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# *In vitro* Antioxidant Properties of Mango Powder Produced from Blends of *Brokin* and *Julie* Cultivars Fortified with Yellow Pea protein hydrolysate

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**Abstract** The aim of the this study was to evaluate the *in vitro* antioxidant potential of mango powder prepared from blends of *Brokin* and *Julie* cultivars fortified with Yellow Pea protein hydrolysates. Six blends were used: *$B_{CI}$, *$J_{CII}$, *$B_{80}J_{20}$, *$B_{70}J_{30}$, *$B_{60}J_{40}$ and *$B_{50}J_{50}$. The fortified blends were derived by obtaining 90% of each of the original unfortified blend and incorporating to each of them, 10% of Yellow pea protein hydrolysate (YPPH). The results of the antioxidant assays were: % inhibition for DPPH radical scavenging activity includes 0.70(*$B_{CI}$)-5.08%(*$B_{70}J_{30}$), 2.24(*$J_{CII}$)-8.69%(*$B_{60}J_{40}$), 7.79 (*$J_{CII}$)-12.46% (*$B_{CI}$), 12.42 (*$B_{60}J_{40}$)-21.87% (*$B_{CI}$), 20.60 (*$B_{60}J_{40}$)-38.57% (*$B_{CI}$) and 30.08 (*$B_{60}J_{40}$)-69.64% (*$B_{CI}$)} at dose concentration levels of 62.50µg $mL^{-1}$, 125.00µg $mL^{-1}$, 250.00µg/mL, 500.00µg $mL^{-1}$, 1000.00µg $mL^{-1}$ and 2000.00µg $mL^{-1}$ respectively; Metal chelating abilities include 51.84 (*$B_{60}J_{40}$)-66.98 (*$B_{80}J_{20}$), 64.49 (*$J_{CII}$)-72.64 (*$B_{50}J_{50}$), 65.11 (*$B_{60}J_{40}$)-73.58 (*$B_{80}J_{20}$), 67.95 (*$J_{CII}$)-74.59 (*$B_{70}J_{30}$) and 60.37(*$J_{CII}$)-75.44% (*$B_{70}J_{30}$)} at concentrations of 50.00-250.00µg $mL^{-1}$ accordingly; ferric reducing antioxidant power (265.42-411.08µg.AAE $mg^{-1}$) at those concentration level of 1000.00µg $mL^{-1}$ The % inhibition of hydroxyl radical scavenging activity showed that among all the samples at the concentration range of 62.50-2000.00µg $mL^{-1}$ respectively, samples, *$B_{80}J_{20}$ and *$B_{50}J_{50}$ exhibited highest and lowest radical scavenging activities accordingly. Clearly, the best antioxidant activity was observed to be metal chelating ability assay. Based on the aforementioned results, it is concluded that the fortified mango powder samples have good antioxidant activity *in vitro* which can treat and manage chronic disease condition such as oxidative stress.

***Keywords:*** *Brokin mango powder, Julie mango powder, Yellow Pea protein hydrolysate, in vitro antioxidant properties, oxidative stress*

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## 1. Introduction

Fruits and vegetables, being rich sources of nutrients, have been a subject of interest due to their potential health benefits in preventing several chronic diseases [1]. The health benefits of fruits and vegetables have been attributed partly to the compounds having antioxidant capacity and an ability to overcome oxidative stress by neutralizing the over production of oxidant species [2]. If Reactive Oxygen Species (ROS) are not removed, their accumulation overcomes the cellular reparative abilities, causing the collapse of cellular functions and can result in the generation of pathological states related to aging, cancer, atherosclerosis, heart attack, stroke, and diabetes [3]. It is well established fact that among all tropical fruits that are known, mango is the king [4]. This is not surprising in view of the inherent distinguishable unique features it has. It is a fruit that is tasty, reveled for its juicy nature and its delightful flavor. The world production of mango is estimated at 42 million tons per year [5]. Nigeria ranks 8th [6] with total production of 730,000 metric tons [7]. An overview of states in Nigeria showed that Benue State regarded as the '*Food Basket of the Nation'* is the 1st among mango league of producing states. Other states in Nigeria that are well known for mango production include; Jigawa, Plateau, Kebbi, Niger, Kaduna, Kano, Bauchi, Sokoto, Adamawa, Taraba and Federal Capital Territory (FCT) [7]. In Nigeria, postharvest losses of fruits and vegetables amounts to 35-45% of the annual production [8]. There is an economic sense in preserving seasonal fruits during peak season by transforming them into shelf stable different fruit products. Mango powders fortified

