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Antimicrobial Effect of Drinkable Lugol Solution

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Highlights:

- Antimicrobial effect
- Using Lugol's solution as a supplement
- Infection agents

Keywords:

- Multidrug resistance
- Lugol (İodine)
- Antibiotic sensitivity

ABSTRACT:

The death rates due to infection in patients receiving long-term antibiotic treatment and hospitalized patients are quite alarming. Treatment of multidrug-resistant strains of *Acinetobacter baumannii, Klebsiella pneumoniae, Escherichia coli, Pseudomonas aeruginosa* and Staphylococcus aureus, which cause widespread nosocomial infection, has become a global problem. Alternative treatment methods are needed for these species, which use all their resistance mechanisms day by day. Recently, it has been seen that iodine (lugol) solution has been used in the treatment of many infections. Significant results are observed, especially for nosocomial and wound infections. The literature on the antimicrobial effect of Lugol solution is very limited. In order to scientifically support such treatments, we aimed to investigate the antimicrobial effect of lugol on resistant bacteria in a laboratory environment. In our laboratory, we have previously isolated *Acinetobacter baumannii, Shigella sonneii, Escherichia coli, Pseudomonas aeruginosa* and *Staphylococcus aureus* strains with known antibiotic resistance. For these isolates, the agar dilution method prepared with different concentrations of lugol and the Broth Microdilution Method were used. As a result of our study, it was observed that drinkable Lugol solution affects multidrug-resistant microorganisms at very low concentrations. Thus, Lugol's success in infection treatments will be scientifically supported.

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INTRODUCTION

The use of antiseptics for treatment purposes dates back to ancient times. Many deaths were prevented by using them as sprays on wounds in the 1800s (Grønseth et al., 2022). However, the possibility that it would cause antiseptic toxicity later prevented it from being treated. Antibiotic discoveries for treatment have begun, and the use of antibiotics has been preferred. The prevalence of multidrug-resistant bacteria increased after antibiotic use. To prevent this, antiseptics have since become a hot topic. Antiseptics do not target a specific area as antibiotics do. Since it targets many regions in microorganisms, it prevents the development of antimicrobial resistance.

Iodine has long been known as an antimicrobial agent. Several clinical studies have also shown the efficacy of iodine and povidone iodine (PI) in oral hygiene. However, limited studies have been performed on iodine's effect on infections (Tam et al., 2006). Polyvinylpyrrolidone-iodine is a widely used antiseptic introduced by Shelanski and Shelanski in 1956. It is a water-soluble compound that results from the combination of molecular iodine and polyvinylpyrrolidone (Siggia,1957). The preparations of polyvinylpyrrolidone-iodine commercially available are povidone-iodine solution, scrub, ointment, and foam; of these, the solution is the most commonly used. The 10 percent polyvinylpyrrolidone-iodine solution generally contains 90 percent water, 8.5 percent polyvinylpyrrolidone,1 percent available iodine, and iodide (Zamora, 1986). The content of Lugol's solution consists of 85% water, 5% iodine and 10% potassium iodide.

The spread of multidrug-resistant microorganisms emerging in hospitals has become a cause for concern. Bacteria with intense antibiotic resistance are a serious problem for society. In addition, the increase in the death rate due to difficulty in treatment poses both financial loss and a great difficulty for healthcare professionals (Ibrahim et al., 2021). For this reason, new alternatives are being sought to prevent antibiotic resistance. Lugol solution is used as an antiseptic in skin, wound and soft tissue infections. No therapeutic use has been found for *Staphylococcus aureus, Escherichia coli, Acinetobacter baumannii* and *Pseudomonas aeruginosa*, which cause nosocomial infections. In addition, no literature has been found in which Lugol's solution is included in the treatment of multidrug-resistant mycoorganisms. Therefore, in our study, we aimed to see to what extent the antimicrobial effect of Lugol's solution was on strains with multidrug resistance. At the same time, we believe that we will provide a scientific basis for the drinkable Lugol solution used in daily life and will contribute to the literature.

MATERIALS AND METHODS

Bacteria Isolation

In order to revive our *Acinetobacter baumannii, Escherichia coli, Pseudomonas aeruginosa, Shigella sonnei* and *Staphylococcus aureus* isolates, which were previously diagnosed with the VITEK 2 device in Iğdır State Hospital, they were planted in 5% sheep blood agar. It was incubated at 37ºC for 18-24 h.

Lugol's solution

2% Lugol solution iodine drops (law material) were commercially available. Then, 1% Lugol solution was prepared with DNAse free.

