International Journal of Biological and Pharmaceutical Sciences Archive

ISSN: 0799-6616 (Online)

Journal homepage: https://ijbpsa.com/

(Research Article)

Check for updates

Assessing the effectiveness of turmeric and tamarind leaves extract cream as anti-aging *in vivo*

Bambang Admadi Harsojuwono *, I Ketut Satriawan and Sri Mulyani

Department of Agroindustrial Technology, Faculty of Agricultural Technology, Udayana University, Badung (80361), Bali, Indonesia.

International Journal of Biological and Pharmaceutical Sciences Archive, 2023, 05(02), 068–075

Publication history: Received on 21 April 2023; revised on 31 May 2023; accepted on 03 June 2023

Article DOI: https://doi.org/10.53771/ijbpsa.2023.5.2.0049

Abstract

Turmeric and tamarind leaves extract creams have potential as anti-aging agents, but their use has never been studied *in vivo*. Therefore the purpose of this study was to determine the effectiveness of turmeric and tamarind leaves extract cream in maintaining skin quality *in vivo*. This research was carried out by applying turmeric and tamarind leaf extract cream to 24 volunteers (having received approval from the ethical commission). Variables observed included sebum content, pore size, pigment content, moisture or moisture content, elasticity and collagenase content. The results showed that turmeric and tamarind leaves extract cream can increase sebum content up to 31.1%, reduce pore size to 0.017 mm, increase pigment content up to 13.2%, moisture up to 54.2%, elasticity up to 21.4% and collagenase content up to 37.3%.

Keywords: Anti-aging; Turmeric extract; Tamarind leaves extract; Cream; *In vivo*

1. Introduction

The development of cosmetics made from natural raw materials is important in addition to not having side effects, it is also in great demand by the community. The results of the research by Layli et al [1], show that to improve appearance, health and satisfaction, most women prefer traditional cosmetics derived from natural ingredients rather than synthetic ingredients. Tafsia [2] showed that 82.22% of women chose traditional cosmetics, while 17.78% did not use traditional cosmetics.

Natural ingredients in the form of turmeric and tamarind leaves have been developed as anti-aging ingredients to inhibit skin damage or aging [3]. Extrinsic skin aging (chronologic skin aging), is characterized by the onset of wrinkles, xerosis, dullness, and the appearance of benign tumors on the skin. The appearance of wrinkles is due to reduced collagen content because it is degraded along with the increase in matrix metalloproteinase (MMP) enzymes and cytokine release [4]. Furthermore, the accumulation of reactive oxygen species (ROS) plays an important role in the skin aging process, namely damaging enzymatic antioxidants and non-enzymatic antioxidants [5]. Prevention of extrinsic skin aging is done by inhibiting the increase in matrix metalloproteinase enzymes and providing antioxidants [6]. Therefore, the provision of creams containing antioxidants will be able to defend the skin from free radicals so the process of damaging and aging of the skin is inhibited.

Mulyani et al. [3], showed that turmeric extract and tamarind leaves cream has the ability as anti-collagenases and antioxidants, with vitamin C content of 3.5 mg / 100 g and phenolic reaching 0.12 mg GAE/ml [7]. According to Thring, et al. [8], phenolic compounds with a range of 0.05-0.26 mg GAE/ml, has great potency as antioxidants, so they have prospects for development as anti-aging. *In vivo* Veronica officinalis extract cream with IC50=105.93 µg/ml, applied for 56 days was able to reduce 66% wrinkles on the skin [9].

* Corresponding author: Bambang Admadi Harsojuwono

The problem is that the use of cream from turmeric and tamarind leaves extract has never been studied *In vivo*, especially related to effectiveness as an anti-aging. Therefore, the purpose of this study was to determine effectiveness of turmeric and tamarind leaves extract cream as an anti-aging *In vivo*.

2. Material and methods

2.1. Material

Research materials include turmeric, tamarind leaves, ethanol, stearic acid, triethanolamine (TEA), Virgin Coconut Oil (VCO), mineral oil, cetyl alcohol, methyl paraben, propyl paraben, sodium metabisulfite, EDTA, aquadest, moisturizer conditioner, placebo cream (base cream). The equipment used includes Centrifuge, Vortex, Oven, Hitachi 912 Spectrophotometer, and skin analyzer.