with Yellow Pea protein hydrolysates can serve as a vehicle for promoting new food product developments through creation of new food formulations such as shelf stable constituents of new health drinks, custard powder, cereal flakes, yoghurt, ice cream, mango shakes and so much more. Tropical fruit consumption is increasing on the domestic and international markets due to the growing recognition of its nutritional and therapeutic value [9]. Mango is a super fruit that is acknowledged to be very rich in fiber, including bioactive compounds such as phenolics, provitamin A, carotenoids and vitamin C. Overtime different methods have been used to evaluate the antioxidant properties of several food-derived antioxidants. In the present study, bioactive mango powder blends were developed from two local cultivars from Benue State, North-Central Nigeria, *Brokin* and *Julie* Mangoes whose powder blends were fortified with Yellow Pea protein hydrolysate. The two mango cultivars are very sweet and so were selected because they are the most consumed. In addition, the *Brokin* cultivar is bigger and juicier than the *Julie* cultivar. Six blend formulations were designed and analyzed for *in vitro* antioxidant properties using antioxidant evaluation systems such as DPPH Scavenging activity, Metal ions chelating activity, Ferric reducing power and Hydroxyl scavenging activity.

# 2. Materiel and Methods

## 2.1. Source of Raw Materials

The two mango cultivars, *Brokin* and *Julie* were purchased from a mango orchard in Vandeikya Local Government Area, Benue State and Yellow Pea protein hydrolysates was prepared in the laboratory of Professor Rotimi E. Aluko of Department of Food and Human Nutritional Sciences, University of Manitoba, Winnipeg, Manitoba, Canada.

### 2.1.1. Yellow Pea protein isolates (YPPI)

85% protein content, from Nutri-Pea Ltd Canada, was used in this research work to prepare Yellow Pea protein hydrolysates.

## 2.2. Drying Experiment for Mango Powder Production

The method adopted by Harnkarnsujarit and Charoenrein [10] was followed with slight modification. Preliminary screening by picking only fresh, fully matured and attractive mangoes for laboratory evaluations was carried out. The fruits were placed in suitable containers equipped with running water and properly washed to detach dirt, adhering materials like fruit exudates and to reduce microbial load. The mango peels were removed manually with the help of a stainless steel knife. The fleshy part of the mango fruits were cut into 1cm cubes with the stones removed by manual means, and freeze dried using a freeze dryer (Lyotrap model with accessories). The mango pieces were frozen at -35°C and held for 2 h to achieve ice crystallization and subsequently dried at shelf temperature of -40°C. Then all samples were stored in evacuated desiccators for 3 days, which contain silica gel to remove residual water. After that, the samples were considered to have zero 'water content'. Subsequently, the freeze dried mango pieces were packed in aluminum foil in a partial vacuum and were grounded using a stainless steel grinder. The ground particles were again passed through a series of reduction for further sieving to obtain a refined powder. The sieved powder was packed separately in some selected suitable packaging materials for further analyses. The packaging materials were coded and stored at room temperature.

## 2.3. Preparation of Yellow Pea Protein Hydrolysates (YPPH)

Pea protein hydrolysate was prepared as follows according to a previously described method by Pownall *et al.* [11]. A 6.0% (w/v) aqueous slurry of Yellow Pea protein isolate (PPI) was heated to 37°C and adjusted to pH 2.0 followed by addition of pepsin (5%, w/w pea protein basis). The temperature and pH of the slurry was maintained at constant values for 2 h, after which the hydrolysis by pepsin was stopped by adjusting the pH to 7.5 and adding pancreatin (5%, w/w pea protein basis). This enzyme was allowed to run for four (4h) ensuring that the temperature and pH of the slurry will be maintained at constant values for this period. After 4h, the slurry was heated at 95°C for 15 min and cooled to room temperature. The cooled reaction mixture hydrolysate will be centrifuged at 10000g for 25 min, and the clear supernatant was further sequentially passed through ultrafiltration membranes with 1 kDa and 3 kDa molecular weight cutoff. The resulting permeates was collected, freeze-dried and labeled as YPPH for further use.