Broth microdilution method of lugol's solution

The antimicrobial activity of the diluted Lugol solution was evaluated by the broth microdilution mL method. 100 microliters of brain heart infusion (BHI) was added to each well. 100µl of Lugol's 1% dilution was added to the first well. Thus, dilutions were made at concentrations of 5µg/mL,

2.5µg/mL, 1.25µg/mL, 0.625µg/mL, 0.312µg/mL, 0.156µg/mL and 0.078µg/mL starting from the first well. Finally, the wells were inoculated with 0.5 McFarland turbid microorganisms. The first well with no growth after incubation was considered the MIC value.

Disc diffusion method of lugol's solution

The 2% Lugol solution we obtained commercially was reduced to 1% concentration with DNAse free. Stock solution was prepared. Mueller Hinton Agar was prepared by pouring 20 mL into each petri dish. Passages from our stock bacteria were cultured twice and their antimicrobial susceptibility was evaluated using the disk diffusion method from the subculture. Bacteria were prepared to 0.5 Mc Farland turbidity and plated evenly on Müller hinton agar. Disks impregnated with 10 µl Lugol were placed on it. Zone diameters formed after 18-24 h. of incubation were measured and evaluated in 37 cases. The experiment was repeated 3 times and the averages were evaluated. DMSO solvent was used as a negative control.

Antibiotic resistance of strains

Tablo 1. Antibiotic Resistance of Multidrug Resistant İsolates

	Cefta	Trim/ Sulfa	Mero	Amik	Genta	Cipro	İmi	Col
A. baumannii	+	+	+	+	+	+	+	-
E. coli	+	+	-	-	-	+	-	-
P. aeruginosa	+	-	+	+	+	-	+	-
S. sonnei	+	+	-	-	-	+	+	-
S. aureus	+	+	-	-	-	+	-	-

*****Ceftazidime (Seft), Trimethoprim/ Sulfamethoxazole (Trim/Sulfa), Meropenem (Mero), Amikacin (Ami), Gentamicin (Genta), Ciprofloxacin (Cipro), İmipenem (İmi), Colistin (Col)

RESULTS AND DISCUSSION

Multidrug-resistant *Acinetobacter baumannii, Escherichia coli, Pseudomonas aeruginosa, Shigella sonnei and Staphylococcus aureus* strains isolated from clinical samples at Iğdır State Hospital were randomly selected and included in the study.

Lugol's solution showed a significant antimicrobial effect against multi drug resistance bacteria. Our strains are hospital isolates and all have multiple antibiotic resistance. The MIC values of Lugol's solution 1%'lik for *Acinetobacter baumannii, Escherichia coli, Pseudomonas aureginosa, Shigella sonnei* and *Staphylococcus aureus* strains with high antibiotic resistance are given in Table 2 and Figure 1.

Table 2. Antimicrobial Effect of Multidrug Resistant Strains

Strain	MİC	Disk Difüzyon
Escherichia coli	0.312 µg/mL	10 mm
Acinetobacter baumannii	0.156 µg/mL	10 mm
Pseudomonas aeruginosa	0.156 µg/mL	10 mm
Staphylococcus aureus	0.156 µg/mL	13 mm
Shigella sonnei	0.156 µg/mL	12 mm

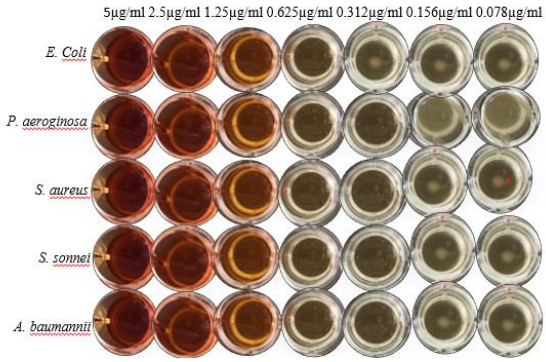

Figure 1. MIC Values of Multidrug Resistant Strains

According to the results of 1% Lugol solution, 4 different concentrations (1, 10^{-1}, 10^{-2}, 10^{-3}) were studied according to the disk diffusion method. The best result was found at 10^{-1} dilution. As seen in the table, it had the same effect with 10 mm diameter for *Acinetobacter baumannii*, *Escherichia coli*, *Pseudomonas aeruginosa*. A diameter of 12mm-13mm was effective for *Shigella sonnei and Staphylococcus aureus*, respectively (Figure 2).

