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ORIGINAL PAPER

Total phenolic contents, total flavonoid contents and antioxidant activities of endocarp and mesocarp of Malaysian local *Durio Zibethinus* (*Musang King* and *Durian Kampung Peel*)

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Abstract

This study aimed to determine and compare the total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activities of mesocarp and endocarp peel extract from two Malaysian local varieties of *Durio Zibethinus*, *Durian Kampung* (DK), and *Musang King* (MK). The extracts were analysed using Folin-Ciocalteu method for total phenolic content, aluminium chloride colorimetric method for total flavonoid content, as well as 2,2-diphenyl-1-picrylhydrazyl (DPPH), and ferric reducing antioxidant power (FRAP) assays for antioxidant activities. Findings showed *Durian Kampung* endocarp, and mesocarp peel extract had significantly higher total phenolic content (6.261 ± 0.442 mg GAE/g; 5.129 ± 0.162 mg GAE/g), total flavonoid content (32.30 ± 1.691 mg QE/g; 27.84 ± 0.701 mg QE/g), and DPPH radical scavenging activity IC_{50} value (39.52 ± 1.400ug/mL; 46.62 ± 0.563 µg/mL) respectively compared to *Musang King*. Eventhough FRAP values of all the peel extracts showing no significant differences ($p > 0.05$), *Durian Kampung* endocarp consistently had the highest value of FRAP (1965.0 ± 174.6 µM TE/g), followed by its mesocarp (1923.2 ± 135.9 µM TE/g). The results showed *Durian Kampung* had better bioactivity than *Musang King*, and the endocarp peel had better bioactivity than the mesocarp peel. The total phenolic content, and total flavonoid content were strongly correlated with antioxidant DPPH IC_{50} value (TPC; $r = -0.964$, TFC; $r = -0.939$), and moderately correlated with FRAP value (TPC; $r = 0.658$, TFC; $r = 0.613$), indicating that the antioxidant activities of durian peel extract are contributed by phenolic, and flavonoid. This finding can serve as baseline data for the antioxidant properties of Malaysian durian waste, and further studies are encouraged, as it is a potential nutraceutical source with good antioxidant qualities.

Keywords DPPH · Antioxidant · FRAP · Durian · Phenolic · Flavonoids

Introduction

According to epidemiology research, eating a lot of fruit helps reduce chronic conditions like diabetes, cancer, atherosclerosis, inflammation, and coronary heart disease [1]. Durian, scientifically known as *Durio Zibethinus*, belongs to the family *Bombacaceae* within the genus *Durio*. The term 'durian' is derived from the Malay word *'duri*,' which means thorn, describing the outer layer of the durian peel. *Durio Zibethinus* is prevalent in Southeast Asian countries

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like Malaysia, the Philippines, Indonesia, Singapore, and Thailand. However, its historical origins trace back to Peninsular Malaysia [2]. There are 139 different cultivars of *Durio Zibethinus* known to exist in Malaysia, and they differ in terms of flavour, texture, scent, and flesh colour [3]. A study by Nordin et al. [4] revealed that the composition of *Durio zibethinus* includes 20–35% pulp, 5–15% seeds, and the largest portion, the peel, which constitutes 55–66%. Durian peel, a byproduct of durian consumption, despite its promising therapeutic potential, is frequently discarded as agricultural waste, contributing significantly to environmental pollution as it is difficult to decompose naturally [5]. This issue is exacerbated when discarded peels are burnt, resulting in air pollution and raising significant environmental concerns [6].

Table 1 shows that there were numerous bioactive compounds and exceptional antioxidant activities found in the fruit peels, including *Durio zibethinus* peel [7–14]. Feng et al. [1] identified 16 known phenolics, including propacin, and four novel phenolics in *Durio zibethinus* peel from China. These compounds demonstrated antioxidant and nitric oxide (NO) inhibitory effects. Additionally, Huang et al. [15] found that propacin from Chinese *Durio zibethinus* peel exhibited strong anti-inflammatory properties by inhibiting the expression of iNOS, COX-2, and ROS, thereby maintaining mitochondrial integrity through the NF-κB and MAPK signalling pathways in macrophages stimulated by lipopolysaccharides (LPS). A study by Muhtadi et al. [16] identified that the extracts of Indonesian *Durio zibethinus* peels may contain flavonoids such as catechin, quercetin, and EGCG, as well as polyphenols and tannins that have antidiabetic properties, causing blood glucose reduction in male white rats induced with alloxan. He et al. [17] found that Golden Pillow *Durio zibethinus* peels from China had quercetin, catechin, rutin, centaureidin-3-glucoside and procyanidin that exerted anti-inflammatory effect by increasing the SOD activities, reducing the expression of ROS, MDA, and LDH levels, and inhibiting cell apoptosis through regulating the expression of genes and proteins related to mitochondrial pathway apoptosis.

However, a limited study was done on locally available *Durio zibethinus* in Malaysia, despite Malaysia being one of the largest exporters. To our knowledge, there was no study has been conducted on the antioxidant properties of Musang *King* peel and *Durian Kampung* peel. Previous studies related to *Musang King* commonly discuss the effect of freezing on durian pulp, nutrient content, and quality of Musang King [18, 19], and the suitable storage temperature