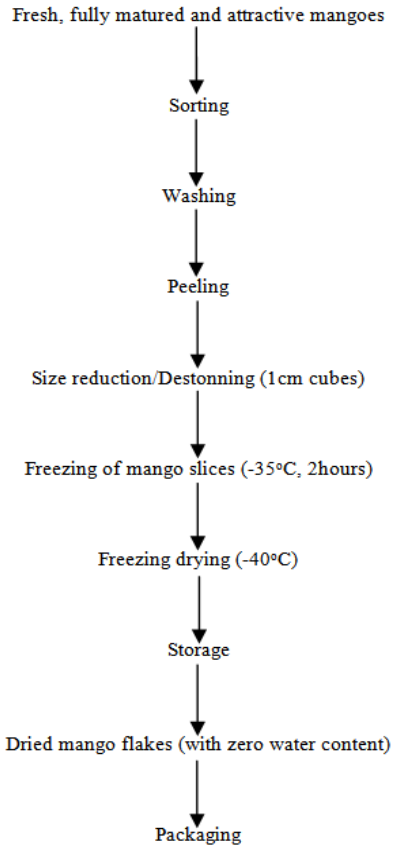


**Figure 1.** Procedure for freeze drying mangoes **(**Source: Modified method of Harnkarnsujarit and Charoenrein [10])

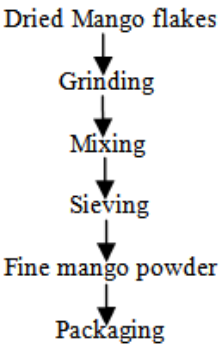


**Figure 2.** Flow diagram for the production of Mango powders (Source: Modified method of Akhter *et al.* [12].*)*

**Table 1. Blend formulations of *Brokin* and *Julie* mango powder fortified with Yellow Pea protein hydrolysates**

| Sample | *Brokin* Mango Powder (BMP) (%) | *Julie* Mango Powder (JMP) (%) |
|---|---|---|
| *$B_{CI}$ & *$J_{CII}$ | 100 | 100 |
| *$B_{80}J_{20}$ | 80 | 20 |
| *$B_{70}J_{30}$ | 70 | 30 |
| *$B_{60}J_{40}$ | 60 | 40 |
| *$B_{50}J_{50}$ | 50 | 50 |

*$B_{CI}$= 90% of (100% *Brokin* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$J_{CII}$= 90% of (100% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{80}J_{20}$= 90% of (80% *Brokin* mango powder: 20% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{70}J_{30}$= 90% of (70% *Brokin* mango powder: 30% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{60}J_{40}$= 90% of (60% *Brokin* mango powder: 40% *Julie* mango powder): 10% Yellow Pea protein hydrolysate
(YPPH)
*$B_{50}J_{50}$= 90% of (50% *Brokin* mango powder: 50% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)

## 2.4. *In vitro* Antioxidant Properties of Bioactive Mango Powder Blends

### 2.4.1. DPPH radical scavenging assay

The The free radical-scavenging ability of the samples against DPPH free radical was measured by measuring the decrease in absorbance of methanolic DPPH solution at 517 nm in the presence of each sample as described by Shodehinde and Oboh [13]. Briefly, 1mL of different concentrations (2000, 1000, 500, 250, 125 and 62.50µg $mL^{-1}$) of samples were added to 1 mL of 0.4 mmol $L^{-1}$ methanolic solution containing DPPH radicals. The mixture was left in the dark for 30 min and the absorbance was measured at 516 nm in the spectrophotometer (JENWAY 6305). The DPPH free radical scavenging ability was subsequently calculated by comparing the results of the test with those of the control (not treated with the test sample). The ability of the samples to scavenge was calculated relative to the control using the formula.

$$\% \text{ Inhibition} = \left\{\left[\left(A_C - A_S\right)/A_C\right] \text{ x } 100\right\} \quad (1)$$

Where $A_C$ represents absorbance of control, $A_S$ represents absorbance of test sample.

