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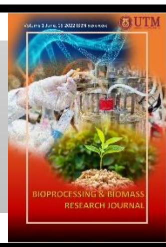

Review Article

Phytochemical Compounds, Therapeutic Effects and Antimicrobial Properties of Gac Fruit (*Momordica cochinchinensis* Spreng.)

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ABSTRACT

Gac fruit, a tropical fruit native to Southeast Asia, has been traditionally used in Asian cuisines as a red coloring agent and to maintain health. It is particularly rich in carotenoids, such as lycopene and β-carotene, and phenolics and flavonoids, which contribute to its antioxidant, antimicrobial, anticancer, and anti-inflammatory properties. The fruit's versatility is evident in its diverse processing options, transforming its aril, pulp, seed, and peel into powders or encapsulated oil products. Despite its nutritional potential, Gac fruit remains underutilized, with limited research on its therapeutic and antimicrobial properties, though preliminary studies show significant promise. This review synthesizes current scientific literature on the phytochemical composition, therapeutic effects of Gac fruit, and processing methods. Recent studies also have highlighted Gac fruit's potential antimicrobial activities, largely due to its rich polyphenol content. The findings highlight a clear relationship between the high polyphenols content of Gac fruit and its antimicrobial properties. This review integrates traditional knowledge with modern scientific insights, positioning Gac fruit as a valuable functional food and a promising tool for antimicrobial food ingredients. In the future, further research is needed to establish the mechanisms of the Gac fruit as a therapeutic and antimicrobial agent, as this area is currently underexplored.

INTRODUCTION

Gac fruit, scientifically known as *Momordica cochinchinensis* Spreng, is a member of the *Curcubitaceae* family. It was first discovered in Vietnam in 1790 (Vuong, 2000). The Cochinchina region in northern Vietnam is where the species name Cochinchinensis derives from (Franke et al., 2006). Its natural habitat encompasses Northeastern Australia, South, and Southeast Asia. The bright orange, spiky Gac fruit, also called cochinchin gourd, baby jackfruit, sweet gourd, or spiny bitter gourd, is easily grown in tropical regions like Thailand, Vietnam, Laos, and China. In Malaysia, the Gac fruit is commonly referred to as Tepurang whereas in Indonesia it is known as Torobok. Meanwhile, in Cambodia, it is known as Makkao, in Thailand as Fak-Khao, in China as Moc Niet Tu, and in India as Golkara. (Thavamany et al., 2020).

Over the past few decades, Gac fruit has gained recognition as one of the richest natural sources of

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carotenoids, surpassing many other commonly known fruits and vegetables. This is attributed to the remarkably high concentrations of β-carotene and lycopene found in its oily seed membrane (Chuyen et al., 2015; Ishida et al., 2004). Aoki et al. (2002) found the lycopene content of Gac fruit to be five times greater than tomatoes whereas, Kandlakunta et al. (2008) reported that its β-carotene content was eight times higher than the amount found in carrots. Lycopene is often linked to lowering the risk of various cancers, including lung, prostate, and digestive tract cancers. Besides, the human body transforms β-carotene into vitamin A, which is crucial for the health and proper growth of cell membranes (Kubola & Siriamornpun, 2011).

In addition to being rich in phytochemicals, some studies have reported that Gac fruit extract exhibits significant antimicrobial activity (Tinrat and Sila-Asna et al., 2016, Tinrat et al., 2014, Tinrat, 2014, Putta et al., 2014, Innun, 2013). The polyphenol compounds are responsible for its microbial inhibition by several mechanisms. Therefore, this review aims to provide a comprehensive summary of Gac fruit's traditional uses, phytochemical composition, therapeutic effects, and processing techniques. It specifically highlights the strong relationship between high polyphenol content with its antimicrobial properties and its emerging potential as a therapeutic and antimicrobial agent.

ANATOMY OF GAC FRUIT AND CULTIVATION

Gac fruit is a perennial dioecious species where it has separate female and male flowering on separate plants and the vines of the plant can extend as long as 6 meters long along a fence. The plants typically begin to flower approximately two months after planting. Female flowers display a bulge at the base where the undeveloped fruit forms, while male flowers have lighter-colored petals that create a more open bloom (Parks et al., 2013). Gac fruits often ripen about 9 to 10 weeks post-pollination. The fruit is primarily cultivated from roots, seeds, and branches.

