



## Antioxidant Activity of Mango Peel Powder (Mangifera Indical.)

### KEYWORDS

free radicals, antioxidants, dietary components, mango peel.

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**ABSTRACT** Technological way of living in this modern era has significantly increased the amount of free radicals produced in the body thereby increasing the challenge of the body's antioxidant defense system. Excessive production of free radicals contributes to several degenerative chronic illnesses such as cancers and cardiovascular diseases. A number of dietary factors are found to possess antioxidant activity and one among them is mango peel due to the presence of various bioactive components. The present study was carried out to exploit the antioxidant activity of mango peel using five extracts namely acetone, ethanol, petroleum ether, chloroform and aqueous. In vitro antioxidant activity were assessed using DPPH, FRAP and Hydrogen Peroxide assay. Among the extracts analysed it was found that acetone extract exhibited greatest antioxidant activity. The result of the study therefore indicates the use of mango peel as a food ingredient.

### INTRODUCTION

Oxygen is considered to be absolutely essential for the life of aerobic organism. Free radicals or reactive oxygen species (ROS) as well as reactive nitrogen species (RNS) are produced in the body when cells use oxygen to generate energy from cellular redox process (Tiwari, 2004). Free radicals are an atom or molecule that bears an unpaired electron and is extremely reactive, which is capable of engaging in rapid change reaction that destabilize other molecules and generate many more free radicals resulting in oxidative stress. Free radicals damages biomolecules such as proteins, lipids, carbohydrates and breaks DNA strand resulting in cell mutation (Halliwell, 2007). They occupy key role in the etiology of several chronic diseases like arthritis, cancer and atherosclerosis.

Antioxidants are substances that interact and stabilize free radicals and protect cells from damage caused by free radicals. The antioxidants present in nature may be either exogenous or endogenous. The endogenous antioxidants can be classified as enzymatic and non-enzymatic. The non-enzymatic antioxidants are also divided into metabolic antioxidants and nutrient antioxidants. Nutrient antioxidant belonging to exogenous antioxidants are compounds which cannot be produced in the body and must be provided through foods such as vitamin E, vitamin C, carotenoids, and trace elements (Willcox et al., 2004). Mango peel is one such novel food source that possesses antioxidant property.

Mango peels a major by-products composes approximately 7–24% of the total weight a mango fruit (Iqbal et al., 2009; Kim et al., 2012). Recently, mango peels have attracted considerable attention in the scientific community due to their high content of valuable compounds, such as phytochemicals, polyphenols, carotenoids, enzymes, vitamin E and vitamin C, which have predominant functional

and antioxidant properties (Ajila et al., 2007). The present study was carried out to determine the antioxidant activity of mango peel as today's nutraceutical trend is shifting towards finding the impact of natural antioxidants that stabilize food and maximize health impact and to document the use of mango peel as a natural food ingredients.

**METHODOLOGY:** The study was carried out in the following phases:

#### Phase I: Collection and Authentication of Fruit

Fresh mangoes bright yellow in colour with no bruises were purchased from a local market. The fruits were washed under running water and wiped with clean cloth to dry before peeling. The peels were removed using a sterilized sharp knife. On an average around 200 g of peel was obtained from 1 Kg of mango.

#### Phase II: Preparation of peel extract

Preparation of peel extracts were carried out by the method described by (Janarthanam and Sumathi, 2010). One gram of dried mango peel powder were extracted with 20ml of aqueous, ethanol, acetone, and chloroform and petroleum ether and soaked overnight at room temperature. The samples were then filtered through Whatman. No.1 paper in a Buchner funnel. The filtered solution was evaporated under vacuum in a rota-vator at 40°C to a constant weight and then dissolved in respective solvents. The dissolving rate of the extracts was approximately 100%.

#### Phase III: Determination of antioxidant activity

##### 1. DPPH Assay

##### Qualitative analysis of antioxidant activity using DPPH assay

The free radical scavenging activity was measured using

DPPH assay described by (George et al., 1996). 50 µl of test sample was taken in the micro titre plate along with 100µl of 0.1 methanol and 1,1 – diphenyl 2 – picrylhydrazyl (DPPH). This was incubated for 30 minutes, in dark condition. The test sample was observed for discolouration from purple to yellow or pale pink.

