourced locally.

### 2.2.1. Phytochemical screening of Mango kernel extracts

The mango kernel extracts were screened for their phytochemical constituents using the methods as described by Trease & Evans, (2002).

### 2.2.2. Determination of flavonoids

Two (2g) grams of the kernel sample was extracted repeatedly with 100ml of 80% aqueous methanol at room temperature. The whole solution was filtered through Whatman filter paper No. 42 (125mm). The filtrate was later transferred into a crucible and evaporated into dryness over a waterbath and weighed to a constant weight

#### Calculation:

$$\% \text{Flavonoids} = \frac{(\text{Weight of Crucible} + \text{Residue}) - (\text{Weight of Crucible})}{\text{Weight of Sample Analyzed}} \times 100$$

### 2.2.3. Determination of alkaloid

Two (2g) grams of the sample was weighed into a 250-ml beaker and 200ml of 20% acetic acid in ethanol was added and covered and allowed to stand for 4 hours at 25°C. This was filtered with filter paper No. 42 and the filtrate was concentrated using a water bath (Mettler) to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was completed. The whole solution was allowed to settle and the precipitate collected and washed with dilute  $\text{NH}_4\text{OH}$  (1% ammonia solution), and thereafter, filtered with pre-weighed filter paper. The residues on the filter paper were titrated with excess  $\text{AgNO}_3$ , washed with 0.02N KCN, and dried in the oven at 80°C.

**Calculation:**

$$\% \text{ Weight of Alkaloids} = \frac{(\text{Weight of filter paper with residue}) - (\text{Weight of filter paper})}{\text{Weight of sample analyzed}} \times 100$$

**2.2.4. Determination of saponin**

Two (2g) grams of the sample was put into 20% acetic acid in ethanol and allowed to stand in a waterbath at 50°C for 24 hours. This was filtered and the extract was concentrated using a waterbath to one-quarter of the original volume. Concentrated NH<sub>4</sub>OH was added drop-wise to the extract until precipitation was completed. The whole solution was allowed to settle and the precipitate was collected by filtration and weighed. The saponin content was weighed and calculated in percentage:

$$\% \text{ Weight of Saponins} = \frac{(\text{Weight of filter paper with residue}) - (\text{Weight of filter paper})}{\text{Weight of sample analyzed}} \times 100$$

**2.2.5. Determination of total phenolic compounds**

One (1g) gram of the sample was boiled with 50ml of ether for the extraction of the phenolic component for 15 min. A portion of 5 ml of the extract was pipette into a 50-ml flask, then 10ml of distilled water was added. Two milliliters (2ml) of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added. The samples were made up to mark with distilled water and left to react for 30 min for colour development. This was then measured with spectrophotometer at 505nm.

**Calculation:**

$$\% \text{ Phenol contents} = \frac{\text{Absorbant of sample}}{\text{Absorbance of standard}} \times \text{Concentration of standard}$$

**2.2.6. Determination of tannin**

Two (2g) grams of the crushed sample in a conical flask was added 100 ml of petroleum ether and covered for 24 hours. The sample was then filtered and allowed to stand for 15 minutes thus allowing petroleum ether to evaporate. It was then re-extracted by soaking in 100 ml of 10 % acetic acid in ethanol for 4 hrs. The sample was then filtered and the filtrate collected. A 25 ml aliquot of NH<sub>4</sub>OH was added to the filtrate to precipitate the alkaloids. The alkaloids were heated with electric hot plate to remove some of the NH<sub>4</sub>OH still in solution. The remaining volume was measured to be 33ml, and thereafter, 5ml of this was pipette and 20ml of ethanol added to it. The mixture was titrated with 0.1M NAOH using phenolphthalein as indicator until a pink end point is reached. The absorbance was measured in UV-spectrophotometer (Gaerenesys 10-S, USA), at 12nm

wavelength with 10 minutes. Blank sample was then prepared and measured at same wavelength. A standard was primed using tannic acid to obtain 100ppm and read.

### 2.2.7. Determination of oxalate

Two (2g) grams of the sample was weighed into a 250-ml volumetric flask and suspended in 190 ml of distilled water. Ten milliliter (10ml) of 6M HCl was added and the suspension digested at 100°C for 1 hr, cooled, and then made up to 250ml mark before filtration. Duplicate portion of 125ml of the filtrate were measured into beakers and 4 drops of methyl red indicator added. This was followed by the addition of NH<sub>4</sub>OH solution (drop-wise) until the test solution changed from salmon pink to a faint yellow colour (ph 4-4.5). Each portion of the duplicate sample was heated to 90°C, cooled and filtered to remove precipitate containing ferrous ion. The filtrate was again heated to 90°C and 10ml of 5% CaCl<sub>2</sub> solution added while being stirred constantly. After heating, it was cooled and left overnight at 25°C. The solution was then centrifuged at 2500rpm for 5minutes. The supernatant was decanted and the precipitate completely dissolved in 10ml of 20% H<sub>2</sub>SO<sub>4</sub> solution. It was made up to 300 ml. Aliquots of 125ml of the filtrate was heated until near boiling and then titrated against 0.05M standard KMNO<sub>4</sub> solution to a faint pink color which persists for 30s.

$$\text{Oxalate} = \frac{T \times (V_{me})(D_f)}{(M_e) \times (M_f)} \times \left(\frac{\text{Mg}}{100}\right)$$

Where T = titre value

V<sub>me</sub>= volume-mass equivalent (i.e 1ml of 0.05M KMNO<sub>4</sub> solution is equivalent to 0.00225g anhydrous oxalic acid). D<sub>f</sub> = Dilution factor (2.4), M<sub>e</sub> = Molar equivalent of KMNO<sub>4</sub>, M<sub>f</sub> = Mass of sample used.

### 2.2.8. Determination of phytate

A portion of 0.2g of the sample was weighed into different 250-ml conical flasks. Each sample was soaked in 100ml of 2% concentrated HCL for 3hours, and then filtered. Fifty milliliter (50ml) of each filtrate was laced in 250-ml beaker and 100ml distilled water added to each sample. Ten milliliter (10ml) of 0.3% ammonium thiocynate solution was added as indicator and titrated with standard iron (III) chloride solution which contained 0.00195g iron per ml.

$$\text{Phytica Acid} = \frac{\text{Titre value} \times 0.00195 \times 1.19}{\text{Weight of sample}} \times 100$$

**2.2.9. Determination of heamagglutinin:**

Two grams (2g) of the sample was added 20 ml of 0.9% NaCl and the suspension shaken vigorously for 1 min. The supernatant were left to stand for 1 hr, and then centrifuged at 2000 rpm for 10min and the suspension filtered. The supernatants in each were collected and used as crude agglutination extract. Absorbance was read at 420nm.

$$\text{Conc. Heamagglutnin} = \frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} \times \text{Conc. of standard}$$

**2.2.10. Determination of cardiac glycoside**

Two (2g) grams of the sample were weighed into beaker and 100ml of distilled water were added. The sample was soaked for 3 hours and filtered to collect the filtrate. To 2ml extract of the sample, was added 1ml of 2% solution of 3,5-DNS (Dinitro salicylic acid) in methanol and 1 ml of 5% aqueous NaOH. The mixture was boiled for 2 minutes (until brick-red precipitate was observed) and the boiled sample was filtered. The weight of the filter paper was measured before filtration. The filter paper with the absorbed residue was dried in an oven at 105°C till dryness and weight of the filter paper with residue was noted.

$$\% \text{ Cardiac glycoside} = \frac{(\text{Weight of filter paper with residue}) - (\text{Weight of filter papaer})}{\text{Weight of sample analyzed}} \times 100$$