### 2.4.2. Chelation of metal ions assay

The $Fe^{2+}$ chelating ability of the samples was determined using the method described by Nwanna *et al.* [14]. Freshly prepared 500 µmol $L^{-1}$ $FeSO_4$ (150 µL) was added to a reaction mixture containing 168 µL of 0.1 mol/L Tris-HCl (pH 7.4), 218 µL saline and the samples (50-250µL). The reaction mixture was incubated for 5 min, before the addition of 13 µL of 0.25% 1, 10-phenanthroline (w/v). The absorbance was subsequently measured at 510 nm in a spectrophotometer (JENWAY 6305). The $Fe^{2+}$ chelating ability was subsequently calculated with respect to the control.

$$\% \text{ } Fe^{2+} \text{chelating ability} = \left\{\left[\left(A_C - A_S\right)/A_C\right] \text{ x } 100\right\} \quad (2)$$

### 2.4.3. Ferric reducing antioxidant power (FRAP) assay

The reducing property of each sample was determined by assessing the ability of the test samples to reduce $FeCl_3$ solution as described by Oboh *et al.* [15]. 2.5 mL aliquot was mixed with 2.5 mL 200 mM sodium phosphate buffer (pH 6.6) and 2.5 mL 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 minutes and then 2.5 mL 10% trichloroacetic acid was added. This mixture was centrifuged at 650 rpm for 10 min. 5 mL of the supernatant was mixed with an equal volume of water and 1 mL 0.1% ferric chloride. The absorbance was measured at 700 nm in the spectrophotometer (JENWAY 6305). The ferric reducing antioxidant property was subsequently calculated as ascorbic acid equivalent.

### 2.4.4. Hydroxyl radical scavenging assay

The scavenging activity of samples on hydroxyl radical was measured according to the method of Pavithra and Vadivukkarasi [16]. Various concentrations (50-2000µg $mL^{-1}$) of the samples were added with 1.0 mL of iron-EDTA solution, 0.5 mL of EDTA solution (0.018%), and 1.0 mL of DMSO (0.85% DMSO (v/v) in 0.1 M phosphate buffer, pH 7.4) sequentially. The reaction was initiated by adding 0.5 mL of ascorbic acid (0.22%) and incubated at 80–90°C for 15 min in a water bath. After incubation the reaction was terminated by the addition of 1.0 mL of ice-cold TCA (17.5% w/v), 3mL of Nash reagent was added and left at room temperature for 15 min. The reaction mixture without sample was used as control. The intensity of the colour formed was measured spectrophotometrically at 412 nm against reagent blank. The % hydroxyl radical scavenging activity was calculated by the following formula: Hydroxyl Radical

$$\text{Scavenging} = \left\{\left[\left(A_C - A_S\right)/A_C\right] \text{ x } 100\right\} \quad (3)$$

Where $A_C$ represents absorbance of control, $A_S$ represents absorbance of test sample.

## 2.5. Statistical Analysis

The method described by Shodehinde and Oboh [13] with slight modification was followed for the statistical

analysis. The results were expressed as means ± standard deviation of triplicate determinations. Statistical analysis was done using IBM SPSS (Statistical Analysis Software 20.0). One way analysis of variance was used to analyze the results and Duncan test was used for the post hoc. Findings were considered statistically significant when the P value was less than 0.05.

## 3. Results and Discussion

### 3.1. Antioxidant Properties of the Blend Formulations Fortified with Yellow Pea Protein Hydrolysate

The result of the antioxidant assays, DPPH radical scavenging activity, Chelation of metal ions, Ferric reducing power and Hydroxyl radical scavenging activity are shown in Table 2 to Table 5.

