

Phytochemistry and Pharmacology of *Mangifera pajang*: An Iconic Fruit of Sabah, Malaysia

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ABSTRACT

Mangifera pajang Kostermans of the mango family (Anacardiaceae) is endemic to the lowland rain forests of Borneo. Although growing wild in the forest, trees are planted in orchards and home gardens due to increasing demand for the fruits which are among the largest of the genus. Fruits are oval in shape, and have a characteristic rough and brown skin. In Sabah, *M. pajang* or bambangan has ethno-cultural significance, and has become an iconic fruit among the Kadazan-Dusun people, who have developed various traditional cuisines using fresh and preserved fruits. Phytochemical investigations on the edible fruit pulp, peel and kernel of *M. pajang* showed that they are rich in flavonoids and phenolic acids. From the edible pulp, flavonoids (daidzein, genistein, hesperidin, kaempferol, naringin, luteolin and quercetin), phenolic acids (caffeic acid, chlorogenic acid and p-coumaric acid), carotenoids (α -carotene and β -carotene), and xanthophylls (cryptoxanthin and cis-cryptoxanthin) have been reported. Pharmacological properties of bambangan fruits include antioxidant, antibacterial, anticancer and cytopro-

TECTIVE activities. A clinical trial at Universiti Putra Malaysia (UPM) has demonstrated the health benefits of regular consumption of bambangan fruit juice. Treated subjects showed significant improvement in certain cardiovascular biochemical parameters that can safeguard against cardiovascular diseases. Traditional and functional food products from bambangan fruits are being developed in Sabah.

Key words: Bornean mango, Bambangan, Phytochemical constituents, Pharmacological properties, Clinical trial.

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DOI : 10.5530/srp.2017.1.15

INTRODUCTION

This synopsis updates the current knowledge on the ethno-cultural significance, phytochemical constituents and pharmacological properties of *Mangifera pajang*. In the Plant List of 2013, a total of 133 species of *Mangifera* have been listed of which only eight are accepted species names while the majority remains as unresolved names, including that of *M. pajang*.¹ Unlike *Mangifera indica* (Indian mango) that is widely documented, there is only a book chapter on *M. pajang*,² and a review on the fruits of *M. pajang* as a source of functional food and medicine.³ There are no reviews on the genus of *Mangifera*.

Mangifera pajang Kostermans of the family (Anacardiaceae) is endemic to the lowland rain forests of Borneo comprising Sabah, Sarawak, Brunei and Kalimantan.²⁻⁴ Commonly known as the Bornean mango, this endemic mango species is locally known as bambangan in Sabah and Brunei, mawang in Sarawak, and embang in Kalimantan. Growing more than 30 m tall, trees have a cylindrical bole (30-70 cm in diameter) with smooth or broadly fissured grey bark (Figure 1). Mature leaves are elliptic-oblong, coriaceous and dark green, while young leaves are bronze to pink.² Flowers have five petals that are purplish-red on the inside and pinkish white on the outside (Figure 2).

Although growing wild in the forest, trees of *M. pajang* have been cultivated by the local Kadazan-Dusun people in Sabah.⁵ Currently, bambangan trees are planted in orchards and in the backyards of homes due to increasing demand for the fruits.

Weighing 0.5-1.0 kg each, fruits of *M. pajang* are among the largest in the genus.^{2,3,5} They are oval in shape, and have a characteristic rough and brown skin (Figure 2), unlike other *Mangifera* species which have a smooth, green, yellow, orange or red skin. During the fruiting season of bambangan (October to February), individual trees bear hundreds of fruits. The edible portion i.e. the pulp which represents 60-65% in total weight⁶ is aromatic, juicy, bright yellow in colour, sweet-sour in taste and



Figure 1: A magnificent tree of *Mangifera pajang*.

fibrous in texture.⁷ The aroma is strongly that of *M. indica* (mango). The pulp of *M. pajang* fruits is usually eaten fresh after removing the peel and the kernel discarded.³ The peel, which is also aromatic, is sometimes used in cooking local dishes such as curry. In Sarawak, young leaves of *M. pajang*, sold in local markets, are blanched and consumed as vegetable with chilli and shrimp paste (sambal belacan) or fermented durian (tempoyak).²

