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Antimicrobials Activity, Antioxidants Activity and Analysis of Active Extract Chemical Compounds Content of Moringa (*Moringa oleifera* Lam.) Leaf

***[1]Masfria, [1]Ginda Haro, [1]Nerdy, [2]Vriezka Mierza, [1]Henny Sri Wahyuni, [1]Yade M Permata**

[1]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Sumatera Utara, Medan, North Sumatera, Indonesia
[2]Department of Biological Pharmacy, Faculty of Pharmacy, University of Sumatera Utara, Medan, North Sumatera, Indonesia

Abstract : Background : For developing countries the emergence of strains of bacteria resistant to antibiotics in infectious diseases is an important issue. Handling of infectious diseases is not only increasing health care costs, in addition to resulting in increased mortality due to antibiotic required is not available. Antioxidant chemical compounds derived from plants have a great potential in solving the imbalance between oxidative stress with some degenerative diseases are growing rapidly at this time. Moringa (*Moringa oleifera* Lam.) is one of the plants that have chemical compounds are bioactive polyphenols among others. Moringa leaf has been used as a traditional medicine to treat various diseases.

Method : Plant material washed with water, drained and then dried in the drying cabinet. Phytochemical screening carried out on moringa leaf powder simplicia includes examining chemical compounds. Extraction was done by maceration method using an organic solvent based on multilevel polarity, from low to high polarity solvent hexane, ethyl acetate and methanol. Test bacteria used are gram-positive bacteria (*Staphylococcus aureus*) ATCC 25923 and gram-negative bacteria (*Escherichia coli*) ATCC 25922 as well as microscopic fungus (*Candida albicans*)ATCC 10231 by agar diffusion wells method. Testing of antioxidant activity using free radical catching methods using 1,1-diphenyl-2-picrylhidrazil (DPPH) as free radicals. Analysis of chemical compounds carried by thin layer chromatography (TLC). The stationary phase used silica gel. The mobile phase used was hexane: ethyl acetate (7: 3) with visualization agent 50% sulfuric acid in methanol, Liebermann-Bourchard, Dragendorff and5% iron (III) chloride.

Results : Screening results of Moringa leaf powder simplicia showed positive results of chemical compounds alkaloids, glycosides, anthraquinone glycosides, flavonoids, steroid, tannins and saponins. The test results demonstrate the antimicrobial activity of the three extracts of Moringa leaves provide inhibition against *Staphylococcus aureus* and *Escherichia coli*, but did not provide barriers against microscopic fungus *Candida albicans*. The antioxidant activity of the all extract of Moringa leaves show IC50 < 50 µg/mL and categorizing them very strong antibacterial and the highest antimicrobial activity demonstrated by the methanol extract. The results of chemical analysis for each extract showed chemical compounds alkaloid, steroids and polyphenols.

Background

Moringa (*Moringa oleifera* Lam.) is one of the plants that have chemical compounds are bioactive. The leaves is one part of the Moringa plant is the most important because it contains a variety of bioactive compounds that have pharmacological effects[1]. Moringa plant belongs to the genus Moringaceae tribe consisting of 13 types and can be grown throughout the tropical and subtropical countries, particularly widespread in Asia and Africa[2]. Among the 13 species of the studied biological activity of the antimicrobial and antioxidant is a type of *Moringa oleifera* Lam. Experts traditional medicine in India says that there are 300 types of diseases that can be prevented and cured using Moringa leaves[3]. Traditionally, all parts of the Moringa plant is used for various purposes to treatment and nutrition. Moringa leaves contain a lot of protein and beta-carotene are known as antioxidants, so it is widely used to meet the nutritional needs of the food. The use of Moringa leaves as traditional medicine used to treat a variety of diseases including malaria, typhoid fever, a disease caused by parasites, arthritis, swelling, ulcers, skin diseases, urinary tract disease, hypertension, diabetes, management of symptoms decreased immunity of AIDS, cardiac stimulant and contraception. Use of a thriving community is traditionally directly consume fresh leaves or using botanicals in extract form or the result of a extraction process. Moringa leaves contain the compound of vitamins, carotenoids, polyphenols, phenolic acids, flavonoids, alkaloids, glucosinolates, isothiocyanates, tannins, saponins and oxalic[1]. Flavonoids can be efficacious as antidiabetic[4], antimalarial[5], anticancer[5,6,7] and antibacterial[8].