**2.2.11. Determination of terpenoids**

Five (5g) grams of sample ( $w_1$ ) was taken and soaked in 100ml of ethanol for 24 hours, filtered and then extracted with 10ml of petroleum ether using separating funnel. The ether extract was separated in pre-weighed glass vials and waited for its complete drying ( $w_f$ ). Ether was evaporated and the yield (%) of total terpenoids contents was measured usinguv-spectrophotometer (gaerenesys 10-S, USA), and calculated as follows:

$$\text{Terpenoids} = \frac{\text{Weight loss}}{\text{Weight of sample}} \times 100$$

**2.3 Anti-oxidant screening of Mango Kernel extracts****2.3.1 Super-Oxide Dismutase (SOD)**

The assay mixture contained 1.2ml of sodium pyrophosphate buffer, 0.1ml of phenazinemethosulfate(PMS), 3ml of nitrobluetetrazolium(NBT), 0.2ml of the sample supernatant and water in total volume of 2.8ml. The reaction was initiated by addition of 0.2ml of NADH. The mixture was incubated at 30°C for 90s. A 1ml portion of glacial acetic

acid was added, shaken vigorously and 4.0ml of n-butanol was added and allowed to stand for 10mins. Its absorbance was measured at 560nm with UV-spectrophotometer (gaerenesys 10-S, USA). One unit (U) of SOD activity was defined as the amount of enzyme causing 50% inhibition of photochemical reduction of nitrobluetetrazolium.

### 2.3.2 Determination of DPPH

Two (2g) grams of the sample was dissolved in 10ml of distilled water. An aliquot of 0.2ml of the solution was pipette and mixed with 1.8ml of 0.1 Mm DPPH solutions in methanol and left in the dark at room temperature for 60mins. The absorbance of the mixture was measured at wavelength of 517nm with UV-Spectrophotometer using methanol as a blank.

$$\text{DPPH} = \frac{A_c - A_s}{A_c} \times 100$$

Where  $A_c$  = Absorbance of control,  $A_s$  = Absorbance of sample

### 2.3.3 Determination of anti-radical scavenging activity

2,2'-azino-bis-3-ethyl benzothiazoline-6-sulphonic acid (ABTS) radical cation was made by reacting 7mM ABTS solution with 2.45mM ammonium persulphate and the mixture was allowed to stand in the dark at room temperature for 12-16 hours before use. Aliquot of 0.5ml of the honey sample was added to 0.3ml of ABTS solution and the final volume was made up to 1ml with ethanol. The absorbance was read at 745nm in UV-Spectrophotometer and percent scavenging activity was calculated with the formula:

$$\% \text{Scavenging Activity} = \frac{A_c - A_s}{A_c} \times 100$$

Where

$A_c$  = Absorbance of control (ABTS radical + ethanol),  $A_s$  = Absorbance of ABTS radical + sample extract

### 2.3.4 Determination of vitamin C

This was determined by the titrimetric method as reported by Kirk & Sawyer (1991). A weighted sample was homogenized in 6% ethylene diaminetetraacetic acid (EDTA) solution. The homogenate was filtered and used for the analysis. Twenty milliliter (20ml) of 30% KI solution was added to the homogenate, followed by 100ml of distilled water and 1ml of 1% starch solution. The mixture was subsequently titrated against 0.1M  $\text{CuSO}_4$  solution. The end point was marked by a black coloration. A reagent blank was also titrated. Vitamin content was calculated based on the relationship below.

1ml of 0.1 mole  $\text{CuSO}_4$  = 0.88Mg vitamin C



$$\text{Vitamin C mg/100} = \frac{100 \times 0.88 \times \text{titre} - \text{blank}}{\text{Weight of sample}}$$

### 2.3.5. Determination of catalase (CAT)

A 20% homogenate of the sample was prepared in phosphate buffer. The homogenate was centrifuged and the supernatant was used for the enzyme assay. H<sub>2</sub>O<sub>2</sub>-phosphate buffer (3.0ml) was taken in an experimental cuvette, followed by the rapid addition of 40µl of enzyme sample and mixed thoroughly. The time required for a decrease in absorbance by 0.05 units was recorded at 240nm in a spectrophotometer (Genesys 10-S, USA). The enzyme solution containing H<sub>2</sub>O<sub>2</sub>-free phosphate buffer served as control. One enzyme unit was calculated as the amount of enzyme required to decrease the absorbance at 240nm by 0.05 units.

$$\text{Catalase} = \frac{\text{Total reaction volume} \times \text{Absorbance of volume}}{\text{Extinction coefficient} \times \text{sample volume}}$$

### 2.3.6 Determination of Glutathione Reductase

A 20% homogenate of sample was prepared in 0.1M phosphate buffer (ph 6.5) from the sample, clarified by centrifugation and the supernatant was used for the assay. To 3.0ml of pyrogallol solution, 0.1ml of glutathione (GSH) was dispensed, 0.1ml of the enzyme extract (sample) was added and the spectrophotometer reading was adjusted to zero at 430 nm. To the test cuvette, 0.5ml of H<sub>2</sub>O<sub>2</sub> was added and mixed. The change in absorbance was recorded every 30 seconds up to 2 minutes in a spectrophotometer.

$$\text{Glutathione } (\mu\text{mol/ml}) = \frac{\text{Absorbance of volume} \times \text{Total reaction volume}}{\text{Extinction coefficient} \times \text{sample volume}}$$

### 2.3.7 Determination of peroxidase

A 20% homogenate was prepared in 0.1M phosphate buffer (ph 6.5), clarified by centrifugation and the supernatant was used for the assay. To 3.0ml of pyrogallol solution, 0.1ml of the enzyme sample was added and the spectrophotometer reading was adjusted to zero at 430 nm. To the test cuvette, 0.5ml of H<sub>2</sub>O<sub>2</sub> was added and mixed. The change in absorbance was recorded every 30 seconds for up to 3 minutes in a spectrophotometer (Genesys 10-S, USA). One unit of peroxidase is defined as the change in absorbance/minute at 430nm.

$$\text{Peroxidase} = \frac{\text{Total reaction volume} \times \text{Absorbance of sample}}{\text{Extinction coefficient} \times \text{sample volume}}$$

### 2.3.7 Ferric Reducing Anti-oxidant Property (FRAP)

Ten (10g) grams of each of the samples were extracted with 50ml of 80% methanol and allowed to stand for 4 hours. Stock solutions used were 300mm acetate buffer (3.1g sodium acetate + 16ml acetone), ph 3.6, 10ml of 10M 2,4,6- tripyridyltriazine (TPTZ) solution, 40ml of 4M HCl and 20ml of 20M ferric chloride. The working solutions were prepared by mixing 25ml acetate buffer, 2.5ml TPTZ and 2.5ml ferric chloride and the temperature of the solution rose to 37°C for 30 minutes. Two (2ml) milliliters of each the extracted samples were pipette into a 10-ml test tube containing 2.8ml of the prepared FRAP reagents and allowed to stand in the dark for 30 minutes, for colour development. Absorbance of the coloured extracts was determined with UV-Spectrophotometer at a wavelength of 593nm. Standard curves were plotted using aqueous solution of ferric chloride at different concentrations, ranging from 20 – 100mg/l, as shown in Table 1 and Fig. 1. FRAP concentration were measured with the equation of the graph.

