Inhibitory Power of Moringa (*Moringa oleifera*) Seed Extract from Various Levels of Maturity on *Eschericia coli* bacteria

Rostiati Rahmatu
Agrotechnology study Program
Faculty of Agriculture
Tadulako University
Palu, Indonesia
grostiatirahmatu@gmail.com

Adehani
Agrotechnology study Program
Faculty of Agriculture
Tadulako University
Palu, Indonesia

Nur Alam
Agrotechnology study Program
Faculty of Agriculture
Tadulako University
Palu, Indonesia

Moh.Nur Sangaji
Agrotechnology study Program
Faculty of Agriculture
Tadulako University
Palu, Indonesia

Rahmi
Agrotechnology study Program
Faculty of Agriculture
Tadulako University
Palu, Indonesia

***Abstract*— This study aims to obtain the maturity level of certain Moringa seeds that can inhibit the development of E. coli bacteria. This study used a Completely Randomized Design (CRD) consisting of one factor with the treatment of the maturity level of moringa seeds namely K1 = young seed extract, K2 = medium seed extract, K3 = old seed extract and each treatment repeated 5 times to obtain 15 units. The results showed the inhibition of Moringa Oleifera Lamk Seed Extract from Different Maturity Levels against E. Coli Bacteria gave a very significant effect on water content, ash content, protein content, vitamin C levels, phenol levels, and bacterial tests. Moringa seed extract was effective in inhibiting the growth of E. coli bacteria at the maturity level of medium moringa seeds obtained the highest value of 23.50 mm, young moringa seeds obtained a value of 22.70 mm, old moringa seeds obtained a value of 21.20 mm. All maturity levels of Moringa seeds, young, medium and old can produce inhibitory zones in very strong categories.**

Keywords*— *Moringa and Escherichia coli Bacteria.

I. INTRODUCTION

The Moringa plant is a plant that has many species and much potential, so it is very familiar to the Indonesian tongue, especially to the people of Central Sulawesi. Arora et al. (2013), there are 750,000 species on earth, and only 10% have the potential to be used. This plant is known, because of its enormous benefits for the human body, especially for health. Rahayu (2019) that Moringa leaves and other parts are widely used for herbal ingredients, which are processed in the form of tea, powder, and capsules. Moringa leaves and seeds are utilized because they contain essential components such as vitamin C 220 mg/100gr (fresh leaves and seeds) and 17.3 mg/100gr (dry condition). Other components are alkaloids, flavonoids, phenolics, triterpenoids/steroids, and tannins, which function as cancer drugs, anti-tumor, antiseptics, lower cholesterol, antioxidants, antibacterial, anti-fungal, and others (Arora et al. (2013). Palupi et al.'s research results (2021), the extract of the n-hexane fraction of coreopsis polyzoan acts as the most potent antibacterial because it can inhibit the growth of Staphylococcus aureus and E.coli. These extracts are almost the same as the results of a study by Rostiati et al. (2018). Moringa seed extract can also inhibit the growth of E. coli.

According to Nasir et al. (2010), Moringa seeds contain 7.9% water, 30.8% oil, 38.3% protein, 6.5% ash, and 4.5% fiber. Tall. The excellent content contained in Moringa seeds has yet to be utilized optimally because many seeds are just wasted. So far, Moringa seeds have only been used as raw material for vegetables. This condition is fascinating to study by utilizing Moringa seed extracts of different maturity levels against the inhibition of E.coli bacteria.

Escherichia coli is a bacterium that is part of the normal microflora in the digestive tract of humans and warm-blooded animals. E. coli refers to heterotrophic bacteria that obtain food in the form of organic substances from their environment because they cannot prepare the organic substances they need themselves. Organic substances are obtained from the remains of other organisms. These bacteria decompose organic substances in food into inorganic substances, namely CO2, H2O, energy, and minerals. In the environment, these putrefactive bacteria function as decomposers and provide plant nutrients (Kusuma, 2010).