In recent times, many epidemiological studies have confirmed that intake of exogenous antioxidants is effective in the prevention of a number of human diseases which have been implicated to be due to oxidative stress [17]. Antioxidant assays have been used to investigate antioxidant potential of bioactive compounds in several food materials. Different antioxidant assays yield different results and therefore, multiple methods are needed to obtain the most accurate antioxidant profile [18]. The study investigated antioxidant and free radical scavenging potential of the fortified mango powder formulated blends based on their ability to (i) scavenge non biological stable free radical (DPPH), (ii) chelate metal ions, (iii) reduce ferric to ferrous ion and (iv) scavenge biologically important oxidant such as $^{\bullet}OH$. The mango samples fortified with YPPH were tested for antioxidant in four different *in vitro* assays namely; DPPH radical scavenging activity (Table 2), chelation of metal ions (Table 3), ferric reducing ability (Table 4) and $^{\bullet}OH$ radical scavenging activity (Table 5). Out of the four assays, the highest antioxidant activity was obtained with the metal chelating ability assay. More so, the scavenging activity of dose concentration levels of 62.50µg/mL-2000.00µg $mL^{-1}$ against hydroxyl radical was more effective than the DPPH radical. This difference may likely be as a result of the different mechanisms used by antioxidant to inactivate the radicals tested. The bioactive compounds which might be responsible for the scavenging activity in the present study are the vitamins, essential metals, phenolic components in the mango powder blends and the amino acids supplementation offered by the Yellow Pea protein hydrolysate. It is evident from Table 2 that at concentration of 62.50µg $mL^{-1}$, the test samples had a very poor DPPH radical scavenging activity which improved with increase in concentration (µg $mL^{-1}$) with the test samples showing the strongest radical scavenging profile at the concentration level of 2000.00µg $mL^{-1}$. The fortified mango powder test samples, on interacting with DPPH, might have transferred an electron to it, thus neutralizing its free radical nature as reported by Adjimani and Asare [19]. Ifesan [20] reported 74.70±0.31mg $100g^{-1}$ and 98.10±0.43 mg $100g^{-1}$ for the mango seed flour and mango kernel flour respectively. Sogi *et al*. [21] reported that mango kernel had higher antioxidant or scavenging activity ranging from 1310.70 to 1799.5µmol TE $g^{-1}$ dry basis. These were higher than the values accounted for in the current research. This may be as a result of the disparities in the parts of mango being evaluated and the amount of bioactive constituents found in them. The results obtained from this study for DPPH radical scavenging activity were lower than those reported for mango peels from *Paparanda*, *Julie* and *Peter* cultivars [22]. Table 3 clearly revealed that all the samples showed very good metal chelating abilities when compared with the Na-EDTA (Standard) in a dose-dependent manner between 50.00-250.00µg $mL^{-1}$ The metal chelating effect were in this order; Na-EDTA>$*B_{80}J_{20}$ >$*J_{CII}$ >$*B_{70}J_{30}$> $*B_{CI} \geq *B_{50}J_{50}$>$*B_{60}J_{40}$ at the concentration of 50.00µg/mL, Na-EDTA>$*B_{50}J_{50}$>$*B_{70}J_{30}$>$*B_{80}J_{20}$>$*B_{60}J_{40} \geq *B_{CI} > *J_{CII}$ at the concentration of 100.00µg/mL, Na-EDTA>$*B_{80}J_{20} \geq *B_{50}J_{50}$ >$*B_{70}J_{30}$>$*J_{CII} \geq *B_{CI}$>$*B_{60}J_{40}$ at the concentration of 150.00µg/mL, Na-EDTA>$*B_{70}J_{30} \geq *B_{60}J_{40} \geq *B_{CI}$>$*B_{50}J_{50}$> $*B_{80}J_{20}$>$*J_{CII}$ at the concentration of 200.00µg $mL^{-1}$ and Na-EDTA>$*B_{70}J_{30}$>$*B_{80}J_{20}$>$*B_{50}J_{50}$>$*B_{60}J_{40}$>$*B_{CI}$>$*J_{CII}$ at the concentration of 250.00µg $mL^{-1}$ accordingly. The observed iron chelating property of the test samples may be beneficial in providing immunity to the cellular constituents against oxidative damage. A therapeutic approach that requires improving antioxidant potentials may be required in effective management of oxidative stress condition should it arise. This may be accomplished by either reducing the possibility of metal interacting with critical biomolecules and inducing oxidative damage, or bolstering the cells antioxidant defenses through endogenous supplementation of antioxidant molecules [23]. In the present study, the ferric reducing abilities of the six samples were in the decreasing order as follows: $*B_{70}J_{30}$>$*B_{80}J_{20}$>$*B_{50}J_{50}$>$*B_{CI} \geq *B_{60}J_{40}$>$*J_{CII}$ as shown in Table 4. The samples exhibited good reducing abilities at a dose concentration of 1000.00µg $mL^{-1}$. Sogi *et al*. [21] reported that mango kernel had antioxidant or scavenging activity ranging from 666 to 942 µmol TE $g^{-1}$ dry basis in terms of ascorbic acid equivalent using the FRAP assay. While, Ifesan [20] reported 52.65±0.62mg $100g^{-1}$ and 72.18±0.35 mg $100g^{-1}$ for mango seed flour and mango kernel flour accordingly. These disparities from the established values accounted for in the current study may be attributable to the differing parts of the mango being examined and also the differing amount of bioactive compounds present in them. Hydroxyl radical is an extremely reactive free radical formed in biological systems and has been implicated as a highly damaging species in free radical pathology, capable of damaging almost every molecule found in living cells [23]. Dose concentration levels of between 1000.00 – 2000.00µg $mL^{-1}$ as shown in Table 5 produced quite a low to moderate hydroxyl radical scavenging activities in most of the experimental samples respectively.