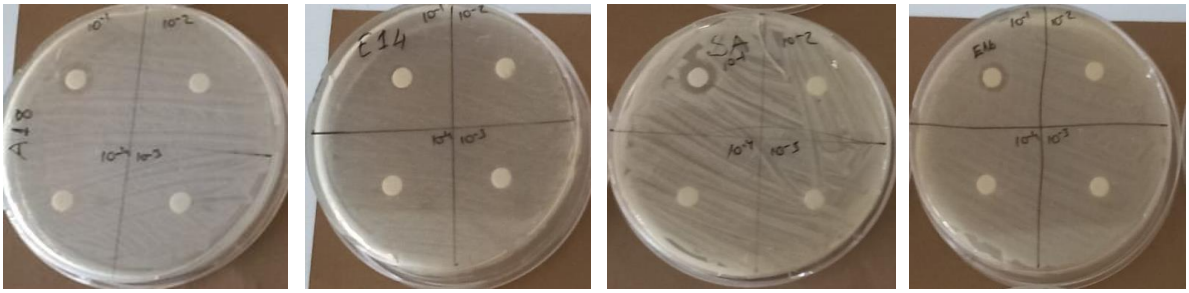

Figure 2. Disc Diffusion of Multidrug-Resistant Strains

Lugol (iodine) has been used as an antiseptic for many years. Nowadays, it has started to be preferred among infection treatments (Hendley et al., 1978). In the 1900s, it was preferred in the treatment of drug-resistant spirochetes and inactivation of contaminated viruses on hand (Akatsu and Hideyo, 1917). In the study conducted by Hendley et al., the iodine (lugol) solution was tested for virucidal activity against rhinovirus type different minute. A 1% aqueous iodine solution was very effective in eliminating infectious rhinovirus from fingertip rinses (Hendley et al., 1978). For a long time, it was used only in wound treatments. As a result of increased antibiotic resistance, the healing time of diseases has been prolonged. People are resorting to alternative ways to get treatment without medication.

In 1867 Joseph Lister published a paper in The Lancet on the application of antiseptics, which paved the way for antiseptic surgery. It saved thousands of patients from lethal infections acquired during and after surgery (Toledo., 2010).

Lugol (iodine), which has a very wide spectrum of action, is effective against multidrug resistant bacteria, *Pseudomonas aeruginosa*, *Mycobacterium tuberculosis*, fungi, viruses and protozoa (Akbulut., 2023). *Streptococcus mutans* is the nightmare of tooth biofilm. In Avshalom Tam's study, they found that it prevented the biofilm formed by *Streptococcus mutans* on the teeth (Tam et al., 2006). In the analysis performed on Candida albicans and Candida glabrata, it was concluded that lugol reduces cellular viability in a dose-dependent (Du et al., 2023).

The cytotoxic Lugol effect is characterized by increased oxidative stress and decreased superoxide dismutase and catalase enzyme activities. In a study comparing the bactericidal and

cytotoxic effects of hydrogen peroxide (H_2O_2), povidone iodine (PVP-I) and Lugol's solution, they reported that high concentrations of Lugol had a bactericidal effect on *E-coli* (Tonoyan et al., 2018).

The antibacterial and antibiofilm effects of lugol against methicillin-resistant *Staphylococcus aureus* found on the skin were investigated. They used 5% Lugol solution for MRSA on mouse skin. They observed that after lugol applied for 5 days, MRSA was significantly reduced and the biofilm was eradicated (Grønseth et al., 2022).

The increasing occurrence of antibiotic resistance in pathogenic bacteria, coupled with a dramatic decline in the number of newly approved antibiotics, represents a major societal challenge.

It is frequently encountered in the treatment of nosocomial and gastrointestinal infections, especially in social media, which has become popular culture. When we investigate this situation academically, we encounter very little data. It is evidence-based in the treatment of wound and skin infections (Grønseth et al., 2017). However, there is no literature in which multidrug-resistant *Acinetobacter baumannii, Shigella sonnei, Escherichia coli, Pseudomonas aureginosa* and *Staphylococcus aureus* strains, which are frequently encountered in hospital infections, are studied together.

CONCLUSION

There are feedback that individual people using it have been cured, but no experimental results have been found in the laboratory environment for nosocomial infection agents such as *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. Antiviral, antibacterial and antifungal effects have been shown in limited studies. There is insufficient data regarding Lugol's solution to prove that it will contribute to alternative treatment by preventing antibiotic resistance. We think that it should be tested on cytotoxicity and live experimental animals.

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Conflict of Interest

The article authors declare that there is no conflict of interest between them

Author’s Contributions

The authors declare that they have contributed equally to the article.

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