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Review Article

A Review of the Antimicrobial Properties of three Selected Underutilized Fruits of Malaysia

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ABSTRACT

Fruits have many important biological effects such as antioxidant, antitumor, antimutagenic and antimicrobial properties[1]. This advantage also applies to the Malaysian fruits including underutilized fruits. Underutilized fruits are fruits that are rarely eaten, unknown and unfamiliar because some of the species only exist at a certain region[2]. Antibiotic resistance can be minimized by using new compounds that are not based on the existing synthetic antimicrobial agents[3]. Thus, natural antimicrobials seem to be the most promising answer to many of the increasing concerns regarding antibiotic resistance and could yield better results than antimicrobials from the combinatorial chemistry and other synthetic procedures[4]. This review paper emphasizes the antimicrobial characteristics possessed by three underutilized fruits namely *Phyllanthus acidus (P. acidus), Averrhoa bilimbi (A. bilimbi)* and *Passiflora edulis (P. edulis)* so that they can be used as natural antibiotic drugs and natural preservatives in processed foods. These three fruits are commonly known as "cermai", "belimbing buluh" and "markisa" respectively in Malaysia.

Keywords: antimicrobial, *Averrhoa bilimbi, Passiflora edulis, Phyllanthus acidus*, underutilized fruits.

INTRODUCTION

Underutilized fruits are neither grown commercially on a large scale nor traded widely but they are cultivated, traded and consumed locally[5]. There are very few people that know about the underutilized fruits and these fruits are usually grown on a small scale or found in the wild[6]. Malaysia is one of the countries in the world that has an abundant diversity of underutilized fruits either growing wild or planted in the villages of Peninsular Malaysia, Sabah and Sarawak. Malaysia is reported to house more than 500 species of cultivated, underutilized and rare fruit species. Sabah and Sarawak are known to have more than 200 species of edible underutilized fruits[2]. Antimicrobial agents are the chemical compounds or substances that can inhibit the bacterial growth in order to improve the quality and prolonged the shelf-life of food products[7]. Antibiotic resistance is a rising problem worldwide in the recent decades[8]. One-half of all deaths which occur in tropical countries are mainly caused by infectious diseases[9]. Recently, there is an increase of multiple drug resistance in human pathogenic microorganisms as a result of the indiscriminate use of commercial antimicrobial drugs. This problem has encouraged many researchers and manufacturers to search for new, safe and effective microbial agents from alternative natural resources like plant products. The demands for natural products are increasing from consumers as they become more concerned about the safety of synthetic preservatives used in food[10]. In Malaysia, underutilized fruits are economically less important but they have a potential for commercial exploitation[6]. The characteristic of some underutilized fruits that are acidic in nature and have a stringent taste resulted in the fruits not being able to penetrate the market if they are sold in a fresh form. Thus, product development and diversification on underutilized fruits need to be done to enhance the value of the products for medicinal, nutritional and economic uses[11]. Underutilized fruits will have better commercial potential when it was used to make jam, jelly, pickle and beverages[12]. Other than that, the high price of underutilized fruits products cause the inability of underutilized fruits to penetrate the local market. Underutilized fruit have limited availability and seasonality and time and work force requirements for harvesting the fruits cause the underutilized fruits to have high prices[13].

Botanical Description

P. acidus is believed to have originated from Madagascar and belong to the *Euphorbiaceae* family[14]. It has been widely dispersed throughout Asia, the Caribbean, Central and South America[15]. The tree can grow 2-9 m tall with spreading, dense and bushy branch. At the end of each branch, there are greenish branchlets with 15-30 cm long. The green leaves occur on short petioles with ovate shape and are 2-7.5 cm long. The flowers of *P. acidus* are small. This plant grows in a grape-like cluster and has a greenish-yellow smooth skin with a single hard stone inside. The fruit is crispy, juicy and very sour. In Malaysia, the ripe or unripe *P. acidus* fruits are used to make juice, cooked and served as relish, syrup or sweet preserve and also jam. Other than eaten fresh, this fruit is also often eaten as candied or pickled in salt. In the Philippines, the fruits are used to make vinegar while in India and Indonesia, the