**Table 2. DPPH scavenging activity of the mango powder samples (YPPH fortified)**

| Samples | Concentration (μg $mL^{-1}$) | | | | | |
|---|---|---|---|---|---|---|
| | 62.50 | 125.00 | 250.00 | 500.00 | 1000.00 | 2000.00 |
| Standard | 77.39±0.34$^{a}$ | 80.57±1.26$^{a}$ | 81.75±0.29$^{a}$ | 82.0±0.17$^{a}$ | 82.59±0.06$^{a}$ | 84.24±0.05$^{a}$ |
| *$B_{CI}$ | 0.07±0.05$^{f}$ | 4.59±0.09$^{d}$ | 12.46±0.16$^{b}$ | 21.87±0.10$^{b}$ | 38.57±0.32$^{b}$ | 69.64±0.05$^{b}$ |
| *$J_{CII}$ | 0.08±0.06$^{f}$ | 2.24±0.05$^{f}$ | 7.79±0.07$^{g}$ | 20.45±0.06$^{c}$ | 32.59±0.06$^{c}$ | 50.97±0.10$^{c}$ |
| *$B_{80}J_{20}$ | 1.49±0.08$^{d}$ | 2.84±0.05$^{e,f}$ | 8.29±0.29$^{f}$ | 13.98±0.16$^{e}$ | 20.73±0.06$^{f}$ | 31.44±0.10$^{f}$ |
| *$B_{70}J_{30}$ | 5.08±0.04$^{b}$ | 6.51±0.06$^{c}$ | 9.36±0.06$^{d}$ | 15.27±0.06$^{d}$ | 24.54±0.12$^{d}$ | 38.27±0.06$^{d}$ |
| *$B_{60}J_{40}$ | 0.42±0.07$^{e}$ | 8.69±0.05$^{b}$ | 11.63±0.04$^{c}$ | 12.42±0.04$^{f}$ | 20.60±0.05$^{f}$ | 30.08±0.06$^{g}$ |
| *$B_{50}J_{50}$ | 2.07±0.00$^{c}$ | 3.59±0.08$^{e}$ | 9.00±0.06$^{e}$ | 13.90±0.06$^{e}$ | 22.08±0.08$^{e}$ | 34.79±0.23$^{e}$ |

*Values are Mean ± Standard Deviation (SD) of triplicate determinations and are presented in percentages (%). Numbers followed by different superscripted letter in the same column do differ significantly by Duncan's multiple range test ($p< 0.05$).
Standard= Ascorbic acid
*$B_{CI}$= 90% of (100% *Brokin* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$J_{CII}$= 90% of (100% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{80}J_{20}$= 90% of (80% *Brokin* mango powder: 20% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{70}J_{30}$= 90% of (70% *Brokin* mango powder: 30% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{60}J_{40}$= 90% of (60% *Brokin* mango powder: 40% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{50}J_{50}$= 90% of (50% *Brokin* mango powder: 50% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH).

