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Original Article

Comparison between *Gardenia jasminoides* J. Ellis and *Celosia cristata* Linn. against *Aggregatibacter actinomycetemcomitans*: in vitro study

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ABSTRACT

Aggregatibacter actinomycetemcomitans (Aa) is one of the causes of periodontitis. Although severe periodontitis can be treated locally with chlorhexidine, prolonged usage of the medication can have negative consequences. Consequently, to take over for chlorhexidine, other options are required. The purpose of this study is to determine which plants have a stronger antibacterial effect on Aa as well as if *Gardenia jasminoides* J. Ellis (GJ) and *Celosia cristata* Linn. (CC) have any effect at all. GJ flowers (GJF) and CC flowers (CCF) were the materials utilised in this study which, together with a 96% ethanol solvent and the maceration process, is turned into a thick liquid extract. This study employed the microdilution and the agar dilution. Tukey's post-hoc analysis was used for multiple comparisons and one-way analysis of variance was used to examine differences across all data sets. 6.25% GJF extract (GJFE) and 25% CCF extract (CCFE) showed a drastic decrease in optical density, and colony forming units that did not form at all were observed in 12.5% GJFE and 25% CCFE. The antibacterial activity of GJ and CC against Aa is comparable to 2% chlorhexidine gluconate, notwithstanding this fact. With a minimum inhibitory concentration (MIC) of 6.25% (0.527 ± 0.013) and a minimum bactericidal concentration (MBC) of 12.5% (0.00 ± 0.000), GJ was the plant with the superior antibacterial action on Aa.

Keywords: *Gardenia jasminoides* J. Ellis, *Celosia cristata* Linn., *Aggregatibacter actinomycetemcomitans*, Microdilution, Agar dilution

Introduction

Asia is a continent that boasts large grasslands, abundant wildlife, distinct and varied ecosystems, and a range of ecological and evolutionary processes [1]. Making herbal medicines, known as herbal medicine in Indonesia, TCM in China, and Ayurvedic medicine in India, is one way that Asia's biodiversity and environment are put to use [2]. There is proof that two Asian herbal plants, *Gardenia jasminoides* J. Ellis (GJ) and *Celosia cristata* Linn. (CC), exhibit antibacterial properties. It has been demonstrated that both plants possess antibacterial properties against the facultative aerobic gram-positive pathogenic bacteria *Staphylococcus aureus* [3, 4].

In the human oral cavity, facultative anaerobic gram-negative pathogenic bacteria called *Aggregatibacter actinomycetemcomitans* (Aa) are present [5]. Aa is one of the causes of periodontitis, particularly the 40.9% prevalence of generalised aggressive periodontitis and the 81.8% prevalence of localised aggressive periodontitis [6]. Local therapy is one of the more aggressive periodontitis therapies available. Chlorhexidine is the local treatment used for aggressive periodontitis; nevertheless, long-term use of chlorhexidine can result in xerostomia, subjective colour changes, numbness and pain in the mouth and tongue, and changes in taste [7]. Consequently, to take over for chlorhexidine, other options are required.

Access this article online	
Website: www.japer.in	**E-ISSN:** 2249-3379

How to cite this article: Widjaja R, Felicia J, Kurnia C, Rahmawati DY. Comparison between *Gardenia jasminoides* J. Ellis and *Celosia cristata* Linn. against *Aggregatibacter actinomycetemcomitans*: in vitro study. J Adv Pharm Educ Res. 2025;15(1):25-8. https://doi.org/10.51847/tVHfXK7wc3

The purpose of this study is to determine which plants have a stronger antibacterial effect on Aa as well as if GJ and CC have any effect at all.

Materials and Methods

The ConPhyMP writing guide was used to write this article. In 2023, this research was carried out.