Table 1 Antioxidant potential of fruit peels

Author Citation	Fruit	Country	Antioxidant Properties and Activities
Charoenphun & Klangbud [7]	*Durio zibethinus* peel	Thailand	*Monthong* durian peel ethanolic extract exhibited total phenolic content of 3471.98 ± 141.06 mg GAE/g, ABTS radical scavenging activity of 18.03 ± 1.06 ug/mL, NO scavenging activity of 145.80 ± 54.13 ug/mL and superoxide radical scavenging activity of 60.25 ± 23.74 ug/mL. *Chanee* durian peel ethanolic extract exhibited total phenolic content of 3576.74 ± 259.99 mg GAE/g, ABTS radical scavenging activity of 10.48 ± 0.26 ug/mL, NO scavenging activity of 357.70 ± 149.60 ug/mL and superoxide radical scavenging activity of 88.55 ± 7.97ug/mL
Tran et al. [8]	*Durio zibethinus* peel	Vietnam	Chloroform fraction of durian peel ethanolic extract exhibited highest total flavonoid content (271.11 mg QE/g), DPPH radical scavenging activity (IC_{50} of 38.72 µg) and FRAP iron reducing ability of 307.88 µg Vit C/g. HPLC analysis also found highest quercetin in this extract, 1006.19 µg/g
Gondi and Rao [9]	*Mangifera indica L.* peel	India	*Badami* mango peel ethanolic extract exhibited total phenolic content of 83.0 ± 1.5 mg/g, flavonoids of 8.0 ± 0.7 mg/g, and DPPH radical scavenging activity, EC_{50} of 4.0 ± 0.08 µg
Yongliang et al. [10]	*Nephelium lappaceum* peel	China	Purified rambutan peel ethanolic extract exhibited total phenolic of 877.11 mg GAE/g, DPPH radical scavenging activity (IC_{50} of 1.67 ug/mL), ABTS scavenging activity (IC_{50} of 1.14 ug/mL), FRAP iron reducing ability (IC_{50} of 0.87 ug/mL) than its crude extract. Both extracts contain a high amount of geranin.
Ponce-Sánchez et al. [11]	*Annona squamosa L.* peel	Mexico	Apple peel methanolic extract exhibited total phenolic content of 5000.13 ± 30.44 mg GAE/100 g, total flavonoid content of 82.04 ± 1.2 mg QE/100 g and ABTS scavenging activity of 55.23 ± 0.43 mmol TE/100 mg
Kalantari et al. [12]	*Punica granatum* peel	Iran	Pomegranate peel extract exhibited around 72.65 mg GAE/g DW to 163.09 mg GAE/g DW of total phenolic content, 13.60 mg QE/g DW to 104.18 mg QE/g DW of total flavonoid content, 47.11–80.04% of DPPH radical scavenging activity, and 17.76–23.06% hydrogen peroxide radical scavenging activity. HPLC analysis also found that the highest phenolic acid was chlorogenic acid in this extract.
Zain et al. [13]	*Durio Zibethinus* Peel	Malaysia	Durian peel 100% ethanolic extract exhibited around 72.65 mg GAE/g DW of total phenolic content, IC_{50} of 96.91 ± 1.09 µg/mL for DPPH, and IC_{50} of 4.53 ± 0.21 for α-glucosidase inhibitory activity. UHPLC analysis also found that the durian peel 100% ethanolic extract consists of β-sitosterol, maslinic acid, kaempferol, and heteroflavone B.
Liang et al. [14]	*Durio Zibethinus* Peel	China	ECTs from durian peel consist of B-type procyanidin oligomers and (epi)catechin that exhibited IC_{50} of 71.28 ± 2.01 ug/mL for DPPH, IC_{50} of 82.53 ± 3.13 ug/mL for ABTS, IC_{50} of 28.26 ± 0.83 ug/mL of β-carotene bleaching assay and FRAP iron reducing ability of 6.66 ± 0.91 mmol AAE/g.

of *Musang King* [20, 21]. Meanwhile, previous studies on *Durian Kampung* mainly focus on the physicochemical content of the flesh instead of its peel [22]. Therefore, this study aimed to determine and compare total phenolic content, total flavonoid content, and antioxidant activities (DPPH assay and FRAP assay) of endoderm and mesoderm peel extract from *Musang King* and *Durian Kampung*.

Materials and methods

Sample Preparation

Durian samples (*Musang King* and *Durian Kampung*) were purchased at a local fruit stall at Bandar Baru Bangi and Serdang, Selangor in July 2023. The maturity of both durian samples was based on commercial maturity. The inner layer of the peel for each durian was separated into mesocarp and endocarp, which were then categorised as *Durian Kampung* endocarp (DK-E), *Durian Kampung* mesocarp (DK-M), *Musang King* endocarp (MK-E), and *Musang King* mesocarp (MK-M) as shown in Fig. 1. The overall experimental design was summarised in Fig. 2. The drying method was adapted from Liang et al. [14] with slight modifications. The mesocarp and endocarp were cut into pieces, with sizes between 2 and 3 centimetres. Then, the samples were stored in a -80 °C freezer for 24 h before freeze-drying. Then, the samples were freeze-dried (Virtis, NY, USA) for 24 h. The freeze-dried samples were ground into fine particles using a blender (Torrington, CT, USA) before they passed through a sieve with 40 mesh and stored at -20 °C until the following analysis.

Sample extraction

The samples were extracted using a maceration method adapted from Charoenphun & Klangbud [7]. Firstly, 5 g of the ground samples were macerated with 50 mL of 95% ethanol (99.5%) solution (1:10 v/v ratio) for 1 h. This process of extraction was done thrice. Next, the liquid from the macerated solution was filtered using Whatman No. 1 filter paper. Then, the filtered solution was evaporated using a Laborata 4000 rotary evaporator (Heidolph, Japan) to obtain the highly concentrated extract of the peels. Then, the concentrated extract was weighed and kept at -80 °C. The extraction yield was obtained by using formula (1):

$$Percentage\ yield = \frac{Weight\ of\ extracts\ (g)}{Weight\ of\ dry\ samples\ (g)} \times 100 \quad (1)$$