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young leaves are cooked as greens. In the Indian traditional medicine, the fruit are taken as a liver tonic to enrich the blood. The leaves with the addition of pepper are used as apoultice on sciatia, lumbago and rheumatism. Cathartic can be cured by eating the seeds and the syrup of *P. acidus* can relieve stomach ache. Besides Malay gooseberry, this fruit also have other common names like ma-yom (Thailand), iba (Philippines), chalmeri and harpharoi (India), kantuet (Cambodia) and chum ruot (Vietnam)[15]. *A. bilimbi* is believed to have originated from the Moluccas Island of Indonesia and it belongs to the *Oxalidaceae* family[16]. The fruit can be found throughout the Asian countries, Brazil and Cuba. The tree is medium in size with 5-10 m in height but can reached up to 20 m. Each branch can have up to 45 leaflets with 30-60 cm long and the leaflets are oblong-pointed tips. The flowers have five petals, dark red in colour, small and fragrant. The fruits develop in clusters and the shape is oblong and nearly cylindrical with 4-10 cm long. The fruit changes colour from bright-green to yellowish-green and to ivory or nearly white when ripe. The skin of *A. bilimbi* is very thin, glossy, soft and tender[17]. *A. bilimbi* that has been dried or freshly harvested is sometimes added into Malaysian dishes such as curries and chutney to give a sour taste[18]. Besides food, the Malays use the fruit to clean the blade of a dagger and as a mordant in the preparation of an orange dye for silk fabrics. In traditional medicine of Malaysia, the leaves of *A. bilimbi* are used to treat venereal disease, cough, rectal inflammation and as an afterbirth tonic. In the Philippines, the leaves are used to make poultice and applied on itches, swelling of mumps, skin eruptions and insect bites[17]. The common names for *A. bilimbi* are kamias (Philippines), blimblim (French), belimbing buluh (Malaysia) and kaling pring (Thailand)[16]. *P. edulis* is a native species of Brazil. It was then distributed widely to other countries such as South America, the Caribbean, Asia, Africa, India and Australia during the 19th Century. This plant belongs to the *Passifloraceae* family[14]. *P. edulis* are semi woody vine species and can grow up to 15 m. The vines have a medium to large-toothed size of leaves. The tendrils are green in colour. The flowers have five white sepals surrounded by a threadlike crown in the centre. The fruits are nearly round or ovoid in shape and are 4 cm to 7.5 cm in diameter. The fruit has a tough, smooth and waxy skin. The skin colour ranges from green to purple. The pulp is very juicy and orange in colour. There are many small, hard, black and pitted seeds inside the fruit. This fruit can be eaten fresh by cutting the fruit into half and scooping out the pulp or eaten with fruit salads, ice cream or fruit juice. The fruit can also be used to make tropical fruit cocktail, passion fruit sherbet, jelly and jam. There are several common names of the passion fruit depending on which country they exist. It is called as granadilla (English), limangkan (Laos), parcha or marflora (Philippines), maracuja or morada (Spanish) and maracuya peroba (Portugeuse)[15].

Nutritional Composition

The proximate fruit composition of *P. acidus* per 100 g of edible portion was presented in Table 1[15]. The minerals of the flesh of *P. acidus* were also determined[15]. Calcium, iron and phosphorus are identified at levels of 5, 0.4 and 23 mg/100 g of edible portion respectively. The vitamins found in *P. acidus* are ascorbic acid, thiamine and riboflavin at 8, 0.01 and 0.05 mg/100g of edible portion respectively. The proximate fruit composition of *A. bilimbi* was represented in Table 2[15]. The minerals found in the flesh of *A. bilimbi* are calcium, iron, phosphorus, potassium and sodium at 5, 0.6, 13, 130 and 4 mg/100 g of edible portion respectively. The vitamins present are ascorbic acid, thiamine, riboflavin, niacin and vitamin A with 35, 0.02, 0.04, 0.02 and 105 mg/100 g of edible portion respectively. Table 3 shows the proximate composition of *P. edulis* per 100 g of edible portion[15]. Calcium, phosphorus and iron are minerals found in *P. edulis* at 3.6, 12.5 and 0.24 mg/100 g of edible portion respectively. Ascorbic acid, riboflavin, niacin and vitamin A are some vitamins found in *P. edulis at* 29.8, 0.131, 1.46 and 717 mg/100 g of edible portion respectively.