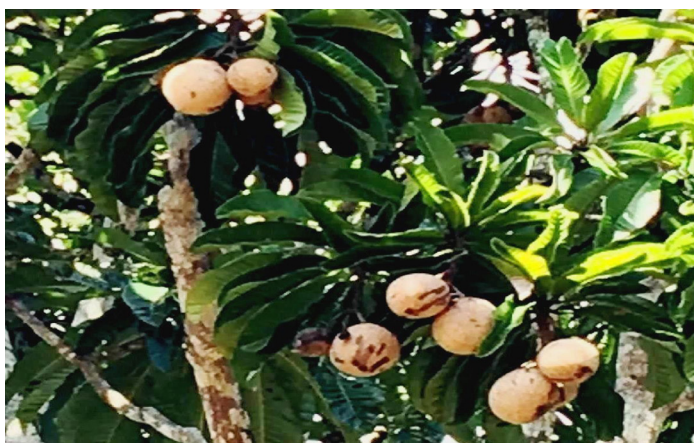
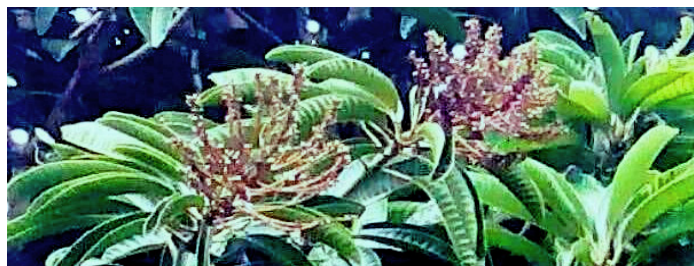


Figure 2: Flowers (top) and fruits (bottom) of *Mangifera pajang*.

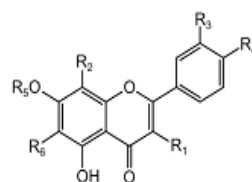
Fermented *M. pajang* fruits or jeruk bambangan is a popular food item among the Kadazan-Dusun community in Sabah.^{2,3} Fruits are cut into cubes with salt and grated kernel added, and left to ferment at 28–30°C in tightly-closed containers for 7–10 days. Available in most local markets and with a life-span of several weeks, the pickle is usually eaten with rice as an appetiser. Although fruiting of *M. pajang* is seasonal, the fermentation process makes the product available throughout the year. Lactic and citric acid are the major organic acids produced during fermentation.⁸ Main microorganisms involved are lactic acid bacteria of *Lactobacillus plantarum* and *Pediococcus pentosaceus*, and yeasts of *Candida krusei* and *Kloeckera apis*.

The most well-known traditional cuisine of the Kadazan-Dusun community in Sabah is hinava. (Figure 3), a salad-like dish consisting of pieces of raw fish marinated in lime or lemon juice and garnished with grated bambangan kernel, chilli, ginger and onion.² Sometimes, slices of bitter melon are added. Hinava is simple to prepare and yet its taste enriched with the aroma of bambangan is so exquisite that it leaves behind fond memories.

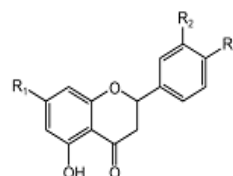
Phytochemical Constituents

Extracts most of the phytochemical investigations on *M. pajang* are conducted on the fruits (Table 1). Flavonoids and phenolic acids are the two main classes of compounds found in the fruit peel, kernel and pulp. Selected flavonoids detected in the fruits of *M. pajang* are shown in Figure 3.

Flavonoids are widely distributed in plants and they constitute the largest group of phenolic compounds. They are 15-carbon compounds with a C6-C3-C6 skeleton consisting of two benzene rings which are connected by an oxygen-containing pyrene ring.¹⁷ As a group of compounds, flavonoids can be further divided into six major classes, namely, flavonols, flavones, flavanones, flavanols, anthocyanins and isoflavones. Phenolic



Compound	R ₁	R ₂	R ₃	R ₄	R ₅	R ₆
Luteolin	H	H	OH	OH	H	H
Kaempferol	OH	H	H	OH	H	H
Quercetin	OH	H	OH	OH	H	H
Diosmin	H	H	OH	OCH ₃	ORut	H
Rutin	OH	H	OH	OH	ORut	H



Compound	R ₁	R ₂	R ₃
Naringin	ONeo	H	OH
Hesperidin	ORut	OH	OCH ₃

Rut = rutinose; Neo = neohesperidose

Figure 3: Molecular structures of flavonoids found in bambangan fruits.

acids are aromatic phenols with one carboxylic acid functionality and they consist of two groups, namely, hydroxybenzoic acids (HBAs) and

hydroxycinnamic acids (HCAs).¹⁷ HBAs have a C6-C1 structure that is derived from benzoic acid. Examples are *p*-hydroxybenzoic, protocatechuic, vanillic, syringic and gallic acids. HCAs have a C6-C3 structure that is derived from cinnamic acid. Examples are *p*-coumaric acid, caffeic acid, ferulic acid and sinapic acid.