When young, the fruit is green in color, turning dark orange or red as it ripens (refer to **Figure 1**). The fleshy, red, oily membrane called aril that surrounds the seed of the Gac fruit is edible, but the fruit's exterior shell is not palatable. Rather than being sweet, pulp and aril have a mushy texture and a moderate flavor (Pinthong et al., 2019). The ripe fruit is typically harvested in an outdoor cultivation environment from August to February. Although the fruit is firm at the time of harvest, it softens rapidly afterward, which can pose challenges for transportation and may affect its shelf life (Win et al., 2015).

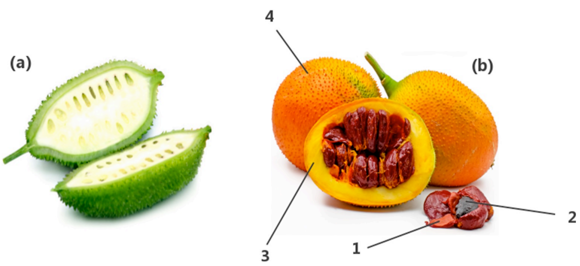

Figure 1 The structure of the Gac fruit is depicted in (a) a young fruit and (b) a moderately ripe fruit, showing the following components: 1. Aril, 2. Seed, 3. Pulp, 4. Peel with spines (Do et al., 2019).

Nevertheless, Gac fruit is presently cultivated as a commercial crop on a limited scale primarily in Thailand and Vietnam (Ariyarathna & Gamage, 2024, Tran & Parksa, 2022). The main challenge in its agricultural practice is differentiating between male and female plants, as the differences in features are only obvious when the flowers have bloomed. Several strategies have been identified to improve large-scale cultivation of Gac fruit. These include propagating and cultivating female plants that produce fruit, increasing pollen availability by preserving pollen, boosting fruit set via hand pollination, managing fruit size during plant growth, and controlling fruit quality in postharvest handling (Tran & Parks, 2022).

TRADITIONAL USAGE OF GAC FRUIT

Gac is widely believed to have health benefits and has been traditionally used as both a food source and folk medicine in Southeast Asia. Various parts of the Gac plant, including its oil, seeds, and roots, are commonly utilized in traditional medicine practices (Nhung et al., 2010). The beneficial compounds found in Gac, such as carotenoids, omega-3 fatty acids, α-tocopherol, flavonoids, and polyphenols, offer notable health benefits for humans. Thus, Gac fruit has been given the nickname "heaven's fruit" or "super fruit" due to the rich phytonutrient content present in all its parts of the fruit, including the aril, seeds, pulp, and peel, as well as its medicinal and pharmaceutical properties (Parks et al., 2013; Wong et al., 2004). As modern research continues to uncover its benefits, the Gac fruit remains a symbol of traditional wisdom and natural wellness in Southeast Asian cultures.

The vibrant red color of ripe Gac aril is often utilized as a food colorant in the preparation of red glutinous rice, known as xoi Gac (refer to **Figure 2**). In Vietnam, this meal is typically offered during special occasions like weddings and Lunar New Year celebrations (Ishida et al., 2004). Moreover, Chuyen et al. (2015) Stated that intake of Gac fruit aril oil as a supplement can help prevent xerophthalmia which is often referred to as night blindness and dry eyes, which primarily result from a deficiency in provitamin A. Besides, for the treatment of tinea, swelling, and inflammatory scrofula, grounded Gac seeds are applied externally and administered orally. Conversely, the roots of the Gac plant are also widely utilized in Vietnam to induce urination and blood circulation (Tran et al., 2016). Additionally, the seeds of the fruit, known as mubiezhi play a significant role in treating breast cancer in traditional Chinese medicine (Zheng et al., 2015).

Beyond its medicinal uses, Gac fruit is valued as a culinary ingredient. In Thailand, the young green Gac fruit is added to a curry or cooked with chili paste to make their dishes more scrumptious (Kubola & Siriamornpun, 2011). It is often mixed with rice, incorporated into soups, or used in desserts and beverages. Furthermore, Gac fruit is also utilized as a traditional treatment for degeneration of the macula, arthritis, and cardiovascular illness (Burke et al., 2005). Whereas the seeds have been traditionally used to cure various skin conditions and promote wound healing. These traditional healing properties are supported by the active compounds found in Gac fruit, such as carotenoids for maintaining eye health, phenolic compounds with anti-inflammatory and antioxidant effects, and antimicrobial activity for wound healing.

