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Research Article

Enhancing the Antibacterial Efficacy of Dishwashing Detergents Using *Myrtus communis* L. Leaf Extract

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Abstract

Background: This study investigates the increase in the antibacterial activity of dishwashing detergents with the addition of the leaf extract of *Myrtus communis* L.
Methods: The screening of antimicrobial activity against *Enterococcus faecalis*, *Bacillus subtilis, Escherichia coli,* and *Klebsiella pneumoniae* was done by the Kirby-Bauer disk diffusion method.
Results: The results revealed that the extract of *M. communis* at 25 mg/ml exhibited a good inhibitory effect against all the species of bacteria tested, but the lowest concentration of detergent that showed a significant inhibitory effect was 0.78 mg/ml. The synergetic effect of 25 mg/ml of plant extract and 0.78 mg/ml detergent created the largest inhibition zones of all the tested bacteria. These results suggest that *M. communis* L. could develop into a natural product that would increase the antimicrobial effectiveness of dishwashing detergents as an alternative and effective method of preventing the growth of pathogenic bacteria as part of domestic and industrial hygiene management.

1. Introduction

Traditional medicine has been using plants since time immemorial to prevent and treat many diseases in the human body. Hundreds of years of folk knowledge of medicinal flora contributed to the discovery and advancement of modern pharmaceuticals. Natural products of plant origin have been revealed to be therapeutically useful and have been adopted into traditional medicine [1, 2].

Myrtus communis L. (myrtle) is an aromatic evergreen shrub in the family Myrtaceae. It grows in the Mediterranean region and is valued not only as a culinary, but also as an aromatic and medicinal plant. It was discovered that Myrtle leaves contain such bioactive compounds as essential oils, flavonoids, tannins, and phenolic acids that are known to possess antimicrobial, antioxidant, anti-inflammatory, and other pharmacological effects [3, 4]. Myrtucommulone, quercetin derivatives, and ellagic acid are also found in the leaves and are attributed to high antibacterial activity levels [5, 6].

Bacterial contamination at both domestic and industrial levels is a major problem for the health of the population. The most widespread pathogenic bacteria include *Escherichia coli, Klebsiella pneumoniae, Enterococcus faecalis*, and *Bacillus subtilis* and are usually associated with foodborne infections, nosocomial infections, and surface biofilm [7, 8]. Biofilms not only facilitate the survival of bacteria in unfavorable conditions but also increase their resistance to disinfectants and antibiotics, which complicates the management of infection occurrence.

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The dishwashing detergents are one of the most used cleaners in the kitchen and are used to clean the kitchen utensils and surfaces of grease, food remnants, and microbial contamination. They mostly consist of surfactants, builders, bleaching agents, and enzymes, and assist in cleaning and antimicrobial effect [9]. However, excessive use of synthetic detergents and incorrect use may lead to environmental hazards and even the choice of resistant bacteria to biocides [10, 11].

In a bid to solve these problems, the addition of natural antibacterial agents into the composition of detergents has become a trend as a long-term solution. Plant extracts and those with proven antimicrobial action can bring efficacy to detergents and reduce the application of high concentrations of chemical compounds. In this respect, the *M. communis* L. leaf extract could be considered a prospective natural additive as it is a broad-spectrum antibacterial agent with an environmentally friendly profile.

The objective of the present study is to test the antibacterial activity of *M. communis* L. leaf extract, commercial dishwashing detergent, and a combination of these two against some of the pathogenic bacteria. It also focuses on the potential synergism between the application of the plant extract and low concentration of detergent to achieve the optimal antibacterial activity, therefore, offering a safer and more eco-friendly means of addressing hygiene in homes and industries.

2. Materials and Methods

2.1. Plant material collection and preparation

In November 2023, fresh leaves of *Myrtus communis* L. were harvested in Al-Samawa City, Iraq. Faculty of Agriculture, Al-Muthanna University, taxonomically identified and authenticated the plant species. The leaves were cleaned twice with tap water to eliminate debris and shade-dried until it was about 80 percent dry, and then oven-dried at 50°C to finish the drying process. The dried leaves were pulverized into a fine powder using an electric blender and stored in airtight containers until extraction.

2.2. Extraction procedure

Fifty grams of dried leaf powder were mixed with 450 ml of ethanol (1:10 w/v) and incubated in a shaker at room temperature for 48 h. The mixture was filtered through Whatman No. 1 filter paper, and the filtrate was left at room temperature for solvent evaporation. The resulting crude extract was collected, weighed, and stored at 4°C until further use.

