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108 <https://oamjms.eu/index.php/mjms/index> Scientific Foundation SPIROSKI, Skopje, Republic of Macedonia Open Access Macedonian Journal of Medical Sciences. 2022 Jan 03 10(T8):108-113. <https://doi.org/10.3889/oamjms.2022.9499> eISSN: 1857-9655 Category: T8 –"APHNI: Health Improvement Strategies Post Pandemic Covid-19" Section: Pharmacology Formulation, Evaluation of Physical Properties, and In Vitro Antioxidant Activity Test of Moringa Leaf (*Moringa oleifera* L.) Ethanolic Extract Capsules Annisa 1* , Sucianingsih 1 , Riswan 1 , Emelda 1 , Kusumawardhani 1 , Fauzi 1 , Daru Estiningsih 1 , Muhammad Abdurrahman Munir 1 , Marisa Yansiani 1 , Hamam Hadi 2 , Mika Matsuzaki 3 1Department of Pharmacy, Faculty of Health Science, Alma Ata University, Yogyakarta, Indonesia; 2Department of Public Health, Graduate School, Faculty of Health Sciences, Alma Ata University, Yogyakarta, Indonesia; 3Department of Human Nutrition, International Health, Johns Hopkins Bloomberg School of Public Health, Baltimore, Maryland, USA Abstract BACKGROUND: Supplements that contain antioxidants may enhance prevention and treatment effects of a wide range of diseases including COVID-19.

Quercetin, a flavonoid compound, is a natural antioxidant that can neutralize free radicals. AIM: The present study was conducted to formulate Moringa leaf (*Moringa oleifera* L.) ethanol extract capsules and to determine the quercetin antioxidant activity levels of Moringa ethanol extract capsule formulations. MATERIALS AND METHODS: tested total levels solutions concentrations 20, 60, 70, 100 ppm thin-layer graph y method. of properties 96% Moringa ethanol capsules moisture test, angle repose granule property capsule uniformity and disintegration test.

Antioxidant test the 2,2-diphenyl-1-picryl-hydrazyl-hydrate method with two samples, namely, 96% Moringa leaf ethanol extract capsules with formulas I, II, and III, quercetin

as a comparison. RESULTS: results the of Moringa ethanol capsules that II (polyvinylpyrrolidone 50 mg) had good physical properties. Testing the antioxidant activity of capsules of the ethanol extract Moringa formulas II, III, obtained values 44.0 ppm, ppm, ppm, and 4.80 ppm, respectively.

CONCLUSION: evaluation the extract of Moringa formula met parameters a good evaluation requirement had strong activity from acquisition the IC50 antioxidant of Moringa extract may able improve immune and clinical trials need to be carried out on patients to become candidates for prevention and therapeutic supplement for a range of diseases including COVID-19. Edited by: Sinisa Stojanoski Citation: Fatmawati A, Sucianingsih D, Riswan R, Emelda E, Kusumawardhani N, Fauzi R, Estiningsih D, Munir MA, Yansiani M, Hadi H, Matsuzaki M.

Formulation, Evaluation of Physical Properties, and In Vitro Antioxidant Activity Test of Moringa Leaf (Moringa oleifera L.) Ethanolic Extract Capsules. Open-Access Maced J Med Sci. 2022 Jan 03; 10(T8):108-113. <https://doi.org/10.3889/oamjms.2022.9499>
Keywords: Antioxidant; Moringa oleifera L; Capsule; COVID-19 *Correspondence: Annisa Fatmawati, Department of Pharmacy, Faculty of Health Science, Alma Ata University; Yogyakarta, Indonesia. E-mail: annisafatma20@almaata.ac.id Received: 13-Oct-2021 Revised: 21-Nov-2021 Accepted: 02-Dec-2021 Copyright: © 2022 Annisa Fatmawati, Depita Sucianingsih, Revalina Riswan, Emelda Emelda, Nurul Kusumawardhani, Rizal Fauzi, Daru Estiningsih, Muhammad Abdurrahman Munir, Marisa Yansiani, Hamam Hadi, Mika Matsuzaki Funding: This research did not receive any financial support Competing Interest: The authors have declared that no competing interest exists Open Access: This is an open-access article distributed under the terms of the Creative Commons Attribution- NonCommercial 4.0 International License (CC BY-NC 4.0) Introduction table due compounds [3], [4]. Flavonoid are [3]. Recent states flavonoids improve immune in degenerative and infectious by such COVID-19 [5].