Antimicrobial Properties

The antimicrobial properties of the fruit part of *P. acidus* were investigated. The methanol extract of the fruit part of *P. acidus* was evaluated against 13 pathogenic bacteria at 400 µg/disc and 800 µg/disc. At higher concentrations, the results showed moderate to good zone of inhibition when tested on *Shigella dysenteriae, Salmonella typhi, Staphylococcus aureus, Bacillus subtilis* and *Bacillus megaterium*[19]. The methanolic extracts of leaves and fruit of *P. acidus* showed positive antimicrobial activity on *Proteus vulgaris, Shigella boydii, Shigella flexneri, Klebsiella aerogenes* and *Corney bacterium*[20]. The chloroform extract of *P. acidus* fruit was reported to exhibit an antimicrobial activity on four bacteria which are *S. dysenteriae, S. aureus, Escherichia coli* and *Sarcina lutea* at the concentration of 500 µg/disc[21]. In another study, the petroleum ether extracts of *P. acidus* fruit at the concentration of 500 µg/disc was also found to exhibit an antimicrobial activity on *S. typhi, S. aureus, Bacillus cereus, B. megaterium* and *E. coli*[22]. The antimicrobial properties of the leaf part of *P. acidus* on *Candida albicans, E. coli* and *S. aureus* was investigated. The hexane, dichloromethane, ethyl acetate and ethanol extract of the leaf showed a positive antimicrobial activity and the highest inhibition was possessed by the ethanol extract[23]. The methanol extract of the leaf part of *P. acidus* showed an inhibition zone when tested on *Micrococcus luteus* but there was no inhibition zone when tested on *S. aureus, B. subtilis, Pseudomonas aeruginosa, S. typhi, Aspergillus niger* and *Penicilium notatum*[24]. The ethanol extract of the bark of *P. acidus* showed a positive antimicrobial activity on *E. coli, S. typhi* and *Vibrio cholera* but showed a negative antimicrobial activity on *B. megaterium, S. aureus, B. subtilis, P. aeruginosa* and *S. dysenteriae*[25]. The methanol extract of the leaf of *P. acidus* possessed an antimicrobial activity when tested on *B. subtilis, E. coli, S. typhi, Klebsiella pneumonia* and *Proteus mirabilis* at the concentration of 12.5 mg/ml[26]. The 80% methanolic extract of the leaf of *P. acidus* showed a positive antimicrobial activity when tested on *E. coli, S. aureus, K. pneumonia, Bacillus spizizenii, Bacillus licheniformis* and *Pseudomonas stutzeri* at the concentration of 2.5 mg/ml[27]. The methanol extract of the fruit part of *P. acidus*

Table 1: Proximate fruit composition of *P. acidus* per 100 g of edible portion.

Fruit content	Percentage (%)
Water	91.7
Energy (kcal)	28
Protein	0.7
Lipid	0.52
Carbohydrate	6.4
Fibre	0.6
Ash	0.51

Table 2: Proximate fruit composition of *A. bilimbi* per 100 g of edible portion.

Fruit content	Percentage (%)
Edible pulp	86
Water	92.5
Energy (kcal)	27
Protein	0.61
Carbohydrate	6.3
Fibre	0.6
Ash	0.3
Fat	0.3

Table 3: Proximate fruit composition of *P. edulis* per 100 g of edible portion.