2.3. Preparation for test concentrations

Eight concentrations of *M. communis* L. leaf extract were prepared in distilled water: 100, 50, 25, 12.5, 6.25, 3.12, 1.56, and 0.78 mg/ml. The same concentration range was prepared for the commercial dishwashing detergent. For combination assays, 25 mg/ml of plant extract was mixed with 0.78 mg/ml of dishwashing detergent.

2.4. Bacterial strains and culture conditions

Bacterial isolates (*Bacillus subtilis, Klebsiella pneumoniae, Escherichia coli,* and *Enterococcus faecalis*) were obtained from the Department of Biology, College of Science, Al-Muthanna University. Strains were purified using selective chromogenic media: Bacillus chromogenic agar for *B. subtilis*, UTI Hichrome agar for *K. pneumoniae* and *E. coli*, and Hichrome Enterococcus agar base for *E. faecalis*. Pure colonies were maintained on nutrient agar slants at 4°C.

2.5. Antibacterial activity assay

The antibacterial activity of the plant extract, dishwashing detergent, and their combination was evaluated using the Kirby–Bauer disk diffusion method following CLSI (2020) guidelines. Overnight bacterial cultures were adjusted to a 0.5 McFarland standard in 0.9% sterile saline. Bacterial suspensions were inoculated on the Mueller-Hinton agar with a sterile cotton swab. The 6 mm filter paper discs were sterile and impregnated with 100 μ L of each concentration of the test and placed on the agar surface on which the test was inoculated. Discs loaded with sterile saline served as negative controls. Plates were incubated at 37°C for 24 h, and inhibition zones (mm) were measured. All experiments were performed in duplicate to ensure reproducibility.

3. Results and Discussion

The antibacterial activity of *Myrtus communis* L. leaf extract, commercial dishwashing detergent, and their combination against *Bacillus subtilis, Klebsiella pneumoniae, Escherichia coli,* and *Enterococcus faecalis* was evaluated using the Kirby–Bauer disk diffusion method.

3.1. Antibacterial activity of *M. communis* L. leaf extract

As shown in Table 1, high concentrations of *M. communis* leaf extract (100, 50, and 25 mg/ml) exhibited strong inhibitory effects against all tested bacterial species, with inhibition zones ranging from 16 to 25 mm. The concentration of 25 mg/ml was identified as the lowest uniform concentration that inhibited all bacterial species (*B. subtilis* – 16 mm, *K. pneumoniae* – 20 mm, *E. coli* – 18 mm, *E. faecalis* – 20 mm).

Lower concentrations (12.5 mg/ml and below) showed inconsistent antibacterial effects, inhibiting only certain species, which confirms the concentration-dependent activity of the extract. These findings align with previous reports on the broad-spectrum antibacterial potential of *M. communis*, largely attributed to its phenolic compounds, flavonoids, and essential oils [3, 5] Figure 1.

Table 1: Antibacterial activity of *Myrtus communis* L. leaf extract at different concentrations against pathogenic bacteria based on inhibition zone diameters (mm)

Concentration (mg/ml)	B. subtilis	K. pneumoniae	E. coli	E. faecalis
100	19	22	18	25
50	19	21	16	22
25	16	20	18	20
12.5	7	12	9	10
6.25	0	0	9	0
3.12	0	0	0	9
1.56	0	0	0	7
0.78	0	0	0	0