**Table 3. % $Fe^{2+}$ chelating ability of the mango powder samples (YPPH fortified)**

| Samples | Concentration (μg $mL^{-1}$) | | | | |
|---|---|---|---|---|---|
| | 50.00 | 100.00 | 150.00 | 200.00 | 250.00 |
| Standard | 76.43±0.05$^{a}$ | 81.12±0.17$^{a}$ | 82.07±0.10$^{a}$ | 83.02±0.07$^{a}$ | 87.75±0.01$^{a}$ |
| *$B_{CI}$ | 55.03±0.54$^{e}$ | 66.97±0.30$^{d}$ | 67.94±0.15$^{d}$ | 74.52±0.11$^{b}$ | 62.58±0.55$^{f}$ |
| *$J_{CII}$ | 63.21±0.19$^{c}$ | 64.49±0.39$^{e}$ | 67.94±0.17$^{d}$ | 67.95±0.19$^{e}$ | 60.37±0.16$^{g}$ |
| *$B_{80}J_{20}$ | 66.98±0.27$^{b}$ | 67.94±0.16$^{c}$ | 73.58±0.18$^{b}$ | 69.82±0.08$^{d}$ | 72.64±0.08$^{c}$ |
| *$B_{70}J_{30}$ | 58.49±0.57$^{d}$ | 72.64±0.11$^{b}$ | 69.82±0.18$^{c}$ | 74.59±0.11$^{b}$ | 75.44±0.17$^{b}$ |
| *$B_{60}J_{40}$ | 51.84±0.52$^{f}$ | 66.98±0.19$^{d}$ | 65.11±0.23$^{e}$ | 74.54±0.11$^{b}$ | 67.93±0.16$^{e}$ |
| *$B_{50}J_{50}$ | 54.74±0.33$^{e}$ | 72.64±0.19$^{b}$ | 73.57±0.09$^{b}$ | 71.68±0.11$^{c}$ | 70.74±0.05$^{d}$ |

*Values are Mean ± Standard Deviation (SD) of triplicate determinations and are presented in percentages (%). Numbers followed by different superscripted letter in the same column do differ significantly by Duncan's multiple range test ($p< 0.05$).
Standard= Ascorbic acid
*$B_{CI}$= 90% of (100% *Brokin* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$J_{CII}$= 90% of (100% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{80}J_{20}$= 90% of (80% *Brokin* mango powder: 20% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{70}J_{30}$= 90% of (70% *Brokin* mango powder: 30% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{60}J_{40}$= 90% of (60% *Brokin* mango powder: 40% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{50}J_{50}$= 90% of (50% *Brokin* mango powder: 50% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH).

**Table 4. Ferric reducing antioxidant power of the mango powder samples (YPPH fortified)**

| Samples | Concentration (μg $mL^{-1}$) |
|---|---|
| | 1000.00 |
| *$B_{CI}$ | 311.88±2.93$^{d}$ |
| *$J_{CII}$ | 265.42±0.29$^{e}$ |
| *$B_{80}J_{20}$ | 377.81±0.10$^{b}$ |
| *$B_{70}J_{30}$ | 411.08±3.82$^{a}$ |
| *$B_{60}J_{40}$ | 308.19±2.07$^{d}$ |
| *$B_{50}J_{50}$ | 362.57±2.24$^{c}$ |

