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Antibacterial And Antibiofilm Activity of *Escherichia coli* From Plants Containing Flavonoids - A Mini-Review

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Abstract: Biofilms are associated with infections in humans which have significant reproduction rates due to their resistance to infection. Traditional medicines are generally considered more comfortable because they have relatively fewer side effects than modern medicines. This study aims to determine the antibiofilm and antibacterial potential of plants containing flavonoids against *Escherichia coli* bacteria. This research is a literature review. A literature review is used to collect data obtained from journals, the internet, and other libraries. After conducting a literature review, the results are that plants containing flavonoids can be antibacterial and antibiofilm against *Escherichia coli* bacteria. Based on the results obtained after conducting a literature review, it is stated that flavonoids have the activity of inhibiting and destroying biofilms.

Keywords: Biofilm; Antibacterial; Flavonoid; *Escherichia coli*

1. Introduction

Biofilms are a collection of cells of microorganisms, mainly bacteria, that stick tightly to a surface accompanied by organic materials and are enveloped by an extracellular polymer matrix released by bacteria [1–3]. Plaque control is the removal of plaque, the microbiome, and prevention of its accumulation to the surface of the teeth and surrounding areas [4,5]. Plaque control also limits the preparation of calculus. Plaque can be removed by mechanical cleaning and chemical inhibition. Mechanical cleaning can be tried by brushing teeth. Instead, chemically can be tried with mouthwash. One of the purposes of brushing teeth is to limit the development of plaque bacteria [6,7]. The use of folk remedies, in general, is considered more convenient because it has relatively fewer side effects than modern medicine [8,9]. The preparation of biofilms begins on the abiotic surface in the presence of an electrostatic force between the substratum and the bacterial surface intermediated by the presence of adhesin forces [10,11]. At the same time, the attachment of bacteria on the biotic surface is intermediated by the relationship of bacterial proteins with extracellular proteins or carbohydrates on the surface of tissues. It relies on the hydrophobicity of the cell membrane [12,13]. *Escherichia coli* is a gram-negative, unspayed stem germ in the form of the pediatric flagellum, ± 1.1–1, 5 μm x 0, 2– 0, 6 μm in diameter. *Escherichia coli can* survive in a medium that easily creates gases and acids from glucose and ferments lactose [14,15].

The movement of these germs is motile, not motile, and in pediatrics, there are aerobic

and anaerobic facultative [16]. Flavonoids are secondary metabolites of polyphenols, are found widely in plants and foods, and have various bioactive effects, including anti-viral, anti-inflammatory, cardioprotective, antidiabetic, anticancer, anti-aging, antioxidant, and others [17–19]. Flavonoid compounds are polyphenol compounds with 15 carbon atoms arranged in the configuration of C6-C3-C6. The carbon skeleton consists of two C6 groups (substituted benzene rings) connected by a three-carbon aliphatic chain. Flavonoids are found in plants, which produce melamine patterned yellow, red, orange, blue, and purple from fruits, flowers, and leaves [20,21]. Flavonoids are listed in the water-soluble polyphenol family [22–24]. Bioactive flavonoids are considered phytochemicals, especially in food, which have biological properties for humans at large [22,25–27]. Microbial biotransformation methods to create flavonoids are important because they create new flavonoids that are not present in nature. The primary responses throughout microbial biotransformation are hydroxylation, dehydroxylation, O-methylation, O-demethylation, glycosylation, deglycosylation, dehydrogenation, hydrogenation, cyclization, and carbonyl reduction. *Cunninghamella*, *Penicillium*, and *Strain Aspergillus* are well known for flavonoid biotransformation, and they can perform almost any response with excellent results. Structural biofilms are microcolonies, planktonic cells of germs found in the Extracellular Polymeric Substance (EPS) matrix [28,29]. Structural biofilms consist of one or more categories of germs, depending on the type of germ. Biofilms are composed of 10-25% germ cells and an EPS matrix of 79- 90%. The EPS matrix protects germ cells from poor area conditions, such as UV radiation, turnover of PH values, and drying. The general composition of EPS is polysaccharides, proteins, nucleic acids, lipids, phospholipids, and hazmat substances. The EPS molecular matrix is required for communication between quorum sensing cells [11,30,31]. Quorum sensing is a process that works for germs to speak by secreting the signal molecules spoken by autoinducers [32]. primary mediators of infection, with an estimated 80% of the incidence of infection related to the formation [33].