Infection is a common disease in the tropics[9]. Tuberculosis, gonorrhea, malaria and ear infections in children is part of the disease taking antibiotic therapy. Currently, many traditional drugs are effective and the benefits almost identical to modern medicine, especially for the purpose of replacing antibiotics[10]. For developing countries the emergence of strains of bacteria resistant to antibiotics in infectious diseases is an important issue. The World Health Organization (WHO) has conducted research on 30 types of infectious diseases and it is known that many strains of bacteria that cause infections that are resistant to antibiotics. Handling of infectious diseases are not only increase healthcare costs because it requires the handling of a combination of antibiotics, in addition to resulting in increased mortality due to antibiotic is not available[11]. Based on data from the World Health Organization (WHO) in the field of treatment, bacterial resistance caused by the use of antibiotic drugs has now become a major problem for the health of the world's population[9,12].

Oxidative stress is the most important cause in some cases of the disease that often develops today such as cancer, diabetes mellitus, atherosclerosis, cardiovascular disease, premature aging and inflammation that is the result of an imbalance between the formation and neutralization of prooxidant[13]. Oxidative stress is characterized by the emergence of the first free radicals that seek stability by taking a pair of electrons contained in biological macromolecules, such as proteins, lipids and DNA contained in human cells healthy so causing proteins and DNA is damaged[14]. The compounds that can protect human cells from free radicals, among others tocopherol, ascorbic acid and glutathione. Antioxidant chemical compounds derived from plants have a great potential in solving the imbalance between oxidative stress with some degenerative diseases are growing rapidly at this time, because it is necessary a plant chemical compounds that have potent antioxidant properties but cytotoxic weak. Many of the chemical compounds of plants that contain compounds called polyphenols have an effect as a catcher of free radicals, including flavonoids, polyphenols, tannins and phenolic terpenes[15].

Materials and Methods

The study was conducted at the PhytochemistryLaboratory, Microbiology Laboratory and Research Laboratory, Faculty of Pharmacy, University of Sumatera Utara.Plant material used in this study was moringa plant obtained from the Jalan Jamin Ginting Nomor 10,Tanah Seribu, Binjai Selatan, Sumatera Utara. Intake of plant material is done purposively ie without comparing with similar plants in other areas.Identification of plants is done in "Herbarium Bogoriense" Botany Field Research Center for Biology, Indonesian Institute of Sciences - Bogor. Identification started by sending a portion of plants that allow it to be sent, such as leaves, stems, roots and flowers. The cover letter that is included does not include the name of the local area test plants.Fresh Moringa leaves obtained was washed with water, drained and then dried in the drying cabinet at a temperature of ± 40 ° C (room temperature of drying cabinet no more than 50ºC). Moringa leaf dry declared if when crushed, direct material brittle and shattered. Moringa leaves dried crushed using a blender, then the resulting powder is stored in a place protected from sunlight and moisture.

Simplicia Phytochemical Screening

Phytochemical screening carried out on moringa leaf powder simplicia includes examining chemical compounds alkaloids, flavonoids, glycosides, anthraquinone glycosides, saponins[16]; tannins, triterpenoids and steroids[17].

Extraction

Extraction was done by stratified maceration method that has been modified using an organic solvent based on multilevel polarity, from low to high polarity solvent hexane, ethyl acetate and methanol. 1 kg of powdered moringa leaf botanicals are macerated in advance with the solvent hexane for 3 days, then filtered, do it continuously until the filtrate obtained is clear and colorless. The resulting dregs do the same thing for the solvent ethyl acetate and methanol[18].