<b>Table 1: Calibration curve for Ferric Reducing Anti-oxidant Property</b>	
<b>Concentration</b>	<b>Absorbance</b>
0	0.000
20	0.286
40	0.387
60	0.501
80	0.695
100	0.802

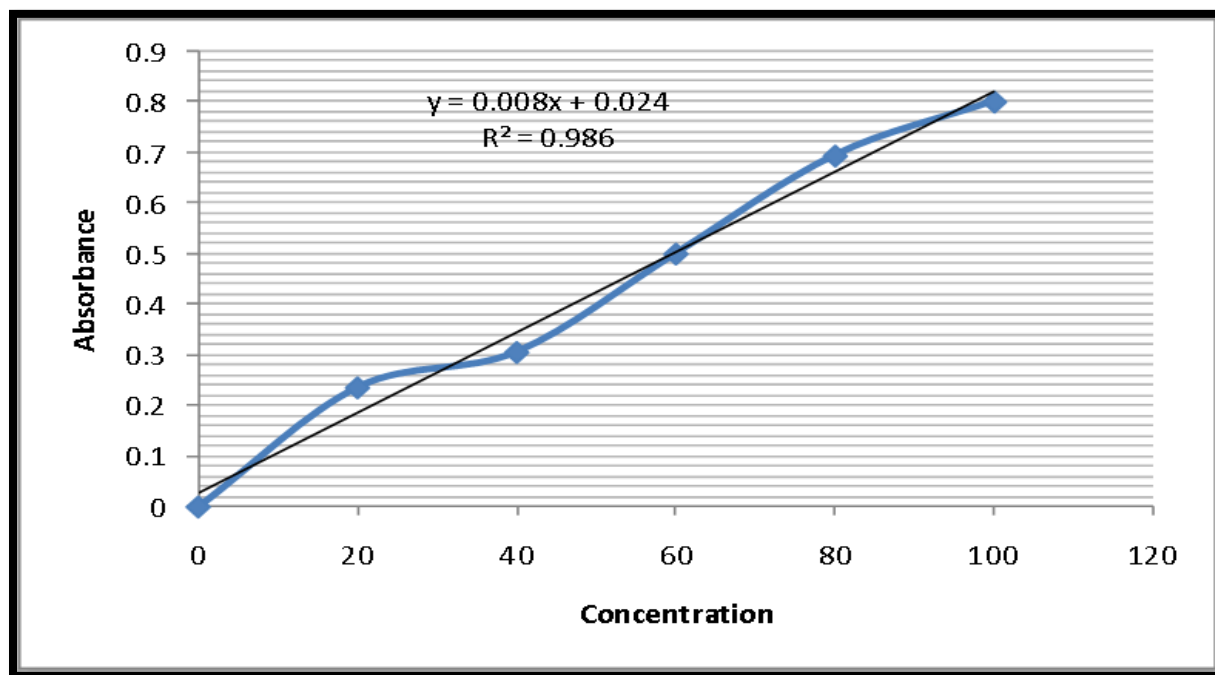


Fig. 1: Standard curves of aqueous solution of ferric chloride at different concentrations

**Equation of curve**,  $Y = 0.008x + 0.024$ , where  $y$  = absorbance of sample

$X$  = concentration

$$X = \frac{y - 0.024}{0.008}$$

0.008

$$\text{Safida} = \frac{0.198 - 0.024}{0.008} = 21.750 \text{ mg/l}$$

$$\text{Cherry} = \frac{0.105 - 0.024}{0.008} = 10.125 \text{ mg/l}$$

$$\text{German} = \frac{0.192 - 0.024}{0.008} = 21 \text{ mg/l}$$

$$\text{Fazli} = \frac{0.512 - 0.024}{0.008} = 61 \text{ mg/l}$$

#### 2.4 Statistical Analysis

The studies were done in triplicate. The results were presented as mean  $\pm$  standard deviation (SD) from three sovereign analyses. Statistical analysis of antioxidant activity was performed by one-way ANOVA using origin, and statistical analysis of stability test was performed by paired T-test, using same program. The same letter indicates that the

difference between the mean values was not statistically significant, but variables with different letters were significantly different at  $P < 0.05$ .

### 3.0 Results and Discussion

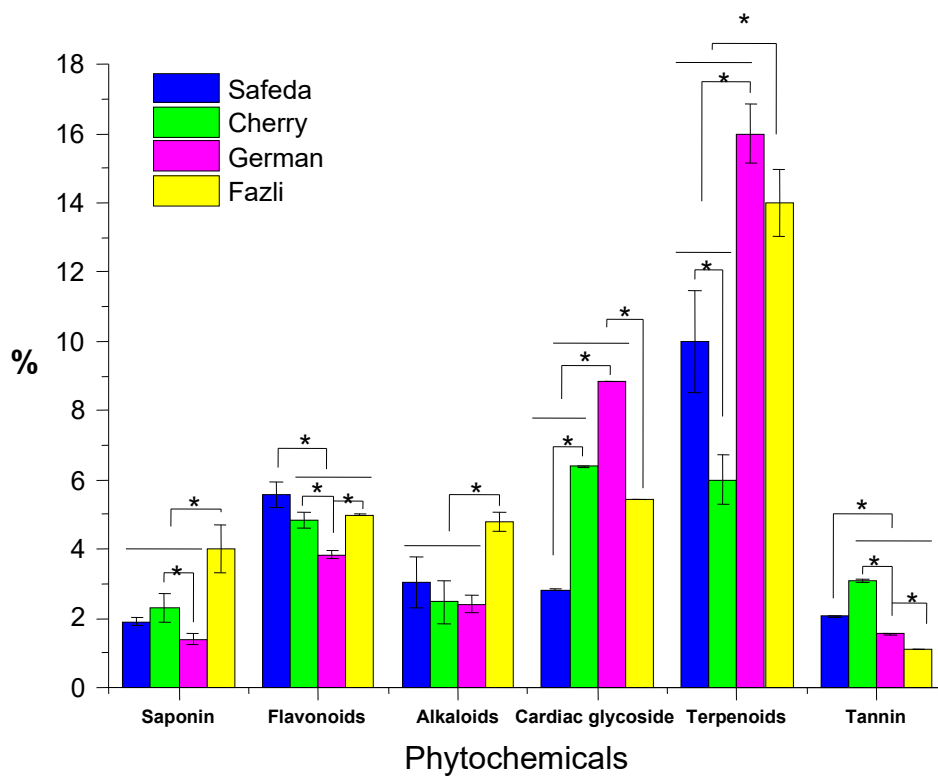


Fig. 2: Analysis of varietal differences in phyto chemical contents of selected mango kernel

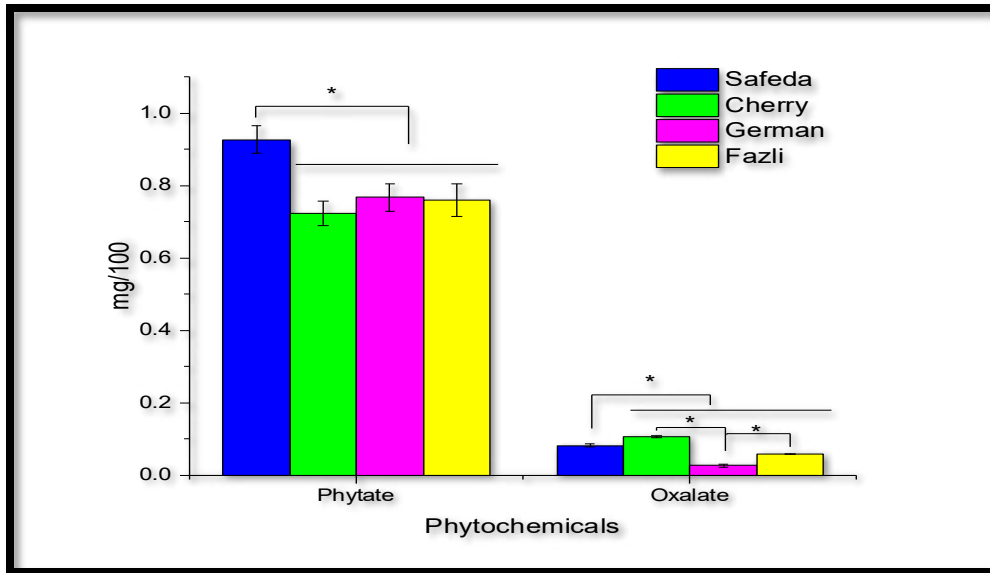


Fig. 3: Comparative result of varietal differences in phytate and oxalate content of selected mango kernel