Figure 2 Xoi Gac, a type of Vietnamese red glutinous rice prepared with Gac aril and flavored with coconut milk.

PHYTOCHEMICAL COMPOSITION OF GAC FRUIT

Lately, Gac fruit has been gaining international attention due to its rich phytochemical profile and potential health benefits, with studies highlighting its potential to enhance skin health, promote cardiovascular wellness, and support overall immune function. Known for its striking red arils and spiky exterior, the unique compositions of Gac fruit include significant amounts of carotenoids, particularly lycopene, and β-carotene, alongside a diverse array of other bioactive compounds such as flavonoids, phenolics, saponins, and essential fatty acids. Therefore, consumption of this fruit regularly, which has a high concentration of bioactive components, enhances basic nutrition and helps prevent disease (Tran et al., 2016).

Carotenoids, especially β-carotene, and lycopene, are highly concentrated in the aril of the Gac fruit with a total carotenoid content of 78.00 mg/100 g fresh weight (FW) (Tran et al., 2016). It is also reported that the highest carotenoid content in decreasing order would be aril followed by pulp and peel of the Gac fruit (Aoki et al., 2002). Moreover, the total carotenoid content is stated to be the highest in fully ripe Gac fruits in comparison to medium-ripe ones which are still green in color. Therefore, fruit maturity is an important factor in achieving the highest yield of carotenoids. **Table 1** summarizes the carotenoid content of Gac aril reported in different studies. In addition, carotenoids are mostly sensitive to heat and light thereby, storage conditions and transportation above -20°C can lead to the degradation and isomerization of carotenoids (Vuong et al., 2006).

Table 1 Carotenoid levels of Gac aril across various studies.

Total Carotenoids (mg/ 100 g FW)	Lycopene (mg/ 100 g FW)	β-carotene (mg/ 100 g FW)	References
48.10	38.00	10.10	Aoki et al. (2002)
294.50	222.70	71.80	Ishida et al. (2004)
49.70	40.80	8.30	Vuong et al. (2006)
410.70	372.80	37.90	Nhung et al. (2010)
1502.00	702.00	800.00	Kubola & Siriamornpun (2011)
78.00	45.00	33.00	Tran et al. (2016)

Apart from β-carotene and lycopene, significant amounts of carotenoids such as zeaxanthin, β-cryptoxanthin, and lutein have also been found in Gac fruit. Kubola and Siriamornpun (2011) discovered that lutein was present in every part of the fruit, however, the peel and pulp of the fruit contained the highest quantities with 12,480 and 1,448 µg/g FW, accordingly. This finding implies that rather than being thrown away as a by-product during the processing of Gac fruit, the peel and pulp of the plant should be considered as a possible source of lutein.

Polyphenolics are a diverse group of naturally occurring constituents found in plants, known for their potent antioxidant activities, that are essential for protecting the body against oxidative stress and associated chronic diseases. In Gac fruit, polyphenolics are present in various forms, including phenolic acids and tannins. Flavonoids, a subclass of polyphenolics, are another essential group of bioactive compounds in Gac fruit which can be further categorized into several subgroups, such as flavanols, flavones, flavanones, and anthocyanins. Based on findings by Kubola and Siriamornpun (2011), the total flavonoid content (TFC) was identified to be highest in peel followed by aril, pulp, and seed, consecutively. Meanwhile, the total phenolic content (TPC) values were discovered to be highest in aril then only followed by peel, pulp, and lastly seed. Hence, there is a correlation between the total flavonoid phenolic content of Gac fruit and its antioxidant activity. **Table 2** illustrates a comparison of TPC, TFC, and FRAP across various parts of the Gac fruit from a study by (Kubola & Siriamornpun, 2011). Recent review papers have also collected and summarized the antioxidant activities of Gac fruit phenolic compounds which further strengthens this claim (Mat et al., 2024, Ariyarathna & Gamage, 2024).

Table 2 Comparison of total phenolic content (TPC), total flavonoid content (TFC), and Ferric reducing antioxidant power (FRAP) of various Gac fruit fractions (Kubola & Siriamornpun, 2011).