In to activity, compounds also antiviral that potentially severe from diseases COVID-19 The virus affects immune which trigger diseases, and infectious of virus [7]. occurs characterized by white blood cells that will respond to production cytokines. will to cell so they trigger [8]. According recent by et al . quercetin have potential be against SARS-CoV-2 which may prevent severe illnesses from COVID-19 by reducing inflammation [9]. Furthermore, studies suggested quercetin may be able to reduce inflammation caused by COVID-19 and help prevent hospitalization due to COVID-19. Moringa (Moringa oleifera Open Access Maced J Med Sci. 2022 Jan 03; 10(T8):108-113.

109 Moringa Moringa Moringa Moringa formulate Moringa (DPPH) method. Methods Materials The used Moringa powder from Market, Yogyakarta, 96% (Bratachem), shell

DPPH (Merck), quercetin (Merck), aerosil, polyvinylpyrrolidone (PVP-K30), lactose, acid, pro toluene, acetate, silica GF-254 The used Pyrex Oven UN Spectrophotometry Scientific UV-Vis), Camag Chromato graph y (TLC) Scanner. Collection and extraction procedure Moringa powder M. oleifera was obtained Beringharjo Yogyakarta September Moringa powder 500 g dissolved 96% 2.5 L 5), for × h, remuneration time.

filtrate was over water or using a rotary 60 o C the extract formed be by the yield [12]. Formulation Moringa extract (MLECs) made mixing Moringa ethanol with powdered into mortar pestle homogeneous. dissolve polyvinylpyrrolidone with 96% ethanol until it dissolves evenly and mix it into the containing ethanol of Moringa leaves a The granules then using a number 10 mesh sieve and dried at 40–60°C for 1 h in the oven. The granules were then sieved using a number mesh and put capsule number 0 [16]. The MLEC formulation is listed in Table 1.

Furthermore, evaluation of physical properties includes moisture content (MC) (%), angle of rest (degrees), flow rate weight (g), breakdown test (minutes) [17]. Table 1: Formulation capsule of Moringa leaf extract

Ingredients	Function	Formula F I (mg)	F II (mg)	F III (mg)
Moringa extract	Active compound	250	250	250
Aerosil	Glidant	10	10	10
Polyvinylpyrrolidone	Binders	25	50	75
Avicel	Disintegrant	50	50	50
Lactose	Filler	165	140	110
Total		500	500	500

Information: FI (Formula I), FII (Formula II), and F III (Formula III). Determination of total flavonoid levels in Moringa leaf capsules by TLC densitometry 20–100 o 10 acetate: [18].

Test of antioxidant activity of MLEC with DPPH method Preparation 100 ppm solution carried by 10 mg DPPH then dissolved 96% to mark a mL volumetric flask and as a mother liquor placed in a dark glass bottle. The blank solution was made using 1.0 mL of ppm solution dissolved 96% 110 <https://oamjms.eu/index.php/mjms/index> ethanol a mL flask, solvent the mark. stand 30 min measure absorbance a of nm. 100 ppm quercetin mother liquor was prepared by weighing 10 mg of powder dissolved 96% while and in 100 mL flask to make 100 ppm quercetin solution. Furthermore, variations concentrations 2 ppm, ppm, ppm, 8 ppm, 10 ppm made.

of concentrations much 1 ml, added mL 100 ppm solution, and for 30 min in a dark room [19]. % Reduction = $((\text{Abs control} - \text{Abs assay}) / \text{Abs Control}) \times 100\%$ Abs: Absorbance Equation The of reduction percentage. The solution made a concentration 1000 ppm weighing capsule powder to calculation sample preparation and dissolved using 96% ethanol as solvent. Add ethanol the in 25 mL flask, until Then a of solution namely, 200; 300; and ppm the liquor of 1000 ppm. Solutions of various concentrations as much as 1 ml, each added 1.0 mL of 100 ppm DPPH solution, vortexed, and incubated for 30 min in the flask covered.

The reduction of (Moringa Leaf Capsule), was by Equation that be by regression linier The value calculated entering number as in linear equation $bx + a$) [19]. Results

Evaluation of physical properties The extract was by calculating percentage as Table 2 shows results the of Moringa ethanol capsules several such as calculation % granule of granule rate, weight and capsule time with formulas using different amounts of PVP [17]. Table 2: Results of evaluation of Moringa leaf ethanol extract capsules **Formula MC** (%) AR (degrees) FR (g/s) WU (g) BTT (min) I 1.75 ± 0.06 35.17 ± 2.14 4.72 ± 0.16 0.4656 ± 0.5412 4.12 ± 0.15 II 3.62 ± 0.00 30.27 ± 2.67 5.93 ± 0.11 0.4678 ± 0.5436 2.42 ± 0.30 III 4.57 ± 0.00 38.07 ± 2.00 2.95 ± 0.03 0.4665 ± 0.5419 5.25 ± 0.20 Information: MC: Moisture content, AR: Angle of rest, FR: Flow rate, WU: Weight uniformity, BTT: Breakdown time test.

