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Antioxidant Properties of the By-product Indonesian Favourable Fruits

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Fruit seed waste is part of the fruit that cannot be eaten, abundant and its use is still limited so that it is often considered garbage by the community. One effort to reduce waste is by utilising metabolism as secondary as fruit seed waste. This research determined the phytochemistry, total phenolic, total flavonoid and antioxidant of *Dimocarpus longan lour var diamond river, Mangifera indica var podang, Cucumis melo L var retikulus,* and *Parcia americana var miki mentega* seeds. It had been extracted using 3 various solvents (n-hexane, methanolic, and ethyl acetate). The flavonoid was found in all fruit seeds methanol extract. Saponin and tanin were found in all fruit seeds methanol extract and *M. indica var podang* and *P. americana var miki mentega* ethyl acetate extract. The alkaloid was found in all fruit seeds in ethyl acetate and *n*-hexane extract. The Folin-Ciocalteu assay was employed to evaluate the overall phenol content, with a range that took away 22.6 - 142.17 mg/g GAE. The highest total phenolic was found in *M. indica var podang* seed with methanolic extract. Total flavonoid compounds were determined by aluminium chloride calorimetric, with range 20.32 - 40.27 mg/g Quercetin Equivalent. The highest total flavonoid was found in *M. indica var podang* seed with methanolic extract. The potential antioxidant activities were assessed using the DPPH (diphenyl picryl hydrazyl) radical scavenging capacity, which showed a range of 15.23 to 87.23 percent activity of scavenging , the highest percentage of DPPH radical scavenging was acquired in *M. indica var podang* seed with methanolic extract, with the best IC_{50} 65.75 µg/mL.

Keywords: fruit seed; total phenol; total flavonoid; antioxidant; DPPH radical scavenging

I. INTRODUCTION

Solar UV radiation on living cells can create diversities of danger such as a chemical photograph, photo-isomerisation, and photo-oxidation. Photo oxidation reaction takes place as an outcome of the release reactive oxygen species (ROS) in the form, hydroxyl radicals (OH), hydrogen peroxide (H_2O_2) and superoxide anion (O_2^-), by a chromosphere that absorbs ultraviolet light (Gadri *et. al.,* 2012; Safitri *et al.,* 2016). All of ROS accomplished of reacting with membrane lipids, nucleic acids, proteins and enzymes, and other small molecules, resulting in cellular destruction (Kumar, 2014) and it likewise can be involved in numerous diseases like cancer, neurodegenerative, diabetes (Garaguso & Nardini, 2015). Guard against the harm of ROS, humans have evolved a very refined and complicated antioxidant safeguard system (Kumar, 2014).

Antioxidants can be incorporated *in vivo*; the body has confidence in several internal immunity systems. The enzyme, for example, reduced superoxide dismutase (SOD) and glutathione (GSH), assimilate oxidises poison halfway and demand micronutrient cofactor, for example, selenium, iron, copper, zinc, and manganese for ideal catalytic activity. Intake and assimilation of these essential trace minerals may downturn with ageing, and it could be concerned with the internal antioxidant immunity system. The inclusion of

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supplement with external antioxidants or an advance of internal antioxidant immunity of the corpse has been established to be an encouraging strategy of responding to the abominable belongings of stress caused by oxidation (Kasote *et al.,* 2013). Plants have long been a point of supply to the external antioxidant.

The plant incorporates and collects a variety of secondary metabolites of low and high molecular weight. These metabolites are likewise essential to alteration of plants to climate (Baier *et. al.,* 2005; Kasote *et al.,* 2015). Some secondary antioxidant metabolites take place fundamentally, others are constructed in feedback to biotic and abiotic emphasis circumstances (Nicholso *et. al.,* 1992; Bailey *et. al.,* 2005; Kasote *et al.,* 2015). The amassing of phenolic compounds ahead with the improvement of phenylpropanoid metabolism has been attended below diverse ecological emphasis circumstances (Michalak, 2006; Kasote *et al.,* 2015). In plants, phenolic can deed as an antioxidant by contributing electron to guaiacol-type for H_2O_2 detoxification was produced under high-emphasis conditions (Sakihama *et. al.,* 2002; Kasote *et al.,* 2015). Phenolic likewise supports safeguard adjacent UV emission over the capability to tramp radical.