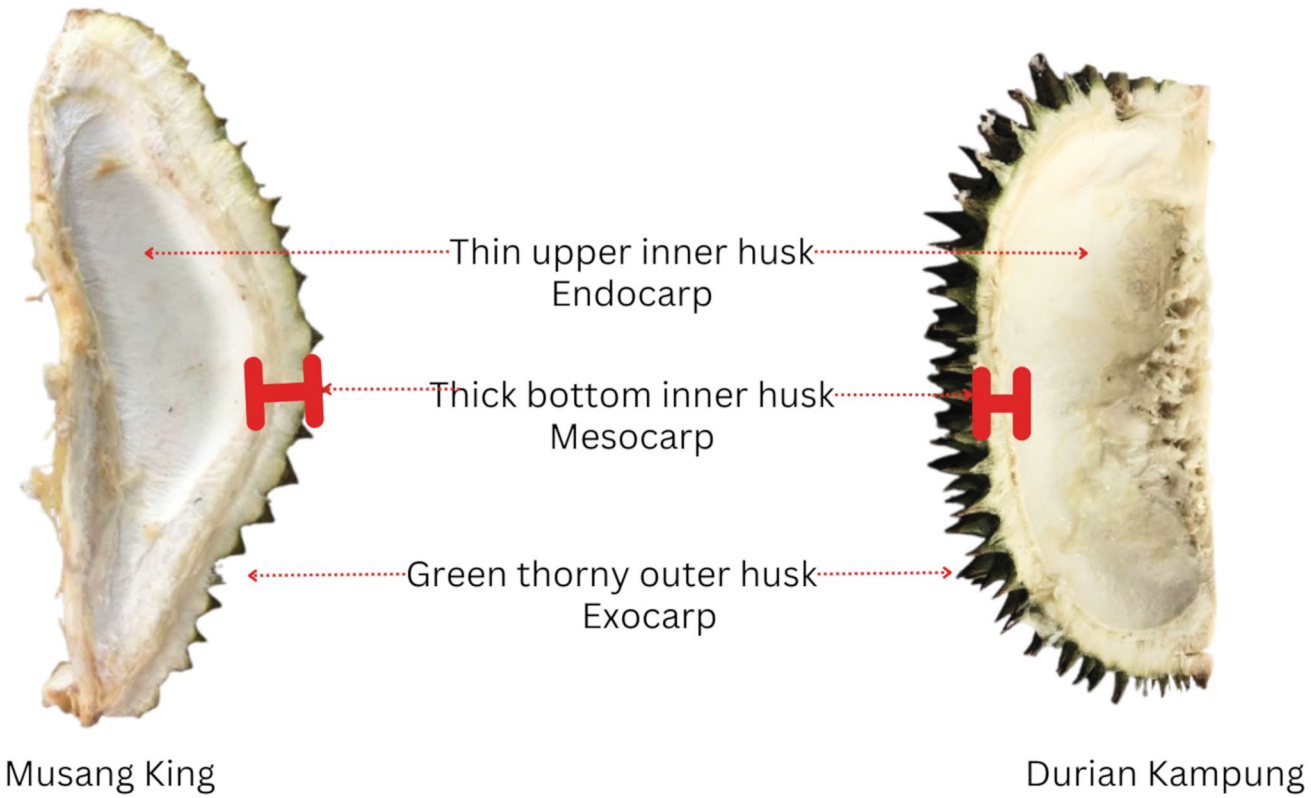

Fig. 1 Cross section of durian husk segment

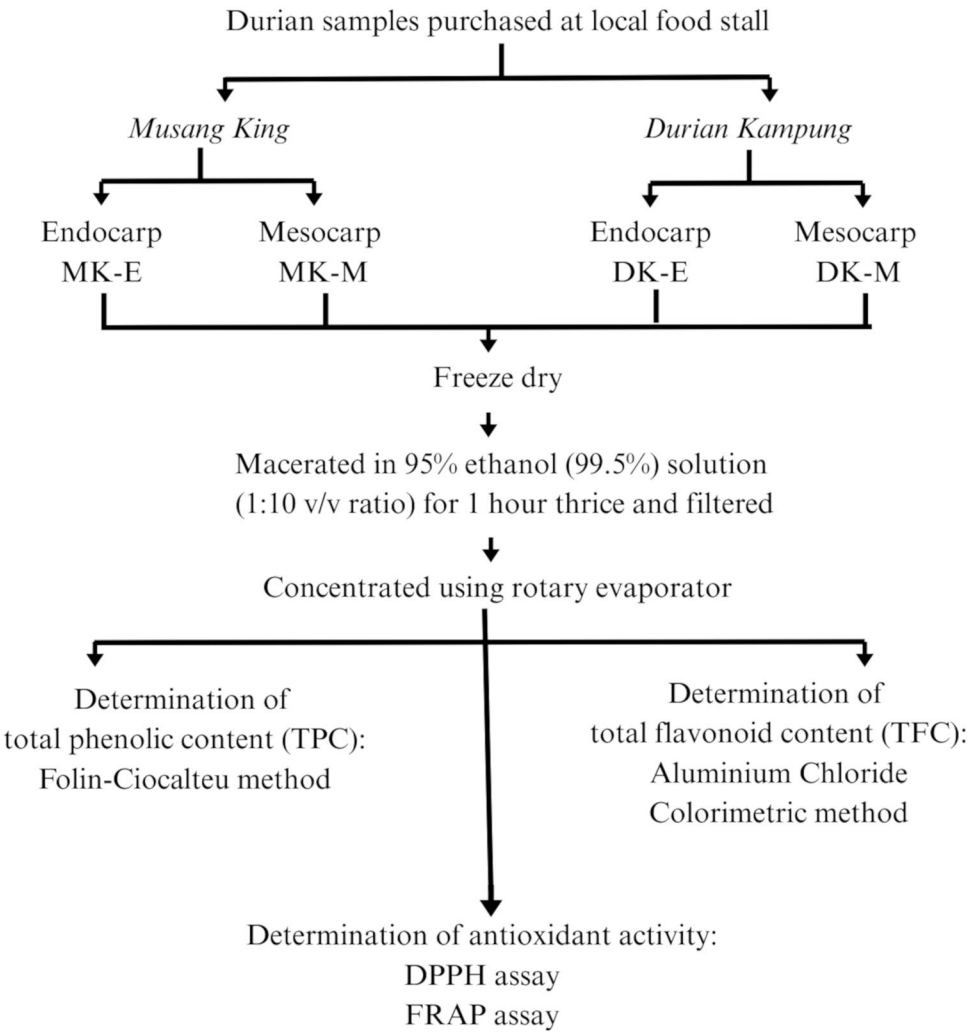

Fig. 2 Schematic diagram of the study

Total flavonoid content (TFC)

The total flavonoid content was determined using the aluminium chloride colorimetric method from Muhtadi and Ningrum [22]. Firstly, in a 10 mL volumetric flask, 0.5 mL of sample was mixed with 4.0 mL of distilled water and 0.3 mL of 5% (w/v) sodium nitrite ($NaNO_2$). After 5 min, 0.3 mL of 10% (w/v) aluminium chloride ($AlCl_3$) was added to the mixture and left again for another 5 min. Next, 4.0 mL of 1 M sodium hydroxide (NaOH) and distilled water were added to reach the boundary mark in the volumetric flask. The absorbance value of the mixture was determined using a spectrophotometer at a wavelength of 510 nm. A standard curve ($y=0.4161x+0.0781$, $R^2=0.9941$) was prepared using quercetin with different concentrations (0.05 mg/mL–0.5 mg/mL) against the absorbance of samples. Then, the total flavonoid content was calculated using formula (2):

$$Total\ flavonoid\ content = \frac{c \times v}{m} \quad (2)$$

C = concentration of quercetin from the standard curve (mg/mL).
V = volume of extract used in the assay (mL).
m = weight of extract (g).
The result was expressed as mg QE/g.