Gac Fruit Fractions	TPC (mg GAE/g)	TFC (mg RE/g)	FRAP (µmol $FeSO_4$/g)
Peel (Green)	2.80 ± 0.23	2.80 ± 0.14	471.92 ± 3.27
Peel (Yellow)	2.31 ± 0.20	2.25 ± 0.07	383.44 ± 6.94
Peel (Red)	1.90 ± 0.35	1.79 ± 0.23	213.87 ± 1.71
Pulp (Green)	2.62 ± 0.27	1.58 ± 0.12	466.36 ± 5.14
Pulp (Yellow)	2.60 ± 0.09	1.88 ± 0.24	301.08 ± 2.87
Pulp (Red)	1.52 ± 0.13	1.69 ± 0.07	74.02 ± 7.11
Aril	4.29 ± 0.15	2.09 ± 0.11	531.17 ± 1.89
Seed (Medium Ripe)	1.63 ± 0.22	1.57 ± 0.09	36.31 ± 3.42
Seed (Fully Ripe)	2.39 ± 0.26	1.88 ± 0.10	46.49 ± 1.59

Furthermore, a combination of saturated, unsaturated, poly-, and mono-unsaturated fatty acids can be found in Gac fruit. It was found that from 102.00 mg/g FW of Gac fruit, 70% of the total fatty acids were unsaturated, whereas 50% were polyunsaturated, respectively (Vuong et al., 2002). An investigation conducted by Bruno et al. (2018) revealed that the concentration of oleic acid in Gac fruit was greater with 44.5% of total fatty acids than that of watermelon and tomato, which had concentrations of 20.7% and 2.5%, respectively. Therefore, it can be applied as an additional source of oleic acid, alongside common sources like coconut, soy, and palm. These aril-derived fatty acids from Gac fruit are essential for the uptake of fat-soluble elements through low-fat diets, especially carotenoids. Therefore, Gac fruit is considered highly nutritional as the intake of Gac oil consisting of fatty acids along with carotenoids about 2 mL/day raises the consumption of β-carotene and essential fatty acids while lowering the consumption of saturated fatty acids (Vuong & King, 2003).

In addition, Gac fruit is an exceptional source of α-tocopherol, the most active form of Vitamin E in biological systems. This vitamin is primarily located in the fruit's arils, where it is found alongside high concentrations of carotenoids such as lycopene and β-carotene. Thus, the presence of these fat-soluble compounds in a lipid-rich matrix enhances their bioavailability, making Gac fruit an efficient source of Vitamin E. Vuong et al. (2006) Reported that the average concentration of α-tocopherol in Gac aril was higher with 7.6 mg/100 g of FW in comparison to the pulp or even other Momordica genus fruits such as the bitter melon (*M. charantia*). Besides, vitamin E is also crucial in preserving the fruit's polyunsaturated oil from oxidizing. Hence, the recommended daily intake of 15 mg of vitamin E can be successfully achieved by consuming vitamin E-rich Gac fruit.

THERAPEUTIC EFFECTS OF GAC FRUIT

In recent years, scientific research has begun to uncover the pharmacological activities of Gac fruit, highlighting its potential as a valuable resource in both traditional and modern medicine. The fruit is rich in bioactive compounds, including carotenoids, polyphenols, flavonoids, vitamins, and essential fatty acids. Research also indicates that Gac fruit exhibits a range of pharmacological activities, including antioxidative, antimicrobial, anti-inflammatory, and anti-cancer properties (Ariyarathna et al., 2024). These activities are attributed to its unique phytochemical profile, which interacts synergistically to promote health and prevent disease. As research continues to explore these diverse pharmacological activities, Gac fruit emerges as a promising candidate for the development of nutraceuticals and functional foods aimed at improving health and preventing diseases.

The phytochemicals in Gac fruit are renowned for their high nutritional value as the fruit's aril and the products derived from it have been shown to possess exceptionally high antioxidant properties due to their remarkably high concentrations of carotenoids, particularly lycopene. The antioxidant activities were assessed using diphenyl-picrylhydrazyl (DPPH) radical scavenging, ferric reducing antioxidant power (FRAP), and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) methods. Research on various parts of the Gac fruit, including the aril, peel, pulp, and seed, revealed that the aril exhibited the highest antioxidant activities with 531.17 ± 1.89 µmol $FeSO_4$/g. This suggests that the antioxidant activity of aril is likely attributed to the highest TPC of 4.29 ± 0.15 mg GAE/g (Kubola & Siriamornpun, 2011).