2.2. Method

2.2.1. Making turmeric and tamarind leaves extract cream

Simplicity turmeric and tamarind leaves moisture content of 10 ± 1 % were made into powder size of 80 mesh. Furthermore, each simplicity powder was extracted with 96% ethanol solvent macerated with a ratio of ingredients: solvent (1: 6). Maceration is carried out twice each for 24 hours, the filtrate is evaporated using an evaporator, until turmeric extract and tamarind leaf extract are obtained, then mixed with the ratio of turmeric extract: tamarind leaf extract = 10: 2.

Table 1 Turmeric extract and tamarind leaf cream formula

Material	**Weight**
Stearic Acid (g)	10.9
VCO (g)	3.64
Mineral oil (g)	2.27
Cetyl Alcohol (g)	0.91
Span 80 (g)	2.80
Tween 80 (g)	2.20
Moisturizer conditioner (g)	10
Turmeric and tamarind leaf extract (g)	0.2
Water addition up to (g)	100

The oil phase consists of stearic acid, VCO, mineral oil,cetyl alcohol, moisture conditioner, tween 80, and span 80. The water phase is aquadest, all materials are weighed and put into beaker glasses according to their phases. The oil phase and water phase are heated to 65°C in a water bath. Heating is done until all ingredients melt then removed from the water bath and inserted turmeric extract and tamarind leaves are (10:2). Further, the water phase is added to the oil phase while mixing until a homogeneous cream with HLB 10 is obtained. The formula as shown in Table 1.

2.2.2. Testing the effectiveness of turmeric and tamarind leaves extract cream on the skin

Selection of research subjects and determination of volunteers

The study subjects were twenty-four (24) women aged 45 - 65 years without a history of serious illness or allergy to cosmetics or pregnant women. All subjects had signed a statement letter of consent given by the researcher (Informed Consent) before the study began.

Treatment of volunteers

The study used a randomized group design, with two types of treatment, namely given turmeric tamarind leaves extract cream and placebo cream. The study was supervised by a cosmetic dermatologist and approved by an ethics committee on human testing. The study sample were twenty-four female subjects aged 45 - 65 years without a history of serious

illness or allergies to cosmetics or pregnant women. The subjects who were smeared with cream were the volunteers' arms which were divided into 2 groups, namely placebo and turmeric extract and tamarind leaf treatment, each group of 12 volunteers. All research subjects had given a written statement in Informed Consent before the study was conducted. The subjects were smeared with cream containing turmeric extract and tamarind leaves for 28 days. Clinical evaluation and measurements were performed at H0 (before treatment), H14 (day 14) and H28 (day 28). The anti-wrinkle effect of turmeric extract, tamarind leaf and placebo cream on skin wrinkles were evaluated using Skin Analyzer. Observations include sebum content, pigment, pore size, acne count, elasticity, moisture, and collagen content.

2.2.3. Data analysis

The data obtained were tabulated and analyzed descriptively quantitatively and qualitatively.

3. Results and discussion

The effectiveness or ability of turmeric and tamarind leaves extract cream in its application to volunteers is shown from the results of sebum content, pigment, elasticity, pore size, number of acnes, moisture, and collagen content as described as follows.

3.1. Sebum content

The results of the sebum content analysis in Table 2 showed that it tended to be an increase in sebum content in skin given turmeric and tamarind leaves extract cream for 28 days compared to placebo and control. On day 28 the skin sebum content in the control treatment was 35.2%, placebo of 35.3%, and turmeric and tamarind leaves extract cream treatment was 36.2%.

Basically, sebum is a yellowish oily substance produced by oil glands in the skin. According to Senka [10], this skin sebum consists of various components, namely fatty acids, squalene, cholesterol, and wax esters which function to maintain skin moisture, protect the skin from UV and prevent bacterial growth.