Antimicrobial Activity Testing

The bacteria used are gram-positive bacteria (*Staphylococcus aureus*) ATCC 25923 and gram-negative bacteria (*Escherichia coli*) ATCC 25922 as well as microscopic fungus (*Candida albicans*)ATCC 10231 were obtained from the MicrobiologyLaboratory, Faculty of Pharmacy, University of Sumatera Utara with each concentration culture test 106 cfu / ml which has been likened to the turbidity standard solution of Mac Farland. Testing Antimicrobial Activity agar diffusion method with wells modifications. A total of 10 ml test medium Mueller Hinton Agar (MHA, Merck®) put in a sterile petri dish, left for some time until the media hardens, then put 0.1 ml suspension culture and then poured 20 ml test media MHA were homogenized and immediately planted metal reservoir the distance that has been set, and then allowed to stand up to the media to solidify. Each reservoir metal is taken, then each of the wells that have been formed included 0.1 ml test solution with varying concentrations of solvent DMSO and ethanol. Tests done 3 times with measured each clear area around the well indicating the test culture growth inhibitory region[19].

Testing Antioxidant Activity

Testing of antioxidant activity using free radical fishing methods using 1,1-diphenyl-2-picrylhidrazil (DPPH) as free radicals. The arrest marked by the free radical DPPH color change from purple to yellow with IC50 values obtained a parameter in determining antioxidant activity[15].

Content Analysis of Chemical Compounds

Analysis of chemical compounds of hexane, ethyl acetate and methanol extract carried by thin layer chromatography (TLC). The stationary phase used aluminum plate TLC silica gel 60 F254 (Merck®). The mobile phase used was hexane: ethyl acetate (7: 3) (pro analysis, Merck®) with visualization agent 50% sulfuric acid in methanol, Liebermann-Bourchard, Dragendorff and5% iron (III) chloride[20].

Results and Discussion

Moringa leaf identification results conducted in "Herbarium Bogoriense" Botany Field, Research Center for Biology, Indonesian Institute of Sciences - Bogor, stating that the plants used in this study is the plant leaves of Moringa (*Moringa oleifera* Lam.) on Moringa family.3.6 kg Fresh moringa leaves obtained 1.2 kg after drying. Maceration sesults from 500 g simplicia moringa leaves using a solvent hexane, ethyl acetate and methanol respectively obtained 21.2599 grams of hexane extract, 11.9204 grams of ethyl acetate extract and 25.1028 gramsofmethanol extract.

Figure 1. a. Fresh Moringa leaves; b. Moringa Leaf Simplicia; c. Moringa leaf powder Simplicia

Screening results simplicia Moringa leaf powder showed positive results against the class of chemical compounds alkaloids, glycosides, anthraquinone glycosides, flavonoids, steroid, tannins and saponins. Results of phytochemical screening simplicia powder can be seen in Table 1 below.

Table 1. Results of phytochemical screening Powder Simplicia

No	Screening	Reagent	Results (color / sediment)	Conclusion
1.	Alkaloids	Dragendorff Bouchardat Mayer	(+)orange-brown (+) yellow-kecoklatan (+) turbidity and a white precipitate	(+)
2.	Flavonoids	Zn + concentrated hydrochloric acid Mg + concentrated hydrochloric acid	(+) red	(+)
3.	Glikosida	Molish Fehling LB	(+)purple rings (+) brick red precipitate (+) red	(+)
4.	Saponin	Hot water / shaken	(+)foam	(+)
5.	Antrakuinon glikosida	NaOH	(+)intensively red on NaOH layer	(+)
6.	Tanin	$FeCl_3$ 1%	(+) green	(+)
7.	Triterpenoid/Steroid	Liebermann-Burchard	(+)bluish green	(+) steroid

Description: (+) = containing the compound, (-) = Do not contain the compound

Moringa leaf simplicia powder is added to the reagent Dragendorff give precipitates brownish orange color, with a reagent Bouchardat give precipitates brownish yellow color, while the white precipitate formed with Mayer reagent and turbidity, it indicates the presence of alkaloid compounds. The addition of Mg powder and Zn powder with concentrated hydrochloric acid gives the red color, indicating the presence of flavonoids. Screening glycoside reagent is indicated by the addition of concentrated sulfuric acid molisch and which formed a ring of purple, while the addition of Fehling (Fehling A and B) as much precipitation of red brick. Molisch reagent is a reagent commonly used for identification of carbohydrates, in this case is sugar and Fehling reagent used to identify the presence of sugar, especially is reducingsugar. Test glycosides aglycone of the compound produced is shown by the formation of a red color indicating the presence of compounds triterpenoid on its aglycone bond. Screening saponins produce a stable foam with a height of 3 cm foam and does not disappear with the addition of HCl 2 N. Examination of anthraquinone glycosides, form in the yellow benzene showed anthraquinone aglycone and intense red color of the NaOH layer. The addition of 1% $FeCl_3$ gives green indicating tannin. Tannin formed complex with iron (III) chloride solution ($FeCl_3$) produces black blue color to green colors indicate the presence of phenolic compounds[21]. The addition of Liebermann-Burchard