Total phenolic content (TPC)

The Folin-Ciocalteu method was applied to determine total phenolic content according to Charoenphun and Klangbud [7] with minor adjustments. Firstly, 2.5 mL Folin-Ciocalteu reagent (10%, v/v) was mixed with 0.5 mL sample. Then, 2.0 mL sodium carbonate solution (Na_2CO_3) was added, and the mixture was incubated at room temperature for 120 min. The absorbance of samples was measured using a spectrophotometer at a wavelength of 740 nm. A standard curve ($y=4.3083x+0.0268$, $R^2=0.9676$) was prepared using different concentrations of gallic acid (0.05 mg/mL– 0.5 mg/mL) against the absorbance of samples. The phenolic content was calculated using formula (3):

$$Total\ phenolic\ content = \frac{c \times v}{m}, \quad (3)$$

where
C = concentration of gallic acid from the standard curve (mg/mL).
V = volume of extract used in the assay (mL).
m = weight of extract (g).
The result was expressed as mg GAE/g.

DPPH radical scavenging assay

The DPPH assay assessed antioxidant activity using a guideline from Xiao et al. [23] with some modifications. Firstly, an ethanolic DPPH solution was prepared by dissolving 7.89 mg of 2,2-diphenyl-1-picrylhydrazyl (DPPH) with 100 mL of ethanol (99.5%) and kept in a dark place for 2 h. Next, 1.0 mL of 2,2-diphenyl-1-picrylhydrazyl (DPPH) solution was mixed with 0.8 mL of Tris-HCl buffer (pH 6.8) and 0.2 mL of durian peel extracts. This mixture was kept for 30 min in the dark at room temperature. The absorbance of the solution was determined at 517 nm. This experiment used 1.2 mL of ethanol (99.5%) and 0.8 mL of Tris-HCl buffer as a blank. Meanwhile, 1.2 mL of ethanolic DPPH solution with 0.8 mL of Tris-HCl buffer was used as a control. The radical scavenging activity will be obtained using formula (4):

$$\%RSA : \frac{Abs\ control - Abs\ sample}{Abs\ control} \times 100 \quad (4)$$

$Abs_{control}$ = Absorbance of control solution (DPPH + Tris-HCl buffer).
Abs_{sample} = Absorbance of the sample (DPPH solution + Tris-HCl buffer + durian peel extract).

The standard curve was plotted with the Y-axis representing the percentage of radical scavenging activity, while the X-axis represented different concentrations of the durian peel extract samples (20–100 µg/mL). The IC_{50} (x value) was calculated from the standard curve equation by substituting the y value with the inhibition ratio (50%).

Ferric ion reducing antioxidant power assay

This determination of ferric ion reducing antioxidant power (FRAP) assay uses a guideline described by Xiao et al. [23]. Firstly, the FRAP solution was freshly prepared by mixing acetic acid buffer, 2,4,6-tris (1-pyridyl)-5-triazine (TPTZ) solution, and iron (III) chloride ($FeCl_3$) with a ratio of 10:1:1, v/v/v. The FRAP solution was incubated at 37 °C. Then, 20 µL of sample was mixed with 180 µL of the FRAP solution before being incubated at 37 °C in dark conditions for 15 min. The absorbance was measured using a microplate reader at 595 nm wavelength. Distilled water was used as a blank while Trolox was used as the standard curve ($y = 0.0115x + 0.1106$, $R^2 = 0.9921$) with concentrations ranging from 20 µM/mL -100 µM/mL. The FRAP value will be calculated using formula (5):

$$FRAP\ value = \frac{c \times v}{m}, \quad (5)$$

where
C = concentration of Trolox from standard curve (µM/mL).
V = volume of sample used (mL).
m = weight of the extracts (g).
The result was expressed as µM TE/g.

Statistical analysis

The analysis was done in triplicate, and IBM Statistical Package for Social Sciences (SPSS) version 26 was used to analyse the data obtained from the experiment. The results were expressed as mean ± standard deviation. One-way Analysis of Variance (ANOVA) and Post-hoc Tukey HSD test with significance level set at $p < 0.05$ was used to compare the mean differences between endoderm and mesoderm peel extract from different durian cultivars. Meanwhile, Pearson's correlation was used to identify the association between total phenolic content and total flavonoid content with antioxidant activity in DPPH and FRAP assays, where a significant level was set at $p < 0.05$.

Results and discussion

Sample extraction

Figure 3 shows the extraction yield of *Musang King* and *Durian Kampung* peel extracts. There were statistically significant differences in the extraction yield between mesocarp and endocarp peel extract from the two durian cultivars ($p < 0.01$). The highest extraction yield was found in MK-M (29.13 ± 0.153%), followed by DK-E (27.10 ± 0.100%), MK-E (22.73 ± 0.252%), and DK-M (21.57 ± 0.153%). The *Musang King* mesocarp had a significantly higher extraction yield than its endocarp. In contrast, the *Durian Kampung* mesocarp had a significantly lower yield than its endocarp.

Total flavonoid content (TFC)

Figure 4 shows the flavonoid content of Musang King and Durian Kampung peel extracts. There were statistically significant differences in total flavonoid content between