Wulandari [11] explained that too little sebum content causes the skin to dry and susceptible to premature aging. Meanwhile, excessive sebum content and accompanied by accumulated of dead skin cells, the pores of the face will be clogged and trigger bacteria to develop around the hair follicles. Table 4 shows that turmeric extract and tamarind leaves cream can increase the activity of oil glands to produce sebum so that its levels increase. This is accordance with the opinion of Hou et al [12], who explain that anti-aging cosmetics increase sebum levels and improve skin dryness.

Table 2 The sebum content (%) during application of the turmeric and tamarind leaves extract cream on the skin

Group	Day 0	Day 14	Day 28
Control	35.2	35.2	35.2
Placebo	35.3	35.3	35.3
The treatment uses cream from turmeric and tamarind leaves extract	35.3	35.8	36.2

3.2. Pigment content

Table 3 Pigment content (%) during application of turmeric and tamarind leaves extract cream on the skin

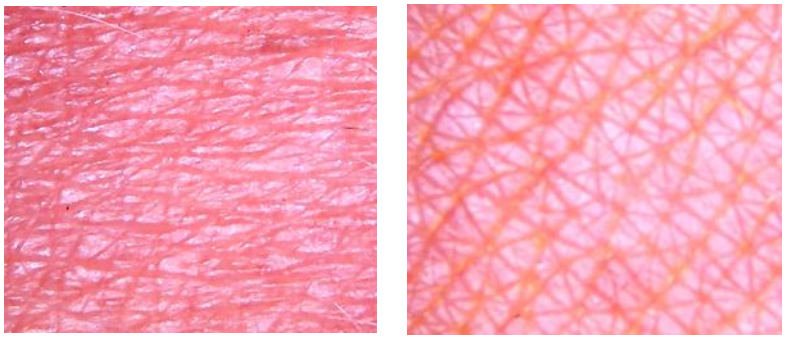
Group	Day 0	Day14	Day 28
Control	8.1	8.2	8.1
Placebo	8.2	8.4	8.3
The treatment uses cream from turmeric and tamarind leaves extract	8.1	11.3	13.2

The results of pigment content analysis can be seen in Table 3, which shows an increase in brighter pigment in skin treated with turmeric and tamarind leaves extract cream for 28 days compared with the skin applied placebo and control. On the 28th day, the skin pigment content in the control treatment was 8.1%, placebo was 8.3%, while the

turmeric and tamarind leaves extract cream treatment was 13.2%. Meanwhile, Figure 1 shows the appearance of skin pigment with placebo application and cream application of turmeric and tamarind leaves extract.

Basically, skin pigment is affected by the presence of melamine substances produced by melanocyte cells. When melanocyte cells are damaged, it will interfere the production of melamine. Based on Table 5 and Figure 1, showed that the skin which were given turmeric and tamarind leaves extract cream appears to have increased pigment content and is brighter and clearer than placebo. This shows that turmeric and tamarind leaves extract cream which contain lots of antioxidants can inhibit melanocyte cell damage. Baumann and Allemann [13, 14] explain that antioxidants can inhibit the activity of tyrosinase enzyme therefore the melanin production is inhibited by decreasing the content of o-quinone which makes bright skin in normal people.

According to Rattanawiwatpong et al [15], vitamin C has anti-aging effects on the skin, not only as a powerful antioxidant and mediator of photo-damage and melanogenesis, but also plays a role in supporting collagen biosynthesis and stability, which provides a renewal and anti-inflammatory effect. Topical use of 15% L-ascorbic acid combined with 1% alpha-tocopherol has been shown to provide more significant protection against sunburn skin cell formation compared to L-ascorbic acid or 1% alpha-tocopherol alone. Meanwhile, according to Kembuan et al [16], in melamine synthesis, antioxidant compounds require a lot of oxygen and can convert dark oxidized melanin into reduced melanin that is slightly pale in colour.



a b

Figure 1 Skin pigment display (a) placebo application (b) turmeric and tamarind leaves extract cream application

3.3. Elasticity

The results of elasticity analysis can be seen in Table 4, which shows that skin given turmeric and tamarind leaves extract cream increased its elasticity compared to control treatment and placebo. On day 28, skin elasticity in the control treatment was 15.3%, placebo of 16.6% while the turmeric treatment and tamarind leaves extract cream increased to 21.4%.