2. Antibacterial And Antibiofilm Activity from Plants

The research results were conducted by reviewing the literature on the effects of flavonoids on antibacterial and antibiofilm with a narrative review approach.

Table 1. Reviewing the literature.

Plant species	Research Methods	Results and Conclusions
Gigantochloaapus	The NA (Sodium agar) disc paper diffusion method is used in this study. The presence of antibacterials can be seen in minimum levels of bland (KHM) formed.	The most effective extract of bamboo shoots as antibacterial *Escherichia coli* and *Staphylococcus Aureus* is a concentration of 150 mg/ml [34].
Citrofortunella microcarpa	The disc paper diffusion method is a method for testing antibacterial activity, antibacterial activity indicated by the presence of the diameter of the bland zone formed.	Kalamansi orange leaf extract (*C.microcarpa*) affects the growth of *S. aureus* and *E. coli* bacteria. This is seen from significant values (P<0.01). The antibacterial ability of kalamansi orange leaf ethanol extract against both bacteria is moderate, with a bland zone diameter of 7.20 and 5.73 mm at a concentration of 40% [35].
Samanea saman	Extraction is done by the method of maceration and partition. The separation of compounds is done by column chromatography, while antibacterial activity tests with disc diffusion methods and identification are performed with Ultraviolet-visible (UV-vis) and Infrared	Flavonoid compounds in the n-butanol extract of trembesi leaves have antibacterial activity against *E.coli* bacteria [36].

Plant species	Research Methods	Results and Conclusions
	spectrophotometers.	
Syzygium aromaticum	The research began with extraction using maceration techniques, then separated compounds based on polarity levels through fractionation of ethanol, n-hexane, and ethyl acetate. The results of fractionation are tested to the test bacteria. Identification of secondary metabolite compounds of clove leaves using Gas Chromatography-Mass Spectrometry (GC-MS).	The results showed that the ethyl acetate fraction of clove leaves is the most potent antibacterial fraction against *E. coli* and *S. aureus* with a KHM value of 10% and a bland zone of ±17mm through an inhibitory mechanism of action suspected of making holes in bacterial cell membranes [37].
Gnetum gnemon	This study used well diffusion methods with concentrations of 10% b/v, 20% b/v, 30% b/v, 40% b/v, 50% b/v, 60% b/v, 70% b/v, 80% b/v, 90% b/v, and 100% b/v	The observations showed that ethanol extract of melinjo leaves (*Gnetum gnemon L.*) has activity and effectiveness against *Escherichia coli* bacteria [38].
Mangifera indica	This study used maceration extraction methods and then isolated ethanol extract to obtain flavonoid isolates using the liquid partition method. Testing of antibacterial activity is carried out by the paper disc diffusion method	antibacterials show the presence of antibacterial barriers. Flavonoid isolates have a more potent inhibitor than mango leaf ethanol extract [8].
Holothuria scabra	Experimental laboratories with Complete Random Design (RAL).	The study of bioactivity tests of sea cucumber extract killing *H. atra* as antibacterial *S. aureus* and are as follows: The best solvent for extracting bioactive compounds in sea cucumber *H. atra* is ethanol, based on the results of phytochemical tests, namely flavonoids by 0.65%, phenols by 0.42%, saponins by 0.30%, and alkaloids by 0.27%. The difference in the concentration of sea cucumber extract *H. atra* has a real influence on the diameter value of the habitat zone. The higher the concentration of extracts, the greater the diameter of the bland zone formed. Sea cucumber killing (*H. atra*) has weak antibacterial abilities at concentrations of 2.5% and 5% and is moderate at a concentration of 7.5% [39].
Zingiber officinale Roscoe	This study includes an experimental laboratory study with a complete random design (RAL) consisting of 7 treatments with three repeats.	The study results obtained that ginger rhizome ethanol extract has antibacterial activity against *S. aureus* and *E. coli*. At concentrations of 20%, 40%, and 80% are more effective in inhibiting the growth of both bacteria. The higher the concentration level of the extract, the diameter of the bland zone of bacterial growth is also greater. Inhibition of bacterial growth is suspected to influence the content of ginger rhizomes [40].
Nymphaeae alba	The Kirby-Bauer disc diffusion method was used in this study.	Antibacterial tests have shown that extracts of white lotus leaves, stems, and rhizomes have the highest attack value against bacteria, while the diameter of the habitat zone is 20 mm each against *S. aureus* bacteria. Extracts of acetone and ethyl acetate of white lotus leaves, stems, and rhizomes have no inhibition against *E. coli* bacteria [41].
Citrus	The total mass of the biofilm was measured by washing dishes with phosphate buffer (0Æ1 mol l)1, pH 7Æ4) and staining with 3% crystal violet (Fisher, Hanover Park, IL, USA) for 20 minutes, with a Bacterial Culture of E. coli O157. : H7 strains ATCC 43895 and *V. harveyi* BB120 which were diluted with new CFA or LM media. The diluted cultures were placed on 96-well microtiter plates. All wells received either	The results showed the potential modulation of bacterial cell communication, *E. coli* biofilm O157:H7, and virulence *V. harveyi*, by flavonoids, especially naringenin, quercetin, synergetic, and apigenin. Among flavonoids tested, naringenin appeared as valuable as an antibiofilm [20].