The fusion of flavonoids is commonly known to be persuaded due to UV accentuation, depressed temperature or low temperature, and few nutrient statuses, that capable of an attribute to the absorption of UV rays, radical tramp, and alloy sequestering agents (Michalak, 2006; Winkel-Shirley, 2002; Rivero *et. al.,* 2001; Kasote *et al*., 2015). Flavonoids, an enormous group of plant total phenolic, are produced in plant tissues, such as fruits, vegetables, seeds and leaves in nearly tremendous concentration (Ciou *et al.,* 2008).

The fruit of *D. longan lour*, *M. indica*, *P. americana*, and *C. melo L* has been the greatest consumed and exported flesh fruits worldwide mainly in Indonesia. It has great economic benefits and is cultivated in assorted regions of the world because of alteration diverse kinds of soil and humidity. The primary wastes assembled throughout these fruits processing are seeds and peels in the variety of 35-60 precents (Namngam *et al.,* 2018). Fruit seeds can be conceivably abused as antioxidant promoters and nutraceuticals since this by-product might consist of a great number of bioactive constituents. Extract of *D. longan* seed was recorded to wield effective antioxidant activities on scavenging radicals (Zheng *et. al.,* 2009; Soong & Barlow, 2004; Chen *et al.,* 2014). Mango seed kernel presented antioxidant achieve as an outcome of polyphenols, phytosterols and microelements such as zinc, copper, and selenium (Soong & Barlow, 2004; Kraur & Brar, 2017). Antioxidant activity and phenolic content of seeds of avocado pear were resulted to be higher than 70% (Soong and Barlow, 2004). Oriental melon seeds were recorded to detachment therapeutic achieve such as antioxidant, anti-inflammatory, and analgesic properties (Gill *et al.,* 2009).

Nevertheless, further research on the antioxidant in abundant fruit seeds in the other countries have been done, but its Indonesian local varieties are not studied yet. The object of this research was to scrutinise antioxidant belongings involve total phenolic, total flavonoids of Indonesian local diversification fruit seeds. It is advised that the microclimate may be pretentious to the antioxidant properties of the constituents.

II. MATERIALS AND METHOD

A. Material

1. Chemical

All reagents and chemicals were purchased in high grade from Merck, including, methanol, dichloromethane, *n*-hexane, DMSO (dimethyl sulfoxide), ethyl acetate, H2SO4 (sulfuric acid) ferric chloride, HCl (hydrochloric acid), glacial acetic acid, potassium iodide, chloroform, and iodine. TCI, Tokyo, Japan (Tokyo Chemical Industries) supplied the 2,2-diphenyl-1 picrylhydrazyl (DPPH). Wako Pure Chemical Industries, Osaka, Japan supplied the Gallic acid. Folin-Ciocalteu and quercetin were supplied from Merck, Darmstadt, Germany.

2. Fruit seed material and preparation of the sample

Fruit seeds (*D. longan lour var diamond river, P. americana var miki mentega, M. indica var podang, C. melo var rerikulus*) was gathered from a home farm in the Kediri-East Java region, Indonesia, in January 2018. Fruit seeds were collected. The distilled water was used to wash fruit seeds then reduce to little pieces, dried in an air dryer set to 30 °C overnight, and grind into powder with a grinder.

B. Methods

1. Extraction of fruit seeds

The sample fruit seeds powder (20 g) was pour into 350ml Erlenmeyer flask, dissolved in 200 mL of solvents (*n*-hexane, ethyl acetate, and methanol), and then wrapped separately and tightly with aluminium foil, after which it was extracted and shaken in anticipation of 24 hours at 180 rpm. The suspensions were then filtered through filter paper by Whatman number one. The supernatants were extracted and evaporated in a rotary evaporator while maintaining a temperature of 68 °C particularly for *n*-hexane extract, 77 °C for the extract of ethyl acetate, and 65 °C for an extract of methanol to obtain dry extracts. Solvent-free extracts were transferred into extraction vials and stored at 4 °C for later use.