Basically, skin elasticity is the ability of the skin to stretch and return to its original shape. Halodoc [17] explains that reduced elasticity causes sagging, wrinkled and rough skin. It is further explained that the elasticity, strength and suppleness of the skin are highly dependent on the collagen content of the skin. Ganceviciene et al [18] explain that there are two main groups of agents that can be used as components of anti-aging creams, namely antioxidants and cell regulators.

Table 4 Elasticity (%) during application of turmeric and tamarind leaves extract cream on the skin

Group	Day 0	Day 14	Day 28
Control	15.3	15.4	15.3
Placebo	15.4	16.5	16.6
The treatment uses cream from turmeric and tamarind leaves extract	15.3	19.3	21.4

According to Bissett et al. [19], antioxidants, such as vitamins, polyphenols and flavonoids reduce collagen degradation by reducing FR concentrations in tissues. Vitamins C, B3, and E are the most important antioxidants due to their ability to penetrate the skin through their small molecular weight. This is according to the opinion of Halodoc [17], cream

containing antioxidants can absorb into the deep layers of the skin so antioxidants can neutralize free radicals and increase collagen production. Ganceviciene et al [18] explain that cell regulators, such as retinol, peptides, and growth factors (GF), have a direct effect on collagen metabolism and affect collagen production.

3.4. Pore size

The results of pore size analysis can be seen in Table 5 which shows that skin pores smeared with turmeric and tamarind leaves extract cream, decreased in size compared to placebo and control. On the 28th day, the skin pores in the control treatment measured of 0.022 mm, placebo of 0.021 and turmeric extract and tamarind leaves cream of 0.017 mm.

Table 5 Pore size (mm) during application of turmeric and tamarind leaves extract cream on the skin

Group	Day 0	Day 14	Day 28
Control	0.021	0.022	0.022
Placebo	0.022	0.021	0.021
The treatment uses cream from turmeric and tamarind leaves extract	0.021	0.019	0.017

Principally, skin pores are openings (holes) of pilosebaceous follicles. The size of skin pores will increase along the increase of age, this is one of the characteristics of skin aging [20]. Reduction in pore size of skin applied to turmeric extract and tamarind cream for 28 days in Table 7, according to research by Iskandar et al [21]. Iskandar et al. [21] showed that patchouli oil micro emulsion preparations as anti-aging can reduce pore size, accelerate the increase in water content, skin smoothness, reduce blemishes and wrinkles [22]. Meanwhile, Park et al [23] showed that the use of an antiaging mask for 4 weeks with moisturizing and antioxidant content was able to shrink the size of pores and eliminate wrinkles. The results of Hanum and Laila's [24] research show that anti-aging containing *andalima*n alcohol extract can maintain skin moisture, skin flatness, reduce pore size and reduce the number of dark spots and wrinkles.

3.5. Number of acnes

The results of the analysis of the number of acnes as shown in Table 8. Table 8 shows that the absence of acne in the control treatment, placebo and application of turmeric and tamarind leaves extract cream on the skin of the volunteers.

Fundamentally, acne is a disorder of the skin associated with excess oil (sebum) production [25]. Table 6 shows that there was no acne on the volunteers, possibly due to volunteers doing regular skin treatments or cleanses. According to Wulandari [11], acne will appear when excess oil production and skin pores are clogged with dead skin cells and dirt. Meanwhile, Hanum and Laila [24] explained that the use of anti-aging containing *andaliman* alcohol extract for 4 weeks can prevent and cure acne.

Table 6 The numbers of acnes during the application of turmeric cream and tamarind leaves extract on the skin

Group	Day 0	Day 14	Day 28
Control	0	0	0
Placebo	0	0	0
The treatment uses cream from turmeric and tamarind leaves extract	0	0	0

3.6. Moisture

The results of moisture analysis are shown in Table 7, which shows that skin moisture in turmeric and tamarind leaves extract cream treatment tends to be higher than control and placebo treatment. The control showed no change in humidity until day 28, while the placebo treatment increased by 49.3% and the turmeric extract cream treatment and tamarind leaves caused an increase of 54.2%.