Fruit content	Percentage (%)
Moisture	85.6
Energy (kJ)	213
Protein	0.39
Fat	0.05
Carbohydrate	13.6
Fibre	0.04

possessed an antimicrobial activity when tested on *E. coli, S. aureus, B. cereus, B. subtilis, P. vulgaris, Alcaligenes faecalis, Enterobacter cloacae, Pseudomonas fluorescens, Serratia marcescens, Arthrobacter globiformis, Bacillus coagulans, Micrococcus roseus, Mycobacterium phlei, Mycobacterium rodochrus* and *Mycobacterium smegmatis*[28]. The methanol extract of the fruit part of *P. acidus* showed a positive antimicrobial activity when tested on *E. coli, S. aureus, B. cereus* and *Salmonella* spp. The extract also possessed positive a antifungal activity on *C. albicans* and *A. niger*[29]. There is an antimicrobial activity of the aqueous extracts of leaves and fruits of *A. bilimbi*. The aqueous extracts of both parts showed positive results when tested on the Gram-positive bacteria which are *S. aureus, Staphylococcus epidermis, B. cereus, Corneybacterium diphtheriae* and *Kochuria rhizophilia* at 100 mg/ml. Extracts from both parts also indicated positive results on the Gram-negative bacteria which are *S. typhi, Citrobacter fuendii* and *Aeromonas hydrophila.* In the same study, 100 mg/ml of the chloroform extracts of leaves and fruits of *A. bilimbi* also demonstrated an antimicrobial activity on the Gram-positive bacteria namely *S. aureus, S. epidermis, B. cereus, C. diphtheriae* and *K. rhizophilia* and the Gram-negative bacteria of *S. typhi, C. fuendii, A. hydrophila* and *P. vulgaris*[30]. Other than that, the methanol extract of leaves and fruits of *A. bilimbi* produced an antimicrobial activity on *S. aureus, B. megaterium, B. subtilis, S. lutea, Salmonella paratyphi, S. typhi, E. coli, S. dysenteriae, S. boydii, Vibro parahemolyticus, Vibrio mimicus* and *P. aeruginosa* at the concentration of 400 µg/disc[31]. Besides that, the methanol, hexane, carbon tetra chloride and chloroform extracts of the bark of *A. bilimbi* at the concentration of 400 µg/disc showed positive results on *B. subtilis, S. aureus, E. coli* and *V. mimicus.* The extracts also produced a positive antifungal activity on *C. albicans, A. niger* and *Sacharomyces cerevacae*[32]. The methanol extract of the fruit part of *A. bilimbi* showed a positive antimicrobial activity when tested on *S. aureus* and *B. cereus* and it also possessed a positive antifungal activity on *C. albicans* and *A. niger*[29]. The ethanol extract of the leaves of *A. bilimbi* produced a positive antimicrobial activity when tested on *B. megaterium, B. cereus, S. typhi, E. coli, S. dysenteriae, P. aeruginosa, S. paratyphi* and *V. cholera.* The extract also showed a positive antifungal activity on *C. albicans, Blastomyces dermatitidis, Cryptococcus neoformans, Pityrosporum ovale* and *Trichophyton* spp.[33]. The ethanol, methanol and aqueous extracts of leaves of *A. bilimbi* produced inhibition zones when tested on *B. subtilis, B. cereus, S. typhi, E. coli, V. cholera* and *P. mirabilis.* The aqueous extracts resulted in the maximum efficacy against most the bacteria compared to the ethanol and methanol extracts[34]. The methanol extract of the fruit part of *A. bilimbi* showed a negative antimicrobial activity when tested on *E. coli, P. aeruginosa* and *S. aureus* and also a negative antifungal activity on *A. niger* and *C. albicans* at 102.4 mg/ml[35]. The aqueous, chloroform and methanolic extracts of leaves of *P. edulis* showed a positive antimicrobial activity on *E. coli, B. subtilis, S. aureus, P. aeruginosa, S. typhi* and *K. pneumonia* at the concentration of 200 µg/disc. Among the three extracts studied, the methanolic extract exhibited the highest antimicrobial activity against the selected bacteria compared to the aqueous and chloroform extract[36]. In another study, the leaves and callus of *P. edulis* were examined using solvents of benzene, methanol, ethanol, isopropanol, chloroform and petroleum ether. Among these extracts, the chloroform extract of leaf and callus showed the maximum antimicrobial activity at the concentration of 250 µg/disc while extracts from benzene, petroleum ether and isopropanol were ineffective to inhibit the selected bacteria. The bacteria used in this study were *S. aureus, P. aeruginosa, K. aerogenes, Aeromonas spp., Serratia* and *E. coli*[37]. The methanolic extract of leaves of *P. edulis* showed positive results of antimicrobial activity on *S. aureus, Staphylococcus faecalis, B. subtilis, E. coli, P. vulgaris* and *S. typhi* at 200 µg/disc[38]. The leaf, stem and fruit of *P. edulis* were screened for antimicrobial activity using hexane, water, ethyl acetate and ethanol as solvents on *B. subtilis, S. aureus, P. aeruginosa, S. paratyphi, K. pneumonia* and *E. coli.* All the extracts produced an antimicrobial activity on the pathogenic bacteria tested but extract of hexane was the most effective[39]. The peel of the passion fruit also tested positive for antimicrobial activity on *S. aureus, B. cereus, P. vulgaris, S. typhi, B. subtilis, E. coli* and *P. aeruginosa* by using petroleum ether, chloroform and ethanol as solvents. The petroleum ether and chloroform extracts did not exhibit any antimicrobial