2. Phytochemical screening of Fruit seed

Phytochemical's screening of a variety of extracts fruit seeds was done qualitatively to detect the existence of saponins, flavonoids, tannins, triterpenoids, and alkaloids.

i. Analysed of alkaloids

Dragendorff's test was used to detect alkaloids: two mg extract of fruit seeds were added to five mL of distilled water, after that, 2 M HCl (hydrochloric acid) was added until the acid reaction occurs., and one mL of Dragendorff's reagent was added. The presence of alkaloid was indicated by the formation of a red-orange precipitate. (Sariwati *et. al.,* 2019; Abdulahi *et. al.,* 2013; Joshi *et. al.,* 2013; Iqbal *et al.,* 2015).

ii. Test for saponins

The fruit seeds extracts (0.5 g) were flustered into 10 mL aqua distillation within a glass of 15 tubes. The presence of saponins is expressed by the formation of foaming that lasts for 5 minutes after being heated up (Sariwati *et. al.,* 2019; Iqbal *et. al.,* 2015; Banso & Adeyamo, 2006).

iii. Test for tannins

The fruit seeds extracts (0.5 g) were mixed with 10 mL aqua distillation and then separated in order to refine, adding almost no drops of 5% $FeCl_3$ (ferric chloride). Black or turquoise formations indicate the presence of tannins (Sariwati *et. al.,* 2019; Iqbal *et. al.,* 2015; Banso & Adeyamo, 2006).

iv. Analysed of steroids

Boiling fruit seed extracts (0.5 g) in 10 mL ($CHCl_3$) chloroform and filtered, after which 1 mL of acetate acid (CH_3COOH) and only a few drops 37% H_2SO_4 (sulphuric acid) were added to the filtrate. Steroids were indicated by a green ring (Sariwati *et. al.,* 2019; Samejo *et al.,* 2013).

v. Test for flavonoids

The methanol (0.5 mL) was used to dissolve 0.5 mg fruit seeds extract, then fewer drops of diluted (NaOH) sodium hydroxide solution were added. The yellow formation followed by the addition of very little sulfuric acid renders the extracts colourless, indicating the existence of flavonoids (Sariwati *et. al.,* 2019; Alabri *et al.,* 2014).

vi. Test for triterpenoids

The fruit seed extracts (5 mL), 2 mL chloroform, and fewer drops of H_2SO_4 were mixed. The presence of triterpenoids was expressed by the creation of the blue-green ring. (Sariwati *et. al.,* 2019; Samejo *et al.,* 2013).

3. Phenolic content in total

The 100 L of extracts (20 mg fruit seed extract diluted in 3% HCL and 60% methanol) was added with 2 mL sodium carbonate. After 3 minutes, the Folin-Ciocalteu reagent was added to the mixture. After 30 minutes of standing, the absorbance at 725 nm was measured. The standard curve was plotted by utilising Gallic acid 20, 40, 60, 80, and 100 µg/mL. Phenolic content in total precipitate indicated the amount of (GAE) Gallic acid equivalent in milligrams per gram of extract (Sariwati *et. al.,* 2019; Tsai *et al.,* 2009).

4. Total Flavonoid Content Determination

Total flavonoid content was measured using a modified version of Kaur and Mondal's (2014) aluminium chloride colorimetry test. In brief, 125 L of crude extracts was combined with 625 µL of deionised water and 37.5 µL of 5% sodium nitrite. After allowing the mixture to stand for six minutes, 75 L of 10% aluminium chloride-6-hydrate was added. After five minutes, 250 µL sodium hydroxide solution was added, followed by 137.5 µL deionised water, which was mixed. The absorbance at 510 nm was immediately measured on the blanks (obtained by replacing the plant extract with deionised water). Measurements were corrected for standard

curves for prepared quercetin solutions (25, 30, 35, 40, 45 μg/mL). TFC expressed in mg of quercetin equivalents (QE) per 100-g dry weight (DW) (Sariwati *et al.,* 2019).

