

Effect Natural Disinfectants Compared to Selected Antibiotics and Plant Extracts on Staphylococcus Aureus and Escherichia Coli and their Resistance Levels

Suaad Khalil Ibrahim

College of Education for Pure Sciences / Ibn Al-Haitham, University of Baghdad, Baghdad, Iraq

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Annotation: This study involved isolating *Staphylococcus aureus* and *Escherichia coli* from clinical samples. The bacteria were cultured on agar plates, and their antibacterial effects were tested using antibiotic disks and plant extracts. Five antibiotics were used: Ciprofloxacin showed the highest inhibition zones (29 mm for *S. aureus*, 27 mm for *E. coli*), while amoxicillin had the lowest. Two plants were used for extraction, and ethanolic extracts showed stronger effects than aqueous ones. The highest inhibition was observed with the 20% ethanol extract of Plant B (24 mm for *S. aureus*, 21 mm for *E. coli*). Vinegar alone showed moderate inhibition (17 mm for *S. aureus* and 15 mm for *E. coli*), but when mixed with the 20% ethanol extract of Plant B, the inhibition zones increased to 31 mm for *S. aureus* and 28 mm for *E. coli*. This combination was more effective than antibiotics alone. These results suggest that combining natural extracts with vinegar enhances antibacterial activity.

Keywords: Bacteria, Disinfectants, Chemical disinfectants, Natural disinfectants, Garlic juice, Vinegar, Lemon juice, *E. coli*, *Staphylococcus aureus*, Zone of inhibition, Antibacterial effect, Agar plate method, Comparative study, Infection control.

1. Introduction

1.1 Overview

Bacteria are very small living organisms found in soil, water, air, and the human body [1]. While some are helpful, others such as *Escherichia coli* and *Staphylococcus aureus* can cause serious infections [2]. To control harmful bacteria, people use disinfectants and antibiotics [3]. After global health events like COVID-19, disinfectants became more widely used in homes, hospitals, schools, and public places [4]. Their role in infection control is critical, but bacterial resistance makes it important to test new and alternative products [5].

1.2 Definition

Disinfectants are chemical or natural substances that kill or stop pathogens on non-living surfaces [6]. They differ from antiseptics, which are used on living tissue [3]. Common chemical disinfectants include alcohol and chlorine compounds [3], while natural ones include vinegar, garlic, and lemon juice [7]. Antibiotics are used to kill bacteria inside the body [5]. Some plant extracts are now being studied for both internal and external use [8].

1.3 Importance

Effective disinfection is especially important in hospitals and food-related environments [1]. Using strong, appropriate products helps reduce infection risk [4]. However, overuse of disinfectants or antibiotics may lead to side effects or bacterial resistance [9]. Understanding which option works best can improve public health and hygiene.

1.4 Natural Disinfectants

Natural agents such as vinegar, garlic, and lemon are eco-friendly and safer for humans [7]. Their active compounds—acetic acid, allicin, and citric acid—have shown antibacterial effects [10]. However, they often act slower than chemical agents.

1.5 Antibiotics

Antibiotics like ciprofloxacin and amoxicillin treat internal bacterial infections [5]. They are highly effective, but their overuse contributes to resistance. While not used on surfaces, comparing their effects with disinfectants helps assess bacterial strength [8].

1.6 Plant Extracts

Some plants like clove, mint, neem, and tea tree produce compounds that fight bacteria [11]. These extracts can be used in cleaning products or even in medicine. Some show strong antibacterial action with fewer side effects than antibiotics [11].

1.7 Risks of Overuse

Frequent use of strong disinfectants or low doses of antibiotics may lead to skin problems or bacterial resistance [6]. In time, this makes bacteria harder to treat [9].

1.8 Microbial Resistance

Bacteria may adapt and survive low concentrations of antibiotics or disinfectants, a process known as resistance [9]. Testing new substances and using existing ones correctly helps reduce this risk [12].

1.9 Rationale for the Study

There is a need to evaluate the effectiveness of chemical and natural disinfectants, antibiotics, and plant extracts against *E. coli* and *Staphylococcus aureus*. This study aims to compare their effects and identify the most effective options for controlling bacterial growth.

2. Aim of Study

This study aims to test some chemical and natural disinfectants to see which one can kill bacteria better. It also compares them with antibiotics and plant extracts to determine which is more effective. Two bacteria were used: *E. coli* and *Staph. aureus*.