The peel of *M. pajang* is rich in flavonoids (catechin, daidzein, diosmin, epicatechin, hesperidin, kaempferol, naringin, luteolin, morin, quercetin and rutin), and in phenolic acids (chlorogenic acid, *p*-coumaric acid, ellagic acid, ferulic acid, gallic acid, 4-hydroxybenzoic acid, protocatechuic acid, pyrogallol and vanillic acid).^{10,13,15} Dominant phenolic acids in the fruit peel are gallic acid (21 mg/g) and *p*-coumaric acid (13 mg/g).¹⁰ Aromatic esters, carotenoids and xanthophylls are also reported in the peel.^{9,11} Of particular interest is mangiferin found in the peel of *M. pajang*. Mangiferin or C-glycosyl xanthone is a natural xanthone that bears a catechol moiety and possesses an array of pharmacological properties such as antioxidant, anti-inflammatory, anti-diabetic, immunomodulatory, anti-tumour, antipyretic and neuroprotective activities.^{18,19} The mangiferin content of 4.8 mg/g in the peel of *M. pajang*¹⁰ is substantially higher than that in the peel (2.4 mg/g)¹⁸ and leaves (0.02–0.04 mg/g)¹⁹ of *M. indica*. Phenolic acids (caffeic acid, chlorogenic acid, *p*-coumaric acid, ferulic acid, gallic acid and sinapic acid), and flavonoids (diosmin, hesperidin, naringin and rutin) are detected in the kernel.¹⁵ Phytochemical analysis revealed the dominance of ferulic acid (5334 µg/g) and diosmin (2386 µg/g) in the kernel.^{5,15}

From the edible pulp, flavonoids (daidzein, genistein, hesperidin, kaempferol, naringin, luteolin and quercetin), phenolic acids (caffeic acid, chlorogenic acid and *p*-coumaric acid), carotenoids (α -carotene and β -carotene), and xanthophylls (cryptoxanthin and *cis*-cryptoxan-

Table 1: Classes and names of compounds from *Mangifera pajang*

Compound Class and Name	Plant Part	Reference
Aromatic Esters		
Benzaldehyde	Kernel	[9]
Benzyl alcohol	Kernel	[9]
Ethyl gallate	Peel	[9]
Methyl gallate	Kernel, peel, leaf	[9,10]
Carotenoids		
α -Carotene	Pulp, peel	[11]
β -Carotene	Pulp, peel, juice	[11,12]
Flavonoids		
Catechin	Peel	[10,13]
Daidzein	Pulp, peel	[10,14]
Diosmin	Peel	[10,15]
Epicatechin	Peel	[13]
Genistein	Pulp	[14]
Hesperidin	Pulp, kernel, peel	[15]
Kaempferol	Pulp, peel	[10,15]
Naringin	Pulp, kernel, peel	[15]
Luteolin	Pulp, peel	[15]
Morin	Peel	[10]
Quercetin	Pulp, peel	[15]
Quercitrin	Leaf	[9]
Rutin	Kernel, peel	[10,13,15]
Phenolic Acids		
Caffeic acid	Pulp, kernel	[15]
Chlorogenic acid	Pulp, kernel, peel	[10,15]
<i>trans</i> -Cinnamic acid	Juice	[16]
<i>p</i> -Coumaric acid	Pulp, kernel, peel	[10,15]
Ellagic acid	Peel	[10]
Ferulic acid	Kernel, peel	[10,15]
Gallic acid	Kernel, peel, juice	[10,13,15,16]
4-Hydroxybenzoic acid	Peel	[10]
Protocatechuic acid	Peel	[10]
Pyrogalllic acid	Peel	[13]
Synapic acid	Kernel, juice	[15,16]
Vanillic acid	Peel	[10]
Sterols		
β -Sitosterol	Stem bark	[9]
Stigmasterol	Stem bark	[9]
Terpenoids		
3 β ,23-Dihydroxy-cycloart-24-ene-26-oic acid	Stem bark	[9]
3 β -Hydroxy-cycloart-24-ene-26-oic acid	Stem bark	[9]
Lupenone	Leaf	[9]
Lupeol	Leaf, stem bark	[9]
Mangiferonic acid	Stem bark	[9]
<i>trans</i> -Sobrerol	Stem bark	[9]