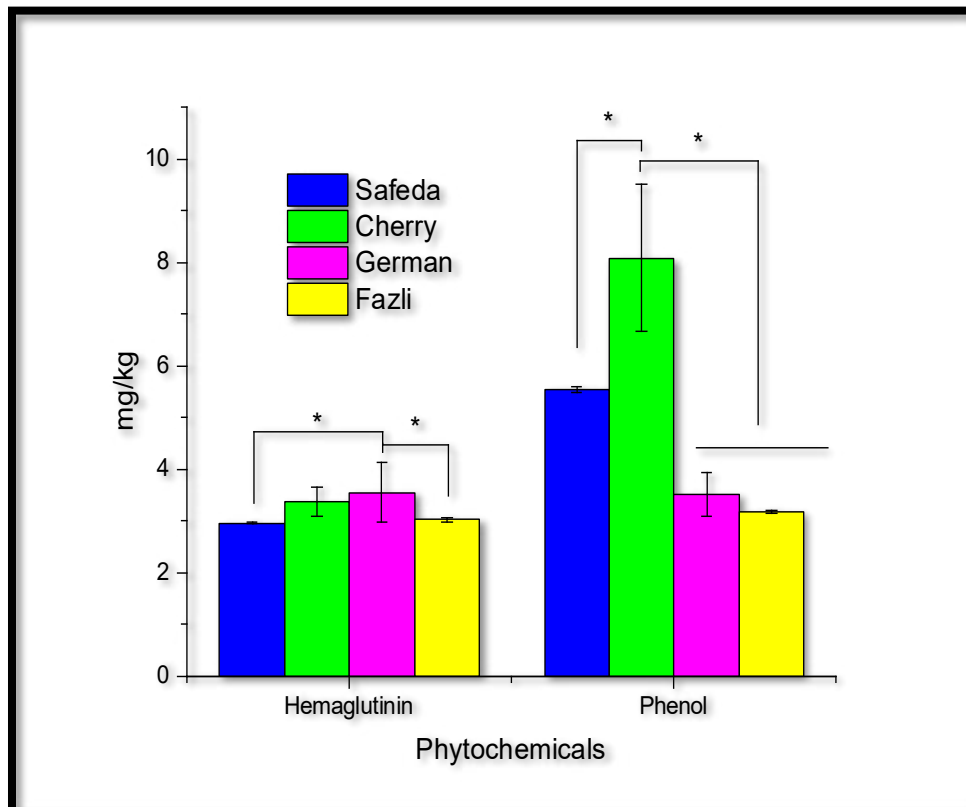


Fig. 4: Analysis of varietal differences in hemagglutinin and phenol content of selected mango kernel

Table 2: Antioxidant properties of different varieties of Mango (*Mangifera indica*) kernel by different methods

	Safeda	Cherry	German	Falzi
<b>Superoxide Dismutase (SOD) (unit enzyme)</b>	0.116±0.023 <sup>d</sup>	0.156±0.023 <sup>c</sup>	0.267±0.024 <sup>a</sup>	0.214±0.020 <sup>b</sup>
<b>DPPH (%)</b>	36.82±2.574 <sup>c</sup>	50.51±2.135 <sup>a</sup>	16.73±1.032 <sup>d</sup>	47.36±1.923 <sup>b</sup>
<b>Anti-radical scavenging activity (%)</b>	21.72±2.432 <sup>c</sup>	41.61±2.77 <sup>a</sup>	20.07±1.513 <sup>c</sup>	32.89±2.673 <sup>b</sup>
<b>Catalase (µmol/ml)</b>	2.1±0.131 <sup>b</sup>	1.8±0.128 <sup>c</sup>	1.5±0.141 <sup>d</sup>	2.5±0.337 <sup>a</sup>
<b>Glutathione Reductase (µmol/ml)</b>	0.514±0.091 <sup>b</sup>	1.135±0.078 <sup>c</sup>	1.215±0.064 <sup>c</sup>	1.916±0.093 <sup>a</sup>
<b>Lipid Peroxidase (µmol/ml)</b>	1.076±0.065 <sup>a</sup>	0.983±0.047 <sup>b</sup>	0.956±0.051 <sup>b</sup>	0.928±0.054 <sup>b</sup>
<b>Vitamin C mg/100</b>	178.879±5.48 <sup>c</sup>	210.358±6.16 <sup>a</sup>	153.039±4.29 <sup>d</sup>	126.708±5.24 <sup>b</sup>
<b>Ferric Reducing Antioxidant Property (mg/ml)</b>	21.75±0.75 <sup>b</sup>	10.125±0.625 <sup>c</sup>	21±0.5 <sup>b</sup>	61±1 <sup>a</sup>

Data are presented as mean ± SD.

Mean values with same letter are not statistically significant from one another, but Mean values with different letters are significantly different from one another ( $P < 0.05$ ).

Scientific awareness of varietal differences in antioxidant and bioactive constituents of mango kernel is essential, not only for therapeutic and nutritional purposes, but also because such information may be of value in discovering good sources of economic materials such as flavonoids, phenols, tannin, heamagglutinin, saponins, and essential oils precursors for the synthesis of complex chemical substances (Gbadamosiet *al.*, 2011). The result from the analysis and quantitative determination of varietal differences in percentage and concentration yields of bioactive constituents of mango seed indicated the presence of tannin, terpenoids, cardiac glycoside, flavonoids, phenols, heamagglutinin, oxalate, alkaloid and phytate (Figs. 2-4). These phytochemicals are believed to play pivotal roles in human

health and exhibit a wide range of biological effects such as effect on cell differentiation, maintenance of DNA repairs, increase detoxifying activities of enzymes, improve in apoptosis of cancerous cells etc, as consequence of their antioxidant properties (Monika *et al*, 2020). The presence of tannin, phenols, and flavonoids, among others suggest the dispersive nature of these phytochemicals present in the seeds of mango. These bioactive compounds have been reported to be free radical scavengers and inhibitors of lipid peroxidation (Gbadamosiet *al.*, 2011). Cherry mango seed contains the highest concentration of tannin, oxalate and phenols as shown in Figs.2 - 4.

The antioxidant properties of tannin, phenolic acids and flavonoids are due to their redox properties, ability to chelate metals and quenching of singlet oxygen, as reported by Kim *et al.*, (2011).