activity while the ethanol extract showed an antimicrobial activity against all bacteria except for *E. coli*[40]. The leaf and stem were tested for antimicrobial activity using petroleum ether and chloroform on *B. megaterium, B. subtilis, S. aureus, S. lutea, E. coli, P. aeruginosa, S. paratyphi, S. typhi, S. dysenteriae, S. boydii, V. mimicus* and *V. parahemolyticus*. The petroleum ether extract from the leaf parts of *P. edulis* showed a negative antimicrobial activity on all microorganisms but the chloroform extract of the leaf of *P. edulis* showed a positive antimicrobial activity on all microorganisms. Meanwhile, the petroleum ether and chloroform extracts of the stem of *P. edulis* showed a positive antimicrobial activity on all microorganisms. In comparison, the chloroform extract was more effective as an antimicrobial compared to the petroleum ether extract and the stems were also more effective compared to the leaves of the passion fruit[41]. The aqueous and phosphate buffer saline extract of passion fruit pulp showed a positive antimicrobial activity on *Streptococcus mutans* with the aqueous extract found to be more effective compared to the phosphate buffer saline extract[42]. The leaf and stem of the passion fruit were tested for antimicrobial activity with three types of solvents which are petroleum ether, acetone and methanol. All the extracts showed a positive antimicrobial activity on *B. subtilis, S. aureus, B. cereus, E. coli, P. aeruginosa, Listeria monocytogenes, Streptococcus gallolyticus* and *Klebsiella oxytoca*. The methanolic extract of leaf and stem were the most effective extract with leaves being more effective compared to the stem of passion fruit[43]. In addition, the leaf of passion fruit were screened for antimicrobial activity with petroleum ether, chloroform, ethyl acetate, methanol and aqueous extract on *B. subtilis, S. aureus, E. coli, S. typhi, P. aeruginosa, K. pneumonia* and *Streptococcus pyrogens*. All extracts showed a positive antimicrobial activity on the tested microorganisms and the most effective extract is methanol[44]. Besides, the aqueous peel extract of passion fruit showed a positive antifungal activity on *A. niger, Aspergillus flavus, Rhizopus stolonifera* and *Penicillium marneffei*. The extract also showed positive antimicrobial activity on *P. aeruginosa* and *Erwinia carotovora*[45].

CONCLUSION

Many parts of *P. acidus, A. bilimbi* and *P. edulis* have been tested for their antimicrobial activity. The results showed that any part of the selected underutilized fruits have an antimicrobial activity against wide range of Gram-positive and Gram-negative bacteria. These potential effects of underutilized fruits may be useful for further studies. It also can be used for the natural antibiotic drugs and natural preservatives in processed foods instead of chemically produced antibiotic and preservatives as they may have side effects.

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