#### Quantitative analysis of antioxidant activity using DPPH assay

The antioxidant activity was determined using DPPH assay where DPPH was used as a free radical. 100µl of the test samples were mixed with 2.7 ml of methanol and then 200µl of 0.1 % of methanol DPPH was added. The suspension was incubated for 30 minutes in dark condition. Initially absorption of blank containing the same amount of methanol of DPPH solution was prepared and used as control. Subsequently at every 5 minutes interval, the absorption maxima of the solution was measured using UV double beam spectra scan at 517 nm. Antioxidant activity was compared with known a synthetic standard of (0.16%) of Butyric Hydrated Toulene (BHT). The ability of the test sample to scavenge DPPH radical was calculated by the following equation:

Percentage of inhibition was calculated using the formula:

$$\frac{\text{Absorbance in control} - \text{Absorbance in sample}}{\text{Absorbance in control}} \times 100$$

#### 2. Ferric reducing antioxidant power (FRAP)

FRAP assay was carried using the standard method described by (Benzie and Strain, 1996). FRAP assay is based on the ability of antioxidants to reduce Fe<sup>3+</sup> to Fe<sup>2+</sup> in the presence of TPTZ, forming an intense blue Fe<sup>2+</sup>-TPTZ complex with an absorption maximum at 593 nm. This reaction is pH-dependent (optimum pH 3.6). 0.1 ml of the test sample was added to 3.0 ml FRAP reagent [10 parts 300 mM sodium acetate buffer at pH 3.6, 1 part 10 mM TPTZ (2,4,6- tripyridyl-s-triazine) in 40 mM HCl and 1 part 20 mM FeCl<sub>3</sub>. The reaction mixture was incubated at 37°C for 10 min and then the absorbance was measured at 593 nm. FeSO<sub>4</sub> (100 to 1000 µM ml<sup>-1</sup>) was used as a positive control (Thondre et al., 2011). The antioxidant capacity was based on the ability to reduce ferric ions of sample and was calculated from the linear calibration curve and expressed as FeSO<sub>4</sub> equivalents per gram of extracted compound.

#### 3. Hydrogen peroxide Scavenging Assay

A solution of hydrogen peroxide (20 mM) was prepared in phosphate buffer saline (PBS at pH 7.4). The test sample and standards in methanol (1 ml) were added to 2 ml of hydrogen peroxide solutions in PBS. After 10 minutes, the absorbance was measured at 230 nm against a blank solution. The percentage of inhibition was calculated and the scavenging activity of crude extract, column fraction and compound was compared.

$$\% \text{ of Scavenged } H_2O_2 = \left[ \frac{A_0 - A_1}{A_0} \right] \times 100$$

A<sub>0</sub> = Absorbance of control  
A<sub>1</sub> = Absorbance of Sample/ Standard

### RESULTS

#### Antioxidant activity using DPPH assay

Scavenging activity of free radical such as 1,1-diphenyl-2-picrylhydrazyl (DPPH) has been widely used to evaluate the antioxidant activity of natural products from plant sources. Qualitative analysis of antioxidant by DPPH assay

clearly reveals that aqueous, acetone and ethanol extract had the ability of scavenging free radicals which was evidenced by change of DPPH colour from purple to yellow thus indicating its anti-oxidant property (table 1).

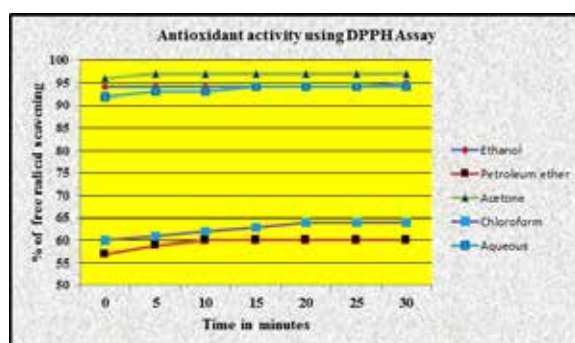
**Table 1**  
Qualitative antioxidant activity of mango peel powder (Mangifera indica L.) using DPPH Assay

Extract	Antioxidant activity
Aqueous	++
Acetone	++
Ethanol	++
Petroleum ether	-
Chloroform	-

The result pertaining to quantitative analysis of antioxidant using DPPH assay is presented in table 2 and depicted graphically in figure 1. DPPH is a stable radical and has been widely used for studying the free radical scavenging activity. It is evident from table 2 that the ability to scavenge free radicals increased as time increased from 0 to 30 minutes. Among the extracts analysed it was found that acetone, aqueous and ethanol extracts possessed greater antioxidant activity due to their potential ability to scavenge more free radicals followed by petroleum ether and chloroform extract which had the ability to scavenge free radicals to a lesser extent.

