Multidisciplinary Cancer Investigation **Original Article**

March 2024, Volume 8, Issue 1

The Antibacterial properties of *psidium guava* to eliminate *Helicobacter pylori*: Preventing Gastric Cancer

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Submitted: 30 June 2024

Revised: 02 September 2024

Accepted: 29 August 2024

e-Published: 10 October 2024

Keywords:
Psidium guava
Medicinal plants
Helicobacter pylori
Cancer

Antibiotic treatment has often failed due to the increased biological resistance of *Helicobacter pylori* (*H. pylori*). Therefore, medicinal plants have become necessary to combat this bacteria. This study aims to investigate the antibacterial properties of psidium guava and focus on the effect of the properties of this plant on *H. pylori* infection to prevent gastric cancer. This study systematically followed the PRISMA guide and searched various international and national databases such as SciELO, PubMed, ProQuest, Google Scholar, Ovid, SID, MediLib, IranDoc, and Cochrane. The search was conducted for articles published in Farsi and English from 2003 to 2023, covering two decades. The search was performed using Persian keywords such as "antibacterial activity," "Psidium guava," and "*Helicobacter pylori*", as well as their English equivalents and various combinations of these words. The articles were evaluated based on their relevance to the study's purpose without statistical analysis. Out of the 368 selected articles, only nine were deemed final. The results showed that most research was focused on the leaves and fruits of *Psidium guava* L (*PGL*). It can be stated that the essential oil of *PGL* leaves exhibits the most biological activity, and the compounds present in this plant make it a promising candidate for producing drugs with antibacterial activity. *PGL* plant extracts have appropriate antimicrobial substances that can be used as a pharmaceutical base or a suitable herbal medicine to fight against *H. pylori*, heal stomach ulcers, and ultimately prevent gastric cancer (GC).

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INTRODUCTION

One of the main challenges of treating bacterial infections is the increase in antibiotic resistance. One of these infections is stomach infection caused by Helicobacter pylori (*H. pylori*), which causes approximately 90% of gastric cancers [1]. *H. pylori* is a prevalent bacterial infection that is crucial in causing gastrointestinal ulcers. It is also a leading cause of chronic and progressive inflammation that can ultimately lead to gastric cancer [2]. *H. pylori* is a gram-negative bacterium found on the stomach's epithelial lining, which usually affects individuals in

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[DOI: 10.61186/mci.8.1.5]

their early years and persists indefinitely if not treated. H. pylori infection is the main cause of three important upper gastrointestinal tract diseases. Among them are duodenal ulcers or gastric ulcers, reported in 1 to 10 percent of affected patients, and GC, which occurs in 0.1 to 3 percent of cases. Additionally, Mucosa-Associated Lymphoid-Tissue (MALT) is another rare disease caused by H. pylori infection, affecting just 0.01 percent of individuals [3].Current treatments with a combination of at least three antibiotics fail in 40% of patients, which is one of the most important challenges in the health field [1].

In recent years, *H. pylori* has become increasingly difficult to treat due to its high antibiotic resistance. H. pylori eradication is the first choice for treatment due to the various complications associated with this bacterium. There are different drugs to treat and destroy this bacteria, and clarithromycin is a key component of some of these drugs [4]. In a study conducted in China, the resistance levels of H. pylori to clarithromycin, metronidazole, and levofloxacin were 37%, 77%, and 33%, respectively [5]. Recently, the World Health Organization has recognized clarithromycin-resistant *H. pylori* as a "high priority" for which more effective drugs are needed [6]. Researchers are exploring various methods to address this issue, including developing new antibiotics, immunotherapy, probiotics, and medicinal plants. Consequently, there is a growing focus on finding more effective ways to treat and eradicate H. pylori, increasing the importance of exploring the potential of medicinal plants.