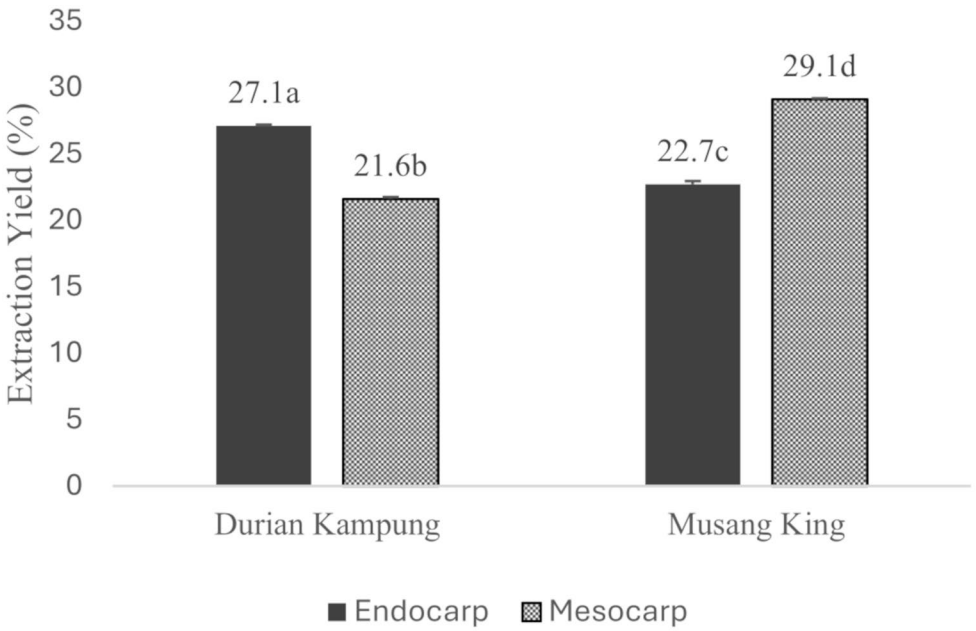

Fig. 3 Extraction yield of Musang King and Durian Kampung peel extracts. Error bars indicate ± standard deviation. Means with different letters are significantly different at $p < 0.05$

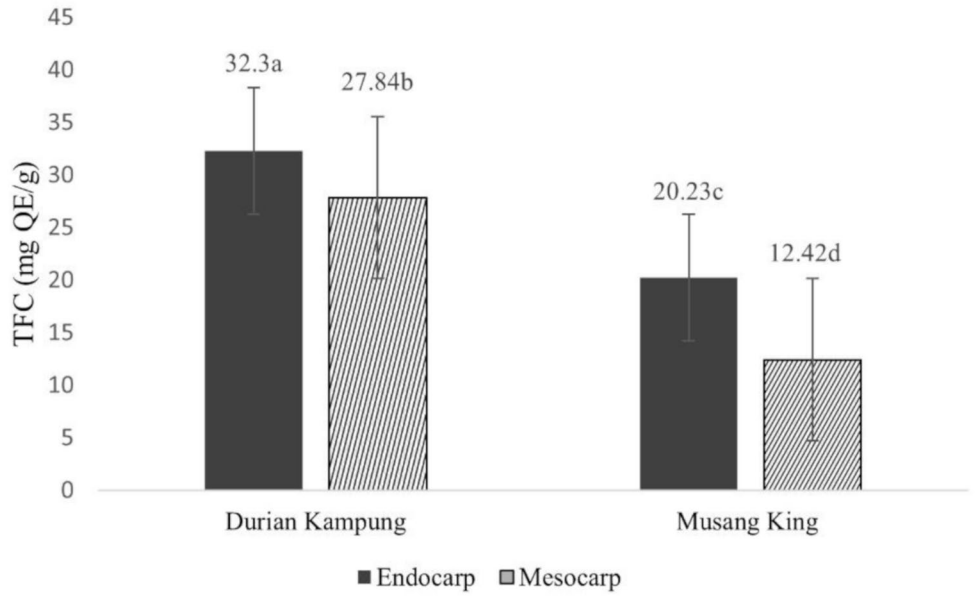

Fig. 4 Total flavonoid content of Musang King and Durian Kampung peel extracts. Error bars indicate ± standard deviation. Means with different letters are significantly different at $p < 0.05$

mesocarp and endocarp peel extract from the two durian cultivars ($p < 0.05$). Results show that DK-E has the highest total flavonoid content with a value of 32.30 ± 1.691 mg QE/g, followed by DK-M (27.84 ± 0.701 mg QE/g), MK-E (20.23 ± 0.559 mg QE/g), and MK-M (12.42 ± 1.326 mg QE/g). Peel extracts from *Durian Kampung* peels had significantly higher total flavonoid content than *Musang King*. Total flavonoid content in endocarp peel extract from both durians was significantly higher than the total flavonoid content in mesocarp peel extract.

The total flavonoid content of both Malaysian *Durian Kampung* peel and *Musang King* peel extracts was lower compared to Vietnamese durian peel in 75% ethanol extract, 82.22 ± 11.11 mg QE/g [8]. Indonesian durian peel extract from *Malika*, *Malon*, and *Monti* cultivars had lower total flavonoid content of 0.321 mg QE/g, 0.324 mg QE/g, and 0.405 mg QE/g, respectively, compared to both Malaysian *Durian Kampung* peel and *Musang King* peel [24]. Meanwhile, Indonesian *Monthong* and *Medan* durian peel extract in 96% ethanol were reported to have 310.30 mg RE/g and 472.80 RE/g, respectively [23], yet cannot be compared to this study due to the difference in the standard used.

One of the factors that may contribute to the difference in total flavonoid content of durian peel is cultivar type and geographical location, as they impact the genetic and physicochemical properties of the fruit [25]. In addition, the type

of solvent used, the polarity of the extraction solvent, and the extraction temperature are the other factors that could contribute to the difference in total flavonoid content [8]. Without proper and optimised extraction, the total flavonoid content observed will be very low, as reported by Masturi et al. [24], which briefly dissolved the durian peel with ethanol upon analysis. Water in the extraction solvent may facilitate the release of hydrophilic antioxidants from plant material, while ethanol alone can reduce the amount of flavonoids since it cannot extract all flavonoids [8].

Total phenolic content (TPC)

Figure 5 shows the total phenolic content of *Musang King* and *Durian Kampung* peel extracts. There were statistically significant differences in total phenolic content between mesocarp and endocarp peel extract from the two durian cultivars ($p < 0.05$). DK-E had the highest total phenolic content with a value of 6.261 ± 0.442 mg GAE/g, followed by DK-M (5.129 ± 0.162 mg GAE/g), MK-E (4.794 ± 0.188 mg GAE/g), and MK-M (3.700 ± 0.069 mg GAE/g). Peel extracts from *Durian Kampung* peels had higher total phenolic content than *Musang King*. The total phenolic content in the endocarp peel extract from both durians was significantly higher than the total phenolic content in the mesocarp extract.

The total phenolic content of Malaysian Durian Kampung peel and Musang King peel extracts was lower than in the previous study. The total phenolic content of ripe *Medan* durian peel and mature *Monthong* durian peel from Indonesia in 96% ethanol extract was 245.38 mg GAE/g and 148.34 mg GAE/g, respectively [23]. The total phenolic content of Malaysian *Red Prawn* durian peel in 0%, 50%, and 100% ethanol/water extract was 74.20 ± 1.02 mg GAE/g, 130.57 ± 1.92 mg GAE/g, and 63.89 ± 2.91 mg GAE/g crude, respectively [13].