Continued

Xanthone			
Mangiferin	Peel	[10,13]	
Xanthophylls			
Cryptoxanthin	Pulp, peel	[11]	
cis-Cryptoxanthin	Pulp, peel	[11]	
Vitamin			
Ascorbic acid	Pulp, juice	[12]	

thin) have been reported.^{4,14} The pulp has 46 mg/100 g of ascorbic acid, 42 mg/100 g of β -carotene and 8.5 mg/100 g of daidzein.^{12,14} Naringin and hesperidin (Figure 3) are two major compounds of the fruit pulp of *M. pajang* with contents of 1450 and 930 μ g/g, respectively.¹⁵ In comparison with the bambangan fruit pulp, the fruit juice powder was three-fold higher (132 mg/100 g) in ascorbic acid content and comparable in β -carotene content (36 mg/100 g). A phytochemical analysis of the bambangan fruit juice showed the presence of gallic acid (1.7 mg/100 mL), vanillic acid (0.5 mg/100 mL) and *trans*-cinnamic acid (0.2 mg/100 mL).¹⁶

Apart from the fruits of *M. pajang*, little work has been done on the other plant parts (Table 1). From the leaves, methyl gallate, quercitrin, lupenone and lupeol, and from the stem bark, β -sitosterol, stigmasterol, mangiferonic acid, *trans*-sobrerol, lupeol, 3 β ,23-dihydroxy-cycloart-24-ene-26-oic acid and 3 β -hydroxy-cycloart-24-ene-26-oic acid have been reported.⁹ Also reported in the leaves of *M. indica*,²⁰ lupeol has anti-inflammatory and anti-arthritic properties.²¹

Polysaccharides from the fruit pulp extract of *M. pajang* have been isolated and separated into neutral and acidic components.²² The molecular weight of the neutral polysaccharide fraction (F1) was 7 kDa while those of the acidic polysaccharide fractions (F2, F3 and F4) were 13, 24 and 9 kDa, respectively. Major monosaccharide components were mannose, arabinose and glucose in F1 and F4; arabinose and rhamnose in F2; and glucose and fructose in F3.

Proximate composition

A study on the proximate composition of 42 varieties of tropical fruits belonging to eight species reported that fruits of *M. pajang* had the lowest fat content of 0.2-0.4%.²³ Protein and carbohydrate contents were 3.1-3.3% and 78-81%, respectively.

Essential oil

From the essential oil *M. pajang* fruits, 50 volatile components were identified⁷. Comprising mainly of monoterpene hydrocarbons (91%) and esters (7.6%), the two major components were α -pinene (67%) and α -phellandrene (11%). The kernel oil comprised 56%, 39% and 5.3% of saturated (SFA), mono-unsaturated (MUFA) and poly-unsaturated (PUFA) fatty acids.²⁴ SFA contained 40% of stearic acid and 16% of palmitic acid while MUFA contained 39% of oleic acid. Low contents of linoleic acid (5.0%) and linolenic acid (0.4%) were detected in PUFA.

PHARMACOLOGICAL PROPERTIES

Antioxidant properties

A study on the antioxidant properties of methanol extracts of *M. pajang* fruits reported the highest total phenolic content (TPC) and free radical scavenging (FRS) activity in the kernel.⁶ Compared to the peel and pulp, the TPC of the kernel was 4.5 and 17 times higher, while

the FRS value of the kernel was 1.1 and 2.3 times stronger, respectively. Several recent studies have been conducted on the antioxidant properties of *M. pajang*. Kernel, stem bark and leaf extracts (petroleum ether, chloroform, ethyl acetate and methanol) and isolated compounds of *M. pajang* showed that ethyl acetate and methanol extracts of the kernel had the strongest FRS activity with IC_{50} values of 7.3 and 8.8 $\mu\text{g/mL}$, respectively.⁹ It was rightly noted that the kernel and peel as by-products of bambangan fruits have stronger antioxidant properties than the pulp.²⁵ None of the stem bark extracts showed any FRS activity while only the ethyl acetate and methanol leaf extracts had weak activity. Isolated from the kernel and leaves of *M. pajang*, methyl gallate was the only compound with potent FRS activity of 6.24 $\mu\text{g/mL}$.