Psidium guava plant (PGL) is a subfamily of Myrtaceae found in tropical and subtropical countries. This plant is native to Central and Latin America and adapts well to soil use. In Iran, it is cultivated in Sistan –Baluchistan, and Hormozgan. PGL is a plant with great diversity that is used as food and a therapeutic agent.[7]. PGL is used to treat symptoms related to the problems and diseases caused by the action of pathogenic/opportunistic microorganisms [8]. In the ongoing battle against bacterial infections, discovering new sources of antimicrobial compounds is crucial. To advance this goal, the present review study was conducted to investigate the antibacterial properties of the PGL plant and determine its effect on H. pylori. Researching the antibacterial properties of medicinal plants, such as psidium guava, could provide insights into potential alternative treatment options for *H. pylori* infection.

METHODS

This study aimed to conduct a systematic review following the PRISMA guide and searching in various international and national databases such as SciELO, PubMed, ProQuest, Google Scholar, Ovid, SID, MediLib, IranDoc, and Cochrane. The search was conducted for articles published in Farsi and English from 2003 to 2023, covering two decades. The search was performed using Persian keywords such as "antibacterial activity," "Psidium guava," and "Helicobacter pylori", as well as their English equivalents and various combinations of these words. The articles were evaluated based on their relevance to the study's purpose without statistical analysis (Table 1).

Two authors checked the articles to ensure the work was carried out and there was a significant match. The Irandoc database was also used to check the theses and dissertations, but no case was found that matched the purpose of the present study.

The inclusion criteria were articles that only dealt with the antibacterial effects of the Psidium guava plant and were related to the study's objectives. Exclusion criteria included articles whose full text was unavailable or presented as a poster, speech, or letter to the editor. Also, the authors qualitatively evaluated

Table 1. Search strategy for articles in PubMed database.

Row	Boolean operators	Result
1	AND OR	**"antibacterial activity"[Title] AND (("psidium"[MeSH Terms] OR "psidium"[All Fields]) AND "guava"[Title]) AND "helicobacter pylori"[Title]**

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[DOI: 10.61186/mci.8.1.5]

the validity of the selected articles in terms of their originality, credibility, and the Journal that published them. The final articles were reviewed according to study type using the PRISMA guidelines in the next step (Figure 1).

In the initial search strategy, articles containing the keywords in the title and abstract were selected based on the research's purpose, and there were 368 articles. Finally, considering the entry and exit criteria of the articles in the research, nine articles, considering all the criteria, were included in the study for review.

RESULTS

The summary of the results of the reviewed studies was classified in the tables in the text of this research. The antibacterial activity of the PGL plant is listed in (Table 2). The study findings suggest that guava leaves and bark possess antibacterial properties.

PGL leaf extract contains high amounts of gallic acid and quercetin, which greatly influence antioxidant activity. 70% ethanol extract of this plant causes high inhibition for H.Pylori. After the phytochemical screening, the multi-herb suspension (combination of guava with herbs of similar function) showed the presence of terpenoids, flavonoids, and tannins that have anti-ulcer activity (Table 3).

The leaves of the PGL plant are used in India as an anti-fever, anti-spasm, and rheumatism. It is used in Colombia, Mexico, Maya, USA, and Mozambique to treat diarrhea and stomachache. In the United States, the leaves are used as an antibiotic [27]. Many of the biological activities of PGL are related to the active chemical compounds of this plant, which are mentioned in Table 4. It has been reported that the