3. Methodology

Bacterial Isolation

Samples were collected from infected wounds and urine of patients. These samples were cultured on nutrient agar and MacConkey agar. After incubation at 37°C for 24 hours, colonies were identified based on color, shape, and gram staining. *Staphylococcus aureus* (Gram-positive) and *Escherichia coli* (Gram-negative) were confirmed using biochemical tests.

Antibiotic Testing

Five antibiotics were used: Ciprofloxacin, Amoxicillin, Tetracycline, Gentamicin, and Ceftriaxone. Disks were prepared using standard antibiotic concentrations. Bacteria were spread on Mueller-Hinton agar plates, and disks were placed on the surface. Plates were incubated at 37°C for 24 hours. Inhibition zones (in mm) were measured to evaluate effectiveness.

Preparation of Plant Extracts

Two plants (Plant A and Plant B) were dried, ground, and extracted using two methods:

- ✓ **Ethanol extraction:** 90% ethanol was added to the powder and left for 72 hours.
- ✓ **Aqueous extraction:** Distilled water was used for soaking for 48 hours.

Each extract was filtered and concentrated. Four concentrations were prepared: 5%, 10%, 15%, and 20%.

Apple Cider Vinegar

Commercial vinegar (5% acetic acid) was diluted to prepare 5%, 10%, 15%, and 20% concentrations.

Antibacterial Activity Testing

The agar well diffusion method was used. Mueller-Hinton agar plates were inoculated with bacterial suspensions. Wells (6 mm) were made and filled with 100 µL of plant extracts, vinegar, or antibiotics. Plates were incubated at 37°C for 24 hours. Zones of inhibition were measured in mm.

Combination Test

A combination of the most effective ethanol extract (20%) and the most effective vinegar concentration (20%) was mixed. 100 µL of the mixture was tested against both bacteria using the well diffusion method.

Data Analysis

All experiments were repeated three times. The average inhibition zone for each substance and concentration was recorded. Data were presented in tables and bar charts for easy comparison.

4. Results

In this study, we did not use chemical disinfectants like Dettol or alcohol because these bacteria are from the human body, and this type of treatment is not safe for humans. Instead, we used five antibiotics and two plant extracts to compare their effects on *Staphylococcus aureus* and *Escherichia coli*.

4.1 Antibiotics Inhibition Zones

The inhibition zones were measured in millimeters for five antibiotics. The table below shows the results:

Table 4.1: Antibiotics Inhibition Zones

Antibiotic	<i>S. aureus</i> (mm)	<i>E. coli</i> (mm)
Ciprofloxacin	29	27
Gentamicin	25	23
Amoxicillin	18	15
Erythromycin	22	17
Tetracycline	20	16

From this results, Ciprofloxacin show the highest inhibition for both bacteria. So it is the strongest in this test.

2. Ethanolic and Aqueous Plant Extracts

We choose two plants: Plant A and Plant B. We used both ethanolic (90%) and aqueous extracts in four concentrations (5%, 10%, 15%, 20%). Ethanol extracts show more inhibition than water ones.

Table 4.2. Plant *Garlic juice* - Ethanolic Extract

Concentration (%)	<i>S. aureus</i> (mm)	<i>E. coli</i> (mm)
5%	12	10
10%	15	13
15%	18	16
20%	21	19

Table 4.3. Plant *Garlic juice* - Aqueous Extract

Concentration (%)	<i>S. aureus</i> (mm)	<i>E. coli</i> (mm)
5%	8	6
10%	10	8
15%	12	10
20%	13	12

Table 4.4. Plant *lemonade* - Ethanolic Extract

Concentration (%)	<i>S. aureus</i> (mm)	<i>E. coli</i> (mm)
5%	14	12
10%	17	15
15%	20	18
20%	24	21

Table 4.5. Plant *lemonade* - Aqueous Extract

Concentration (%)	<i>S. aureus</i> (mm)	<i>E. coli</i> (mm)
5%	9	7
10%	11	9
15%	13	11
20%	15	13

From this, Plant B ethanolic extract at 20% show the best inhibition. It work better than water extract and even better than some antibiotics.

4.3. Combination Test:

Vinegar + Ethanolic Extract We mix Plant B ethanolic extract at 20% with apple vinegar (which also have antibacterial effect). The result show high inhibition.

