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**Research Article** 

# Antimicrobial Activity of Ethanol Extract of Rhizome Turmeric (Curcuma Longa L.) For Growth of Escherichia coli, Staphylococcus aureus and Candida albicans

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# ABSTRACT

**Objective:** The purpose of this study was to determine at what concentration of ethanol extract of rhizome turmeric is active against *Escherichia coli, Staphylococcus aureus* and *Candida albicans*.

**Methods:** Ethanolic extract of rhizome turmeric was tested for phytochemical screening by using standard protocol. Antimicrobial testing was using the diffusion disc method to measure the inhibition zone against the *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans* with various concentration of rhizome turmeric extract (500 mg/mL, 400 mg/mL, 300 mg/mL, 200 mg/mL, 100 mg/mL, 50 mg/mL, 25 mg/mL).

**Results:** Phytochemical screening showed that ethanolic extract of rhizome turmeric contain alkaloids, flavonoids, saponins, tannins, and triterpenoid/steroid. The antimicrobial inhibition of ethanol extract of rhizome turmeric against *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans* microbia at a concentration of 500 mg/mL had a diameter of 15.88 mm, 15.63 mm and 15.22 mm with a strong category.

Conclusions: rhizome turmeric could be use as a novel antimicrobial agent.

Keywords: Extract, antimicrobial, Curcuma longa L, Escherichia coli, Staphylococcus aureus, Candida albicans

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#### **INTRODUCTION**

urmeric (Curcuma longa L.) is a plant of Family Zingiberaceae, generally widely used as a spice and coloring agent in food and is known for medicine. Turmeric is a perennial herb that is widely cultivated in Asia, India, China and other countries with tropical climates<sup>1</sup>. Traditionally, rhizomes of turmeric have been as insecticides, antimicrobials<sup>2</sup>, antidiabetic<sup>3</sup> used rheumatism, body aches, skin diseases, intestinal worms, diarrhea. dyspepsia, inflammation. fever. liver. constipation, leukoderma, and inflammatory colic disorders<sup>4,5</sup>.Components of chemical compounds that found in turmeric consist of curcuminoids, especially curcumin (diferuloymethane, demethoxycurcumin, and bismethoxycurcumin)<sup>6</sup> that potentially as antioxidants, antiinflammatory, anti-platelet, cholesterol, antibacterial and antifungal<sup>7</sup>, alkaloid, saponin, tannin, tannin, stolysic acid, glycoloside<sup>8</sup> .Escherichia coli is an enteric gram-negative bacterium (Enterobactericeae) which belongs to the normal flora found in the human large intestine. This bacterium is

activated. *Escherichia coli* often causes clinical infections such as urinary tract infections, sepsis and meningitis<sup>9</sup>. *Staphylococcus aureus* is one of the important pathogenic bacteria associated with virulence toxins, invasive, and the body's resistance to antibiotics. Staphylococcus aureus infection is a main cause of skin disorders, soft tissue, respiratory, bone, joint, and endovascular disorders<sup>10</sup>. *Candida albicans* is the most common pathogenic fungus that can cause oral candidiasis, gastrointestinal and vaginal<sup>11</sup>. By considering the potentials of turmeric as sources for antimicrobial drugs, the objective of this study is to investigate antibacterial and antifungal activity of turmeric rhizome ethanolic extracts against the most common microbial

#### MATERIAL AND METHODS

#### **Plant preparation**

Fresh Rhizoma turmeric was collected from local area in Pancurbatu Village (Deli Serdang Regency North Sumatra Medan, Indonesia), and authenticated by Indonesian

[5]

Institute of Sciences: Research Center For Biology (No. 2460/MEDA/2019). Voucher specimen was deposited in the Pharmacognosy Laboratory, Sekolah Tinggi Ilmu Kesehatan Senior Medan.

#### **Extraction of Rhizome Turmeric**

The making of turmeric rhizome ethanol extract was using maceration method 96% ethanol (1:10), be weighed 1 Kg turmeric powder and then put into a vessel and dissolved with 96% ethanol as much as 75 parts (3.7 L) in a closed container, left for five days while frequently stirring, filtered, the waste is remastered with the remaining solvent that is 25 parts (1.3 L), the maserat is evaporated with a rotary vaccum evaporator at 40°C, so get the viscous turmeric rhizome ethanol extract.