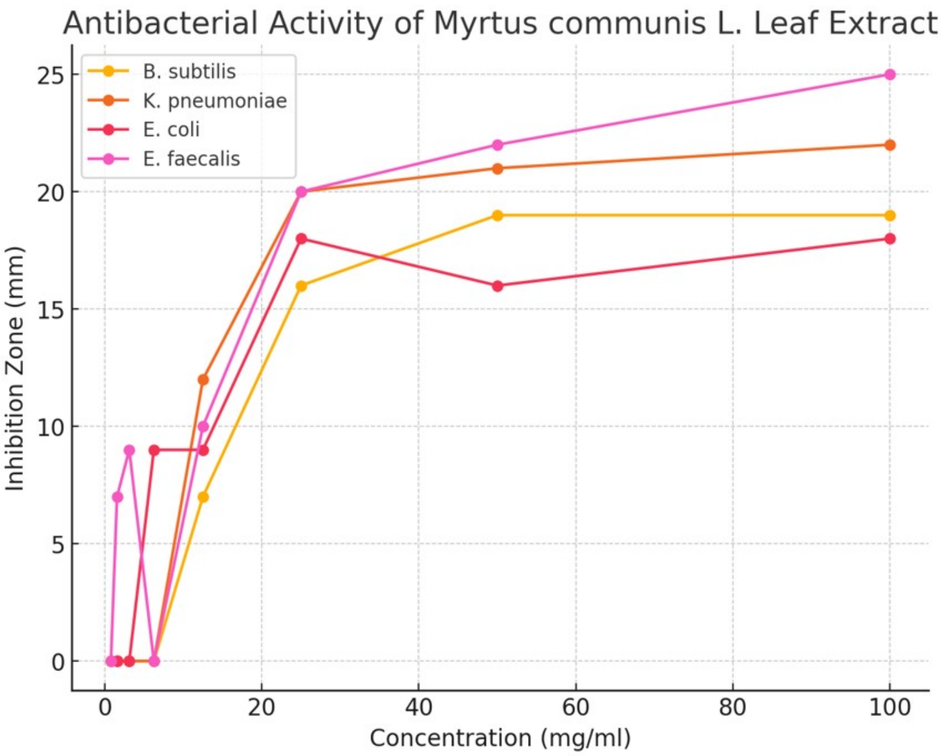

Figure 1: Inhibition zone diameters (mm) of *Myrtus communis* L. leaf extract at varying concentrations against four pathogenic bacterial species

3.2. Antibacterial activity of dishwashing detergent

Results in Table 2 indicate that the dishwashing detergent demonstrated measurable antibacterial activity at all tested concentrations (100–0.78 mg/ml). Even at the lowest concentration of 0.78 mg/ml, significant inhibition was observed (*B. subtilis* – 18 mm, *K. pneumoniae* – 25 mm, *E. coli* – 18 mm, *E. faecalis* – 23 mm). This effect is likely due to surfactants and other active ingredients that disrupt bacterial membranes and denature proteins [12]. Nevertheless, exclusive reliance on synthetic detergents can lead to environmental concerns and the emergence of microbial resistance [10] Figure 2.

Table 2: Antibacterial activity of dishwashing detergent at different concentrations against pathogenic bacteria based on inhibition zone diameters (mm)

Concentration (mg/ml)	B. subtilis	K. pneumoniae	E. coli	E. faecalis
100	31	34	31	38
50	28	29	31	35
25	25	29	27	33
12.5	24	27	22	33
6.25	24	25	20	31
3.12	20	26	19	26
1.56	19	26	18	25
0.78	18	25	18	23

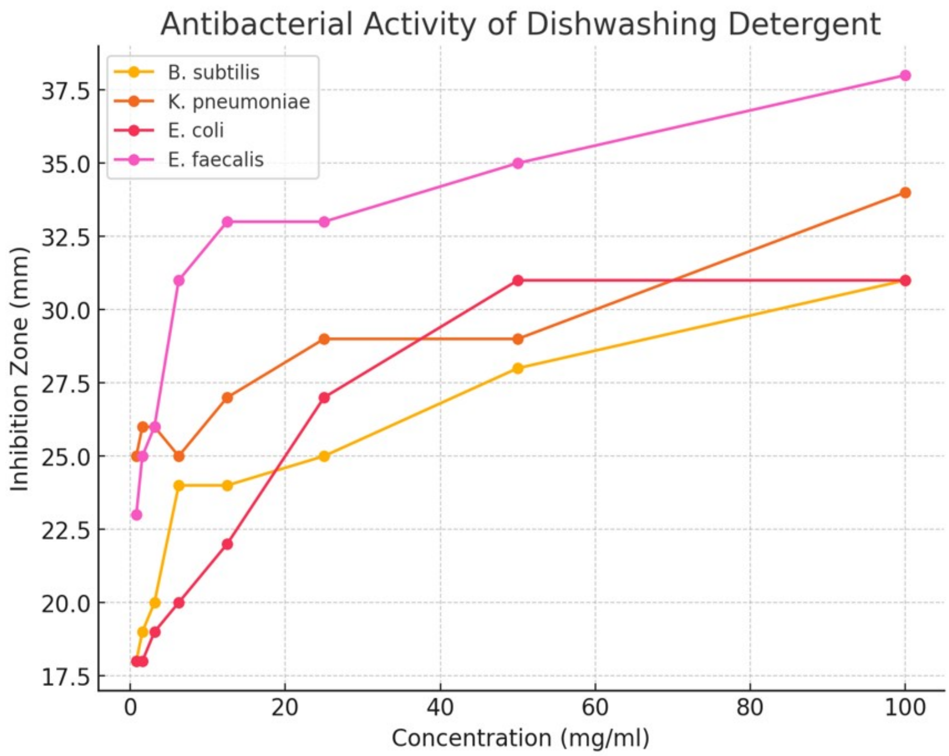

Figure 2: Inhibition zone diameters (mm) of dishwashing detergent at varying concentrations against four pathogenic bacterial species

3.3. Synergistic effect of combining plant extract and detergent

The combination of 25 mg/ml *M. communis* extract with 0.78 mg/ml detergent, as presented in Table 3, resulted in the highest inhibition zones recorded across all treatments (*B. subtilis* – 33 mm, *K. pneumoniae* – 33 mm, E. coli – 28 mm, *E. faecalis* – 35 mm). This improvement suggests a synergistic interaction between the plant extract and detergent, where phenol and flavonoid compounds likely compromise bacterial cell walls, enabling detergent surfactants to penetrate more effectively Figure 3.

