

SPECIFIC AND NON-SPECIFIC QUALITY PARAMETERS OF ETHANOL EXTRACT FROM MORINGA SEEDS (*Moringa oleifera* L.)

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ABSTRACT

Moringa (*Moringa oleifera* L.) is one of the plants that has the potential and is used in traditional medicine. One of the parts of the moringa plant that can be used is moringa seeds, because it contains essential nutrients such as protein, vitamins A, B, K, C, as well as minerals such as calcium, magnesium and iron. In addition, moringa seed extract also has secondary metabolite compounds such as alkaloids, tannins, flavanoids and saponins that have the potential to be medicinal ingredients. This study aims to determine the test results of specific parameters including identity, organoleptic, dissolved compounds in water solvents, dissolved compounds in ethanol solvents and non-specific test results of extracts including moisture content, ash content, and insoluble acid ash content. The design used in this study is descriptive and the data analysis used is univariate analysis. Moringa seed ethanol extract (*Moringa oleifera* L.) was obtained by the extraction method for 5 days and then remacerated. The specific parameters of moringa seed extract include yield of 14.35%, organoleptic yellowish creamy color, distinctive smell of moringa seeds, thick shape and bitter taste, dissolved compounds in water solvent $70,40\% \pm 0,50\%$ and dissolved compounds in ethanol $20,90\% \pm 0,87\%$. The non-specific quality parameters of moringa seed ethanol extract include a moisture content of $16,94\% \pm 0,72\%$, an ash content of $4,92\% \pm 0,08\%$ and an insoluble acid ash content of $0,21\% \pm 0,01\%$. The research results of specific quality parameters of yield, identity, organoleptic, value of water-soluble compounds, soluble compounds in ethanol extract standard. The research results of non-specific parameters of moringa seed extract are total ash content and insoluble ash content of standard extract acids, while the results of moisture content do not meet the standard.

Keywords : Specific parameters, non specific parameters, moringa seeds extract

INTRODUCTION

The utilisation of natural ingredients as traditional medicine in Indonesia has been going on for centuries, as reflected in various historical relics. Authentic evidence of this can be found in a number of ancient manuscripts on lontar leaves Husodo in Java, Usada in Bali and Lontakrak from South Sulawesi. In addition, traditional documentation such as Serta Primbon Jampi, Serat Racikan Boreh Wulang Dalem, and Borobudur Temple reliefs depicting the compounding of plant-based herbal medicine also corroborate the long tradition of natural-based medicine in the archipelago (Sukandar, 2006).

Traditional medicine refers to preparations derived from natural materials such as plants, animals, minerals, or their extract forms (galenic), either singly or in combination, which have been used for generations in accordance with community norms (BPOM RI, 2014). *Moringa* (*Moringa oleifera* L.) is one of the plants used as traditional medicine. All parts of the *Moringa* plant including leaves, seeds, roots and flowers can be used. *Moringa*

leaves contain protein, minerals, beta-carotene and antioxidant compounds that are used in traditional medicine. Moringa seeds have many benefits because they contain most of the oil with high quality fatty acid composition (oleic acid > 70%) and once purified have significant resistance to oxidative degradation (Leone et al., 2016). The nutritional compound content of moringa seeds needs to be extracted with further processing technology. The technology is the extraction method in the processing of traditional medicinal raw materials.

Extraction is a technique of separating soluble compounds from insoluble materials using liquid solvents (Kemenkes RI, 2017). The process of isolating bioactive compounds from plant minerals aims to determine the amount of yield obtained, which is strongly influenced by the extraction method applied. Thermal extraction methods include reflux, soxhletation, digestion, infusion, decoction, while non-thermal extraction methods include maceration and percolation (Kiswandono, 2011). The maceration method is used to extract samples that are relatively impervious to heat. This method is done by simply immersing the sample in a solvent for a period of time, usually 24 hours without heating (Kiswandono, 2011). Ethanol 70% is used as a solvent because it has an intermediate polarity, so it is effective in extracting bioactive compounds with various levels of polarity, both polar and semi-polar to non-polar (Surya & Luhurningtyas, 2021).