#### Phytochemical screening of various rhizome turmeric

The crude extract of rhizome turmeric was screening by using the standard protocol to know the presence of phytochemical compounds. The extracts were subjected to phytochemical tests for determination of plant secondary metabolites such as alkaloid, flavonoid, tannin, saponin, and triterpenoid/steroid<sup>12,13</sup>.

#### **Antimicroba Activity**

Preparation of antimicrobial activity will begin with sterilizing the tools and materials to be used, rejuvenation of microba (Escherichia coli, Staphylococcus aureus Candida albicans), making media, making bacterial suspense, making rhizome turmeric extract test solutions making comparative standard solutions. The and determination of the antimicrobial activity was carried out with sterilized NA media for (Eschericia coli and Staphylococcus aureus) PDA media (Candida albicans) inserted into 20 mL sterile petri dishes each and allowed to condense at room temperature. The media was dropped with 0.1 mL of microba suspension test and flattened using an L bar until smooth and dry. Sterile disc paper with a diameter of 6 mm was dropped with ethanol extract of rhizome turmeric as much as with each concentration of 500 mg/mL, 400 mg/mL, 300 mg/mL, 200 mg/mL, 100 mg/mL, 50 mg/mL, 25 mg/mL and then placed on the media so that the solid that had been dripped with a test

bacterial suspension, DMSO 10% as a negative control, and amoxicilin as a positive control for bacteria and Ketokonazol as a positive control for fungi. Then incubated at 37°C for 24 hours and after incubation the clear zone was measured using calipers, three replications were performed.

### **RESULT AND DISCUSSION**

#### **Phytochemical Screening**

Phytochemical screening results of rhizome turmeric ethanol extract showed different phytochemical compounds (Table 1).

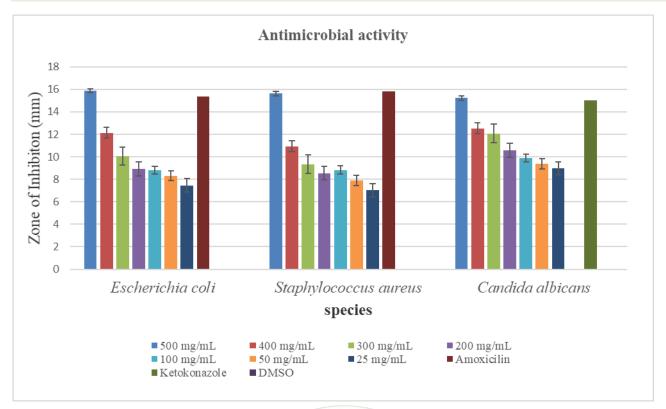
Table: 1. Phytochemical screening of rhizome turmeric ethanolic extract

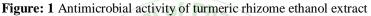
No	Screening	Reagent	Ethanolic Extract
1	Alkaloids	Mayer	Positive
		Dragenddroff	Positive
		Bouchardat	Positive
2	Flavonoids	Mg+HCl+Amyl Alcohol	Positive
3	Saponins	Foaming Test	Positive
4	Tannins	FeCl <sub>3</sub> 1%	Positive
5 Ph	Triterpenoid/ Steroids	Liebermann Bouchard	Positive

#### **Antimicrobial Activity**

Antimicrobial activity testing of ethanol extract of rhizome turmeric was carried out using the disc diffusion method, namely the determination of microbial sensitivity with a particular substance that may have antimicrobial activity using disc paper. Antibacterial testing was carried out with various concentrations of 500 mg/mL, 400 mg/mL, 300 mg/mL, 200 mg/mL, 100 mg/mL, 50 mg/mL, 25 mg/mL DMSO 10% as a negative control, and amoksisilin as a positive control for bacteria and Ketokonazole as a positive control for fungi. The results of testing the antimicrobial activity of ethanol extract of rhizome turmeric against *Escherichia coli, Staphylococcus aureus* and *Candida albicans* can be seen in Table 2 below.

Sample Concentraction	Zone of Inhibiton (mm)			
	Escherichia coli	Staphylococcus aureus	Candida albicans	
500 mg/mL	15.88±4.09	15.63±1.60	15.22±1.21	
400 mg/mL	12.12±3.86	10.92±1.55	12.52±0.47	
300 mg/mL	10.06±0.55	9.33±0.33	12.07±0.55	
200 mg/mL	8.93±0.87	8.85±0.51	10.57±0.33	
100 mg/mL	8.81±0.23	8.83±0.38	9.88±0.15	
50 mg/mL	8.32±1.06	7.88±0.14	9.38±0.45	
25 mg/mL	7.45±0.26	7.03±0.23	8.98±0.06	
Amoxicilin	15.38±0.43	15.80±0.61	-	
Ketokonazole	-	-	15.02±1.36	
DMSO	0	0	0	