The highest concentration of flavonoids and phytate when compared to other varieties where found in Safida seed (Figs.2 and 3). This suggests that Safida mango kernel can play protective roles such as anti-inflammation, anti-oxidant, anti-viral and anti-carcinogenic properties as reported previously (Liu *et al.*, 2005; Egbuna *et al.*, 2015). They can also play a defensive role against oxidative stress in man and in plants (Tsao, 2010). Alkaloids and saponins were found predominantly in Fazli kernel variety, though it also contains sizeable quantities of the other compounds tested for in this study, as could be seen from the Figures above. The result indicates that Fazli kernel can exert pharmaceutical and therapeutic potentials such as anti-atherosclerotic, anti-diabetic, anti-inflammation, pain relievers, tranquilizers, gastro-protective effect, hepatoprotective effect, hypolipidemic and can stimulate nervous systems, as reported by Egbuna, *et al.*(2015). Hemagglutinin, cardiac glucoside and terpenoids were mostly found in German kernel variety (Figs. 2 and 4). The results obtained in this study thus suggest that the concentrations of bioactive compounds in these varieties of mango kernel seeds and most importantly, the overlooked and abandoned mango kernels are invaluable reservoir of bioactive compounds of potential socioeconomic importance (Choudhary *et al.*, 2023).

In this study, SOD, DPPH, scavenging activity assays, Anti-radical scavenging activity assay, Catalase scavenging assay, Glutathione Reductase, Lipid Peroxidase scavenging activity assays, Vitamin C mg/100 and FRAP activity assays were performed to examine the antioxidant properties of selected mango kernel seed extracts as well as those of other bioactive compounds.

SOD is commonly known as the key cellular antioxidant enzyme that catalyzes dismutation of harmful superoxide anion radical to harmless molecular oxygen and hydrogen peroxide, thereby decreasing its deleterious effect on the cells at high concentration. It's known for its therapeutic and physiological protection against free radicals and reactive oxygen species in plants and animals. It was observed that German kernel possessed the highest antioxidant ability 0.267(unit enzyme) followed by Fazli kernel 0.214(unit enzyme), as shown in Table 2

above.

The DPPH radical scavenging assay is a widely accepted method for estimating antioxidant activity due to its efficiency, easiness, reproducibility and the good correlation between DPPH and antioxidant activity (Latif& Anwar, 2011). The DPPH scavenging activity of mango kernel seeds of various varieties as observed in this study ranged from 16.73% to 50.51%(Table 2). The highest value was observed in Cherry variety (50.51%), followed by Felzi (47.36%), while German variety recorded the least (16.73%). The ability of mango kernel seeds to scavenge for free radicals varied significantly ( $P < 0.05$ ) in relation to different varieties. The values of antiradical scavenging properties range from 20.07% - 41.61% and the anti-radical scavenging activity decreases as follows Cherry >Falzi>Safeda> German which is in consonant with DPPH Scavenging Activity Assay.

The antioxidant agent present in the four selected mango kernel seed extracts scavenged the DPPH free radical by donating hydrogen, which resulted in the formation of the non-radical type of DPPH (Kedare& Singh 2011).Anti-radical scavenging activity is attributed to the manner the test compounds (selected mango kernel seed) scavenge the free radicals. The high Anti-radical scavenging activity found in mango kernel could be attributed to the phytochemicals domicile in all the varieties.

Catalase scavenging assay is a universal test that examines the ability of catalase to detoxify hydrogen peroxide to water and oxygen gas. Catalase is a vital enzyme in protecting the cell from oxidative damage caused by reactive oxygen species. The highest catalase scavenging activity was seen in Falzi variety, while the least scavenging activity was seen in German mango kernel seed variety, with the values 2.5 $\mu$ mol/ml and 1.5 $\mu$ mol/ml respectively. Generally, all the varieties assayed showed appreciable catalase antioxidant properties but the values recorded were significantly different among the varieties tested ( $P < 0.05$ ).

In Glutathione Reductase and Lipid Peroxidase scavenging activity assays, it was noted that Falzi, and Safeda, exhibited highest anti-oxidant scavenging proper with values 1.916 ( $\mu$ mol/ml) and 1.076 ( $\mu$ mol/ml) respectively..

The order of FRAP activities based on the principle of reduction of ferric ion ( $Fe^{3+}$ ) complex to ferrous ion ( $Fe^{2+}$ ) complex was as follows: Falzli>Safida> German > Cherry, and there was significant difference in the FRAP activities of Falzi mango kernel and other varieties used in the study. The determined antioxidant property reduced the ferric ion ( $Fe^{3+}$ ) to the ferrous ion ( $Fe^{2+}$ ), which resulted in the formation of a blue complex ( $Fe^{2+}$ /TPTZ) (Gupta 2015). The significance of FRAP activity indicated the presence and potentiality of its antioxidant capacity, as the FRAP assay result was based on the reduction of ferric ions. This is because antioxidants are reducing agents that can donate a single electron or hydrogen for reduction.



#### 4.0 Conclusion

The concentration of phytochemicals and effectiveness of anti-oxidant properties of mango kernel varies greatly among different varieties. A considerable relationship was seen between phytochemicals and antioxidant activities, showing that phytochemicals are the major contributors to antioxidant abilities of mango kernel seeds. Therefore, scientific knowledge of the proportion in every variety is paramount and exploiting all the qualities of mango kernel seeds may offer tremendous solutions to some prevailing pharmaceutical conditions and nutritional challenges. It's clear that the wasting *mangifera indica* seed around us is enriched with potential solutions to some of our health challenges and the extent to which we discover it have great implications to dealing with these challenges. Hence, extensive research on its pharmacodynamics, proper standardization and nutritional effect is necessary to exploit their dietary values and therapeutic uses in combating various prevailing challenges.

#### Author Address:

<sup>1</sup>Research scholar; Department of Biotechnology, Federal University of Technology, Owerri, Imo State, Nigeria

<sup>2,3</sup>Department of Biotechnology, Federal University of Technology, Owerri, Imo State, Nigeria

<sup>4</sup>Department of Biotechnology, Faculty of Sciences, Ebonyi State University, Abakaliki, Nigeria

#### 5. References

1. Beyene, G. And Araya, A. (2015). Review of Mango (*Mangifera indica*) Seed-Kernel Waste as a Diet for Poultry. *Journal of Biology, Agriculture and Healthcare* 5, 11.
2. Choudhary, P., Devi, T. B., Tushir, S., Kasana, C. R., Popatrao, D. S., and Narsaiah K. (2023) Mango Seed Kernel: A Bountiful Source of Nutritional and Bioactive Compounds. *Food and Bioprocess Technology*, 16:289–312
3. Chukwuebuka, E., and Jonathan, C. I.(2015). Biological functions and anti-nutritional effects of phytochemicals in living system. *Journal of pharmacy and biological sciences* 10(2):15-17.
4. Deepshikha Gupta (2015), Methods for Determination of Antioxidant Capacity: A Review. *International Journal of Pharmaceutical Science and Research*. 6(2): 546-566
5. Egbuna, C. And Ifemeje, J. C. (2015). Comparative Studies on the Phytochemical Properties of Five Nigerian Medicinal Plants. *Journal of Advances in Medical and Pharmaceutical Sciences* 6(2):1-12
6. FAO (2015), "FAOSTAT". Food and Agriculture Organization of the United Nations.