The antioxidant activities of pulp and peel were found to decrease from the immature to the ripening stage, whereas the seed's antioxidant activity rose from the mature to the ripening stage. Thus, during the fruit development stage, there was a correlation between the decline in TPC and TFC and the reported drop in antioxidant capacity in DPPH and FRAP (Kubola & Siriamornpun, 2011). In addition, Tinrat et al. (2014) Found that the ripe ethanolic aril extract exhibited the highest antioxidant properties, measuring 4.87 and 0.016 mg ascorbic acid equivalents in the FRAP and DPPH assays, accordingly in comparison to the peel and pulp. Hence, these findings suggest that the antioxidant activities of Gac fruit can be greatly affected by its level of maturity and different parts of the fruit.

Additionally, numerous in vivo and in vitro studies have been conducted to investigate the anticancer properties of Gac fruit and it was found that the seed and aril extracts have shown prominent anticancer activities (Do et al., 2019). The anticancer potential of the seeds has lately drawn attention from fellow researchers due to the identification of unique saponins that include chymotrypsin inhibitors, trypsin inhibitors, and disaccharide chains. Furthermore, Liu et al. (2012) findings reported that the growth of human gastric cancer cells SGC7901 and MKN-28 was inhibited by the seed extract. The mechanism behind the seed extract's antiproliferation properties was attributed to the activation of apoptosis through increased caspase-3 and caspase-9 enzyme activity, poly (ADP-ribose) polymerase, and p53 signal pathways. The anticancer properties of water extract from aril towards breast cancer and melanoma cells were evidenced with IC_{50} concentration values ranging from 0.49 to 0.73 mg/mL. The highest toxicity recorded was 70% and 50% cell death in melanoma cells and breast cancer cells, respectively. The cytotoxicity caused both dose- and time-dependent apoptotic and necrotic cell death (Wimalasiri et al., 2020).

The Gac seed extract also aids in the development of an inhibiting treatment for breast cancer. It was observed that the seed extract successfully prevented ZR-75-30 cancer cells from migrating, adhering, and invading (Zheng et al., 2014). Moreover, the anti-estrogenic properties of Gac aril extract and its underlying method of inducing apoptosis in MCF-7 human breast cancer cells were further studied and the findings reported that the human MCF-7 breast cancer cells are susceptible to the anticancer effects of aril extract through both intrinsic and extrinsic apoptotic pathways (Petchsak & Sripanidkulchai, 2015).

Lycopene, which is abundant in Gac fruit, has also been linked to demonstrating anti-inflammatory properties. It was reported that the Gac seed extract exhibits anti-gastritis properties in rats with ethanol and diclofenac-induced gastritis, according to Jung et al. (2013). Moreover, the seed extract treatment showed higher preventative efficacy in comparison to a conventional rebamipide treatment, a gastroprotective medication that works by enhancing the mucosal defense mechanisms and promoting the healing of the gastric mucosa. Additionally, the Gac seed is a significant source of saponins and triterpenoids. It was reported that Momordica saponin I, one of the extracted triterpenoid saponins, inhibited the synthesis of nitric oxide (NO), the activation of inflammatory signaling proteins via repression, and the transcriptional activation of inflammatory genes (Yu et al., 2017). In addition, lutein in Gac fruit was shown to exhibit a protective effect on the retina from light- and oxidants-induced oxidative stress. The Gac lutein was able to repair the damaged retinal pigment epithelium (RPE) cells to over 80% viability, which is similar to the effects seen when using pure lutein (Nguyen et al., 2024).

The recent study also reported the effect of Gac juice and its probiotic-fermented version on gut health. Both Gac juice and its probiotic-fermented version positively impacted gut health by increasing beneficial bacteria and enhancing the production of short-chain fatty acids (SCFAs). The probiotic-fermented Gac juice showed a stronger effect than non-fermented Gac juice, promoting species diversity, increasing beneficial bacteria like *Akkermansia*, and reducing harmful bacteria like *E. coli*. These results suggest that fermented Gac juice has greater potential as a functional food for supporting gut health and maintaining a balanced microbiota (Marnpae et al., 2024).

PROCESSING METHODS OF GAC FRUIT

The various parts of the Gac fruit, including the peel, seeds, aril, and pulp, are rich in carotenoids, fatty acids, and α-tocopherols. Numerous studies have been conducted to prove the pharmacological effects and health advantages of these bioactive compounds. Fresh Gac aril often needs to be processed to preserve its carotenoid compound as they are highly vulnerable to oxidation and deterioration caused by external factors that include particularly microorganisms, light, oxygen, and temperature. As such, it is critical to preserve, extract, and enhance to properly utilize these potent phytochemical resources. Currently, there are a few viable methods for achieving this, such as drying and oil extraction, and incorporating Gac fruits into other cuisines.