5. Antioxidant capacity (ability to trap DPPH free radicals)

The scavengers of reactive oxygen species in solutions of each extract against DPPH radicals were determined as previously described. The stock solution was 24 mg DPPH diluted in 100 mL methanol, and the absorbance was measured at 517 nanometres. The 33 μL of samples at various concentrations of 10 until 500 μg/mL were mixed with one ml of DPPH stock solution, agitated, and incubated at room temperature for 20 minutes in the dark. Control only with stock solution for DPPH. Scavenger capacity was hypothesized to pay attention to the percentage of DPPH radical scavengers. as a subsequent analogy of radical scavenger inhibition (Amalia and Sariwati., 2019).

$$\% = \frac{\boldsymbol{Control\ Absorbance - Sample\ Absorbance}}{\boldsymbol{Control\ Absorbance}}\ \boldsymbol{x\ 100}$$

The IC50, which reflects the sample's concentration required to capture 50% of the free radicals DPPH, was assessed by accomplishing a variable gradient pairing response model (Sariwati *et. al.,* 2019; Fitriana *et al.,* 2016).

6. Analytical statistics

Values were the median of 3 replicates. The student t-test was used to identify significant differences within or between groups during substrate transformation. The dissimilarity level of five percent ($P < 0.05$) was found to be statistically significant throughout (Sariwati *et al.,* 2019)

III. RESULT AND DISCUSSION

A. Extraction Yields

Extraction yield briefly to the allotment of extracts, which acquired from powdered fruit seeds by utilising a solvent extractions method for further isolation and application. Table 2 presented that among four fruit seeds (*D. longan lour var diamond river, P. americana var miki mentega, M. indica var podang,* and *C. melo var rerikulus*) with three solvents, *P. americana miki mentega* in methanol extract assembled the substantial extraction yields (7.07 g dried extract). However, this yield was not unavoidably different from the *D. longan lour var diamond river* methanol extract (6.26 g dried extract). Its dissimilarity outcome when *P. americana miki mentega* and *D. longan lour var diamond river* extraction with *n*-hexane formed lowest extraction yields (0.38 g and 0.42 g dried extract).

Yield results of maceration extracts with dissimilarity solvent assemble distinct yield percentage (Salamah *et al.,* 2008). Extracted polar solvents were necessarily higher yield than the nonpolar counterpart. *M. indica var podang* with methanol solvent was the highest extracts yields. The effectiveness of methanol as a solvent disclose to its middle polarity, which acknowledges for the solvation of low molecular weight organics compounds acquired the proton table functional groups (for example OH, COOH) (Nguyen *et al.,* 2015). Methanol solvent produced the best extraction results, possibly due to the greater solubility of proteins and carbohydrates in methanol (Do *et. al.,* 2014; Zielinski *et al.,* 2000).

B. Phytochemistry

Table 1 showed the results of the phytochemical screening. Flavonoids and other secondary metabolites were detected in all extracts. The existence of alkaloids was to establish all fruit seeds with ethyl acetate and *n*-hexane extracts. The existence of triterpenoid vanished in all extracts. The existence of tannins was established all fruit seeds methanol extracts and *M. indica var podang* and *P. americana var miki mentega* ethyl acetate extracts, while saponin compound was established in all fruits in methanol and *P. americana var miki mentega* and *C. melo var retikulus* ethyl acetate extract, *D. longan lour var diamond river*, *P. americana var miki mentega, M. indica var podang n*-hexane extracts. Phytochemical screening of dissimilarity extracts of four fruit seeds presented different outcomes, which the existence primary of active chemical compounds such as flavonoids. The comprehensive therapeutic impacts of flavonoids can be greater ascribed to their antioxidant properties (Kumar, 2014). All fruit seeds in methanol extracts assembled secondary metabolites tannin and saponin. Tannins and their derivatives are phenolic compounds considered to be important antioxidants or scavengers of free radicals (Ayoola *et. al.,* 2008; Najafi *et. al.,* 2010; Barile *et. al.,* 2007; Alabri *et.*