Plant species	Research Methods	Results and Conclusions
	0.5% DMSO (control) or the test compound dissolved in DMSO. The plates were monopolized at 26°C for 24 h without shaking.	
Carica papaya L.	Using the Microtiter Plate Biofilm Assay method	Papaya leaf water extract had the highest activity in biofilm degrading by 48.99%. After optimization using the Response Surface method in the best conditions in biofilm degradation by papaya leaf extract (> 52.5%), which is at a temperature condition of 35ºC- 40ºC, the contact time is 57-60 minutes while the concentration is between 75-100% (v / v) [42].
Citrus limon	Biofilms are planted in broth-immersed acrylic discs, inoculated with microbial suspension (106 cells/mL), and incrusted at 37°C/48 hours.	Limonene was tested separately for antimicrobial activity and confirmed bactericidal activity against gram-positive and gram-negative bacteria, including *E. coli* [43].
Vitis vinifera	Orange seed extract (GSE; ES Food, Korea) is dissolved in TSB with 0.05% Tween 80. The double broth dilution method is used to determine the MIC GSE in the 96-hole plate.	These results showed that GSE had anti-biofilm effects on both gram-positive and gram-negative bacteria by reducing EPS production and motility in *S. aureus* and *E. coli* [44].
Camellia sinensis	Cultures are treated with EGCG-S and antibiotics only, and different formulations are followed up with incubation at 37 C for four days. The liquid is removed, the biofilm (if any) is washed and then stained with 0.1% violet crystal (CV) for 24 hours. After tense aspiration and washing 1 PBS final, the biofilm is dried for 24 hours.	Our results identified the optimal formulation for each bacterium, effectively inhibiting the formation of biofilms 95–99%. Analysis of colony-forming units (CFU) and cell viability showed a decrease in live bacteria. These results illustrate egcg-S's potential synergistic agent with antibiotics and an anti-biofilm agent [45].
Leptospermum scoparium	Mh's inhibitory effects on the planktonic cell growth of *E. coli* O157:H7 were determined. In short, *E. coli* O157:H7 is incensed with or without MH (0.1 and 0.2 g/mL) for 1, 3, 6, 12, and 24 hours. After incubation, bacterial growth is measured at a wavelength of 595 nm.	Collectively, the study demonstrates the potential anti-biofilm properties of MH that can be applied to control *Escherichia coli* O157:H7 [46]
Allium cepa L. var. Aggregatum	Testing of the tissue culture plate (TCP) method (also called the quantitative microtiter plate semi test (biofilm test)	Bacterial isolates (a total of nine Gram-negative isolates and five Gram-positive isolates) were isolated from clinical samples. Diffusion tests for wells test the tissue culture plate (TCP) method and the ability of bacteria to attach to the mouth. Epithelial cells are used to evaluate the effect of onions on growth, motility, adhesion, and biofilm formation in desired bacterial species [47].
Malus domestica	Testing of static biofilm formation was conducted at the 96-well polystyrene plate (Fisher Scientific) as previously reported (31). Briefly, the overnight breeding is diluted to OD600 0.05 in LB (300 l) media containing phloretin or other compounds at 0, 5, 10, 25, 50, and 100 g/ml and inculcated for 10 hours without shuffling at 37 °C.	Pathogenic biofilms have been associated with persistent infections due to their high resistance to antimicrobial agents, while commensal biofilms often fortify the host's immune system. Hence, controlling biofilm formation of both pathogenic bacteria and commensal bacteria is important in bacterium-related diseases. We investigated the effect of plant flavonoids on biofilm formation of enterohemorrhagic *Escherichia coli* O157:H7. The antioxidant phloretin, which is abundant in apples, markedly reduced *E. coli* O157:H7 biofilm formation without affecting the growth of planktonic cells, while phloretin did not harm commensal *E. coli* K-12 biofilms. Also, phloretin reduced *E. coli* O157:H7 attachment to human colon epithelial cells. Global transcriptome analyses revealed that