Based on previous research, ethanol extract of moringa seeds using maceration method was reported to contain secondary metabolite compounds such as alkaloid, tannin, saponin and flavonoid compounds (Dising & Pasau, 2022). This proves that the ethanol extract of Moringa seeds (*Moringa oleifera* L.) has potential as a medicinal ingredient, but the extract results need to be tested for quality parameters to obtain a standardised extract. Extract quality parameters include specific and non-specific parameters. Specific parameters include extract identification, organoleptic, and soluble compound content in certain solvents. Meanwhile, non-specific parameters include moisture content, total ash content, acid insoluble ash content, heavy metal contamination, and total plate count (Kemenkes RI, 2017).

There has been no research on the specific and non-specific quality parameters of moringa seed ethanol extract, so researchers are interested in conducting research on "Specific and Non-Specific Quality Parameters of Moringa Seed Ethanol Extract (*Moringa oleifera* L.)".

METHODS

Tools and materials

The equipment used in this study includes analytical scales, grinder, mesh 60 sieve, bowl spoon, plastic, brush, measuring cup jar, funnel, stirring rod, filter paper, flannelette, porcelain cup, waterbath, incubator, ointment pot, label, stopwatch, desiccator, metal staple, crucible, stove or furnace, erlenmeyer, ash-free filter paper, spiritus lamp, magnetic stirrer, clogged flask.

The materials used in this study include 1.5 kg of dried simplisia of moringa seeds, 400 grams of moringa seed simplisia powder and 4000 mL of 70% ethanol, 2 grams of moringa seed thick extract, 1 gram of moringa seed thick extract, ash from total ash determination, moringa seed thick extract, 2.5 grams of moringa seed thick extract, 50 mL of distilled water, 2.5 mL of chloroform, 2.5 grams of moringa seed thick extract, 50 mL of 95% ethanol.

Preparation of moringa seed powder

Dry simplisia of moringa seeds was obtained from Moringa Organik Indonesia, Inc., Blora Regency. The study used 1.5 kg of dried simplisia of moringa seeds that had been

sorted. The dried simplisia of moringa seeds used were separated from impurities and other foreign materials. Then the dried simplisia of moringa seeds that have been selected are ground using a grinder and then sieved using a mesh 60 sieve.

Extraction by maceration method

Moringa seed simplisia powder weighing 400 g was added with 70% ethanol solvent as much as 75% of 4000 mL, namely 3000 mL. The extraction process was carried out by soaking for the first 6 hours accompanied by periodic stirring, then continued to stand for 5 days where the extract was entered every day. After 5 days of filtering with flannel cloth and filter paper, remaceration was carried out for 2 days with a new solvent as much as 25% of 4000 mL, namely 1000 mL.

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Specific quality parameters of extracts

Identity test is done by describing the name of the extract including the name of the extract, the Latin name of the plant, the plant part used, the Indonesian name of the plant, to the plant's identity compound (Depkes RI, 2000). Determination of this identity was carried out by macroscopic identification which was then compared with the literature on Moringa seeds.

Organoleptic test is done by describing the colour, smell, shape and taste of the extract using the senses of sight, smell and taste (Depkes RI, 2000).

Soluble compounds in aqueous solvents were carried out by weighing 2.5 grams of moringa seed extract and then macerating it using 50 mL of water-chloroform LP solution (a mixture of 50 mL of water and 2.5 mL of chloroform P) for 24 hours. The maceration process was carried out in a clogged flask with periodic shaking in the first 6 hours, then left for 18 hours, before filtering. 20 mL of filtrate was evaporated to dryness in a pre-weighed and pre-tempered porcelain dish, the residue was heated at 105 °C to a fixed weight (Baso et al., 2022).

Soluble compounds in ethanol solvent were carried out by weighing 2.5 grams of extract and macerating it in ethanol 50 mL of 95% ethanol for 24 hours using a stoppered flask. Shaking was carried out periodically for the first 6 hours, then the mixture was left for 18 hours, then filtered and 20 mL of filtrate was evaporated to dryness using a pre-weighed and calibrated porcelain cup, the residue was heated to a fixed weight at 105 °C (Baso et al., 2022).