al., 2014; Sekar *et. al.,* 2012; Varahalarao & Kaladhar, 2012). Saponins are called bioactive compounds that sophisticated in plant protection systems because of their antioxidant activity and likewise anti-inflammatory promoters (Samejo *et. al.,* 2013; Najafi *et al.,* 2010). Nevertheless, all fruit seeds in ethyl acetate and *n*-hexane extracts likewise were exposed to other bioactive compounds, an alkaloid. Numerous alkaloids drifted from medicinal plants presented biological activities identical to antimicrobial (Iqbal *et. al.,* 2015; Benbott *et al.,* 2012). The current recorded that *D. longan lour var fenke* seeds from Taiwan consist of great amounts of bioactive constituents, similar to phenolic acids, flavonoids and polysaccharides (Zheng *et al.,* 2009). Egyptian *M. indica* seed karnels consist of varied polyphenols, counting flavonols (Abdalla *et al.,* 2007). *P. americana* from Nigerian consists of phenolic contents and flavonoids Folasade *et al.,* 2016). Brazilian's *C. melo var retikulus* consist of polyphenol, flavonoid, and tannin (Rolim *et al.,* 2018).

Table 1. Phytochemistry of fruit seeds with a different solvent

Treatment	Phytochemistry				
	Saponin	Tanin	Triterpenoid	Alkaloid	Flavonoid
Methanol solvent					
D. longan lour var diamond river	+	+	-	-	+
P. Americana var miki mentega	+	+	-	-	+
M. indica var podang	+	+	-	-	+
C. melo var retikulus	+	+	-	-	+
Ethyl acetate solvent					
D. longan lour var diamond river	-	-	-	+	+
P. Americana var miki mentega	+	+	-	+	+
M. indica var podang	+	+	-	+	+
C. melo var retikulus	-	-	-	+	+
***n*-Hexane solvent**					
D. longan lour var diamond river	-	-	-	+	+
P. Americana var miki mentega	-	-		+	+
M. indica var podang	-	-	-	+	+
C. melo var retikulus	-	-	-	+	+

C. Total Phenolic Compound

The Folin-Ciocalteu assay was carried out to determine the phenol content in total of the fruit seeds. Classification of total phenol content was obtained using the standard curve procedure utilising gallic acid (y = 0.0028x - 0.457; r2 = 0.9916), where y was the optical density and x represents the concentration of the solution of gallic acid (μg/mL), mg/g display as GAE. Table 2 showed the total phenolic content of fruit seed extracts. *M. indica* var podang had the greatest phenolic compounds in total (142.18 mg/g GAE) in methanol extract, one of the different solvents of fruit seed extracts, assiduously adhere to *D. longan lour var diamond river* in methanol extract (130.85 mg/g GAE). However, this result remarkable contrasting with *M. indica var podang* in ethyl acetate extracts assembled total phenol (92.64 mg/g GAE), assiduously adhere to *Parcia americana* in ethyl acetate extract (75.92 mg/g GAE) and the minimum was acquired by all fruit seed in *n*-hexane extract approximately 22 mg/g GAE). One of twelve extracts of fruit seeds, *M. indica var podang* in methanol extract presented the greatest quantity of phenol compound (142.18 mg GAE/g), adhere to *D. longan lour var diamond river* in methanol extract (130.85 mg GAE/g). All fruit in *n*-hexane extract had a little phenolic compound. The convalescence of phenolic compounds in dissimilar samples is affected by the polarity of extracting solvents and solubility of each compound in the solvent apply

for the extraction process (Iloki-Assanga *et. al.,* 2015; Sulaiman *et. al.,* 2011; Alothman *et al.,* 2009). Polarity of the solvent possesses an important rule enlarging phenolic solubility (Naczk & Shahidi, 2006; Medini *et al.,* 2014). This total phenolic compound is made up of a chemical class of compounds found in methanol extract. Aromatic metabolites with one or more acidic phenolic hydroxyl groups are known as phenolic metabolites, they are further subdivided into hydroxycinnamic acids, tannins, anthocyanins, and flavonoids (Unal *et al.,* 2014). Because tannin compounds account for more than half of polyphenol compounds, the capability of reducing Fe (III) in Fe (II) is proportional to tannin's contribution to polyphenol compounds (Bangou *et al.,* 2012).