One of the factors that could affect the total phenolic content of the durian peel was the difference in fruit maturity stages. In a study done by Leontowicz et al. [26], ripe durian pulp had the highest amount of quercetin, anthocyanin, and ascorbic acids; over-ripe durian pulp had the highest amount of polyphenols and flavonoids; meanwhile, mature durian had the highest amount of tannins. To our knowledge, there were no studies have been done on the antioxidant properties of durian peel at different stages of ripening.

Thus, another factor contributing to the difference in total phenolic content was the choice of layers of the peel used in the studies. The exocarp, mesocarp, and endocarp are the three layers of peel found on many fruits [27]. To our knowledge, there was a study reported using both green and white peel [22], two studies reported using only white peel [7, 17], and others did not specify. In this study, the endocarp of *Musang King* and *Durian Kampung* had significantly higher total phenolic content than the mesocarp, which correlates with a previous study done by Moonrungsee et al. [28] that showed the ethanolic extracts of the Nipa Palm fruit endocarp had the highest total phenolic content (102.85 ± 5.73 mg GAE/g) compared to its other layers (exocarp-mesocarp, endosperm, fresh fruits). The higher total phenolic and flavonoid contents found in the endocarp might be contributed by the presence of essential metabolites and gene expression in the shikimate pathway, which is central to synthesizing the phenolic compounds.

In the metabolomic and transcriptomic analysis done by Han et al. [27]. The endocarp of *Sumac* peel was reported to have more flavonoids and the highest rhusflavone, a type of biflavonoid, than the exocarp-mesocarp section. DAHP synthase 2–2, DAHP synthase 1, and most other enzymes

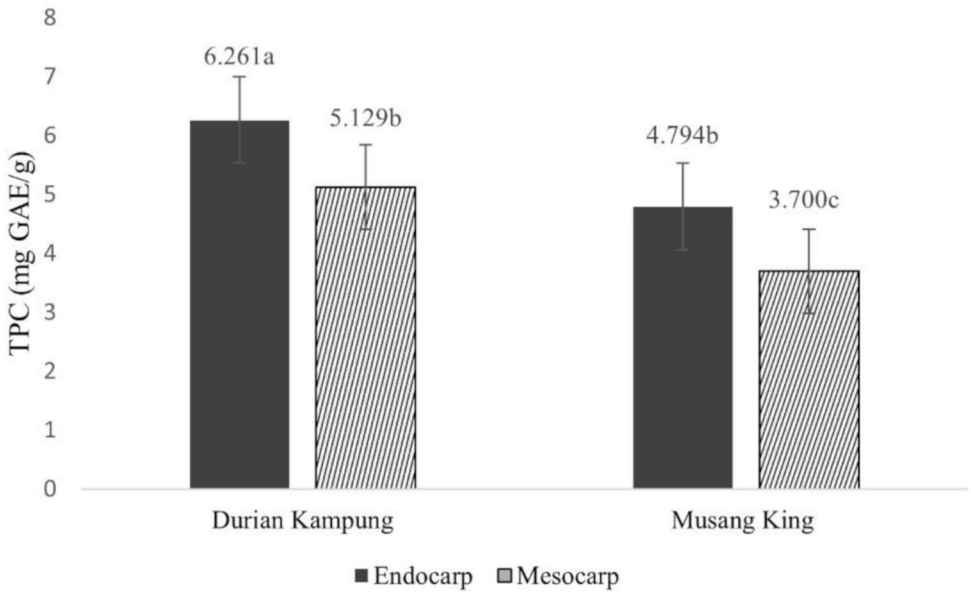

Fig. 5 Total phenolic content of *Musang King* and *Durian Kampung* peel extracts. Error bars indicate ± standard deviation. Means with different letters are significantly different at $p < 0.05$

involved in phenylalanine and tyrosine biosynthesis were expressed at higher levels in the endocarp of *Sumac* fruit than in the exocarp-mesocarp section, indicating higher amounts of phenolic compound synthesis [27]. The high MYB8 expression, increased in the majority of phenylpropane pathway enzymes, high phenylalanine ammonia lyase (PAL), and low expression of MYBC1 in the *Sumac* endocarp contributed to its high flavonoid content or/and high lignification [27]. Thus, 8 peroxidase or laccase transcript found only in *Sumac* endocarp was estimated to catalyse the synthesis of biflavonoids [27].

Furthermore, several previous studies had identified the phenolic compounds in durian peel extracts. For instance, a study by He et al. [17] identified five abundant phenolic contents in durian peel extracts: quercetin, rutin, centaureidin-3-glucoside, catechin, and procyanidin B. A study by Zain et al. [13] identified 20 compounds in durian peel extract, including β-sitosterol, kaempferol, maslinic acid, and heteroflavanone B. A study done by Feng et al. [1] found the durian peel extract contained twenty phenolics that somehow showed potent antioxidant and NO inhibitory activities, including four novels (durianol A, durianol B, durianol C and 5′-methoxy-7′-epi-jatrorin A).

DPPH radical scavenging assay

Results in Fig. 6 indicate a significant difference in IC_{50} value in DPPH assay determination between mesocarp and endocarp peel extract from the two durian cultivars ($p<0.05$). The lowest IC_{50} values were observed in DK-E with a value of 39.52 ± 1.400 µg/mL, followed by Trolox (46.23 ± 3.694 µg/mL) and DK-M (46.62 ± 0.563 µg/mL. The free radical scavenging activity of DK-E was 90.6% at a concentration of 100 µg/mL. This finding indicates that DK-E had the highest scavenging activity against the DPPH radicals. The IC_{50} value of DK-M was not significantly ($p>0.05$) different when compared with the IC_{50} value of MK-E (49.24 ± 0.887 µg/mL). Moreover, the highest IC_{50} value was from MK-M (53.50 ± 1.326 µg/mL), indicating that the scavenging activity for MK-M was significantly the lowest. Peel extracts from *Durian Kampung* peels had higher radical scavenging activity than *Musang King*. Radical scavenging activity in endocarp peel extract from both durians was higher than the radical scavenging activity in mesocarp extract.