II. MATERIALS AND METHODS

This research was conducted at the Agrotechnology Laboratory, Faculty of Agriculture, Tadulako University, Palu. This research was conducted from April to October 2018. The tools used for this research were a blender, porcelain cup, LTE-UK model OP250-U oven at 1050C, desiccator, PW 254 analytical balance, 100 ml volumetric flask, 100 mL Erlenmeyer, Automatic Distillation vapodest and titrator model vapodest 45s GERHARDT, turbosog digestion, kjedahl flask, dropping fins, test tube, autoclave, furnace model L24/11/B 13000C, centrifuge, wellbore pipe, colony counter scan 500, ose needle, and microscope. The materials used for this study were young, medium, and old Moringa seeds taken in Tondo Village, Mantikulore District, Palu City, at an altitude of 0-150 m above sea level (in Muh Saadilah's thesis, 2017), E. Coli, selenium, H2SO4, 25 ml ethanol, distilled water, NaOH, HCl, H3BO3, starch, iodine, crystal violet, safranin, iodine, sodium agar, Sodium Broth, and chloramphenicol.

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This study used a completely randomized design (CRD) which consisted of one factor with the treatment of the maturity level of Moringa seeds, namely K1 = young seed extract, K2 = medium seed extract, K3 = old seed extract, and each treatment was repeated five times to obtain 15 units.

The implementation of this research began with a survey to obtain the location of the Moringa growing area. Two Moringa trees were taken as samples at an altitude of 0-150 m above sea level (Saadilah, 2017) in Tondo Village. Mantikulore District, Palu City. Sampling time is in the morning by taking Moringa fruit from four points (north, south, west, and east). Then the fruit is sorted into young fruit (100 fruit), medium fruit (100 fruit), and old fruit (100 fruit). The fruit from the field was brought to the Laboratory, peeled the Moringa fruit flesh, and then. The seeds were taken and then blended until smooth (≤ 3 minutes), after which they were analyzed for water, ash, protein, Vitamin C, Phenol, and bacterial test.

The data obtained is analyzed using variance F test 5%. If the treatment shows a significant effect, a further test is carried out using the 5% Tukey HSD test.

III. Results and Discussion

Water content

The maturity level of young Moringa seeds has a higher water content compared to the maturity levels of other Moringa seeds. This is because young seeds contain much water, causing a higher water content. Conversely, old seeds contain little water, so the water content is low. The water content in foodstuffs also determines the freshness and durability of these foodstuffs. The high water content will cause microorganisms such as bacteria, molds, and yeast to grow because water is the primary nutrient microorganisms need to grow (Winarno, 1997).

Table 1. Tukey HSD Test Results in 5% Moisture Content of Moringa Seed Extract from Different Maturity Levels

Treatments	Mean	HSD 5%
K1	80.34c	
K2	74.61b	5,05
K3	48.13a	

Ash Content

The degree of maturity of Moringa seeds has a very significant effect on the ash content. This is because the old seeds contain many minerals, causing a higher ash content. On the other hand, easy seeds contain few minerals, so the ash content is low. Ash content can show the total minerals in a food ingredient. Determining total ash content can be used for various purposes, including determining whether the processing is good or not, knowing the material used, and determining the nutritional value of a food ingredient (Astuti, 2011).

Table 2. Tukey HSD 5% Test Results in Ash Content of Moringa Seed Extract from Different Maturity Levels.

Treatment	Mean	HSD 5%
K3	17.52b	
K2	13.62a	3.51
K1	12.04a	

Protein Content

The maturity level of old Moringa seeds has a higher protein content than other Moringa seeds' maturity levels. This is because the drier the material, the higher the protein. Supriadi et al. (2013) reported that a decrease in water content would increase the protein content in the material. The use of heat in foodstuffs can reduce the percentage of water content, which increases the percentage of protein content. The drier the material, the higher the protein content.