This result is comparable to the current record of methanol being demonstrated as a powerful solvent for the extraction of phenolic contents (Iloki-Assanga *et. al.,* 2015; Bangou *et al.,* 2012). *N*-hexane was the minimum total phenolic compound that may be ascribable to the compound of more nonphenolic compounds such as carbohydrate and terpene than in order extract. This outcome resemblance with current work that verifies the greatest total phenol compound of Brazilian's *Cucumis melo var retikulus* was in hydroethanolic (70:30) was acquired 111.72 mg/g GAE (Rolim *et al.,* 2018). It is outcome distinct substantial with current recorded that the total phenol of *M. indica* seeds from Pakistan with methanolic extracts was acquired 45.56 mg/g GAE (Sultana *et al.,* 2012). Chinese *D. longan lour* seeds in ethanolic extracts were acquired 23 mg/g GAE) (Song & Barlow, 2006). *P. americana var mills* from Nigeria in the ethanolic extract was acquired 8.72 mg/g GAE (Folasade *et al.,* 2016). The greater total phenol compound could have a relation with existence of polyamines as a stress indicator in plant (Unal *et al.,* 2014).

D. Total Flavonoid Contents

Total Flavonoid compounds of the fruit seed were analysed with aluminium chloride colourimetry assay. Classification of total flavonoid content was obtained by standard curve procedure utilising quercetin (y = 0.02894x - 0.04823; r2 = 0.9959), where y was optical density, x was concentration of quercetin solution (µg/ml) mg QE / 100g dry Weight (mg/ 100 g QE DW).

Total Flavonoids of Fruit seeds extracts were presented in the Table 2. *M. indica var podang* in methanol extract had the greatest total phenolic compounds (40.27 mg/100g QE Dry Weight (DW)) one of the varied solvent of fruit seeds extracts, assiduously adhere to *D. longan lour var diamond river* in methanol extract (37.54 mg/100g QE DW). However distinct substantial with *M. indica var podang* in ethyl acetate extracts assembled total phenol (37.54 mg/100g QE DW), assiduously adhere to *P. americana* in ethyl acetate extract (34.43 mg/100g QE DW) and the minimum was acquired by all fruit seed in *n*-hexane extract nearly (20 mg/100g QE DW). Flavonoids and flavonols possess antioxidant activity due to radical foraging or sequestering agents (Pnourmoed *et. al.,* 2006; Iloko-Assanga *et al.,* 2015). *M. indica var podang* in methanol extract presented the greatest content of total flavonoid compounds (40.27 mg QE/g), succeeded by *D. longan lour var diamond river* in methanol extract (37.54 mg QE/g). All fruit in *n*-hexane extract had less flavonoid content. A flavonoid is a group of polyphenolic compounds (Tensiska *et al.,* 2007) that have a hydroxyl group substituted in ortho position to the –OH and –OR (Andayani *et al.,* 2008). Methanolic as an efficient solvent for the extraction of flavonoids (Iloki-Assanga *et. al.,* 2015; Siddhuraju & Becker, 2003). Extract of *n*-hexane has fewer absolute flavonoids. Flavonoid may consist of nonpolar contents such as aglycone (Tensiska *et al.,* 2007).

Table 2. Extraction yields, Phenolic content, Flavonoid content, DPPH Radical Scavenging, and IC_{50} of fruit seeds