Table 3: Synergistic antibacterial activity of *Myrtus communis* L. leaf extract (25 mg/ml) combined with dishwashing detergent (0.78 mg/ml) against pathogenic bacteria based on inhibition zone diameters (mm)

Treatment	B. subtilis	K. pneumoniae	E. coli	E. faecalis
0.78 mg/ml detergent + 25 mg/ml extract	33	33	28	35

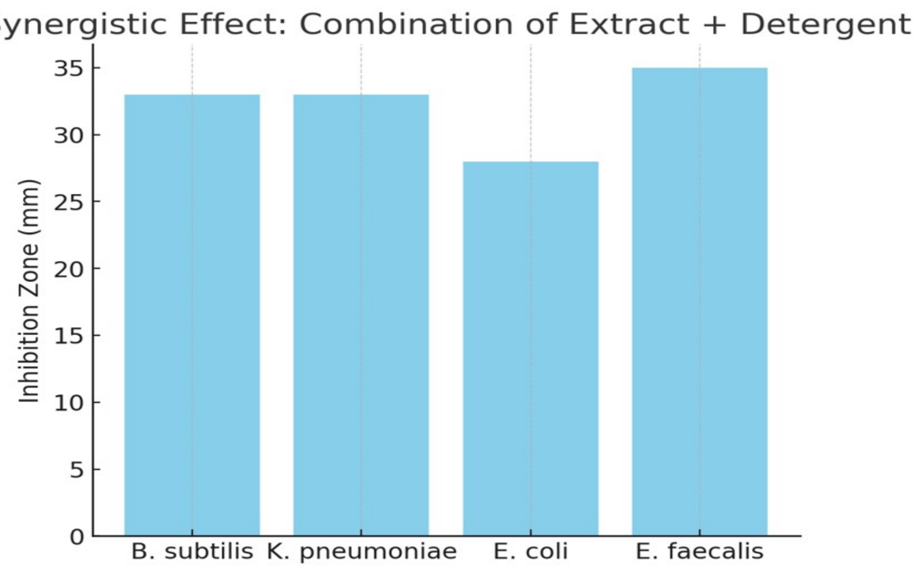

Figure 3: Synergistic antibacterial effect of *Myrtus communis* L. leaf extract (25 mg/ml) combined with dishwashing detergent (0.78 mg/ml) against four pathogenic bacterial species

3.4. Comparative analysis with previous studies

The synergistic gain is in line with previous studies showing that a combination of natural antimicrobials with synthetic compounds leads to a significant increase in bactericidal activity and a decrease in the necessary concentration of chemicals [13]. Not only is such a dual mechanism more effective, but it can even help to avoid the possibility of the development of resistance since it can affect bacterial cells in more than one way [11].

3.5. Applications

These results justify the fact that *M. communis* L. extract has the potential to be used as a natural additive in order to increase the antibacterial effect of a dishwashing detergent. This approach may be applied to home and industrial hygiene, specifically, to avoid food contamination and biofilm development. The second step of the research should be to streamline the extract-to-detergent ratio, determine the stability of the product, and test it against biofilm-forming and antibiotic-resistant strains to render it practically applicable.

4. Conclusion

The incorporation of *Myrtus communis* L. leaf extract into dishwashing detergent formulations significantly enhanced their antibacterial activity against *Bacillus subtilis, Klebsiella pneumoniae, Escherichia coli,* and *Enterococcus faecalis*. The findings of the experiment determined that the greatest zone of inhibition was achieved when 25 mg/ml of the plant extract was combined with 0.78 mg/ml of a detergent, which represented a synergistic effect. The synergetic effect can be explained by the principle that the action of bioactive plant compounds and surfactants in the detergent works synergistically to disrupt bacterial cell walls and membranes better than either compound separately. The findings suggest that the natural plant extract can serve as the ingredient of the cleaning products as a sustainable and efficient way to control pathogenic bacteria, reduce the number of chemicals used, and develop sustainable hygiene practices at home and in industry. More research on the stability of the formulation, cost-saving, and scalability of the application is advised to allow for commercial application.

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