Based on Table 2 and Graph 1, it can be seen that the antimicrobial effect of turmeric rhizome ethanol extract is more effective in inhibiting the growth of the fungus *Candida albicans*. This can be seen from the diameter of inhibitory zones in the fungus Candida albicans at concentrations of 500 mg / mL, 400 mg / mL, 300 mg / mL and 200 mg / mL including strong criteria. For *Escherichia coli* bacteria, it is more effective at concentrations of 500 mg / mL, while *Staphylococcus aureus* is more effective at concentrations of 500 mg / mL. According to Davis and Stout, explain that the criteria for antibacterial inhibition consistency of  $\geq$ 20 mm is very strong, 10-20 mm is strong, 5-10 mm is moderate and  $\leq$  5 mm is weak<sup>14</sup>.

Positive controls using standard amoxicilin with a concentration of 30 mg/mL. Amoxicilin is a broadspectrum bacteriotic antibiotic and a strong bactericide by inhibiting the synthesis of cell walls and bacterial proteins that can cause lysis or death cell<sup>15</sup>. Ketoconazole is used as a positive control in inhibiting the growth of candida fungus. Ketoconazole is an orally active agent with a broad spectrum of activity against yeast, dermatophytes and dimorph fungi. It works on the synthesis of ergosterol in the yeast cell wall and also interferes with the process of respiration in the mitochondria but does not directly involve glycoprotein synthesis and works by inhibiting the germination of candida<sup>16</sup>. The negative control used was DMSO with a concentration of 10%. In this study negative control DMSO has no clear zone, so DMSO can be said to not be able to inhibit bacterial growth.

Ethanol extract 96% of active rhizome turmeric as antimicrobial due to chemical components contained in the extract. Based on phytochemical screening, ethanol extract of rhizome turmeric contain alkaloid, flavonoid, saponin, tannin and triterpenoid/steroid compounds.

The mechanism of action of alkaloids as an antimicrobial is by inhibiting the synthesis of nucleic acids, because it can inhibit the enzymes dihydrofolate reductase and topoisimerase I. Alkaloids can disrupt the constituent components of peptidoglycan on bacterial cells so that the cell wall layers are not formed intact and cause cell death. Another mechanism of antibacterial alkaloids is that the alkaloid component is known as a DNA accelerator and inhibits bacterial cell topoisomerase enzymes<sup>13,17</sup>.

Flavonoids provide bacteriolytic effects, inhibit protein synthesis, DNA synthesis, RNA and damage cell membrane permeability<sup>18</sup>. Flavonoids have antibacterial activity because of the ability of flavonoids to interact with cell membranes and affect cell membrane bioactivity and it has been reported that flavonoids are able to reduce the fluidity of bacterial cell membranes that is directly related to damage to cytoplasmic membranes or indirect damage through autolysis / weakening of the cell wall and consequently osmotic lysis<sup>13,19</sup>.

The mechanism of action of saponin as an antibacterial and antifungal causes lysis of the bacterial cell wall and leakage of AKP (Alkaline Phosphate), an increase in saponin concentration causes the protein to dissolve, causing intercellular compounds to diffuse through the outer membrane and cell wall. This causes the cytoplasm to leak out of the cell resulting in cell death<sup>20</sup>.

Tannin is a water-soluble polyphenol that can precipitate proteins. Tannin has been reported to prevent the development of microorganisms by precipitating microbial proteins and making nutrient proteins unavailable to bacteria<sup>21</sup>. Tannin acts as an antimicrobial with less iron, hydrogen bonds or specific interactions with vital proteins such as enzymes in microbial cells. Herbs that have a tannin component are astringent and are used in the treatment of intestinal disorders such as diarrhea and dysmenorrhea<sup>22</sup>.

Triterpenoids have broad antimicrobial activity against filamentous bacteria, yeast and fungi. Triterpenoids are as antimicrobial because they can damage yeast cell membranes or damage lipid membrane synthesis that effect of membrane permeability resulting in cell leakage components<sup>23</sup>.

#### CONCLUSIONS

The ethanol extract of rhizome turmeric has a group of secondary metabolites including alkaloids, flavonoids, saponins, tannins, and triterpenoid/steroid. The ethanol extract of rhizome turmeric has effective antimicrobial activity against *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans* with a concentration of 500 mg/mL with a strong category.

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