7. Food and Agriculture Organization of the United Nations. (2015). Food loss analysis: causes and solutions Case study on the mango value chain in the Republic of India. Accessed 18th June. 2024
8. Gbadamosi, I.T., Moody, J.O. Lawal and A.M. (2011). Phytochemical screening and proximate analysis of eight ethno botanicals used as antimalaria remedies in Ibadan, Nigeria, Journal of Applied Biosciences. 44: 2967-2971
9. Ibarra, I. P., Ramos, P. A., Hernández, C., & Jacobo, D. A. (2015). Effects of postharvest ripening on the nutraceutical and physicochemical properties of mango (*Mangifera indica* L.) Cvkeitt. Postharvest Biology and Technology. 103: 45-54
10. Jahurul, M. H. A. , Zaidul, I. S. M., Kashif, G., Fahad, Y. A., Kar-Lin, N, Norulaini, N. A. N., Sahena , F., and Mohd Omar, A. K. (2015). Mango (*Mangifera indica* L.) by-products and their valuable components: a review. Food Chemistry, 15:183:173-80
11. Kedare, S.B. Singh, R.P. (2011). Genesis and development of DPPH method of antioxidant assay. Journal of Food Science Technology, 48: 412-422
12. Kim, I., Yang, M., Lee, O. And Kang, S. (2011). Antioxidant activities of hot water extracts from various spices. International Journal of Molecular Sciences. 12(6):4120-4131.
13. Kirk, R. S. And Sawyer, R. (1991). Pearson's Composition and Analysis of Foods, 9th ed. (student edition), England: Addison Wesley Longman Ltd. 33-36
14. Latif, S.; Anwar, F. (2011). Aqueous enzymatic sesame oil and protein extraction. Food Chemistry. 125, 679– 684,
15. Liu, Z. H., Hui, F. M., and Zhang, G. P. (2005). Grain phytic acid in japonica rice as affected by cultivars and environment and its relation to protein content. Food Chemistry 89:59-62
16. Monica, T., Karuna, S. And Renu, K. (2020). Functional and preservative properties of phytochemicals. Phytochemical extraction process, safety assessment, toxicology evaluation, and regulatory issues. 11:341-361.
17. Rigane, G., Sami, M., and Ridha, B. S. (2018). One step purification of 6-shogaol from *zingiber officinale* Rosco, a phenolic compound having a high effectiveness against bacterial strains. Re. Roum. Chim. 63(1): 5-10
18. Rongtsao (2010). Chemistry and biochemistry of dietary polyphenols. National center for Biotechnology information. 2(12):1231-1246
19. Saranyu, K. And Rakrudee, S. (2011). Antioxidant and antibacterial activities of selected varieties of Thai mango seed extract. Pakistan Journal of Pharmaceutical Science. 24(1):37-42
20. Tharanathan, R. N., Yashoda, H. M., and Prabha, T. N. (2006). Mango (*Mangifera indica* L.). The King of Fruits-An Overview. Food Reviews International, 22: 95-123.

21. Tohma, H., Gülçin, I., Bursal, E., Koksal, E., Gören, A. C., and Alwasel, S. (2017). Antioxidant activity and phenolic compounds of ginger (*zingiberofficinalerosc.*) Determined by HPLC-MS/MS. *Journal of Food Measurement and Characterization*.11(2): 556–566,
22. Torres-León, C., Rojas, R., Contreras-Esquivel, J. C., Serna-Cock, L., Belmares-Cerda, R. E., and Aguilar, C. N. (2016). Mango seed: Functional and nutritional properties. *Trends in Food Science & Technology*.109-117
23. Trease, G. And Evans, W. (2002) *Phytochemicals*. In: *Pharmacognosy*, 15th Edn. Saunders Publishers, London, 42-393.

## Appendix

### Table 2. Phytochemicals Multiple Comparisons

LSD

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
Phytate	1.00	2.00	.20350*	.02268	<.001	.1512	.2558
		3.00	.15950*	.02268	<.001	.1072	.2118
		4.00	.16750*	.02268	<.001	.1152	.2198
	2.00	1.00	-.20350*	.02268	<.001	-.2558	-.1512
		3.00	-.04400	.02268	.088	-.0963	.0083
		4.00	-.03600	.02268	.151	-.0883	.0163
	3.00	1.00	-.15950*	.02268	<.001	-.2118	-.1072
		2.00	.04400	.02268	.088	-.0083	.0963
		4.00	.00800	.02268	.733	-.0443	.0603
	4.00	1.00	-.16750*	.02268	<.001	-.2198	-.1152
		2.00	.03600	.02268	.151	-.0163	.0883
		3.00	-.00800	.02268	.733	-.0603	.0443
Oxalate	1.00	2.00	-.02550*	.00167	<.001	-.0294	-.0216
		3.00	.05550*	.00167	<.001	.0516	.0594
		4.00	.02350*	.00167	<.001	.0196	.0274
	2.00	1.00	.02550*	.00167	<.001	.0216	.0294
		3.00	.08100*	.00167	<.001	.0771	.0849
		4.00	.04900*	.00167	<.001	.0451	.0529
	3.00	1.00	-.05550*	.00167	<.001	-.0594	-.0516
		2.00	-.08100*	.00167	<.001	-.0849	-.0771
		4.00	-.03200*	.00167	<.001	-.0359	-.0281
	4.00	1.00	-.02350*	.00167	<.001	-.0274	-.0196
		2.00	-.04900*	.00167	<.001	-.0529	-.0451
		3.00	.03200*	.00167	<.001	.0281	.0359
Heamagglutinin	1.00	2.00	-.41000	.18308	.055	-.8322	.0122
		3.00	-.59300*	.18308	.012	-1.0152	-.1708
		4.00	-.06100	.18308	.748	-.4832	.3612
	2.00	1.00	.41000	.18308	.055	-.0122	.8322
		3.00	-.18300	.18308	.347	-.6052	.2392
		4.00	.34900	.18308	.093	-.0732	.7712
	3.00	1.00	.59300*	.18308	.012	.1708	1.0152
		2.00	.18300	.18308	.347	-.2392	.6052

		4.00	.53200*	.18308	.020	.1098	.9542
	4.00	1.00	.06100	.18308	.748	-.3612	.4832
		2.00	-.34900	.18308	.093	-.7712	.0732
		3.00	-.53200*	.18308	.020	-.9542	-.1098
Phenol	1.00	2.00	-2.55100*	.42932	<.001	-3.5410	-1.5610
		3.00	2.02700*	.42932	.001	1.0370	3.0170
		4.00	2.36500*	.42932	<.001	1.3750	3.3550
	2.00	1.00	2.55100*	.42932	<.001	1.5610	3.5410
		3.00	4.57800*	.42932	<.001	3.5880	5.5680
		4.00	4.91600*	.42932	<.001	3.9260	5.9060
	3.00	1.00	-2.02700*	.42932	.001	-3.0170	-1.0370
		2.00	-4.57800*	.42932	<.001	-5.5680	-3.5880
		4.00	.33800	.42932	.454	-.6520	1.3280
	4.00	1.00	-2.36500*	.42932	<.001	-3.3550	-1.3750
		2.00	-4.91600*	.42932	<.001	-5.9060	-3.9260
		3.00	-.33800	.42932	.454	-1.3280	.6520

\*. The mean difference is significant at the 0.05 level.

Mean and Standard deviation of Phytochemicals of the selected mango varieties

Phytochemicals	Varieties			
	Safeda	Cherry	German	Fazli
<b>Saponin</b>	1.82	2	1.3	3.5
	1.98	2.6	1.5	4.5
<b>Mean</b>	<b>1.9</b>	<b>2.3</b>	<b>1.4</b>	<b>4</b>
<b>SD</b>	<b>0.113137</b>	<b>0.424264</b>	<b>0.141421</b>	<b>0.707107</b>
<b>Flavonoids</b>	5.85	4.98	3.91	4.98
	5.33	4.66	3.77	5
<b>Mean</b>	<b>5.59</b>	<b>4.82</b>	<b>3.84</b>	<b>4.99</b>
<b>SD</b>	<b>0.367696</b>	<b>0.226274</b>	<b>0.098995</b>	<b>0.014142</b>
<b>Alkaloids</b>	2.5	2.04	2.58	4.97
	3.56	2.92	2.22	4.59
<b>Mean</b>	<b>3.03</b>	<b>2.48</b>	<b>2.4</b>	<b>4.78</b>