Total **flavonoid levels in Moringa leaf capsules by** TLC densitometry Spectro-densitometric quercetin performed fluorescence nm Fig ure 1), at theory at nm Table 3 that MLECs contain a high flavonoid 8.97% (w/w), compared **to a thick Moringa leaf extract containing a total** flavonoid not (w/w) as The of regression between grade series the (Fig ure 2) obtained linear regression $Y = 101.21x - 508.19$ $R^2 = 0.9849$. linear equation be to calculate **the total flavonoid content** as quercetin. **Antioxidant activity of capsule** The DPPH wavelength was 517.16 nm. The results of the antioxidant capsules **of Moringa leaf extract** compared with quercetin.

Table 4 shows antioxidant in MLEC and standards the of strong antioxidants (< 50 ppm) [13]. Discussion Moringa was obtained the or method heating, method decomposition the due to Immersion ethanol has antioxidant activity than n-hexane and ethyl acetate solvent [14]. with is sui table **method and solvent for extracting high-quality antioxidant raw** from Moringa for development of and products [21].

The TLC method the of total flavonoid of Moringa capsules chosen because is and when to the spectrophotometric The performance chromatograph y method also option the of content, this has applications, as sample cleaning and the need for expensive solvents and a column period. TLC the of content **because it is a versatile analytical technique that requires** inexpensive instrumentation and expertise [18]. Moringa vitamins, proteins, essential amino carotenoids, and that be [21]. have the of Moringa a in infection pathogens SARS-CoV The modulator immune stimulator activity of Moringa leaves makes it a sui table natural supplement immune **Open Access Maced J Med Sci. 2022 Jan** 03; 10(T8):108-113. 111 against [23].

study an in silico - based showed possibility identifying a inhibitor SARS-CoV-2 from sources, such as Moringa . **The antioxidant properties of the Moringa extract may be**

able to enhance the immune system secondary after Other components in Moringa leaves that may have beneficial impact the system a include pterygospermin and apigenin [24]. Table 3: Total flavonoid capsule formula II Sample replication AUC Quercetin levels % (w/w) X ± SD 1 1618.30 8.49 8.97 ± 0.44 2 1835.77 9.35 3 1761.90 9.06 Das et al M. oleifera M.

oleifera in Moringa [26], Moringa 10–18 with poor dietary choices [28], [29]. Quercetin one the compounds found in Moringa leaves, with strong antioxidant activity. Table 4: Result of antioxidant activity of Moringa leaf capsule Sample Concentration (ppm) Absorbance % inhibition IC 50 (ppm) Category Formula I 100 0.365 56.17 44.0 Very strong 200 0.354 57.49 300 0.349 58.09 400 0.254 69.43 500 0.194 76.72 Formula II 100 0.365 56.17 40.2 Very strong 200 0.346 58.45 300 0.333 59.98 400 0.215 74.20 500 0.185 77.76 Formula III 100 0.368 55.77 46.4 Very strong 200 0.352 57.73 300 0.345 58.49 400 0.213 74.40 500 0.200 75.96 Quercetin 2 0.558 32.97 4.8

Very strong 4 0.403 51.60 6 0.362 56.49 8 0.290 65.18 10 0.252 69.71 In vitro Moringa Moringa stress [31], [32]. Figure 2: Standard curve of quercetin on TLC densitometry Figure 1: The wavelength of the quercetin compound in TLC densitometry 11 <https://oamjms.eu/index.php/mjms/index> Medicinal since or thousands years have important in the of diseases humans. development drugs natural especially medicinal plants, continues to be researched to provide alternative therapies and disease prevention efforts. Moringa from M. oleifera extract have potential be supplement maintain health their content. is known by results the activity Moringa leaf in vitro which strong antioxidant activity.

It is necessary to test the antioxidant activity of Moringa leaf capsule supplements in humans to determine its pharmacological effectiveness clinically. Conclusion All formulas the parameters physical and very antioxidant category. of MLEC formula II is the best capsule evaluation parameters and had a very strong antioxidant activity IC50 of 40.2 ppm, and the total flavonoid content was 8.97 ± 0.44% (w/w). The properties MLEC be for the of and 2 as as improving immune We future to the of Moringa supplements preventing illnesses viral diseases like COVID-19.

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