Materials preparation

GJ flower (GJF) and CC flowers (CCF) were the research materials employed in this study. Harvested from the Petiga Flower Plantation in Bali, GJF are 2-3cm in size, green in colour, and a month old when they are planted in manure-enriched soil. Red-coloured, 7–10 cm in size, and two months old when harvested from Bina Usaha Flora in Cianjur, CCF are planted in manure-enriched soil. The National Research and Innovation Agency in Indonesia recognised both plants.

Extracts preparation

Before preparing the extract, each ingredient is kept out of the damp and covered from the sun until it dries. A thick liquid extract was obtained by evaporating 96% ethanol solvent (EMSURE, Germany) at a ratio of 1:5 using the maceration method for three days at 37°C. The extract was homogenised every day and evaporated for three hours using a rotary evaporator (DLAB, China). To determine the content of each extract, tests were conducted on the secondary metabolite chemicals.

Antibacterial test

Aa was cultured in brain-heart infusion broth (OXOID, England) at 0.5 McFarland standard, or 1.5x108 CFU/ml, before the antibacterial test. In this study, the antibacterial test methods employed were distilled water (ONEMED, Indonesia) as the negative control and 2% chlorhexidine gluconate (ONEMED, Indonesia) as the positive control. Microdilution was used to determine the minimum inhibitory concentration (MIC) and agar dilution to determine the minimum bactericidal concentration (MBC). A spectrophotometer (THERMOFISHER, America) is used to detect the minimal concentration with a sharply falling optical density value, which is used to identify the MIC, and a value of zero colonies is used to determine the MBC.

Data analysis

Tukey's post-hoc analysis was used for multiple comparisons and one-way analysis of variance was used for hypothesis testing on all the data.

Results and Discussion

Numerous chemicals, including flavonoids, saponins, tannins, and steroids, were discovered in several of the extracts used in this study based on qualitative phytochemical testing. **Table 1** shows the qualitative phytochemical data.

Table 1. Qualitative phytochemical results

	GJFE	CCFE
Alkaloid	–	–
Flavonoid	+++	+
Saponin	+	+
Tannin	++	++
Steroid	–	+
Triterpenoid	+	–

One-way analysis of variance was used to statistically analyse all of the data; the findings are shown in **Table 2**. The null hypothesis was rejected based on hypothesis testing using one-way analysis of variance, indicating that GJF extract (GJFE) and CCF extract (CCFE) exhibit antibacterial effects on Aa.

Table 2. Data analysis results using one-way ANOVA

	GJFE	CCFE
p value	<0.001*	<0.001*

The optical density of the 25% CCF extract (0.531 ± 0.007) and the 6.25% GJFE (0.527 ± 0.013) significantly decreased based on microdilution. A significant difference ($p<0.05$) was observed between GJFE and CCFE at concentrations ranging from 50% to 3.125%, according to post-hoc Tukey data analysis. Agar dilution revealed that in 12.5% GJFE (0.00 ± 0.000) and 25% CCFE (0.00 ± 0.000), no colonies grew at all. A significant difference ($p<0.05$) was observed between GJFE and CCFE at concentrations of 6.25% and 3.125%, according to post-hoc Tukey data analysis. **Table 3** displays the overall microdilution and agar dilution data.

Table 3. MIC and MBC in mean ± standard deviation

	MIC	MBC
PC	0.063 ± 0.004*	0.00 ± 0.000*
NC	0.951 ± 0.005	171.00 ± 3.464
GJFE 100%	0.167 ± 0.008	0.00 ± 0.000*
GJFE 50%	0.237 ± 0.004‡	0.00 ± 0.000*
GJFE 25%	0.332 ± 0.012‡	0.00 ± 0.000*
GJFE 12.5%	0.438 ± 0.008‡	0.00 ± 0.000*
GJFE 6.25%	0.527 ± 0.013‡	10.33 ± 1.856‡
GJFE 3.125%	0.863 ± 0.006‡	31.00 ± 1.528‡
CCFE 100%	0.186 ± 0.005	0.00 ± 0.000*
CCFE 50%	0.361 ± 0.008	0.00 ± 0.000*
CCFE 25%	0.531 ± 0.007	0.00 ± 0.000*