Basically, skin moisture is a condition that is influenced by the water content in the skin. Table 9 shows that skin applied with turmeric and tamarind leaves extract cream for 28 days had the highest humidity. This is caused by turmeric and tamarind leaves extract cream contain moisturizing ingredients. According to Prasetya [26], moisturizing ingredients function to soften and smoothen the skin, provide hydration and as a protector from UV rays. If the skin moisture level is low, the skin becomes dry or xerosis cutis. The results of Hanum and Laila's [24] research show that anti-aging

containing *andaliman* alcohol extract can maintain skin moisture. This is in accordance with the research from Tricaesario and Widayati [27], which showed that almond cream effectively increases skin moisture. According to Hou et al [12], the water content in the stratum corneum (SC) in normal skin is approximately around 10% in the outer layer and about 30% in the deeper layer. A decrease in water content in SC to less than 10% will cause the skin to look scaly, rough, and dry.

Table 7 The moisture (%) during application of turmeric and tamarind leaves extract cream on the skin

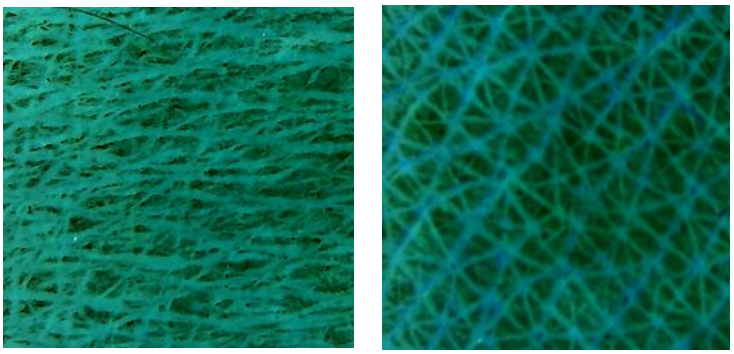
Group	Day 0	Day 14	Day 28
Control	17.2	17.3	17.3
Placebo	17.3	31.7	49.3
The treatment uses cream from turmeric and tamarind leaves extract	17.2	36.2	54.2

3.7. Collagen content

The results of collagen content analysis as shown in Table 8 and Figure 2. Table 10 shows skin collagen levels in control, placebo and treatment of turmeric and tamarind leaves extract cream. Meanwhile, Figure 2 shows the appearance of skin collagen on placebo application and application of turmeric and tamarind leaves extract cream.

Table 8 Collagen levels (%) during application of turmeric and tamarind leaves extract cream on the skin

Group	Day 0	Day 14	Day 28
Control	25.3	25.4	25.3
Placebo	25.4	26.4	26.4
The treatment uses cream from turmeric and tamarind leaves extract	25.4	34.2	37.3



a b

Figure 2 Skin collagen display on (a) placebo application (b) turmeric and tamarind leaves extract cream application

Basically, collagen is a type of fibrous and insoluble protein that is abundant in the human body. This protein is the main foundation of the skin, and the connective tissue that gives it structure and strength [28]. Collagen functions as adhesive that glues the whole body into one whole. In addition, collagen also has benefits in restoring injured body tissues and flexing tendon and skin tissues. According to Savitri and Efendi [29], supple and firm skin is skin that has enough collagen.

Table 10 shows that control and placebo for 28 days showed no noticeable increase in collagen, while skin that received application of turmeric extract and tamarind leaves cream showed high improvement, originally 25.4% increased to 37.3%. According to Savitri and Efendi [29], the presence of antioxidant compounds in the cream causes inhibition of collagenase enzyme activity, so collagen damage is also inhibited. This has an impact on the strength and structure of the skin increasing thus increasing the elasticity and firmness of the skin. This can also be seen in Figure 2, which shows

the collagen structure became more regular and compact with thicker collagen fiber sizes [25]. According to Bissett et al. [19], creams containing antioxidants such as vitamins, polyphenols and flavonoids can reduce collagen degradation by reducing the concentration of FR in tissues. Meanwhile, the presence of cell regulators can improve collagen metabolism so collagen production increases [18].

4. Conclusion

Turmeric and tamarind leaves extract cream can increase sebum content up to 31.1%, reduce pore size to 0.017 mm, increase pigment content up to 13.2%, moisture up to 54.2%, elasticity up to 21.4% and collagenase content up to 37.3%.