Plant species	Research Methods	Results and Conclusions
		phloretin repressed toxin genes, autoinducer-2 importer genes, and dozens of prophage genes in *E. coli* O157:H7 biofilm cells. Electron [48].
Morus nigra L	The antibiofilm activity test consists of a biofilm attachment prevention test, a biofilm inhibition test, and a biofilm destruction test with the assay biofilm microtiter plate method.	Pathogenic biofilms have been associated with persistent infections due to their high resistance to antimicrobial agents, while commensal biofilms often fortify the host's immune system. Hence, controlling biofilm formation of both pathogenic and commensal bacteria is essential in bacterium-related diseases. We investigated the effect of plant flavonoids on biofilm formation of enterohemorrhagic *Escherichia coli* O157:H7. The antioxidant phloretin, which is abundant in apples, markedly reduced *E. coli* O157:H7 biofilm formation without affecting the growth of planktonic cells, while phloretin did not harm commensal *E. coli* K-12 biofilms. Also, phloretin reduced *E. coli* O157:H7 attachment to human colon epithelial cells. Global transcriptome analyses revealed that phloretin repressed toxin genes, autoinducer-2 importer genes, and dozens of prophage genes in\n *E. coli* O157:H7 biofilm cells. Electron microscopy confirmed that phloretin reduced fimbria production in *E. coli* O157:H7. Also, phloretin suppressed the tumor necrosis factor-alpha-induced inflammatory response in vitro using human colonic epithelial cells. Moreover, in the rat model of colitis induced by trinitrobenzene sulfonic acid (TNBS), phloretin significantly ameliorated colon inflammation and body weight loss. Taken together, our results suggest that the antioxidant phloretin also acts as an inhibitor of *E. coli* results show that flavonoids act as inhibitors of the formation of *E. coli* biofilms [49].

3. Conclusions

Sourced from the results obtained after conducting a literature review, reported that flavonoids prove the results of inhibiting effects, avoid attachment, and avoid germs and biofilms against *Escherichia coli* germs. Flavonoids work as antibacterials with several work mechanisms, including limiting the synthesis of nucleic acids, limiting the use of cytoplasmic membranes, and limiting the metabolism of energy from germs. Flavonoids work as antibiofilm by squeezing the manufacture of biofilms by the influence of quorum sensing mechanisms. Sources such as journals, the internet, and other libraries, this review literature research is conducted to provide information about plants that contain flavonoids efficacious as antibacterial and antibiofilm with *Escherichia coli* bacteria. Flavonoid has a bond with its activity as an antibacterial. The mechanism of flavonoids, such as quercetin, is primarily caused by inhibiting DNA gyrase. Sophoraflavone Gram and epigallocatechin gallate have been proposed to limit cytoplasmic membranes, while licochalcones A and C can limit energy metabolism. Flavonoid profiles can limit the expression of the genes lsrACDBF, csgA, csgB, which is the regulator of the manufacture of fimbria in *Escherichia coli* biofilms, resulting in impaired metabolism in the extracellular matrix and causing inhibition of nutrient transport.

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Conflicts of Interest

The authors declare no conflict of interest.

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