One of the oldest techniques for food preservation is the removal of moisture. The advantages of drying Gac fruit include extending its shelf life, lowering storage and transportation expenses, and increasing the likelihood of out-of-season availability of the fruit. Traditionally, fresh Gac arils are typically placed under sunlight until they are dry. Then, the dried arils are packed and kept as traditional medicine or food coloring agent, once the seeds are removed (Vuong et al., 2002). However, this method can lead to substantial losses of lycopene and β-carotene due to degradation from direct sunlight and oxygen exposure. As a result, several processing techniques have been established for Gac fruit, which can be categorized into two main approaches that include, extraction of oil from the aril and drying to produce dried aril powder.

The process of drying modifies the physical, chemical, and biological characteristics of raw materials, including their antioxidant activity, enzymatic activity, flavor, and scent, in addition to lowering their water activity. Thus, the pharmaceutical and food industries must utilize a suitable drying process to preserve the nutritional value, carotenoid contents, and other bioactive compounds present in the Gac fruit. Hence, the physiochemical characteristics, carotenoid content, and antioxidant activities of Gac fruit have been further studied concerning different drying factors and procedures to produce the Gac aril powder.

Pretreatment is often carried out prior drying method to enhance the quality of dried products and speed up the drying process, Studies conducted by Kha et al. (2011) Have examined the effects of pretreatments such as blanching and soaking in ascorbic acid or bisulfite towards the color characteristics, total carotenoid content (TCC), and total antioxidant activity (TAA) in aril powders. It was found that higher drying temperatures resulted in greater losses of TCC and TAA in the powders. Among the methods of blanching and steaming, blanching in citric acid solution, and steaming for 6 min was identified as the most effective pretreatment for preserving TCC in Gac powder dried at 60 °C. Based on research conducted by Tran et al. (2008), the powder created with the enzymatic pre-treatment had less carotenoid content than the powder produced without the enzymatic pre-treatment, using the same drying techniques. At the end of the study, the Gac powder with the highest carotenoid content and brightest color was obtained by freeze-drying whole-seed aril, out of five different drying techniques, including air drying, vacuum drying, spray drying, and oven drying.

It is widely recognized that freeze-drying is an effective technique for preserving the nutritional, functional, and sensory attributes of foods. However, its application in the food industry is limited due to the high operational costs, which are about four to eight times greater than those of hot air drying. In terms of spray drying conditions, high-quality aril powder, characterized by color, TAA, and TCC, can be achieved with an inlet temperature of just 120 °C and a maltodextrin concentration of 10% w/v (Tuyen et al., 2010). Gac peel and pulp make up the largest portion of the fruit by weight and are also rich in carotenoids. Drying is the most appropriate method for processing the peel and pulp into powder as they are easily degradable and perishable without proper storage. The findings by Trirattanapikul and Phoungchandang (2016) Indicate that heat pump-assisted dehumidification drying is the most effective method for drying Gac pulp. Additionally, it is recommended that Gac peel be pretreated with ascorbic acid before drying at 70 °C to reduce carotenoid loss and enhance antioxidant retention.

Moreover, the mechanical pressing method is the most commonly employed technique for recovering oil and carotenoids from Gac aril in the oil processing industry. (Vuong, 2000). Gac aril is first smashed and lightly heated with 5 min of hot steaming before pressing to yield 1 L of oil from 100 kg of whole fresh Gac fruit. According to Vuong & King (2003), a batch duration of 10 to 20 min produced 50% oil with total carotenoid concentrations of 5.77 mg/mL during the first study and 3.19 mg/mL during the second study in Gac oil. Furthermore, various findings have demonstrated that SC-CO_2 extraction is a viable substitute for conventional solvent extraction and mechanical pressing. Studies have been carried out to create models and provide information about the workings and best practices of SC-CO_2-based Gac oil extraction mechanisms. Oil solubility data was mathematically modeled to determine SC-CO_2's oil loading capacity. The findings by Tai and Kim (2014) Demonstrated that, at a temperature of 70 °C, a pressure of 400 bar, and a particular flow rate of 70 kg/h CO_2 per kg of Gac aril, the oil recovery surpassed 95% after 120 min of extraction.