Compared to the fruits of six other wild *Mangifera* species, *M. pajang* ranked first in TPC and ascorbic acid content (AAC), but ranked poorly in FRS and ferric reducing ability.²⁶ The values of TPC (7056 mg/100 g) and AAC (403 mg/100 g) of *M. pajang* were 56 and 3.2 times those of *M. indica*.

The antioxidant properties of fermented bambangan have been studied. Results showed that fermentation at elevated temperature of 35°C yielded higher contents of phenolic compounds and stronger antioxidant activities than fermentation at room temperature of 28°C.²⁷ At 35°C, fermentation was faster and storage at 4°C was stable up to three months.

Antibacterial activity

The study on the antioxidant properties of extracts and isolated compounds of *M. pajang* also tested their antimicrobial activity.⁹ Results showed that most of the extracts and isolated compounds did not show significant inhibition activity towards methicillin-resistant *Staphylococcus aureus* (MRSA), *Pseudomonas aeruginosa*, *Salmonella choleraesuis* and *Bacillus subtilis*. Interestingly, methyl gallate was the only compound that exhibited very strong antibacterial activity against MRSA with a diameter of inhibition zone (DIZ) of 22 mm, comparable to streptomycin with a DIZ of 23 mm. None of the extracts and isolated compounds displayed any antifungal activity against *Candida albicans*, *Aspergillus ochraceus* and *Saccharomyces cerevisiae*. Incidentally, methyl gallate isolated from the leaves of *Anacardium occidentale* (cashew) has also been reported to possess antibacterial activity.²⁸ The compound displayed strong anti-quorum sensing against Gram-negative *Chromobacterium violaceum* by inhibiting violacein production.

Anticancer activity

The cytotoxic properties of *M. pajang* fruit extracts against human cancer cell lines have been reported. Only the kernel and peel extracts, but not the pulp extract, displayed cytotoxic effects.¹⁵ The extracts inhibited the proliferation of liver and ovarian cancer lines with IC_{50} values ranging from 35–92 $\mu\text{g/mL}$. The proliferation of colon cancer cell lines was inhibited only by the kernel extract (63 $\mu\text{g/mL}$). In a related study, the kernel ethanol extract of *M. pajang* was found to have cytotoxic effects on MCF-7 and MDA-MB-231 breast cancer cells with IC_{50} values of 23 and 31 $\mu\text{g/mL}$, respectively.²⁹ Prior to apoptosis, the extract induced sub-G1 cell cycle arrest in MCF-7 cells and G2/M cell cycle arrest in MDA-MB-231 cells. Apoptosis involved caspase-2 and -3 in MCF-7 cells, and on caspase-2, -3 and -9 in MDA-MB-231 cells.

Recently, the cytotoxic properties of extracts and isolated compounds of *M. pajang* have been reported.⁹ Results showed that the kernel had the strongest activity followed by the stem bark and leaves. Extracts of the kernel that exhibited strong cytotoxic activity with IC_{50} values less than 10 $\mu\text{g/mL}$ were methanol (MCF-7, HeLa and HT-29 cells) and ethyl acetate (MCF-7 and HeLa cells). Petroleum ether, chloroform and ethyl acetate extracts of the stem bark showed strong to moderate activity against MCF-7, HeLa and HT-29 cancer cell lines with IC_{50} values rang-

ing from 5–30 $\mu\text{g/mL}$. Moderate activity was shown against MCF-7 cells by the methanol leaf extract, and the petroleum ether leaf extract weakly inhibited HeLa and HT-29 cells. The study also tested the cytotoxic activity of compounds isolated from *M. pajang*.⁹ Isolated from the stem bark, 3 β -hydroxy-cycloart-24-ene-26-oic acid displayed the strongest activity against HeLa cells with an IC_{50} value of 6.3 $\mu\text{g/mL}$. Based on their cytotoxic activity, the compounds can be ranked as follows: 3 β -hydroxy-cycloart-24-ene-26-oic acid > methyl gallate > mangiferonic acid > lupenol > lupenone.

Cytoprotective effects

The protective effects of the *M. pajang* ethanol fruit extracts against oxidative damage caused by *tert*-butyl hydroperoxide in the human hepatocellular HepG2 cells have been investigated.³⁰

Results showed that only the kernel extract and quercetin (positive control) displayed cytoprotective activity in HepG2 cells, with EC_{50} values of 1.2 and 5.3 $\mu\text{g/mL}$, respectively. Expression of glutathione reductase and methionine sulphoxide reductase A were significantly up-regulated by the extract, suggesting its involvement in cytoprotection.