Compliance with ethical standards

Acknowledgments

Acknowledgments are extended to the directorate general of higher education and research & technology, the ministry of education and culture for providing research and publication grants, and the leadership of Udayana University for facilitating this research and publication.

Disclosure of conflict of interest

There is no conflict of interest between the authors and any other party in this publication

Statement of ethical approval

The author states that this research has received approval from the parties involved, especially the volunteers who are the object of this research.

Statement of informed consent

This publication has been approved by the parties that support this research.

References

[1] Layli N, Dilla F, Nurfita. Opinion survey on the use of traditional cosmetics among STIKES Medika Nurul Islam students. Journal of Nursing Science. 2021; 7(2):41-48.

[2] Tafsia CL. Opinion survey on the use of traditional cosmetics among female students of the Faculty of Pharmacy, Sanata Dharma University, Yogyakarta. Thesis. Faculty of Pharmacy, Sanata Dharma University, Yogyakarta. 2017. https://adoc.pub/opinion-survey-about-use-traditional-cosmetic-in-k.html

[3] Mulyani S. Antioxidant synergism of turmeric and tamarind leaves (*Curcuma domestica* Val. - *Tamarindus indica* L.) as active ingredients in cream. Dissertation. Doctoral Study Program of Agricultural Sciences, Faculty of Agriculture, Udayana University. Denpasar. 2017

[4] Yaar, M., Gilchrest B.A. Biochemical and molecule changes in photo aged skin. Photo damage. Blackwell Science. 2008; 168-179.

[5] Joshi LS and Pawar HA. Herbal cosmetics and cosmeceuticals, natural products Chemistry and Research, 2015; 3(2):1-3.

[6] Ardhie AM. 2011. Free radicals and the role of antioxidants in preventing aging. Medician. 2011; 24(1): 4–9.

[7] Mulyani S, Harsojuwono BA, Antara NS, Kencana IN. An assessment of antioxidant characteristics from different ratios of turmeric and tamarind (*Curcuma domestica* Val.-*Tamarindus indica* L.) leaf extracts. Australian Journal of Basic and Applied Sciences, 2016; 10(14):347-353. www.ajbasweb.com/old/ajbas/2016/September/347-353.pdf

[8] Thring TSA, Hili P and Naughton DP. Anti-collagenase, anti-elastase and anti- oxidant activities of extracts from 21 plants. BMC Complementary and Alternative Medicine. 2009; 9:27. http://www.biomedcentral.com/1472-6882/9/27

[9] Lee HY, Ghimeray AK, Yim JH, Chang MS. Antioxidant, collagen synthesis activity in vitro and clinical test on anti-wrinkle activity of formulated cream containing veronica officinalis extract. Journal of Cosmetics, Dermatological Sciences and Applications. 2015; 5:45-51. http://www.scirp.org/journal/jcdsa accessed 20 January 2016

[10] Senka. What is sebum and why is it needed? 2021 Dec 21. [cited 2023 April 10]. Available from https://www.senka.id/senka-stories/apa-itu-sebum

[11] Wulandari LR. What is sebum? Get to know its function for skin health. 2022 Feb 2. [cited 2023 April 10]. Available from https://hellosehat.com/Jadi-kulit/perawatan-kulit/apa-itu-sebum/

[12] Hou X, Wei Z, Zouboulis CC and Ju Q. Aging in the sebaceous gland. Front Cell Dev Biol. 2022; 10: 909694. Doi: 10.3389/fcell.2022.909694

[13] Baumann L, Allemann IB. Depigmenting Agents. In: Baumann L, Saghari S, Weisberg E, editors. Cosmetic Dermatology Principles and Practice (Second Edition). New York: McGraw-Hill Co. 2009; 284-285.