Non Specific quality parameters of extracts

The water content test using the gravimetric method was carried out by weighing approximately 1 gram of thick moringa seed extract, putting it in a porcelain cup that was previously weighed. Next, it was baked at 105°C for 5 hours and weighed. The drying process was continued with weighing every one hour until the weight difference between two consecutive weighings did not exceed 0.25% (Najib et al., 2017)

The total ash content test was carried out by weighing approximately 1.5 grams, the sample was put into a crucible that had previously been weighed and incinerated. Incinerated on a furnace with a temperature of 600 °C until the charcoal runs out after which it is cooled and weighed (Najib et al., 2017).

Determination of acid-insoluble ash content boiled the ash obtained in 25 mL of dilute hydrochloric acid solution for 5 minutes. The insoluble fraction is then filtered using ash-free filter paper, and the residue left behind is rinsed with hot water. Then the filtered ash and filter paper were put in a crucible, then the crucible was heated in a furnace at 600 °C until ash was formed and then weighed the results (Depkes RI, 2000).

RESULT AND DISCUSSION

This study used samples of moringa seed simplisia (*Moringa oleifera* L.) obtained from Moringa Organik Indonesia, Inc., Blora Regency. Moringa seed simplisia was ground into powder which was then macerated with a ratio of ethanol solvent and sample 1: 10 (Sari et al., 2021). The extracts obtained were then tested for specific and non-specific quality parameters. Specific quality parameters of extracts include identity, organoleptic, soluble compounds in water solvents and soluble compounds in ethanol solvents. The test results of specific parameters of the extract can be seen in table 1.

Table 1. Results of Specific Quality Parameters of Moringa Seed Ethanol Extract (*Moringa oleifera* L.)

No.	Specific Quality	Result
1.	Yield	14,35 %
2.	Identity	This research is in accordance with the results of plant identification using Moringa (<i>Moringa oleifera</i> L.) which belongs to the moringaceae family and the parts used are seeds (Semen)
3.	Organoleptic	Colour : Yellowish beige Odour : Moringa seeds Form : Thick Flavour : Bitter
4.	Compounds dissolved in aqueous solvents	70,40% ± 0,50%
5.	Compounds dissolved in ethanol solvent	20,90 % ± 0,87%

The thick extract obtained was 50.40 grams with a yield of 14.35%. Based on previous research, it shows that the reflux method using hexane solvent produces a higher yield of 6.157% compared to the maceration method which only produces 5.256% on 20-25 g powder (Kiswandono, 2011). In the research of Wigunarti et al (2019), it was found that 96% ethanol extract of moringa seeds using maceration method with 250 g powder produced a yield value of 5.509%. According to Ghazali and Yasin (2016), moringa seed samples obtained at 60°C showed the highest extract yield percentage of 36.37%. The difference in yield is influenced by the amount of powder, solvent and extraction method. Reflux tends to be more effective because high temperatures increase the solubility and diffusion of substances (Fajri & Daru, 2022). A high yield indicates an increase in the quantity of extract obtained (Depkes RI, 2000).

Determination of the identity of simplisia and extracts is carried out to obtain objective and specific identification of the name of the material used (Depkes RI, 2000). This study uses moringa plants according to the results of identification (*Moringa oleifera* L.), family Moringaceae, the part used in this study is the seed (semen). Moringa seeds used have the same characteristics, yellowish white inner seeds (kernels) with white epidermis, kernels with the size of soybean seeds (Fajri & Daru, 2022).

Organoleptic testing of the results showed that the ethanol extract of moringa seeds had a yellowish beige colour, the yellowish beige colour was obtained because in previous

studies moringa seeds contained secondary metabolite compounds in the form of flavanoids (Dising & Pasau, 2022). Flavanoids are compounds that have red, blue and yellow dyes in every plant, this is what causes the beige colour of moringa seeds (Masykuroh et al., 2024). Moringa seed extract has a thick form, has a distinctive odour and a bitter taste, the bitter taste in moringa seed extract is due to the presence of saponin and alkaloid compounds that give a bitter taste (Foild et al., 2021). Previous research tests on the specific characteristics of moringa seed simplisia, reported that moringa seeds have a creamy colour with a bitter taste (Swandono et al., 2024).