gives a bluish green color indicates steroid compounds. Moringa leaves contain almost all the phytochemical content, namely polyphenols, phenolic acids, flavonoids, alkaloids, glucosinolates, isothiocyanates, tannins, saponins and oxalic. Phytochemical screening result obtained is equal to what was mentioned by previous research, but the results of the study also found the existence of steroid, anthraquinone glycosides and glycosides[1].

Antimicrobial Activity Test Results

Results of testing the antimicrobial activity of the extract hexane extraction storey maceration using hexane solvent on the growth of Staphylococcus aureus, Escherichia coli and Candida albicans microscopic fungi in Table 2 indicate that the absence of inhibition occurred at concentrations from 200 to 6.25 mg / ml. This is expected because the chemical compounds most nonpolar contained in Moringa leaves drawn on the solvent hexane not provide antimicrobial activity. Chemical compounds nonpolar classes of compounds generally are triterpenoida and steroid free. The type of organic solvent to extract the most widely used chemical compound plant for research purposes antimicrobial is methanol, ethanol and air.Golongan chemical compounds that are often found memberika effect as animikroba include phenol, acids phenolic, quinone, flavones, flavonoids, flavonol tannin and coumarin[19].

Table 2. Results of Testing of Antimicrobial Activity of Hexane Extract against *Staphylococcus aureus*, *Escherichia coli* and *Candida albicans*

Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	0.00	0.00	0.00	0.00
100	0.00	0.00	0.00	0.00
50	0.00	0.00	0.00	0.00
25	0.00	0.00	0.00	0.00
12.5	0.00	0.00	0.00	0.00
6.25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)

Results of testing the antibacterial activity of ethyl acetate extracts maceration extraction using a solvent ethyl acetate-rise on the growth of Staphylococcus aureus and Escherichia coli can be seen in Table 3 and 4 show that the inhibition occurs at concentrations of 200 to 6.25 mg / ml. This is expected because the chemical compounds that are semipolar contents in Moringa leaves drawn in ethyl acetate solvent provides antibacterial activity. Chemical compounds that are semipolar classes of compounds generally are glycosides. The type of organic solvent to extract the most widely used chemical compound plant for research purposes antimicrobial is methanol, ethanol and water[19]. Chemical compounds groups that are often found to give effect as antimicrobial include phenol, acids phenolic, quinone, flavones, flavonoids, flavonol tannin and coumarin. Therefore, when compared with the results of the activity test methanol extract of leaves of Moringa, then the ethyl acetate extract gives lower activity than methanol extract due to less polar solvent ethyl acetate to attract the active chemical compounds that tend to be higher polarity as an antibacterial.

Table 3. Results of Testing of Antimicrobial Activity of Ethyl Acetate Extracts against *Staphylococcus aureus*

Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	13.00	12.50	14.00	13.16
100	12.00	11.00	12.00	11.66
50	12.00	10.00	11.00	11.00
25	10.00	9.50	10.00	9.83
12,5	8.00	7.50	9.00	8.16
6,25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)

Table 4. Results of Testing of Antimicrobial Activity of Ethyl Acetate Extracts Extracts against *Escherichia coli*

Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	12.00	11.00	11.00	11.33
100	11.00	10.00	10.00	10.33
50	10.00	9.00	10.00	9.66
25	9.00	9.00	9.00	9.00
12,5	9.00	8.00	8.00	8.33
6,25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)

Results of testing the antibacterial activity of methanol extract of the extracted storey maceration using methanol to the growth of *Staphylococcus aureus* and *Escherichia coli* can be seen in Table 5 and 6 show the highest inhibitory antibacterial activity among the three extracts of Moringa leaves that occurs at concentrations of 200 to 6.25 mg / ml. This is expected because the chemical compounds that are highly polar contained in Moringa leaves drawn on methanol provides antibacterial activity which is very strong. The type of organic solvent to extract the most widely used chemical compound plant for research purposes antimicrobial is methanol, ethanol and air. Chemical compounds group that are often found to give antimicrobial effect include phenol, acids phenolic, quinone, flavones, flavonoids, flavonol tannin and coumarin[19].