<b>SD</b>	<b>0.749533</b>	<b>0.622254</b>	<b>0.254558</b>	<b>0.268701</b>	
<b>Phytate</b>	<b>0.9</b>	<b>0.7</b>	<b>0.795</b>	<b>0.728</b>	
	<b>0.955</b>	<b>0.748</b>	<b>0.741</b>	<b>0.792</b>	
<b>Mean</b>	<b>0.9275</b>	<b>0.724</b>	<b>0.768</b>	<b>0.76</b>	
<b>SD</b>	<b>0.038891</b>	<b>0.033941</b>	<b>0.038184</b>	<b>0.045255</b>	
<b>Oxalate</b>	<b>0.08</b>	<b>0.106</b>	<b>0.0245</b>	<b>0.0585</b>	
	<b>0.085</b>	<b>0.11</b>	<b>0.0295</b>	<b>0.0595</b>	
<b>Mean</b>	<b>0.0825</b>	<b>0.108</b>	<b>0.027</b>	<b>0.059</b>	
<b>SD</b>	<b>0.0025</b>	<b>0.002</b>	<b>0.0025</b>	<b>0.0005</b>	
<b>Heamagglutinin</b>	<b>2.95</b>	<b>3.57</b>	<b>3.155</b>	<b>3.005</b>	
	<b>2.98</b>	<b>3.18</b>	<b>3.961</b>	<b>3.047</b>	
<b>Mean</b>	<b>2.965</b>	<b>3.375</b>	<b>3.558</b>	<b>3.026</b>	
<b>SD</b>	<b>0.021213</b>	<b>0.275772</b>	<b>0.569928</b>	<b>0.029698</b>	
<b>Phenol</b>	<b>5.5</b>	<b>7.085</b>	<b>3.21</b>	<b>3.154</b>	
	<b>5.58</b>	<b>9.097</b>	<b>3.816</b>	<b>3.196</b>	
<b>Mean</b>	<b>5.54</b>	<b>8.091</b>	<b>3.513</b>	<b>3.175</b>	
<b>SD</b>	<b>0.056569</b>	<b>1.422699</b>	<b>0.428507</b>	<b>0.029698</b>	
<b>Cardiac glycoside</b>	<b>2.8</b>	<b>6.36</b>	<b>8.84</b>	<b>5.43</b>	
	<b>2.86</b>	<b>6.4</b>	<b>8.86</b>	<b>5.45</b>	
<b>Mean</b>	<b>2.83</b>	<b>6.38</b>	<b>8.85</b>	<b>5.44</b>	
<b>SD</b>	<b>0.042426</b>	<b>0.028284</b>	<b>0.014142</b>	<b>0.014142</b>	
<b>Terpenoids</b>	<b>8.95</b>	<b>5.5</b>	<b>15.4</b>	<b>13.3</b>	
	<b>11.05</b>	<b>6.5</b>	<b>16.6</b>	<b>14.7</b>	
<b>Mean</b>	<b>10</b>	<b>6</b>	<b>16</b>	<b>14</b>	
<b>SD</b>	<b>1.484924</b>	<b>0.707107</b>	<b>0.848528</b>	<b>0.989949</b>	

<b>Tannin</b>	2.02	3.06	1.52	1.085	
	2.08	3.1	1.56	1.095	
<b>Mean</b>	<b>2.05</b>	<b>3.08</b>	<b>1.54</b>	<b>1.09</b>	
<b>SD</b>	<b>0.042426</b>	<b>0.028284</b>	<b>0.028284</b>	<b>0.007071</b>	

**Table 4: Antioxidant Multiple Comparisons**

LSD

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
Superoxide Dismutase (SOD)	1.00	2.00	-.04000*	.01289	.015	-.0697	-.0103
		3.00	-.15100*	.01289	<.001	-.1807	-.1213
		4.00	-.09800*	.01289	<.001	-.1277	-.0683
	2.00	1.00	.04000*	.01289	.015	.0103	.0697
		3.00	-.11100*	.01289	<.001	-.1407	-.0813
		4.00	-.05800*	.01289	.002	-.0877	-.0283
	3.00	1.00	.15100*	.01289	<.001	.1213	.1807
		2.00	.11100*	.01289	<.001	.0813	.1407
		4.00	.05300*	.01289	.003	.0233	.0827
	4.00	1.00	.09800*	.01289	<.001	.0683	.1277
		2.00	.05800*	.01289	.002	.0283	.0877
		3.00	-.05300*	.01289	.003	-.0827	-.0233
DPPH	1.00	2.00	-13.69000*	1.15289	<.001	-16.3486	-11.0314
		3.00	20.09000*	1.15289	<.001	17.4314	22.7486
		4.00	-10.54000*	1.15289	<.001	-13.1986	-7.8814
	2.00	1.00	13.69000*	1.15289	<.001	11.0314	16.3486
		3.00	33.78000*	1.15289	<.001	31.1214	36.4386
		4.00	3.15000*	1.15289	.026	.4914	5.8086
	3.00	1.00	-20.09000*	1.15289	<.001	-22.7486	-17.4314
		2.00	-33.78000*	1.15289	<.001	-36.4386	-31.1214
		4.00	-30.63000*	1.15289	<.001	-33.2886	-27.9714
	4.00	1.00	10.54000*	1.15289	<.001	7.8814	13.1986
		2.00	-3.15000*	1.15289	.026	-5.8086	-.4914
		3.00	30.63000*	1.15289	<.001	27.9714	33.2886
Anti radical scavenging activity	1.00	2.00	-19.89000*	1.30815	<.001	-22.9066	-16.8734
		3.00	1.65000	1.30815	.243	-1.3666	4.6666
		4.00	-11.17000*	1.30815	<.001	-14.1866	-8.1534
	2.00	1.00	19.89000*	1.30815	<.001	16.8734	22.9066
		3.00	21.54000*	1.30815	<.001	18.5234	24.5566
		4.00	8.72000*	1.30815	<.001	5.7034	11.7366
	3.00	1.00	-1.65000	1.30815	.243	-4.6666	1.3666
		2.00	-21.54000*	1.30815	<.001	-24.5566	-18.5234
		4.00	-12.82000*	1.30815	<.001	-15.8366	-9.8034
	4.00	1.00	11.17000*	1.30815	<.001	8.1534	14.1866
		2.00	-8.72000*	1.30815	<.001	-11.7366	-5.7034