*Values are Mean ± Standard Deviation (SD) of triplicate determinations and are presented in μg.AAE $mg^{-1}$. Numbers followed by different superscripted letter in the same column do differ significantly by Duncan's multiple range test ($p< 0.05$).
*$B_{CI}$= 90% of (100% *Brokin* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$J_{CII}$= 90% of (100% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{80}J_{20}$= 90% of (80% *Brokin* mango powder: 20% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{70}J_{30}$= 90% of (70% *Brokin* mango powder: 30% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{60}J_{40}$= 90% of (60% *Brokin* mango powder: 40% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{50}J_{50}$= 90% of (50% *Brokin* mango powder: 50% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)

**Table 5. Hydroxyl radical scavenging activity of the mango powder samples (YPPH fortified)**

| Samples | Concentration (μg mL$^{-1}$) | | | | | |
|---|---|---|---|---|---|---|
| | 62.50 | 125.00 | 250.00 | 500.00 | 1000.00 | 2000.00 |
| Standard | 15.29±0.06$^{a}$ | 23.68±0.08$^{a}$ | 33.43±0.17$^{a}$ | 40.32±0.08$^{a}$ | 63.67±0.09$^{a}$ | 92.36±0.07$^{a}$ |
| *$B_{CI}$ | 1.63±0.07$^{d}$ | 8.30±0.08$^{d}$ | 17.35±0.09$^{d}$ | 26.23±0.07$^{d}$ | 45.25±0.06$^{d}$ | 66.44±0.15$^{d}$ |
| *$J_{CII}$ | 3.23±0.09$^{c}$ | 15.83±0.05$^{c}$ | 20.90±0.06$^{c}$ | 27.85±0.05$^{c}$ | 47.91±0.09$^{c}$ | 68.16±0.20$^{c}$ |
| *$B_{80}J_{20}$ | 7.19±0.06$^{b}$ | 17.60±0.03$^{b}$ | 28.33±0.05$^{b}$ | 39.32±0.05$^{b}$ | 55.19±0.07$^{b}$ | 73.68±0.26$^{b}$ |
| *$B_{70}J_{30}$ | 0.55±0.12$^{e}$ | 8.03±0.21$^{e}$ | 15.35±0.10$^{e}$ | 21.32±0.06$^{f}$ | 33.32±0.12$^{f}$ | 55.34±0.10$^{e}$ |
| *$B_{60}J_{40}$ | 0.22±0.08$^{f}$ | 6.55±0.09$^{f}$ | 14.33±0.07$^{f}$ | 22.43±0.04$^{e}$ | 38.38±0.09$^{e}$ | 50.68±0.59$^{f}$ |
| *$B_{50}J_{50}$ | 0.05±0.04$^{g}$ | 4.54±0.06$^{g}$ | 11.35±0.05$^{g}$ | 15.30±0.06$^{g}$ | 20.94±0.04$^{g}$ | 28.13±0.06$^{g}$ |

*Values are Mean ± Standard Deviation (SD) of triplicate determinations and are presented in percentages (%). Numbers followed by the same letter in the same column do not differ significantly by Duncan's multiple rang test ($p< 0.05$).
Standard= Mannitol
*$B_{CI}$= 90% of (100% *Brokin* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$J_{CII}$= 90% of (100% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{80}J_{20}$= 90% of (80% *Brokin* mango powder:20% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{70}J_{30}$= 90% of (70% *Brokin* mango powder:30% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{60}J_{40}$= 90% of (60% *Brokin* mango powder:40% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)
*$B_{50}J_{50}$= 90% of (50% *Brokin* mango powder:50% *Julie* mango powder): 10% Yellow Pea protein hydrolysate (YPPH)

# 4. Conclusion

In the present study, the *in vitro* antioxidant activity of mango powder blends fortified with YPPH has been evaluated. Generally, the result of this study shows that the fortified mango blends have moderate to potent antioxidant activities and/or free radical scavenging activity. The ability of the fortified mango powder formulations to scavenge DPPH and Hydroxyl radicals, reduce ferric metal ions and chelate transition metals ions indicates their potential utilization in the manage metabolic disorders that arise from excessive levels of reactive oxygen species. The bio-fortified mango powders possess the potential for use as raw material for food ingredients that could be used to formulate functional food and neutraceuticals with multifunctional bioactive properties against various free radicals that may cause oxidative stress.

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