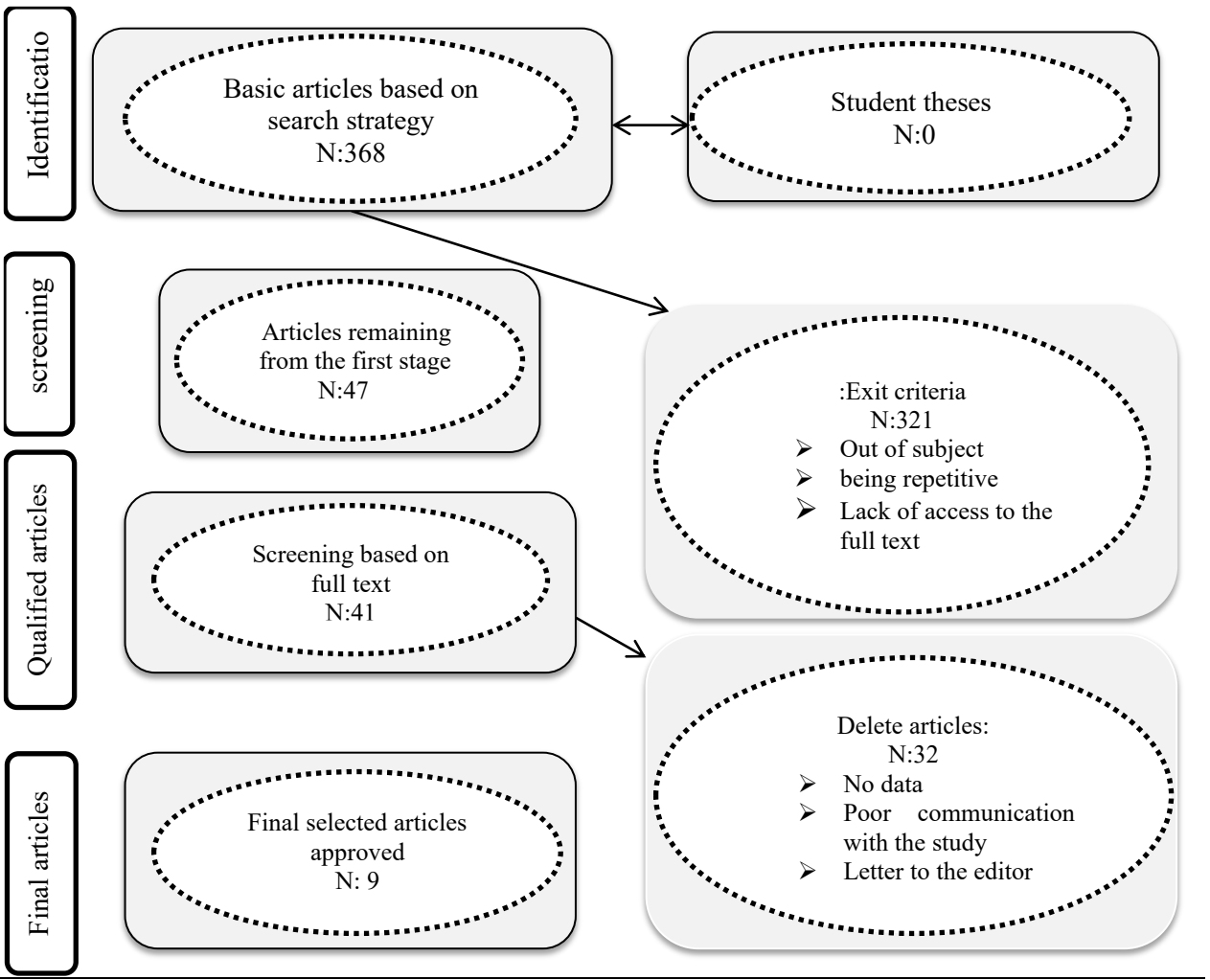


Figure 1. Flowchart of the process of selecting articles and entering the study.

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Table 2. Antibacterial activity of *PGL* (In vivo and In vitro).