Treatment	Bioactivities				
	Extraction Yield (g)	Phenolic content (mg/g) GAE	Flavonoid content (mg/100g) QE	DPPH Radical Scavenging (%)	IC_{50} µg/mL
Methanol solvent					
D. longan lour var diamond river	6.26	130.85 ± 0.28[a]	37.54 ± 0.02[a]	82.20 ± 0.05[a]	68.44
P. americana var miki mentega	2.31	67.68 ± 0.07[b]	30.23 ± 0.03[b]	74.43 ± 0.13[b]	80.34
M. indica var podang	7.07	142.18 ± 0.16[c]	40.27 ± 0.01[c]	87.23 ± 0.03[c]	65.75
C. melo var retikulus	1.84	34.84 ± 0.06[d]	28.60 ± 0.12[d]	66.34 ± 0.21[d]	83.29
Ethyl acetate solvent					
D. longan lour var diamond river	1.56	36.48 ± 0.00[e]	23.78 ± 0.06[e]	36.87 ± 0.01[e]	-
P. Americana var miki mentega	3.83	92.64 ± 0.55[f]	35.31 ± 0.01[f]	80.65 ± 0.04[f]	70.36
M. indica var podang	2.48	75.92 ± 0.10[g]	34.43 ± 0.05[g]	76.23 ± 0.08[g]	76.15
C. melo var retikulus	1.35	36.51 ± 0.01[h]	25.74 ± 0.07[h]	34.86 ± 0.03[h]	-
***n*-Hexane solvent**					
D. longan lour var diamond river	0.42	24.29 ± 0.01[i]	21.72 ± 0.17[i]	17.85 ± 0.07[i]	-
P. Americana var miki mentega	0.38	25.98 ± 0.01[j]	22.18 ± 0.21[j]	18.67 ± 0.04[j]	-
M. indica var podang	1.46	22.62 ± 0.02[k]	20.32 ± 0.11[k]	15.23 ± 0.21[k]	-
C. melo var retikulus	1.14	31.66 ± 0.02[l]	23.56 ± 0.03[l]	22.41 ± 0.17[l]	-

(-) not measured. The data was used to calculate the standard deviation of the mean (n = 3). On each row, data following the same minor letter are significantly different (P< 0.05).

This outcome distinct substantial with current recorded that *P. americana var mills* from Nigeria in the ethanolic extract was acquired 1.72 mg/g QE (Folasade *et al.,* 2016). Brazilian's *C. melo var retikulus* was in hydromethanolic (70:30) was acquired 93.00 mg/g QE (Rolim *et al.,* 2018) Chinese's *D. longan lour var shixia* seeds in ethanolic extracts was acquired 1.94 mg/g Rutin Equivalent) (Teng *et al.,* 2019). Egyptian *M. indica var anacardiaceae* was acquired 3325 mg/g Catechin Equivalent (Aty *et al.,* 2018).

E. DPPH Radical Scavenging and IC_{50}

Total antioxidant potential of varied solvent extracts of fruit seed was analysed by employing the standard curve Gallic acid (y = 34.80 ln (x) – 109.8; r2 = 0.970). The DPPH scavenging capacity of *M. indica var podang* in methanol extract had the greatest (87.23%) one of the varied solvents of fruit seed extracts, assiduously adhere to *D. longan lour var diamond river* in methanol extract (82.20%) was presented in Table 2. It is distinct substantial with *M. indica var podang* in ethyl acetate extracts assembled total phenol (80.65%), closely followed by *P. americana var miki mentega* in ethyl acetate extract (76.23%) and the lowest was acquired by all fruit seed in *n*-hexane extract nearly 20%. Gallic acid, the positive control, inhibited DPPH the most (97.80%). *M. indica* var podang methanol extract was found to be the most effective antioxidant, with IC50 values of

DPPH radicals foraging 65.75 g/mL.

The free radical foraging activity of the fruit seeds extract was analysed by employing the DPPH procedure. The DPPH method works on the principle that antioxidant promoters react with DPPH to form solid free radicals. Hence, the DPPH alteration possesses color slowly. This circumstance suggests the sample consist of antioxidant promoters (Hossain *et al.,* 2014). *M. indica var podang* had the highest DPPH scavenging capacity in methanol extract (87.23%), while the positive control, Gallic acid, had lower inhibition of DPPH (97.80%). The dissimilarity extracts acquired variance as secondary metabolites component as presented by the antioxidant ability outcome (Janet *et al.,* 2015). The presence of flavonoids, tannins, and saponin may contribute to the great antioxidant capacity of methanol extract (Table 1). Flavonoids' antioxidant properties range from the ability to donate a hydrogen atom or transmit an electron to a free radical compounds to the ability to form complexes with metals (Redha, 2010). Saponin compounds are radical scavenger by producing hydrogen peroxide as an intermediate and can donate hydrogen to DPPH radical compound that eliminates radical link reactions (Nurjanah *et. al.,* 2015; Xiong *et al.,* 2010). Tannins have the capabilities to seizure-free radicals. These compounds are very efficacious as an electron donor and hydrogen atom and sequestering metals because it has a hydroxyl group and linked double bonds that authorise the delocalisation of electron (Nurjanah *et al.,* 2015). *N*-hexane extract necessitates a higher concentration to achieve DPPH radical foraging. This is due to the nonpolar solvent's ability to contain only nonpolar compounds, for example, fats, wax, and essential oils. (Nurjanah *et. al.,* 2015; Suratmo, 2009).