[14] Baumann L, Allemann IB. Antioxidants. In: Baumann L, Saghari S, Weisberg E, editors. Cosmetic Dermatology Principles and Practice (Second Edition). New York: McGraw-Hill Co. 2009; 298

[15] Rattanawiwatpong P, Wanitphakdeedecha R, Bumrungpert A, Maiprasert M. Anti-aging and brightening effects of a topical treatment containing vitamin C, vitamin E, and raspberry leaf cell culture extract: A split-face, randomized controlled trial. Journal of cosmetic dermatology. 2020; 19(3):671-676. https://doi.org/10.1111/jocd.13305

[16] Kembuan MV, Wangko S, Tanudjaja GN. The role of vitamin C on skin pigmentation. Journal of Biomedicine, Supplements. 2012; 4(3):13-17

[17] Halodoc. Antioxidant function for skin. 2018 Sep 5. [cited 2023 April 10]. Available from https://www.halodoc.com/artikel/fungsi-antioksidan-untuk-kulit.

[18] Ganceviciene R, Liakou AI, Theodoridis A, Makrantonaki E and Zouboulis CC. Skin anti-aging strategies. Dermato-Endocrinology. 2012; 4(3):308–319. http://dx.doi.org/10.4161/derm.22804

[19] Bissett DL, Miyamoto K, Sun P, Li J, Berge CA. Topical niacinamide reduces yellowing, wrinkling, red blotchiness, and hyper pigmented spots in aging facial skin. Int. J. Cosmetic Sci. 2004; 26:231-8. http://dx.doi.org/10.1111/j.1467-2494.2004.00228.x

[20] Lee S, Cherel M, Gougeon S, Jeong E, Lim JM, Park SG. Identifying patterns behind the changes in skin pores using 3-dimensional measurements and K-means clustering. Skin Res Tech. 2021; 3:1-97. Doi: 10.1111/srt.13082

[21] Iskandar B, Tartilla R, Lukman A, Leny, Surboyo MDC. Anti-aging activity test of patchouli oil micro emulsion (*Pogostemon cablin* Benth.). Pharmaceutical Magazine. 2022; 7(1):52-64. https://doi.org/10.24198/mfarmasetika.v7i1.36464

[22] Vedamurthy M. Antiaging therapies. Indian J. Dermatol Venereol Leprol. 2006; 72:183-186

[23] Park M, Kim H, Kim S, Lee J, Kim S, Byun JW, Hwang-Bo J, Park KH. Changes in skin wrinkles and pores due to long-term mask wear. Skin Research & Technology. 2021; 27(5): 785-788. https://doi.org/10.1111/srt.13019

[24] Hanum TI and Laila L. Evaluation of anti-aging and anti-acne effect of andaliman (*Zanthoxylum acanthopodium* DC.) ethanolic extract peel off gel mask. Asian J. Pharmaceutical and Clinical Research. 2018; 11(1): 90-93. Http://Dx.Doi.Org/10.22159/Ajpcr.2018.V11s1.26576

[25] Azmi N. 2022. 7 types of acne and various ways to deal with them. Hellosehat. 2022 July 5. [cited 2023 April 10]. Available from https://hellosehat.com/Jadi-kulit/jerawat/tipe-jerawat/

[26] Prasetya NL, Get to know the benefits of a moisturizer for facial skin. Aido. 2020 Sept 10. [cited 2023 April 10]. Available from https://aido.id/health-articles/kenali-benefit-moisturizer-bagi-kulit-wajah/detail

[27] Tricaesario C, Widayati RI. Effectiveness of 4% almond oil cream on skin moisture levels. Diponegoro Medical Journal. 2016; 5(4). Http://ejournal-s1.undip.ac.id/index.php/medico

[28] Quamila A. The benefits of collagen for skin and hair health plus its food sources. Hellossehat. 2021 August 5. [cited 2023 April 10]. Available from https://hellosehat.com/Jadi-kulit/perawatan-kulit/untung-kolagen/

[29] Savitri ES and Efendi A. Antioxidant activity and inhibition of the silver nanoparticle collagenase enzyme using red algae (*Gracilaria verrucosa*). Proceedings of SEMNAS BIO 2021, Padang State University. ISBN : 2809-8447

[30] Chatikun, M., and Chiabchalard, A. Thai plants with high antioxidant levels, free radical scavenging activity, anti-tyrosinase and anti-collagenase activity. BMC Complementary and Alternative Medicine. 2017; 17(1), 1–9.