The IC_{50} values of durian *Kampung* peel and durian *Musang King* peel were lower than previous studies, indicating higher antioxidant activity against the DPPH. The study done by Muhtadi and Ningrum [22] showed that the IC_{50} value of the durian peel extracts from *Monthong* and *Medan* cultivars from Indonesia was 78.83 µg/mL and 72.77 µg/mL, in 96% ethanol, respectively. The data is in line with a study from Liang et al. [14] that used extractable condensed tannins from *Monthong* cultivars that showed an IC_{50} value of 71.28 µg/mL, which is close to a study by Muhtadi & Ningrum [22]. Besides that, Chinese durian peel extract was found to have a higher IC_{50} value of 105.37 µg/mL in methanol extract and 372.08 ± 73.02 ug/mL in 70% ethanol extract [1]. A study done by Tran et al. [8] found that the Vietnamese durian is 58.87 ± 1.34 ug/mL in 75% ethanol, meanwhile a study by Zain et al. [13] found that the IC_{50} value of Malaysian durian *Red Prawn* peel extract was 96.91 ± 1.09 µg/mL in 100% ethanol.

Ferric ion reducing antioxidant power assay

Results in Fig. 7 indicate no significant difference in iron reducing ability in FRAP assay determination between

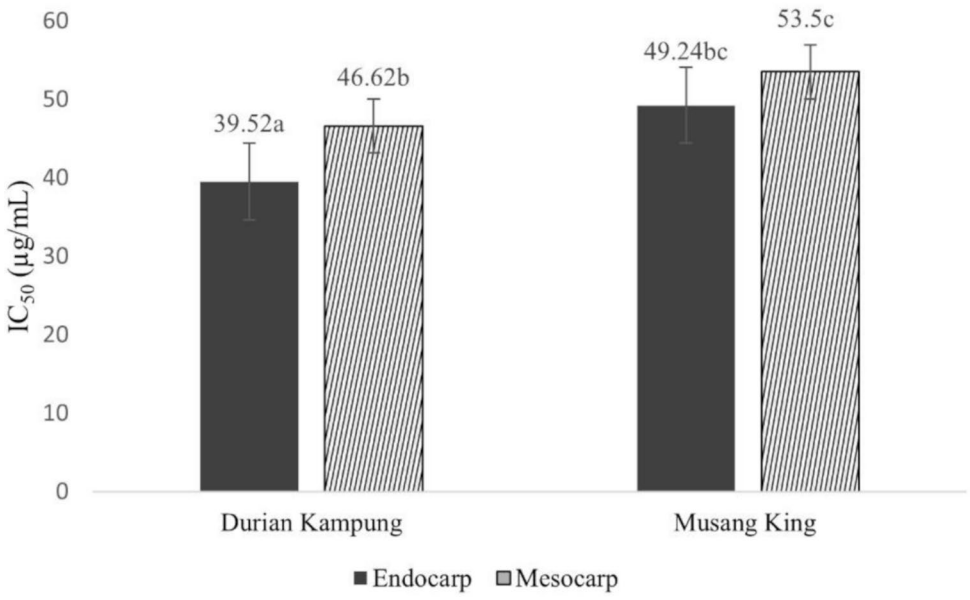

Fig. 6 IC_{50} of Musang King and Durian Kampung peel extracts. Error bars indicate ± standard deviation. Means with different letters are significantly different at $p<0.05$

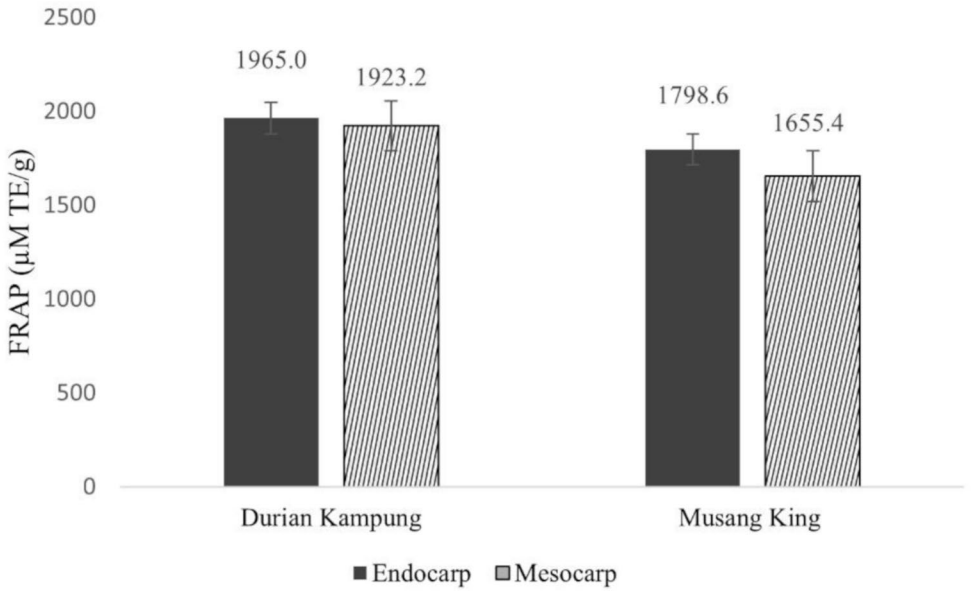

Fig. 7 FRAP value of Musang King and Durian Kampung peel extracts. Error bars indicate ± standard deviation. Means with different letters are significantly different at $p < 0.05$

Table 2 Correlation between total phenolic content and total flavonoid content with antioxidant activities

Antioxidant Activity	Total Phenolic Content (TPC)	Total Flavonoid Content (TFC)
	r	*r*
DPPH (IC_{50})	-0.964**	-0.939**
FRAP	0.658*	0.613*

**Correlations are significant at p-value < 0.01

* Correlations are significant at p-value < 0.05

mesocarp and endocarp peel extract from the two durian cultivars ($p < 0.05$). The highest value of FRAP was found in DK-E (1965.0 ± 174.6 μM TE/g), followed by DK-M (1923.2 ± 135.9 μM TE/g) and MK-E (1798.6 ± 52.73 μM TE/g). In contrast, the lowest value of FRAP was found in MK-M (1655.4 ± 229.4 μM TE/g). Peel extracts from *Durian Kampung* peels had higher iron reducing ability than *Musang King*. The iron-reducing ability in the endocarp peel extract from both durians was higher than that in the mesocarp extract.