Table 5. Results of Testing of Antimicrobial Activity of Methanol Extracts against *Staphylococcus aureus*

Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	15.00	14.00	13.00	14.00
100	13.50	12.10	13.50	13.03
50	11.00	10.00	12.00	11.00
25	10.00	8.11	8.00	8.70
12,5	9.40	6.10	8.00	7.83
6,25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)

Table 6. Results of Testing of Antimicrobial Activity of Methanol Extracts Extracts against *Escherichia coli*

Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	14.00	14.00	15.00	14.33
100	13.00	11.00	14.00	12.66
50	11.00	11.00	11.00	11.00
25	10.00	9.00	9.00	9.33
12,5	9.00	9.00	9.00	9.00
6,25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)

Results of testing the antifungal activity of extracts of ethyl acetate and methanol extraction storey maceration using ethyl acetate solvent and methanol to the growth of microscopic fungus *Candida albicans* in tables 7 and 8 shows that the absence of inhibition occurred at concentrations from 200 to 6.25 mg / ml. It is estimated that both compounds as well as the most polar semipolar contained in Moringa leaves drawn by the solvent ethyl acetate and methanol do not give as antifungal activity.

Table 7. Results of Testing of Antimicrobial Activity of Ethyl Acetate Extracts against *Candida albicans*

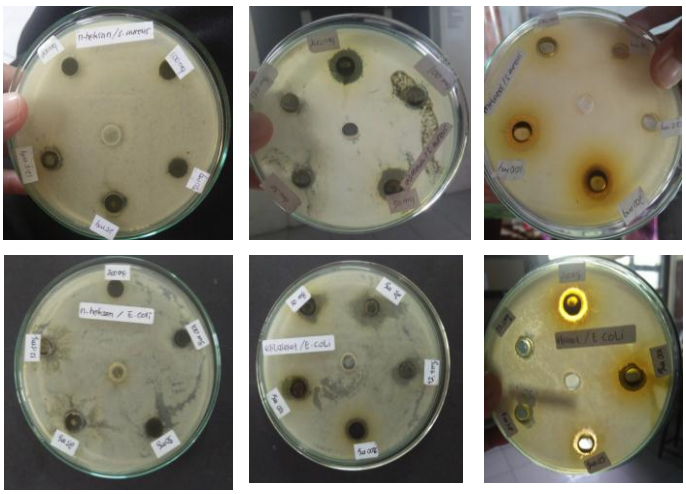
Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	0.00	0.00	0.00	0.00
100	0.00	0.00	0.00	0.00
50	0.00	0.00	0.00	0.00
25	0.00	0.00	0.00	0.00
12,5	0.00	0.00	0.00	0.00
6,25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)

Table 8. Results of Testing of Antimicrobial Activity of Methanol Extracts Extracts against *Candida albicans*

Concentration (mg/ml)	D1 (mm)	D2 (mm)	D3 (mm)	D*(mm)
200	0.00	0.00	0.00	0.00
100	0.00	0.00	0.00	0.00
50	0.00	0.00	0.00	0.00
25	0.00	0.00	0.00	0.00
12,5	0.00	0.00	0.00	0.00
6,25	0.00	0.00	0.00	0.00
Blank	0.00	0.00	0.00	0.00

Description: D: Inhibition diameter, D*: Average inhibition diameter, millimeters (mm)



Antioxidant Activity Test Results

Overall for the third Moringa leaf extract produced from the process of maceration storey, each extract provides strong antioxidant activity with IC50 values < 50 ug / ml. Results of testing the antioxidant activity of the extract ethanol extraction results storied maceration provides the most powerful antioxidant activity compared to the hexane extract and ethylacetate extract. This is expected because the chemical compounds most polar contained in Moringa leaves are extracted in methanol provides the most powerful antioxidant activity. Chemical compounds that are polar in general is a group of compounds called polyphenols, flavonoids and tannins. Many of the chemical compounds of plants that contain compounds called polyphenols have an effect as a catcher of free radicals, including flavonoids, polyphenols, tannins and phenolic terpenes[15].