Table 3. Tukey HSD 5% Test Results in Protein Content of Moringa Seed Extract from Different Maturity Levels

Treatment	Mean	HSD 5%
K3	1667c	
K2	6.59b	1.01
K1	2.95a	

Vitamin C levels

The maturity level of young Moringa seeds has a higher level of vitamin C compared to the maturity levels of other Moringa seeds. This is because the seeds easily contain higher vitamin C than other seeds. Vitamins are organic compounds needed by the body for normal metabolic processes and growth. The human body cannot make vitamins in sufficient quantities. Therefore, it must be obtained from the food consumed (Inayati, 2010).

Vitamin C functions to protect white blood cells from enzymes that are released when digesting bacteria they have swallowed, synthesize of steroid hormones from cholesterol, helps in the formation of collagen, cures canker sores, wound healing processes as well as the body's resistance against infection and stress and as an antioxidant. (Sibagariang, 2010).

Table 4. Tukey HSD Test Results of 5% in Vitamin C Levels of Moringa Seed Extract from Different Maturity Levels.

Treatment	Mean	HSD 5%
K1	2.05b	
K3	1.21a	0.67
K2	1.1a	

Phenol Levels

The maturity level of young Moringa seeds has a higher phenol content compared to the maturity levels of other Moringa seeds.

Phenols and polyphenols contained in plants play an essential role in long-term health, reducing the risk of chronic and degenerative diseases. Phenol compounds can

have antioxidant, antitumor, and antibiotic activities (Apak et al, 2007). Besides that, it also functions as a defense against ultraviolet radiation or self-protection from pathogens, parasites, predators and gives color to plants (Dai and Mumper, 2010).

Phenol compounds are widely used as antioxidants to prevent heart disease, reduce inflammation, reduce the incidence of cancer and diabetes (Khoddami et al, 2013). Phenol compounds have the lowest melting point and a slightly pungent odor. It is also easily soluble in most organic solvents (aromatic hydrocarbons, alcohols and ketones) and slightly less soluble in aliphatic hydrocarbons. Phenols form azeotropic mixtures with water and other substances. Phenol can function to inhibit the activity of bacteria such as Staphylococcus aureus, Bacillus subtilis, and Listeria monocytogenes) and three Gram-negative ones (Escherichia coli, Pseudomonas aeruginosa, and Salmonella enteritidis).

Table 5. Tukey HSD Test Results in 5% Phenol Content of Moringa Seed Extract from Different Maturity Levels

Treatment	Mean	HSD 5%
K1	16.28c	0.37
K2	13.34b	
K3	10.29a	

Bacterial Test

The positive control had a higher inhibition rate than the maturity level of the Moringa seeds. The maturity level of the seeds is increasing, but the maturity level of young and old moringa seeds has decreased inhibition.

The decrease in the number of bacteria inhibited by Moringa seeds indicated that the cotyledons of the Moringa seeds contained 3 important components, namely the antimicrobial substance 4 α L-amnosyloxy benzyl isothiocynate, ben oil and flocculants. Substance 4 α L-amnosyloxy benzyl isothiocynate, ben oil and flocculants are antiseptic, namely a compound that can kill or inhibit the growth or activity of other microorganisms (Erfandi, 2015). The same thing is from the research of Sunartia, Lahaya, Latiefa, and Gondiponb (2022), 40% Moringa leaves can inhibit the activity of S.aureus and E.coli bacteria

Table 6. Tukey HSD Test Results of 5% Moringa Seed Extract from Different Maturity Levels against E.coli Bacteria

Treatment	Mean	HSD 5%
K0	27.93c	17.12
K2	23.50ab	
K1	25.35b	
K3	21,20a	

IV. Conclusion

Based on the results of the discussion, the Inhibitory Power of Moringa Seed Extract from Different Maturity Levels Against E.Coli Bacteria had a very significant effect on water content, ash content, protein content, vitamin C content, phenol content, and bacterial tests. Moringa seed extract was effective in inhibiting the growth of E.coli bacteria at medium maturity level Moringa seeds obtained the highest value of 23.50 mm, young Moringa seeds obtained a value of 22.70 mm, old Moringa seeds obtained a value of 21.20 mm. All maturity levels of Moringa seeds, namely young, medium and old, can produce very strong inhibition zones.

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