Row	Plant/part used	bacteria	Results	Ref.
1	*PGL* / leaf bark, fruit	*Helicobacter pylori Staphylococcus aureus, Pseudomonas aeruginosa, , Streptococcus mitis,*	This study demonstrated that the leaf extract of P. guajava is effective in treating diseases caused by protozoa, fungi, bacteria, and viruses.	[8]
2	*PGL* / leaf	*Staphylococcus aureus*	In the study, a concentration of 5 mg/ml of the extract was effective against MDR bacteria within 10 hours.	[9]
3	*PGL* / leaf	*Enterococcus faecalis, Staphylococcus aureus), freundii, Escherichia coli* and *Pseudomonas sp*	Based on the findings, the PGL leaf extract exhibits excellent antioxidant activity, while the PGL essential oil contains the highest amount of polyphenols. Interestingly, a significant negative linear correlation exists between total polyphenol content (TPC) and antioxidant capacity, as evidenced by the Pearson correlation coefficients. Moreover, the PGL essential oil shows remarkable antibacterial and antifungal activity against all the bacteria and fungi tested in the study.	[10]
4	*PGL* / leaf	*Candida albicans* and *Streptococcus mutans*	The study found that PGLEO exhibited potent anticancer activity of 45.89% at 200 µg/mL, with an IC50 value of 188.98 µg/mL, which was significant ($p < 0.001$) when compared to doxorubicin (47.87%). Additionally, the results suggest that PGLEO has antimicrobial properties and holds promise as a potential anticancer agent.	[11]
5	*PGL* / leaves and bark	*Escherichia coli, Pseudomonas aeruginosa, Bacillus cereus, and Staphylococcus aureus.*	The study findings suggest that guava leaves and bark possess antibacterial properties and may have the potential as alternative treatment options in traditional herbal medicine.	[12]
6	*PGL* / leaves and bark	*V. cholerae*	The researchers determined that the crude aqueous mixture and water-soluble methanolic extract had a minimum inhibitory concentration of 1250 µg/mL and 850 µg/mL, respectively, against 107 CFU/mL V. cholerae in laboratory conditions.	[13]
7	*PGL* / leaf	*Escherichia coli, Staphylococcus aureus* and *Bacillus subtilis*	According to the findings, it was observed that the hydroalcoholic extracts of psidium guava leaves, administered at a dosage of 400 mg/kg, exhibited notable anti-ulcer properties in comparison to the standard medication.	[14]
8	*PGL* / leaf	*S.aureaus* and *E.Coli*	The results show that the plant extract alone can have antimicrobial activity experimentally.	[15]
9	*PGL* / leaf	*Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Escherichia coli,*	Aqueous and ethanol extracts of *PGL* have been suggested as a potential alternative to common antibiotics.	[16]

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10	*PGL* / leaf	*Staphylococcus aureus, Bacillus cereus Salmonella typhi* and *Escherichia coli*	The study found that the antimicrobial activity of guava's methanol extract was the highest against Bacillus cereus, while it was the highest against Salmonella typhi in the ethanolic extract. On the other hand, the lowest antimicrobial activity was observed against Staphylococcus aureus in the ethanolic extract.	[17]
11	*PGL* / leaf	S. sanguinis, S. mitis and Actinomyces sp	The results of both extracts were promising as they exhibited positive anti-adhesion activity and decreased the hydrophobicity of the bacteria's cell surface. This could potentially lead to a reduction in their adhesion to the tooth surface during the initial stages of plaque formation. Additionally, both extracts were found to have the ability to suppress the growth of these bacteria.	11[18]

Table 3. Articles included in the study with the effect of guava leaf extract on *H. pylori.*