The iron-reducing ability of durian peel was determined using the FRAP assay. The FRAP value of durian *Kampung* peel ranged from 1923.2 ± 135.9 μM TE/g (DK-M) to 1965.0 ± 174.6 μM TE/g (DK-M), meanwhile the FRAP value of durian *Musang King* peel ranged from 1655.4 ± 229.4 μM TE/g (MK-M) to 1798.6 ± 52.73 μM TE/g (MK-E). A study by Liang et al. [14] that assessed extractable condensed tannins in the peel of Monthong cultivars reported a higher FRAP value (9.66 mM AAE/g), meanwhile, a study done by Tran et al. [8] reported that FRAP value increased from 40.41 to 307.88 μg AAE/g when increasing their concentration from 0.2 to 1.0 mg/mL. Both results cannot be compared to the FRAP value of durian *Kampung* peel and durian *Musang King* peel, as the standard used differed.

Interestingly, the chloroform fraction of Vietnamese durian ethanolic extracts exhibited superior antioxidant activity, with an IC_{50} value of 33.63 μg/mL, the lowest when compared to its ethanolic crude extract, ethyl acetate fraction, and aqueous fraction, and was closely resembling the IC_{50} of vitamin C (30.60 μg/mL) [8]. A similar trend was found with the FRAP value, where the chloroform fraction of Vietnamese durian ethanolic extract showed the highest iron reducing activity (307.88 μg AAE/g) [8]. The chloroform fraction of Vietnamese durian ethanolic extract also exhibited α-glucosidase inhibition, α-amylase inhibition, and cytotoxicity against HepG-2 and MCF-7 cells [8]. The potent bioactivity of the chloroform fraction may be attributed to its bioactive compounds, such as propacin isomers [1, 15]. In another study done by Huang et al. [15], propacin found in durian peel extract suppressed iNOS, COX-2 and ROS generation, hence controlling mitochondrial integrity, by blocking the LPS-induced immunological response in macrophages via the NF-κB and MAPK signalling pathways. These findings underscore the potential of durian peel extracts, particularly specific fractions, as promising sources of natural antioxidants and therapeutic agents. These findings underscore the potential of durian peel extracts, particularly specific fractions, as promising sources of natural antioxidants and therapeutic agents.

Correlation between total phenolic content and total flavonoid content with antioxidant activity

A strong and negative correlation between TPC and DPPH assay, was statistically significant (r=-0.964, p<0.01). For the TFC, it showed that there was also a strong negative correlation between TFC and DPPH assay, and it was also statistically significant (r=-0.939, p<0.01). In this study, the total phenolic content with antioxidant activity (IC_{50}) showed a higher correlation value (r=-0.964) compared to the total flavonoid content with antioxidant activity (r=-0.939). Furthermore, there was a moderate and positive correlation between total phenolic content and FRAP, which was statistically significant (r=0.658, p<0.05). The moderate positive correlation with a statistically significant value was also observed between total flavonoid content and FRAP (r=0.613, p<0.05). The Pearson correlation test also showed a highly significant value between TPC and TFC with DPPH (p<0.01) compared to TPC and TFC with FRAP. Hence, the antioxidant activity of TPC and TFC in the scavenging of DPPH radical was higher than the antioxidant activity in the FRAP assay.

The strong negative correlation observed between total phenolic content and total flavonoid content with DPPH IC_{50} value shows that the higher the concentration of total phenolic content and total flavonoid content, the lower the DPPH IC_{50} value, which means higher DPPH radical scavenging activity or higher antioxidant activity. The moderate positive correlation between total phenolic and total flavonoid contents with FRAP value means that higher concentrations of total phenolic and total flavonoid contents contribute to higher iron-reducing ability or higher antioxidant activity.

This is consistent with a study done by Tran et al. [8] that shows negative correlations between TPC in durian peel and DPPH inhibition (R=-0.994). A study from Kunarto and Yuniarti Sani [29] also showed consistent results where DPPH inhibition negatively correlates with total phenolic content (r=-0.96) and total flavonoid content (r=-0.92). Total phenolic content in durian peel has a higher correlation with antioxidant activity in DPPH (r=-0.96) compared to the total flavonoid content with antioxidant activity (r=-0.92) [29]. No studies reported on the correlation of total phenolic content and total flavonoid content in durian peel with antioxidant activity in the FRAP assay.

Fruit peels, often considered agricultural by-products, are rich in bioactive compounds such as polyphenols, flavonoids, and carotenoids, which exhibit potent antioxidant properties. These natural antioxidants play a crucial role in neutralising free radicals, thereby reducing oxidative stress, a key contributor to the pathogenesis of chronic diseases such as cardiovascular diseases, diabetes, and certain cancers [13]. Utilising antioxidants derived from fruit peels, such as durian, as a functional dietary component or therapeutic agent offers a sustainable and cost-effective disease prevention and health promotion approach. There is great potential for the studied durian peel to be further analysed in an animal model.

The high antioxidant properties of durian peel extract, as shown in this study was consistent with the preclinical study done on fruit peels, showing that the phenolics and flavonoids content in fruit peels could reduce oxidative stress in the body by reducing the proinflammatory markers and improving the antioxidant enzymes, which eventually led to positive health benefits. A study done by Gondi et al. [30] found that the supplementation of *Mangifera indica L* peel extract significantly improved the CAT, SOD, GPx and GR levels as well as reduced the elevated MDA level in the kidney and liver of diabetic rats. Meanwhile, a study done on diabetic rats showed that the supplementation of *Malus domestica Borkh* peel extract significantly improved SOD, CAT, GPx, and GR levels and reduced the elevated MDA, NF-kB, TNF-α, IL-6, and IL-8 levels in pancreas which eventually improve the blood glucose level [31].

Conclusion

This study was done due to limited data available on the antioxidant potential of Malaysian local durian waste. The total phenolic and flavonoid content in *Musang King* and *Durian Kampung* strongly contributed to the antioxidant. Results showed that all durian peel extracts had high antioxidant potential, especially for *Durian Kampung* endocarp peel extract, which showed the highest total phenolic content, total flavonoid content, and antioxidant activities, suggesting its potential in pharmaceutical applications and agricultural waste management. Further research is needed to fully explore the durian peel, focusing on evaluating its health benefits in in vivo systems as an initial step towards identifying new functional compounds from this discarded part.

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Declarations

Conflict of interest The authors declare no conflict of interest.

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