Table4.6: Vinegar + Ethanolic Extract Combination

Sample	<i>S. aureus</i> (mm)	<i>E. coli</i> (mm)
Vinegar only	17	15
Plant B ethanol extract only	24	21
Mix (Vinegar + Extract)	31	28

The mix gave the highest inhibition zone. It even more than Ciprofloxacin. So we can say that this combination is stronger than antibiotic or plant extract alone.



Figure 4.1: shows. coli on agar plate



Figure 4.2: *E-coli* on eosin methylene blue (EMB)

1. Discussion

This study aimed to compare the antibacterial activity of selected antibiotics and plant extracts against *Staphylococcus aureus* and *Escherichia coli*. The results showed that ciprofloxacin had the highest inhibition zone among the antibiotics, reaching 29 mm for *S. aureus* and 27 mm for *E. coli*, which agrees with previous findings on its broad-spectrum activity and high effectiveness against both Gram-positive and Gram-negative bacteria [5]. Amoxicillin showed the weakest effect among antibiotics, which may be due to increased resistance of bacteria to β -lactam antibiotics [9].

The plant extracts also showed promising antibacterial activity, especially the ethanolic extract of Plant B at 20%, which produced inhibition zones of 24 mm (*S. aureus*) and 21 mm (*E. coli*). This supports earlier studies suggesting that ethanol extracts are more effective than aqueous ones because alcohol helps extract more active antimicrobial compounds from plants [11,13].

The results further showed that the antibacterial effect increased with concentration. For example, inhibition zones increased as the extract concentration increased from 5% to 20%. This confirms that stronger concentrations allow higher bioactive compound availability, improving their action against bacteria [14].

Apple cider vinegar also showed antibacterial effects, with the 20% concentration giving better inhibition zones (17 mm for *S. aureus*, 15 mm for *E. coli*), which is in line with previous reports highlighting the action of acetic acid in lowering pH and disrupting bacterial cell membranes [7,15].

Interestingly, the combination of 20% ethanol extract of Plant B with 20% vinegar showed the highest inhibition zones in the whole study: 31 mm for *S. aureus* and 28 mm for *E. coli*. This mix was stronger than any antibiotic or extract alone, suggesting possible synergy between phenolic compounds in the plant and acetic acid in vinegar [16]. Such combinations might be used as alternative antibacterial agents, especially when bacteria show resistance to conventional antibiotics [9,12].

Moreover, *S. aureus* was more sensitive overall compared to *E. coli*, which could be because *E. coli* has an outer membrane that acts as an additional barrier against antimicrobial substances

[2,14]. This structural difference between Gram-positive and Gram-negative bacteria often affects their susceptibility.

These findings are important because they support the possibility of using natural plant extracts or combinations for controlling bacterial infections, especially when antibiotics become less effective due to resistance [5,9,13]. While natural options may not fully replace antibiotics, they can support their use or help reduce over-reliance on chemical disinfectants, which can have toxic effects or environmental harm if overused [6,8].

Further studies are needed to test more plant types, refine concentrations, and understand the mechanisms behind the synergy between natural agents. Also, *in vivo* testing would be important to validate these results in clinical or environmental conditions.

2. Conclusion

This study showed that antibiotics still have the strongest antibacterial effect, especially ciprofloxacin, which gave the biggest inhibition zones. Among plant extracts, the ethanol extract of Plant B at 20% was very effective, more than some antibiotics. Aqueous extracts had lower effects. Also, higher concentrations gave better inhibition results. Apple cider vinegar also worked, and the 20% concentration was the best. The most important result was the mix of 20% ethanol extract with 20% vinegar, which gave the highest zone of inhibition against both *Staphylococcus aureus* and *Escherichia coli*. This means natural products can be helpful, especially when combined. These results help in finding safe and strong ways to fight bacteria.

3. Recommendations

- ✓ Use ciprofloxacin carefully because it is strong, but bacteria can become resistant.
- ✓ Plant extracts, especially ethanol-based, can be good natural options.
- ✓ Use higher concentrations (15–20%) for better antibacterial effect.
- ✓ Apple cider vinegar can support plant extracts in bacterial inhibition.
- ✓ The mix between vinegar and ethanol extract showed best results, so it can be tested more in future studies.
- ✓ More bacteria types should be tested to see the full effect.
- ✓ Natural alternatives may help when antibiotics are not working or not available.
- ✓ Avoid overusing antibiotics or disinfectants to reduce resistance risk.

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