EFFECT OF POLYPHENOLS ON ANTIMICROBIAL ACTIVITY

Polyphenols, a diverse group of naturally occurring compounds found in fruits, nuts, seeds, vegetables, stems and flowers are well-regarded for their potent antioxidant and antimicrobial capacities. The antimicrobial activity of polyphenols has been extensively researched, with particular focus on their relationship to antimicrobial activities in fruits. This area of study holds significant interest in both the medical and agricultural fields, as understanding this connection can lead to the development of natural antimicrobial agents and improved strategies for disease prevention and treatment. Numerous recent publications have highlighted the efficacy of polyphenols in combating various pathogens.

Putta et al. (2014) Evaluated the relationship between phytochemicals and antimicrobial activity of the aqueous fruit extract of *Momordica cochinchinensis* (AFMC). The findings demonstrated the presence of phenolic compounds, flavonoids, glycosides, and reducing sugars. The antimicrobial activity of AFMC was evaluated in vitro through the agar well diffusion method, testing its effectiveness against *Enterococcus faecalis, Vibrio cholerae, Shigella dysenteriae,* and *Salmonella typhi.* The largest zone of inhibition measured 10 mm against *S. dysenteriae* (Gram-negative) at a concentration of 100 µg of AFMC extract, while the maximum zone of inhibition against *E. faecalis* (Gram-positive) was 9 mm at the same concentration. This indicates that AFMC exhibits significant antimicrobial properties, against Gram-negative bacteria, in comparison to Gram-positive bacteria.

In a study conducted by Tinrat (2014), the antimicrobial properties of the ethanolic extract from three parts of the Gac fruit including peel, pulp, and aril were examined against six pathogenic strains. The findings revealed that the ethanolic extract from the pulp fraction had the highest TPC of 0.205 mg GAE/g FW followed by the aril extract at 0.191 mg GAE/g FW and the peel extract at 0.055 mg GAE/g FW). The extracts from the pulp and peel parts of the fruit exhibited the highest bactericidal activity against *E. coli* ATCC 25922, with a MIC value of 1.562 mg/mL, whereas the aril extract, with an MIC value of 3.125 mg/mL, demonstrated the greatest bactericidal activity against *P. aeruginosa* ATCC 27853. This indicates that the effectiveness of antibacterial properties is proportional to the antioxidant contents of specific parts of the fruit. The MBC values exceed 50 mg/mL against pathogenic strains. The comparative analysis revealed that *E. coli* ATCC 25922, a Gram-negative bacterium, was more susceptible to the peel and pulp extracts than the other strains tested. This suggests that the antimicrobial effectiveness may be related to the specific compounds in the cell walls of different pathogenic strains. Gram-positive bacteria have cell walls with peptidoglycan layers and teichoic acid molecules, while Gram-negative bacteria have a thinner peptidoglycan layer and lack teichoic acid (Silhavy et al., 2010).

Furthermore, Gac fruit's crude aril oil, extracted using a screw press and supercritical carbon dioxide (SC-CO_2) extraction method, demonstrated significant suppression against both Gram-positive and Gram-negative bacteria. The crude extracts and oils exhibited MIC and MBC values ranging from 0.78 to 400.0 mg/mL (Tinrat & Sila-Asna, 2016). This indicates the high efficiency of the SC-CO_2 method in extracting phytochemicals from Gac fruit, as evidenced by its superior antimicrobial effect.

Kubola and Siriamornpun (2011) Reported that phenolic content in the peel and pulp of Gac fruit decreases with maturation (immature > ripe fruit). This finding was further supported by Tinrat (2014) Who examined the aril, pulp, and peel of both, ripe and unripe Gac fruit extracts using various solvents (hexane, methanol, and acetone) to evaluate their antimicrobial and antioxidant properties. The acetone extract of the pulp from unripe Gac fruit showed the highest TPC at 41.60 ± 0.24 mg GAE/100 g FW, which was significantly higher than in other parts of the ripe fruit. Across all solvents, the TPC was highest in the aril, followed by pulp, and then peel. These findings, summarized in **Table 3**, compare the TPC of different Gac fruit fractions extracted using various solvents.

Table 3 Total phenolic content (TPC) of Gac fruit fractions obtained through various extraction solvents (Tinrat, 2014).