Testing the levels of soluble compounds in water and ethanol solvents aims to identify the amount of compounds that can dissolve in water as a polar solvent, as well as in ethanol which represents a solvent with lower polarity (Azis, 2011). The test results showed that the soluble compound in water of moringa seed ethanol extract was $70.40\% \pm 0.50\%$ while the soluble compound in ethanol solvent of moringa seed extract was $20.90\% \pm 0.87\%$. These results can be seen that water soluble compounds are more than soluble compounds in ethanol, this shows that active substances in extracts tend to be more easily extracted in water than ethanol (Utami et al., 2017). This is in line with Moringa seeds, which are known to have water-soluble proteins with low molecular weights, namely 4-alpha-4-ramnosiloxy-benzyl-isothiocinate, which produces many positive charges when dissolved (Haslinah, 2016). In addition, the total soluble compounds in both solvents did not exceed the maximum limit of 100%, in accordance with the established standards (Saifudin et al., 2011).

Non-specific quality parameters of moringa seed ethanol extract (*Moringa oleifera* L.) were tested including water content, ash content and total ash content. The test results of non-specific quality parameters of moringa seed ethanol extract (*Moringa oleifera* L.) can be seen in table 2.

Table 2. Results of Non-specific Quality Parameters of Moringa Seed Ethanol Extract (*Moringa oleifera* L.)

No.	Non-specific quality parameters	Result (σ)	Standard
1.	Moisture content	$16,94\% \pm 0,72\%$	$<10\%$
2.	Ash content	$4,92\% \pm 0,08\%$	$<16,6\%$
3.	Acid insoluble ash content	$0,21\% \pm 0,01\%$	$<0,7\%$

Non-specific quality parameters of water content are used to determine the remaining water content in the extract after the drying or thickening process (Saifudin et al., 2011). The test results of water content in moringa seed extract were $16.94\% \pm 0.72\%$. Water content is strongly influenced by the duration and intensity of drying, the drier the simplisia, the lower the water content (Najib et al., 2017). Previous research testing the water content of moringa seed simplisia obtained results of 11.9%, where these results did not meet the requirements for water content in 10% simplisia (Kiswandono, 2011). The same thing was obtained by research on the water content of chlab seed extract with the result of 13.78%, the water content in the extract exceeding 10% (Isnawati et al., 2013). High moisture content increases the risk of extract damage due to the growth of microorganisms and triggers the decomposition of active compounds through enzymatic reactions, which can reduce extract stability (Saifudin et al., 2011). The range of moisture content also depends on the type of extract, thick extracts usually contain up to 5-30% moisture (Anggraini and Khabibi, 2022).

Determination of total ash content is done by heating the sample at high temperature until all organic components and their derivatives decompose and evaporate, leaving a residue of organic and mineral compounds. Meanwhile, acid insoluble ash content testing aims to assess the presence of inert contaminants such as soil particles or sand in the extract (MOH RI, 2000). Previous research tested the total ash content in moringa seed simplisia with a percentage of 7.75% (Swandono et al., 2024). The results of ash content in

moringa seed ethanol extract obtained $4.9213\% \pm 0.08\%$, these results meet the requirements of the total ash standard which is no more than 16.6% (Depkes RI, 2008). According to Kusumaningrum (2013), the higher the ash content, the more inorganic material in the product.

According to the Indonesian Ministry of Health (2008), the ash content insoluble in acid should not exceed 0.7%. The test results showed that the ash content of the sample was $0.2167\% \pm 0.01\%$, thus fulfilling the standard. Low ash content is expected, as this parameter reflects the presence of inorganic contaminants, including metals that are resistant to high temperatures (Isnawat et al., 2013).

CONCLUSION

Specific quality parameters of moringa seed ethanol extract (*Moringa oleifera* L.) obtained the results of the identity of moringa seeds used included in the Moringaceae family, latin name *Moringa oleifera* L., organoleptic test obtained moringa seed extract has a yellowish beige colour, thick shape, with a smell of has and bitter taste, test of compounds dissolved in water by $70.40\% \pm 0.50\%$ and test of compounds dissolved in ethanol solvent by $20.90\% \pm 0.87\%$. Non-specific quality parameters of moringa seed ethanol extract (*Moringa oleifera* L.) obtained the results of moisture content test of $16.94\% \pm 0.72\%$, ash content test of $4.92\% \pm 0.08\%$ and acid insoluble ash content of $0.2\% \pm 0.01\%$.

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