A compound's IC_{50} was contrarily closed to its antioxidant capacity, as it conveys the amount of antioxidant needed to reduce the concentration of DPPH by 50 percent, which is acquired by interpolation as determined by analysis of linear regression. Fewer IC_{50} suggest a greater antioxidant activity of a compound (Do *et. al.,* 2014; Liu *et al.,* 2009). In this study, *M. indica var podang* in methanol extract presented the optimal antioxidant with IC_{50} has acquired 65.75 µg/mL. These outcomes are constant with phenol bioactive compounds noticed in the extract of methanol involving tannin, saponin, and flavonoids, which revealed that the methanol extract was the higher absolute phenolic compound. Commonly, the majority of antioxidants are phenolic compounds with hydroxyl groups in the ortho position concerning the –OH and –OR (Nurjanah *et. al.,* 2015; Andayani *et al.,* 2008). All fruit seed methanol extract is categorised as a vigorous antioxidant due to IC_{50} rates lower than 100 ppm (Nurjanah *et. al.,* 2015; Molyneux, 2004). This outcome distinct substantial with current recorded that Pakistan's *M. indica* in methanolic have ability DPPH radical scavenging was acquired 48.1 % (Sultana *et al.,* 2012). *P. americana var mills* from Nigeria in the ethanolic extract was acquired 75.45% (Folasade *et al.,* 2016). Brazilian's *C. melo var retikulus* was in hydromethanolic (70:30) was acquired 4.8% (Rolim *et al.,* 2018). Chinese's *D. longan lour var shixia* seeds in ethanolic extracts was acquired 75% (Teng *et al.,* 2019).

F. Correlation Total Phenol, Total Flavonoid and Antioxidant

The correlation within the total phenolic contents, total flavonoid content and antioxidant activity fruit seeds were assessed. There was a relation within the total phenolic compound, total flavonoid compound, and antioxidant activity in addition the IC_{50}. A substantial and be line correlation existed within the antioxidant activity and phenolic content, flavonoid content in a total of fruit seed in methanol extract, thus indicating that phenolic compounds and total flavonoid are major contributors to antioxidant activity (Maizura *et al.,* 2011), a property drifted from their redox capabilities, which can quench and neutralise the free radical (Iloki-Assanga *et. al.,* 2015; Florence *et al.,* 2011). This result is not significantly different from the current record, which assessed the relationship between antioxidant activities and total phenolic of 133 Indiana medicinal plant species (Sarveswaran *et. al.,* 2007; Hardiana *et. al.,* 2012; Bangou *et. al.,* 2012; Nurjanah *et al.,* 2015) mentioned that phenolic compounds familiar to support substantially to the antioxidant activities, the greater the content of phenolic compounds with the antioxidant active.

IV. CONCLUSION

Bioactive compounds are normally accumulated as secondary metabolites in all plant cells including seeds, but their

concentration according to solvent extraction. The existence of phytochemicals such as flavonoids, tannin, saponin presented the antioxidant activities. The *M. indica var podang* in methanolic extract had the highest antioxidant properties: Total phenol 142. 18 mg/g GAE, Total flavonoid content 40.27 mg/g QE, DPPH radical scavenging 87.23 %, and IC50 65.75 µg/mL). This research evinced that *M. indica var podang* that cultivated in Indonesian is the vigorous antioxidant.

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