Gac Fruit Fractions	Unripe/ Ripe	Extraction Method	Total Phenolic Content (mg GAE/ 100g FW)
Gac Peel	Unripe	Solvent Extraction (Methanol)	20.30 ± 0.10
Gac Pulp			4.70 ± 0.00
Gac Aril			35.10 ± 0.10
Gac Peel	Ripe		13.90 ± 0.10
Gac Pulp			23.30 ± 0.10
Gac Aril			33.50 ± 0.10
Gac Peel	Unripe	Solvent Extraction (Acetone)	11.00 ± 0.40
Gac Pulp			41.60 ± 0.24
Gac Aril			16.60 ± 0.34
Gac Peel	Ripe		4.60 ± 0.00
Gac Pulp			8.70 ± 0.10
Gac Aril			23.50 ± 0.10
Gac Peel	Unripe	Solvent Extraction (Hexane)	ND
Gac Pulp			ND
Gac Aril			ND
Gac Peel	Ripe		3.50 ± 0.10
Gac Pulp			3.60 ± 0.00
Gac Aril			4.00 ± 0.10

ND= Not determined.

Meanwhile, the antimicrobial properties of the Gac fruit extracts were evaluated against six pathogens using broth macro-dilution and agar disc diffusion techniques. The acetone extract of unripe pulp was the most effective, particularly against *E. coli* ATCC 25922, showing larger inhibition zones at all concentrations (1, 10, and 100 mg/mL) and a lower MBC of 100 mg/mL compared to other extracts. The methanolic extract of ripe peel was most effective against *S. aureus* ATCC 1216. The MIC for both ripe and unripe fruit extracts ranged from 50 to 100 mg/mL. These findings indicate that both ripe and unripe Gac fruit extracts could be valuable antimicrobial and antioxidant agents for food and pharmaceutical industry applications (Tinrat, 2014).

Innuna (2013) investigated the antimicrobial activity of Gac fruit pulp and aril extracts using 95% ethanol, ether, and distilled water against twelve bacterial strains using the agar disk diffusion method. The pulp extract with distilled exhibited significant inhibition against Staphylococcus aureus (10.0 mm), *Planococcus* sp. (9.2 mm), and *Micrococcus luteus* (9.0 mm). The highest antibacterial activity was observed against *M. luteus* 745 with the ethanolic pulp extract (20.0 mm). Similarly, the aril extract with ethanol demonstrated strong inhibition against *M. luteus* (19.0 mm). In contrast, pulp and aril extracts with ether showed no antimicrobial activity. A study by Oyuntsetseg et al. (2014) demonstrated that Gac fruit seeds possess potent antiviral properties. In vitro tests showed that seed extracts, when used at a high concentration of 0.5%, significantly reduced the infectivity of influenza A virus H3N8. These findings suggest that Gac seeds could be a promising source for the development of new antiviral drugs.

Many studies have reported the potential antimicrobial mechanisms of phytochemicals. Antioxidant compounds like flavonoids exert antibacterial effects by changing the permeability of bacterial cells, disrupting their cell walls, inhibiting nucleic acid synthesis, interfering with the synthesis and expression of proteins, and decreasing enzyme activity (Qi et al., 2022). On the other hand, carotenoids can suppress quorum-sensing genes which responsible for biofilm formation, damage cell wall, and cell membrane, depolarize the cytoplasmic membrane, modulate efflux pumps, and cause accumulation of intracellular reactive oxygen species (Naisi et al., 2023, Karpiński et al., 2022).

CONCLUSION

In conclusion, Gac fruit (*Momordica cochinchinensis* Spreng.) demonstrates significant potential as a source of bioactive compounds with antimicrobial properties and health-promoting benefits, which supports its traditional claims. The fruit's phytochemical profile, rich in carotenoids, flavonoids, and other bioactive, has been linked to its antimicrobial and therapeutic effects, as supported by several studies. However, further evidence-based research is essential to elucidate the precise mechanisms of these compounds and their roles in human health. Addressing potential limitations, such as bioavailability, dosage, and possible side effects, will provide a more comprehensive understanding of Gac fruit's efficacy and safety. The exceptional phytochemical content of Gac fruit, particularly its carotenoids, surpasses that of other plants, yet it remains underutilized as a food source. Strengthening the scientific foundation with these insights could enhance its use in functional foods and therapeutic interventions, highlighting its significant market potential as a valuable ingredient in the growing health and wellness industry, particularly in nutraceuticals and functional foods.

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