Table 9. Testing Results of Antioxidant Activity of Hexane, Ethyl Acetate and Methanol Extract

Test Solution	IC_{50}Value (µg/ml)
N-Hexane Extract	35,07 µg/ml
Ethyl Acetate Extract	33,64 µg/ml
Ethanol Extract	16,40 µg/ml

Content Analysis of Chemical Compounds

The results of the analysis of chemical compounds extract hexane, ethyl acetate and methanol carried by thin layer chromatography (TLC). The stationary phase used aluminum plate TLC silica gel 60 F254 (Merck®). The mobile phase used hexane: ethyl acetate (7: 3) (pro analysis, Merck®) with visualization agent 50% sulfuric acid in methanol to display the entire class of chemical compounds contained in each extract, Liebermann-Bourchard to detect the presence of compounds triterpenes and free steroid, Dragendorff to detect alkaloid class of compounds and 5% iron (III) chloride to detect a class of polyphenolic compounds. Results variety of stains and Rf for each extract can be seen in Table 10, 11 and 12 below.

Table 10. Results of Chromatograms and RfValue of Moringa Leaf Hexane Extract

Mobile phase n-hexane-ethyl acetate (7: 3)	Visual	H_2SO_4 50% in methanol	$FeCl_3$ 5%	Lieberman-Bouchard	Dragendroff
N-HexaneExtract by Graded Maceration	-	0,90 (yl) 0,80 (gr) 0,76 (gr) 0,72 (bl) 0,70 (bl) 0,68 (pk) 0,66 (gr) 0,63 (bl) 0,60 (yl) 0,57 (gr) 0,55 (gr) 0,48 (yl) 0,43 (yl) 0,32 (gr) 0,28 (gr) 0,21 (gr) 0,13 (gr) 0,06 (br) 0,02 (br)	0,90(yl) 0,33(yl) 0,21(gr) 0,06(gr) 0,01(gb)	0,81(gr) 0,73(gy) 0,70(gr) 0,66 (pk) 0,61(gr) 0,55 (p) 0,48(yl) 0,42 (a) 0,37 (a) 0,21 (br) 0,03(gb)	0,71(gr) 0,63(gy) 0,57(gr) 0,51(gr) 0,32(gr) 0,02 (br)

Description: yl = yellow, gr = green, gb = green brown,gy = grey, pk = pink, br= brown, bl = blue

Results TLC n-hexane extract with the mobile phase n-hexane-ethyl acetate (7: 3) using a visualization agent 50% sulfuric acid in methanol provides 19 stains that showed the presence of at least 19 compounds. The appearance with 5% $FeCl_3$visualization agent gave a positive result as much as 3 green color stain showed polyphenol compounds, but at a very low Rf value. Lieberman-Bouchard gave a positive result by 4 stain green color indicates steroid compounds. Dragendroff give positive results as much as 1 positive stain brown color alkaloid.

Table 11. Results of Chromatograms and RfValue of Moringa Leaf Ethyl Acetate Extract

Mobile phase n-hexane-ethyl acetate (7: 3)	Visual	H_2SO_4 50% in methanol	$FeCl_3$ 5%	Lieberman-Bouchard	Dragendroff
Ethyl Acetate Extract by Graded Maceration	-	0,85 (gy) 0,80 (gr) 0,78 (gr) 0,75 (tg) 0,70 (gr) 0,68 (gr) 0,66 (bl) 0,60 (gr) 0,55 (bl) 0,48 (gr) 0,42 (pk) 0,38 (gr) 0,30 (bl) 0,33 (gr) 0,31 (gr) 0,28 (bl) 0,26 (gr) 0,23 (gr) 0,21 (yl) 0,15 (br) 0,07 (bl) 0,05 (yl) 0,03 (br)	0,83 (yl) 0,80 (gr) 0,73 (gr) 0,71 (gy) 0,66 (gr) 0,58 (gr) 0,53 (tg) 0,48 (gy) 0,37 (gy) 0,30 (gy) 0,21 (yl) 0,15 (br) 0,06 (gy) 0.02 (br)	0,85 (yl) 0,78 (gr) 0,75 (lg) 0,72 (yl) 0,68 (gr) 0,60 (gr) 0,55 (yl) 0,47 (gr) 0,43 (kh) 0,38 (gr) 0,28 (gr) 0,25 (gy) 0,13 (yl) 0,06 (yl) 0,02 (tg)	0,82 (gy) 0,77 (yl) 0,68 (gr) 0,66 (gr) 0,61 (yl) 0,48 (br) 0,42 (gr) 0,31 (gr) 0,26 (yl) 0,13 (br) 0,05 (gr) 0,01 (br)