		3.00	12.82000 <sup>*</sup>	1.30815	<.001	9.8034	15.8366
Catalase	1.00	2.00	.30000 <sup>*</sup>	.08165	.006	.1117	.4883
		3.00	.60000 <sup>*</sup>	.08165	<.001	.4117	.7883
		4.00	-.40000 <sup>*</sup>	.08165	.001	-.5883	-.2117
		2.00	1.00	-.30000 <sup>*</sup>	.08165	.006	-.4883
	2.00	3.00	.30000 <sup>*</sup>	.08165	.006	.1117	.4883
		4.00	-.70000 <sup>*</sup>	.08165	<.001	-.8883	-.5117
		3.00	1.00	-.60000 <sup>*</sup>	.08165	<.001	-.7883
	3.00	2.00	-.30000 <sup>*</sup>	.08165	.006	-.4883	-.1117
		4.00	-1.00000 <sup>*</sup>	.08165	<.001	-1.1883	-.8117
		4.00	1.00	.40000 <sup>*</sup>	.08165	.001	.2117
	4.00	2.00	.70000 <sup>*</sup>	.08165	<.001	.5117	.8883
		3.00	1.00000 <sup>*</sup>	.08165	<.001	.8117	1.1883
2.00		1.00	-.62100 <sup>*</sup>	.04916	<.001	-.7344	-.5076
Glutathione Reductase	1.00	3.00	-.70100 <sup>*</sup>	.04916	<.001	-.8144	-.5876
		4.00	-1.40200 <sup>*</sup>	.04916	<.001	-1.5154	-1.2886
		2.00	1.00	.62100 <sup>*</sup>	.04916	<.001	.5076
	2.00	3.00	-.08000	.04916	.142	-.1934	.0334
		4.00	-.78100 <sup>*</sup>	.04916	<.001	-.8944	-.6676
		3.00	1.00	.70100 <sup>*</sup>	.04916	<.001	.5876
	3.00	2.00	.08000	.04916	.142	-.0334	.1934
		4.00	-.70100 <sup>*</sup>	.04916	<.001	-.8144	-.5876
		4.00	1.00	1.40200 <sup>*</sup>	.04916	<.001	1.2886
	4.00	2.00	.78100 <sup>*</sup>	.04916	<.001	.6676	.8944
		3.00	.70100 <sup>*</sup>	.04916	<.001	.5876	.8144
		1.00	2.00	.09300 <sup>*</sup>	.03148	.018	.0204
Lipid Peroxidase	1.00	3.00	.12000 <sup>*</sup>	.03148	.005	.0474	.1926
		4.00	.14800 <sup>*</sup>	.03148	.002	.0754	.2206
		2.00	1.00	-.09300 <sup>*</sup>	.03148	.018	-.1656
	2.00	3.00	.02700	.03148	.416	-.0456	.0996
		4.00	.05500	.03148	.119	-.0176	.1276
		3.00	1.00	-.12000 <sup>*</sup>	.03148	.005	-.1926
	3.00	2.00	-.02700	.03148	.416	-.0996	.0456
		4.00	.02800	.03148	.400	-.0446	.1006
		4.00	1.00	-.14800 <sup>*</sup>	.03148	.002	-.2206
	4.00	2.00	-.05500	.03148	.119	-.1276	.0176

		3.00	-.02800	.03148	.400	-.1006	.0446
Vitamin C mg/100	1.00	2.00	-31.47900 *	3.08284	<.001	-38.5881	-24.3699
		3.00	25.84000 *	3.08284	<.001	18.7309	32.9491
		4.00	52.17100 *	3.08284	<.001	45.0619	59.2801
		2.00	1.00	31.47900 *	3.08284	<.001	24.3699
	2.00	3.00	57.31900 *	3.08284	<.001	50.2099	64.4281
		4.00	83.65000 *	3.08284	<.001	76.5409	90.7591
		3.00	1.00	-25.84000 *	3.08284	<.001	-32.9491
	3.00	2.00	-57.31900 *	3.08284	<.001	-64.4281	-50.2099
		4.00	26.33100 *	3.08284	<.001	19.2219	33.4401
		4.00	1.00	-52.17100 *	3.08284	<.001	-59.2801
	4.00	2.00	-83.65000 *	3.08284	<.001	-90.7591	-76.5409
		3.00	-26.33100 *	3.08284	<.001	-33.4401	-19.2219
2.00		11.62500 *	.60596	<.001	10.2277	13.0223	
Ferric Reducing Anti-oxidant Property (FRAP) Results	1.00	3.00	.75000	.60596	.251	-.6473	2.1473
		4.00	-39.25000 *	.60596	<.001	-40.6473	-37.8527
		2.00	1.00	-11.62500 *	.60596	<.001	-13.0223
	2.00	3.00	-10.87500 *	.60596	<.001	-12.2723	-9.4777
		4.00	-50.87500 *	.60596	<.001	-52.2723	-49.4777
		3.00	1.00	-.75000	.60596	.251	-2.1473
	3.00	2.00	10.87500 *	.60596	<.001	9.4777	12.2723
		4.00	-40.00000 *	.60596	<.001	-41.3973	-38.6027
		4.00	1.00	39.25000 *	.60596	<.001	37.8527
	4.00	2.00	50.87500 *	.60596	<.001	49.4777	52.2723
		3.00	40.00000 *	.60596	<.001	38.6027	41.3973

\*. The mean difference is significant at the 0.05 level.

### Mean and Standard deviation of Antioxidant properties of the selected mango varieties

		Antioxidant properties			
Samples		Safeda	Cherry	German	Falzi
Superoxidase (SOD)	Dismutase	0.116 unit enzyme	0.156 unit enzyme	0.267unit enzyme	0.214 unit enzyme
		0.1	0.14	0.25	0.2

	0.132	0.172	0.284	0.228
<b>Mean</b>	<b>0.116</b>	<b>0.156</b>	<b>0.267</b>	<b>0.214</b>
<b>SD</b>	<b>0.022627</b>	<b>0.022627</b>	<b>0.024042</b>	<b>0.019799</b>
<b>DPPH</b>	35	49	16	46
	38.64	52.02	17.46	48.72
<b>Mean</b>	<b>36.82</b>	<b>50.51</b>	<b>16.73</b>	<b>47.36</b>
<b>SD</b>	<b>2.573869</b>	<b>2.135462</b>	<b>1.032376</b>	<b>1.92333</b>
<b>Anti radical scavenging activity</b>	20	40	19	31
	23.44	43.22	21.14	34.78
<b>Mean</b>	<b>21.72</b>	<b>41.61</b>	<b>20.07</b>	<b>32.89</b>
<b>SD</b>	<b>2.432447</b>	<b>2.276884</b>	<b>1.513209</b>	<b>2.672864</b>
<b>Catalase</b>	2.100umol/ ml	1.778umol/ ml	1.498 umol/ml	2.509umol/ ml
	2	1.7	1.4	2.4
	2.2	1.9	1.6	2.6
<b>Mean</b>	<b>2.1</b>	<b>1.8</b>	<b>1.5</b>	<b>2.5</b>
<b>SD</b>	<b>0.141421</b>	<b>0.141421</b>	<b>0.141421</b>	<b>0.141421</b>
<b>Glutathione Reductase</b>	0.45	1.08	1.16	1.85
	0.578	1.19	1.27	1.982
	<b>0.514</b>	<b>1.135</b>	<b>1.215</b>	<b>1.916</b>
	<b>0.09051</b>	<b>0.077782</b>	<b>0.077782</b>	<b>0.093338</b>
<b>Lipid Peroxidase</b>	1.076umol/ ml	0.983umol/ ml	0.956umol/ ml	0.928umol/ ml
	1.03	0.95	0.92	0.89
	1.122	1.016	0.992	0.966
<b>Mean</b>	<b>1.076</b>	<b>0.983</b>	<b>0.956</b>	<b>0.928</b>
<b>SD</b>	<b>0.065054</b>	<b>0.046669</b>	<b>0.050912</b>	<b>0.05374</b>
<b>Vitamin C mg/100</b>	178.879mg/1 00	210.358mg/1 00	153.539 mg/100	126.713mg/1 00
	175	206	150	123

	182.758	214.716	156.078	130.416
<b>Mean</b>	<b>178.879</b>	<b>210.358</b>	<b>153.039</b>	<b>126.708</b>
<b>SD</b>	<b>5.485734</b>	<b>6.163143</b>	<b>4.297795</b>	<b>5.243904</b>
<b>Ferric Reducing Anti-oxidant Property (FRAP) Results</b>	21.75	10.125	21	61
	21	9.5	20.5	60
	22.5	10.75	21.5	62
<b>Mean</b>	<b>21.75</b>	<b>10.125</b>	<b>21</b>	<b>61</b>
<b>SD</b>	<b>0.612372</b>	<b>0.51031</b>	<b>0.408248</b>	<b>0.816497</b>