**Table 2**  
Quantitative antioxidant activity of mango peel powder (Mangifera indica L.) using DPPH Assay

S.No	Solvents	Time (in minutes)						
		0	5	10	15	20	25	30
1	Ethanol	94	94	94	94	94	94	95
2	Petroleum ether	57	59	60	60	60.3	60.3	60.3
3	Acetone	95.8	96.6	96.6	96.6	96.6	96.6	96.6
4	Chloroform	59.5	61.15	61.9	62.8	63.6	63.6	63.6
5	Aqueous	91.7	93.3	93.3	94.2	94.2	94.2	94.2



**Figure .1.** Antioxidant activity of mango peel powder using DPPH assay

#### Antioxidant activity using FRAP assay

FRAP assay (ferric reducing ability of plasma) has been used extensively in evaluating the total antioxidant power. As acetone extract was found to exhibit the highest antioxidant activity using DPPH assay it was further subjected to FRAP assay. The ferric ion-reducing activity of the acetone extract of mango peel was found to be 1.984 mM FeSO<sub>4</sub> eq (table 3). The higher the frap value the greater is the anti-oxidant activity (Dennog et al., 1999). The result ob-

tained from the study is in par with a previous study carried out by (Imran et al., 2013) which indicated that FRAP value was found to be maximum in the acetone extract of mango peel ( $1.88A \pm 0.19$  mmol/100g).

**Table 3**  
**Antioxidant activity of mango peel powder (*Mangifera indica*.L) using FRAP Assay**

Extract	Concentration ( Gram / Feso4 Equivalent)
Acetone	1.984

#### Antioxidant activity using Hydrogen peroxide assay

The hydrogen peroxide scavenging ability of mango peel and gallic acid that was used as standard is presented in (table 4). From the table it is evident that the acetone extract exhibited 49.04% scavenging activity on hydrogen peroxide while gallic acid exhibited 57.32% hydrogen peroxide scavenging activity.

**Table 4**  
**Antioxidant activity of mango peel powder (*Mangifera indica*.L) using Hydrogen Peroxide Assay**

Sample	Percentage
Standard( Gallic acid)	57.32%
Acetone extract	49.04%

#### DISCUSSION

The edible mango peel has considerable value as a source of polysaccharides (dietary fiber, starch and pectins) and antioxidant pigments (Rocha et al., 2007; Ajila and Prasada, 2008). Peel and pulp contains carotenoids, such as the provitamin A compound, beta-carotene, lutein and alpha-carotene (Gouado et al., 2007). Polyphenols found in mango peels includes quercetin, kaempferol, gallic acid, caffeic acid, catechins, tannins, and the unique mango xanthone, mangiferin play a vital role in counteracting free radicals (Singh et al., 2004; Mahattanatawee et al., 2006; Barreto et al., 2008). An extract of mango branch bark called vimang, contains numerous polyphenols with in vitro antioxidant properties (Rodeiro et al., 2006) and on blood parameters of elderly humans (Pardo et al., 2006).

Two major valuable compounds, namely ethyl gallate and penta-O-galloyl-glucoside, have been isolated from mango peels that had potent ability to scavenge free radicals such as hydroxyl radical and superoxide anion indicating that mango peel can be utilized in experimental and clinical sense (Jiang et al., 2010). Ajila et al., (2007) examined the major antioxidants, such as polyphenols, anthocyanins and carotenoids, in Indian Raspuri and Badami (ripe and raw) mango peel extracts. Their results showed that ripe peels contained higher amounts of anthocyanins (360–365 mg/100 g) and carotenoids (194–436 lg/g) compared to raw peels, while raw mango peels had high polyphenol content (90.18–109.7 mg/g) thus indicating mango peel extracts to be used as a nutraceutical agent.

#### CONCLUSION

Recent researches indicate that natural products derived from plants continues to be a rich therapeutic agent used in treating diseases caused due to oxidative stress. The need for effective, potent pharmacological components possessing antioxidant activity with low or no side effects for use in protective medicine is necessary and this study clearly highlights the use of mango peel powder as a natural functional food supplement due to its antioxidant property and its application in food industry.

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