Description: tg = tosca green, lg = light green, gr = green, gy = grey, yl = yellow, br = brown, gy = green yellow, pk = pink, bl = blue

Results TLC ethyl acetate extract with the mobile phase n-hexane-ethyl acetate (7: 3) using a visualization agent 50% sulfuric acid in methanol provides 23 stains that showed the presence of at least 23 compounds. The appearance with 5% $FeCl_3$visualization agent gave a positive result as 6 green color stain showed polyphenol compounds, but at a very low Rf value. Lieberman-Bouchard give positive results as 8 stain green color shows slightly polar steroid. Dragendroff give positive results as much as 6 stain which 3 stains with brown and 3 stains with yellow shows alkaloid.

Table 11. Results of Chromatograms and RfValue of Moringa Leaf MethanolExtract

Mobile phase n-hexane-ethyl acetate (7: 3)	Visual	H_2SO_4 50% in methanol	$FeCl_3$ 5%	Lieberman-Bouchard	Dragendroff
MethanolExtract by Graded Maceration	-	0,90 (yl) 0,82 (bl) 0,73 (gy) 0,71 (gy) 0,67 (yl) 0,55 (bl) 0,53 (yl) 0,51 (yl) 0,46 (gr) 0,41 (gr) 0,38 (bl)	0,83 (gl) 0,31 (gg) 0,28 (gg) 0,18 (gb) 0,05 (br)	0,90 (gy) 0,82 (gr) 0,67 (yl) 0,55 (yl) 0,46 (yl) 0,38 (gr) 0,33 (gy) 0,26 (gr) 0,03 (br)	0,53 (yl) 0,31 (gr) 0,23 (gr) 0,16 (br) 0,02 (br)

		0,33 (yl) 0,31 (yl) 0,28 (gr) 0,26 (gr) 0,23 (gr) 0,18 (gr) 0,16 (yl) 0,10 (yl) 0,05 (gr) 0,02 (br)			

Description: yl = yellow, bl = blue, gy = grey, gr = green, br = brown, gb = green brown, gl = green yellow, gg = green grey

Results TLC ethanol extract with the mobile phase n-hexane-ethyl acetate (7: 3) using a stain visualization agent 50% sulfuric acid in methanol provides 21 stains that showed the presence of at least 21 compounds. The appearance with 5% $FeCl_3$visualization agent gave 4 stain positively to the polyphenolic compounds with different types of green color. Lieberman-Bouchard gave a positive result as much as 3 stain green color indicates steroid moderately polar compounds. The appearance of Dragendroff provide positive results as much as 3 stains which 1 yellow stain and 2 brown stains indicates high polarity alkaloid compounds.

Conclusions

Screening results of Moringa leaf simplicia powder showed positive results against the class of chemical compounds alkaloids, glycosides, anthraquinone glycosides, flavonoids, steroid, tannins and saponins. The test results demonstrate the antimicrobial activity of the three extracts of Moringa leaves provide inhibition against *Staphylococcus aureus* and *Escherichia coli*, but did not provide barriers against microscopic fungus *Candida albicans*. The antioxidant activity of the extract of leaves of Moringa third show < IC50 value of 50 µg / ml and categorizing them very strong antibacterial and the highest antimicrobial activity demonstrated by methanol extract. The results of chemical analysis for each extract showed alkaloid, steroids and polyphenols chemical compounds.

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