Row	plant/plants	The part to be tested	Method Extraction	Results	Ref.
1	*PGL*	leaf	Ethanol	The inhibitory effect of the PGL extract was evaluated, and the results indicated that PGL had significantly higher effective concentrations (125 µg/mg) and MIC90 (26.6) compared to clarithromycin (1.95 µg/mg) and MIC90 (0.7) against *H. pylori*.	[19]
2	32 types of plants of which PGL was a part.	leaf	chloroform, Petroleum ether, and methanol	The antimicrobial activity of 32 medicinal plants in Malaysia against *Helicobacter pylori* was evaluated using disk diffusion and agar dilution methods. *Psidium guajava* showed significant antibacterial activity.	[20]
3	50 types of plants of which PGL was a part.	Leaf/stem/root/flower (*PGL*(	Ethanol	Based on this research, PGL was classified as moderate against *Helicobacter pylori* among 26 plants tested.	[21]
4	Two types of plants: Terminalia catappa and *PGL*	leaf	Ethanol	The study confirms that PGL and T. catappa leaves have a strong antioxidant potential.	[2]
5	16 types of plants of which *PGL* was a part	leaf	Ethanol	As per the study, most of the tested medicinal plant extracts were found to have cytotoxicity, with PGL being classified as moderate against *H. pylori*.	[22]
6	*PGL* and Chinese Coptis	leaf	Hydroalcoholic	Both extracts obtained from the early hours of incubation showed the ability to inhibit the growth of *H. pylori* strains in culture.	[23]
7	6 types of plants, of which *PGL* was a part	leaf/bark	Ethanol	In addition to its inhibitory effect, the extract of PGL contained the highest amount of polyphenols and effectively inhibited tumor necrosis factor, interleukin-1, and interleukin-6 at both mRNA and protein levels. The extract also exhibited high antioxidant potential, making it a promising candidate for treating gastric ulcers caused by *H. pylori*.	[24]
8	12 types of plants, of	leaf	Hydroalcoholic	After phytochemical screening, the multi-plant suspension showed the presence of terpenoids,	[25]

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	which *PGL* was a part			flavonoids, and tannins that have anti-ulcer activity. The anti-ulcer activity in the model induced by ethanol showed a significant decrease of P<0.05 from the wound.	
9	PGL	leaf	chloroform	The results indicate that phytochemicals in guava leaf extract cause apoptosis in SNU-16 cells, suggesting potential for new strategies in treating human gastric cancer.	[26]

Table 4. Biological activity of some *PGL* compounds

Plant components/ country	Active chemical compounds	Biological activity	Ref.
guava peel, flesh, and seed /China	catechin, galangin, homogentisic acid, gallic acid, kaempferol and cyanidin 3-glucoside	Anticancer and Antioxidant	[29]
Leaf / Taiwan	Ferulic acid, gallic acid, quercetin	Anticoagulant, anti-inflammatory, anti-glucose	[30]
Leaf / India	Main compounds α-terpinyl acetate, trans-caryophyllene, nerolidol, α-cadinol, α-copene and identified minor components of α-humulene.	Pain reliever, anti-inflammatory, antibacterial	[28]
Stem and leaf / Tunisia	The main compounds identified in the stem oil were alpha-humulene, germacrene D, and valenrol, whereas vein florol and trans-caryophyllene were predominant in the leaf oil.	Anti-inflammatory, anti-spasmodic	[31]
Leaf/Egypt	b- Caryophyllene, transnerolidol, global and d-limonene.	Antimicrobial, antidiarrheal, antipyretic, antihypertensive	[32]
Leaf / Thailand	The most active quercetin compound and two flavonoid compounds, quercetin-3-O-glucopyranoside and murine, were found.	Antioxidant	[33]
Leaf / Oman	Most of the leaf oil contains caryophyllene, veiniflorene, farnesin, and limonene.	Antimicrobial, cytotoxic	[34]
Leaf / Korea	The research reveals that guava leaves contain around 60 plant compounds, including phytol, β-eodesmol, α-copene, …. These compounds have the potential to disrupt various signaling pathways associated with the formation of tumors, making them promising candidates for developing cancer prevention and treatment therapies.	Anticancer	[35]

chemical composition of PGL varies by geographic region (seasonal changes and climatic conditions) [28]. Findings show that guava leaves contain many compounds, including phytol, beta-desmol, α-copene, etc. These compounds can potentially disrupt various signaling pathways associated with tumorigenesis, making it a promising candidate for cancer prevention and treatment.