Health benefits

The fruit juice of *M. pajang* has been reported to be rich in gallic acid (1.7 mg/100 mL), and possess strong antioxidant properties such as free radical scavenging, β -carotene bleaching, and inhibition of haemoglobin oxidation and low-density lipoprotein (LDL) oxidation.¹⁶ Daily supplementation of hypercholesterolemic rabbits with *M. pajang* fruit juice powder had positive effects on their cardiovascular biomarkers.³¹ At dosages of 5%, 15%, 25% and 35% of the fruit powder, plasma malondialdehyde and hepatic malondialdehyde were significantly decreased. At 25%, the powder significantly decreased total cholesterol, triglycerides and LDL cholesterol by 37%, 67% and 30%, respectively. The animal experiment led to a clinical trial to test the health benefits of *M. pajang* fruit juice on plasma vitamins and antioxidant enzymes, and on the liver and kidney function of human subjects.³² Bambangan fruit juice in 250 mL bottles was produced from 50 g of fruit powder containing 66 mg of ascorbic acid and 18 mg of β -carotene. The clinical trial involved 32 healthy subjects (12 males and 20 females) aged 24–28 years from the Faculty of Medicine and Health Sciences of Universiti Putra Malaysia (UPM) in Serdang, Selangor, Malaysia. Compared with the placebo, consumption of 250 mL of the fruit juice daily for nine weeks significantly increased the concentration of plasma β -carotene and ascorbic acid. At week 4, the increase was 45% and 28%, respectively. In addition, there was significant improvement in the plasma total antioxidant status with liver and kidney functions unaffected. The study at UPM³² concluded that the consumption of *M. pajang* fruit juice resulted in a significant improvement in certain cardiovascular biochemical parameters, which can safeguard the risk of cardiovascular diseases.

CONCLUSION

In Sabah, *M. pajang* or bambangan has ethno-cultural significance and has become an iconic fruit among the Kadazan-Dusun people. The community has developed various traditional cuisines using fresh and preserved fruits. Phytochemical investigations showed that the pulp, peel and kernel of *M. pajang* fruits are rich in flavonoids and phenolic acids. Pharmacological research affirmed that bambangan fruits possess antioxidant, antibacterial, anticancer and cytoprotective properties. A clinical trial has demonstrated the health benefits of regular consumption of bambangan fruit juice. Since *M. pajang* only fruits once a year, there is great potential for developing commercial products such as fermented bambangan, fruit juice and freeze-dried fruits, which have a much longer lifespan. Traditional and functional food products from bambangan

fruits are being developed in Sabah. Further research is needed on the pharmaceutical applications of by-products such as the fruit peel and kernel, which are abundant in supply following consumption of the pulp.

ACKNOWLEDGEMENT

The authors are grateful to Dauni Seligi, Jamiss Aribin and Fabian Koret, field staff from the Sabah Forestry Department (SFD) in Sandakan, who have been monitoring and photographing the flowering and fruiting of bambangan when they go out on their field trips.

CONFLICT OF INTEREST

No conflict of interest to declare.

ABBREVIATIONS USED

AAC: ascorbic acid content; **DIZ:** diameter of inhibition zone; **FRS:** free radical scavenging; **HBA:** hydroxybenzoic acid; **HCA:** hydroxycinnamic acid; **LDL:** low-density lipoprotein; **MRSA:** methicillin-resistant *Staphylococcus aureus*; **MUFA:** mono-unsaturated fatty acid; **Neo:** neohesperidose; **PUFA:** poly-unsaturated fatty acid; **Rut:** rutinose; **SFA:** saturated fatty acid; **SFD:** Sabah Forestry Department; **TPC:** total phenolic content; **UMS:** Universiti Malaysia Sabah; **UPM:** Universiti Putra Malaysia.

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PICTORIAL ABSTRACT



SUMMARY

- *Mangifera pajang* or bambangan has ethno-cultural significance in Borneo.
- The brown mango has become an iconic fruit among the Kadazan-Dusun people in Sabah who have developed various traditional cuisines using fresh and preserved fruits.
- Phytochemical investigations showed that the pulp, peel and kernel of bambangan fruits are rich in flavonoids and phenolic acids.
- Pharmacological research affirmed that the fruits possess antioxidant, antibacterial, anticancer and cytoprotective properties.
- A clinical trial has demonstrated the health benefits of regular consumption of bambangan fruit juice.

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