CCFE 12.5%	0.718 ± 0.007	12.00 ± 1.155
CCFE 6.25%	0.867 ± 0.007	44.00 ± 4.041
CCFE 3.125%	0.928 ± 0.007	80.33 ± 1.407

*for significant results ($p<0.05$) when compared to every group
‡for significant results ($p<0.05$) when compared to CCFE

Research has shown that, in comparison to CCFE, GJFE is more effective at inhibiting and killing Aa. The GJFE may be the source of this phenomena due to its higher flavonoid and triterpenoid content. The antibacterial properties of flavonoids, saponins, tannins, and steroids, among other secondary metabolite chemicals, have been assessed in several prior research against facultative anaerobic gram-negative bacteria.

Plants, fruit, vegetables, and leaves contain chemicals called flavonoids, which are secondary metabolites that have the potential to be used in the pharmaceutical chemical industry for a variety of purposes, including antibacterial applications [8]. Flavonoids were detected in every extract examined in this investigation. Studies comparing flavonoids to facultative anaerobic gram-negative bacteria have shown that they can prevent the growth of bacterial biofilms [9]. Complex bacterial colonies in an exopolysaccharide matrix adhered to the surface of organisms that cause nosocomial and persistent infections in clinical settings are known as bacterial biofilms [10]. The ultimate state of bacterial death can be brought by inhibiting the production of bacterial biofilms [11].

Plants have a broad family of triterpenoid glycosides and amphiphilic steroids called saponins [12]. All of the examined extracts in this research contain saponins. Saponins work against bacteria by creating pores and damaging bacterial cytoplasmic and protein membranes, which allows intracellular electrolyte leakage. This process is nearly identical to alkaloids [13, 14].

A complex phenolic molecule, tannin may be found in chocolate, berries, plants, and other culinary ingredients [15]. Every extract that was examined in this research included tannin. Tannin can inhibit bacterial cell wall formation by either binding directly to bacterial cell walls or by inactivating enzymes involved in bacterial cell wall synthesis, according to study testing it against facultative anaerobic gram-negative bacteria [16]. Because they offer protective permeability and make a significant structural contribution, bacterial cell walls are vital to the survival of bacteria [17].

Steroids are special substances that are present in practically all plants and have a significant physiological impact on the growth, development, and reproduction of plants [18]. Steroids were only discovered in CCFE in this research. The biological action of hydrophobic steroids can inhibit and kill bacteria because of their ability to interact with bacterial cell membranes [19]. Triterpenoids are biologically active substances that have a variety of biological roles. One of these activities is their ability to protect plants against biotic stress. Steroids were only discovered in GJFE in this investigation [20]. Triterpenoids can affect the structure of peptidoglycans, gene expression, the development of bacterial biofilms, and they can also trigger a stress response in bacteria that can either inhibit or kill the bacteria [21].

Conclusion

The antibacterial activity of GJ and CC against Aa is comparable to 2% chlorhexidine gluconate, notwithstanding this fact. With a MIC of 6.25% (0.527 ± 0.013) and a MBC of 12.5% (0.00 ± 0.000), GJ was the plant with the superior antibacterial action on Aa.

Acknowledgments: Faculty of Dentistry, Maranatha Christian University, Bandung.