DISCUSSION

One of the most common and critical causes of death in modern society is gastrointestinal cancer, one of the main causes of which is *H. pylori* infection [36]. As previously stated, *H. pylori* is considered to be a primary risk factor for the development of gastric cancer [37]. Antibiotics have been very effective in managing microbial infections. However, they are

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expensive and have many side effects. In addition, the threat of antimicrobial resistance has led to the inactivity of various conventional antibiotics. Medicinal plants are used in traditional medicine to control microbial infections since they are affordable and have shown fewer side effects [38]. PGL, commonly known as guava, contains tannins, phenols, saponins, vitamins, fibers, flavonoids, essential oils, fatty acids, etc [39]. Many pharmacological studies with PGL have shown that this plant has anticancer, antioxidant, antimicrobial, analgesic, and antitussive properties [40].

In a 2015 study comparing two plants PGL and Coptis chinensis Franch, for their effect on AGS human gastric cancer cells, the synergy of the mixture of these two plants can prevent chronic gastritis caused by *H. pylori* [23].

In a 2014 study, most plant extracts demonstrated cytotoxic effects. PGL was categorized as a moderate inhibitor against *H. pylori*, indicating that combining this plant with others possessing similar antibacterial properties could yield inhibitory effects. Nevertheless, further investigation is warranted [22].

Notably, most of the PGL extracts in the articles reviewed in the above tables contain phenolic compounds that could be involved in the antibacterial activities observed in this study, some showing antibacterial properties together. The amount of phenolic compounds inside the plant is [41]. In another article, the anti-disease activity of the methanol extract of guava was mentioned due to the percentage of avoided, especially murine glycosides, quercetin glycosides, and quercetin [42]. In this study, the example mentioned in Table No. 2 has been considered as one of the chemical compounds against *H. pylori*.

In many articles, the chemical composition of the obtained extract was of great importance, and in a way, the extract that contained that compound in question showed antibacterial and anti-inflammatory effects. PGL extracts contain chemical compounds of gallic acid and catechin, which may be related to the antimicrobial, anti-inflammatory, and analgesic activities of these extracts [41]. In a study in 2013, researchers investigated the antimicrobial effect of gallic acid and catechin on *H. pylori* culture. Both polyphenols demonstrated strong inhibitory effects on two strains of *H. pylori* [43]. Histochemical or chemical compounds of PGL leaf or fruit extract can treat gastric ulcers. This disease is caused by one of the important causes, *H. pylori* [44].

In a 2019 study, PGL leaves were found to contain high amounts of gallic acid, quercetin, and corilagin based on HPLC analysis. These compounds have demonstrated significant anti-*H. pylori* activities [19]. It is clear from the findings that this plant's main use is in treating digestive disorders, and it has also been used to treat several microbial diseases caused by protozoa, fungi, bacteria, and viruses. A significant gap is that more research should be done on different parts (leaves, stems, roots, fruits, etc) of plants native to Iran and can be used as medicine.

CONCLUSION

The high prevalence of *H. pylori* in the population must be considered, as it often goes undetected unless symptoms require diagnostic tests. This review serves as a good reference for those researching *H. pylori* and inspires further research to improve diagnostic and therapeutic methods to reduce the impact of this common gastrointestinal pathogen. The results of this study show that PGL plant extracts have appropriate antimicrobial substances that can be used as a pharmaceutical base or a suitable herbal medicine to fight against microorganisms such as *H. pylori*. It was found that the good antimicrobial activity of this plant against *H. pylori* is related to its phenolic compounds. However, more research is necessary for efficacy studies in animal models and determining the effective methods of *PGL* plant extracts (including urease inhibition) for *H. pylori* bacteria to access more comprehensive and practical information in the future.

ACKNOWLEDGMENTS

I appreciate and thank all my Zabol University of Medical Sciences colleagues who helped us in this research.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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[DOI: 10.61186/mci.8.1.5]

ETHICS APPROVAL

Not applicable.

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[DOI: 10.61186/mci.8.1.5]