Conflict of interest: None

Financial support: None

Ethics statement: None

References

1. Dhyani S, Sivadas D, Basu O, Karki M. Ecosystem health and risk assessments for high conservation value mountain ecosystems of South Asia: a necessity to guide conservation policies. Anthr Sci. 2022;1(1):211-25.
2. Liana D, Phanumartwiwath A. Leveraging knowledge of Asian herbal medicine and its active compounds as COVID-19 treatment and prevention. J Nat Med. 2022;76(1):20-37.
3. Khalil MAJ, Mansoor RF, Aboud AS, Jazar ZH, Zboon HA. Antibacterial activity of Gardenia jasminoides Ellis leaves extracts on bacteria isolated from burns infected. Int J Sci Res Arch. 2023;9(2):935-40.
4. Sing M, Kumar PS, Shrivastava B, Reddy BP. A comprehensive review of phytochemical and pharmacological overview on celosia cristata for future prospective research. Asian J Pharm Clin Res. 2020;13(12):21-4.
5. Tristano J, Danforth DR, Wargo MJ, Mintz KP. Regulation of adhesin synthesis in aggregatibacter actinomycetemcomitans. Mol Oral Microbiol. 2023;38(3):237-50.
6. Mahalakshmi K, Krishnan P, Chandrasekaran SC. Fimbria-associated protein gene genotypes among periodontitis. Dent Res J. 2018;15(3):185.
7. Poppolo Deus F, Ouanounou A. Chlorhexidine in dentistry: pharmacology, uses, and adverse effects. Int Dent J. 2022;72(3):269-77.
8. Ullah A, Munir S, Badshah SL, Khan N, Ghani L, Poulson BG, et al. Important flavonoids and their role as a therapeutic agent. Molecules. 2020;25(22):5243.

9. Górniak I, Bartoszewski R, Króliczewski J. Comprehensive review of antimicrobial activities of plant flavonoids. Phytochem Rev. 2019;18(1):241-72.
10. Sharma S, Mohler J, Mahajan SD, Schwartz SA, Bruggemann L, Aalinkeel R. Microbial biofilm: a review on formation, infection, antibiotic resistance, control measures, and innovative treatment. Microorganisms. 2023;11(6):1614.
11. Srinivasan R, Santhakumari S, Poonguzhali P, Geetha M, Dyavaiah M, Xiangmin L. Bacterial biofilm inhibition: a focused review on recent therapeutic strategies for combating the biofilm mediated infections. Front Microbiol. 2021;12:676458.
12. Xu P, Yu B. Chemical synthesis of saponins: an update. Adv Carbohydr Chem Biochem. 2021;79:1-62.
13. Amaha ND, Mebrahtu SG, Abdu N. Saponins and their synergistic antibacterial activity with traditional antibiotics against staphylococcus aureus and escherichia coli: review. Qeios. 2022. Available from: https://www.qeios.com/read/YO91ZE
14. Alina P, Kiełb Ewa P, František Z. Antimicrobial activity of saponin-containing plants: review. JDVAR. 2023;12(2):121-7.
15. Singh AK, Kim JY, Lee YS. Phenolic compounds in active packaging and edible films/Coatings: natural bioactive molecules and novel packaging ingredients. Molecules. 2022;27(21):7513.
16. Farha AK, Yang QQ, Kim G, Li HB, Zhu F, Liu HY, et al. Tannins as an alternative to antibiotics. Food Biosci. 2020;38:100751.
17. Zhou J, Cai Y, Liu Y, An H, Deng K, Ashraf MA, et al. Breaking down the cell wall: still an attractive antibacterial strategy. Front Microbiol. 2022;13:952633.
18. Obakan Yerlikaya P, Arısan E, Mehdizadehtapeh L, Uysal Onganer P, Çoker Gürkan A. The use of plant steroids in viral disease treatments: current status and future perspectives. Eur J Biol. 2023;82(1):86-94.
19. Nadaraia NS, Amiranashvili LS, Merlani M, Kakhabrishvili ML, Barbakadze NN, Geronikaki A, et al. Novel antimicrobial agents' discovery among the steroid derivatives. Steroids. 2019;144:52-65.
20. Yang C, Halitschke R, O'Connor SE. OXIDOSQUALENE CYCLASE 1 and 2 influence triterpene biosynthesis and defense in Nicotiana attenuata. Plant Physiol. 2024;194(4):2580-99.
21. Wroński N, Szlaur M, Zawadzka K, Lisowska K. The synergistic effect of triterpenoids and flavonoids-New approaches for treating bacterial infections? Molecules. 2022;27(3):847.