



Antioxidant Activity of Facial Gel Moisturizer Formulation From Various Plants: A Literature Review

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ABSTRACT: Antioxidants are compounds that can neutralize free radicals, which can damage normal cells, fats, and proteins. Various methods, such as DPPH, ABTS, and FRAP, are used to evaluate the capacity of antioxidant compounds against reactive oxygen species (ROS). Skin ageing, caused by free radicals, pollution, and ultraviolet rays, can be mitigated by using antioxidants, one of which is found in cosmetic preparations such as moisturizers. This study aims to determine the antioxidant activity of various plant extracts in the formulation of a moisturizer gel. The method used is a literature review, with sources from the Google Scholar and PubMed databases, which includes articles from 2014 to 2024, resulting in 9 eligible articles out of 20 screened. The review result showed that plants such as star fruit leaf extract (*Averrhoa bilimbi* L.) and green tea leaf (*Camellia sinensis*) have potent antioxidant activity. Starfruit Wuluh (*Averrhoa bilimbi* L.) extracted with 70% ethanol had an IC₅₀ value of 24.78 ppm, while green tea leaf extract (*Camellia sinensis*) with 70% ethanol showed an IC₅₀ value of 5.62 ppm in the gel moisturizer preparation. A high IC₅₀ indicates that the compound contains antioxidants that can protect the skin from free radicals, which are formulated into a moisturizing gel. In conclusion, star fruit leaf extract (*Averrhoa bilimbi* L.) and green tea leaf extract (*Camellia sinensis*) are highly effective ingredients in moisturiser gel products, as they provide significant antioxidant protection to the skin.

Keywords: Antioxidant; Cosmetics; Free Radical; Moisturizer.

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INTRODUCTION

Antioxidants are compounds that can neutralize or absorb free radicals so that they can prevent damage caused by free radicals to normal cells, fats, and proteins. Amthyoxidans have a stable molecular structure, allowing them to donate electrons to molecules and break free radical chain reactions (Pratiwi et al., 2023). DPPH, ABTS, and Ferric Reducing Antioxidant Power (FRAP) are methods used to measure the activity of reactive oxygen species (ROS). ABTS and DPPH differ in ROS activity, whereas FRAP increases ROS activity by the synthesis of Ferri, the leading oxidative agent. The hydrogen atom transfer method is used to measure antioxidant activity by donating hydrogen atoms, such as those measured by ORAC, TRAP, and β -carotene. The electron transfer method is used to reduce ROS activity through electron transfer, including FRAP, CUPRAC, FIC, DMPD, and others (Pisoschi et al., 2016).

As we age, our skin becomes slower to regenerate and eventually shows signs of ageing. Dull, rough, wrinkled skin, dark spots, and reduced elasticity characterize ageing. Skin ageing is caused by free radicals, pollution, and ultraviolet rays (Dewiastuti & Hashanah, 2016). Free radicals can be produced by the body's metabolism, smoking, and by free radical-causing agents found in food and other substances. Free radicals produced in the body can be neutralized by antioxidants present in the body under normal conditions. However, if free radicals in the body continue to increase and cannot be neutralized, antioxidants from outside the body are needed. The need for antioxidants from outside the body can be obtained through the use of cosmetic preparations (Ozil, 2014).

Cosmetics are materials or preparations used on the outside of the human body (epidermis, hair, nails, lips, and external genital organs) or teeth and oral mucous membranes to clean, fragrance, change appearance, and/or improve body odour or protect or maintain the body in good condition (BPOM, 2019). Moisturizer is a basic skincare product, alongside face wash and sunscreen, among other cosmetic preparations. Therefore, it is essential to use a moisturizer to care for our skin. Moisturizer has a gel-like texture that is easily absorbed by the skin, is easy to apply, and leaves no residue after use. Moisturizer gel is considered more effective in moisturizing facial skin than serum and toner products. Moisturizer gel has a mechanism of action through four basic mechanisms that restore water content in the skin, namely occlusive, humectant, hydrophilic matrix, and photoprotection.

Occlusive agents function to reduce transepidermal water loss (TEWL) by forming a hydrophobic barrier layer over the skin surface, thereby preventing the evaporation of water from the subcutaneous tissue and trapping water in the uppermost layers of the skin (Levin et al., 2011). Humectants are hygroscopic substances that attract water and moisture. When humectants are present on the skin, water from the dermis is absorbed into the epidermis. Emollients are chemicals that fill the spaces between corneocytes and provide a sense of softness and pliability (Lee, T., & Friedman, 2016).

Today's moisturizers contain skin-repairing ingredients in addition to the traditional moisturizing components. The most common ingredients are ceramides, free fatty acids, and cholesterol, which help replace deficient lipids in skin diseases characterized by barrier disruption, such as eczema and psoriasis (Zeichner & Del Rosso, 2016). The composition of ceramides in moisturizing preparations distinguishes them from facial toners and serums.

The solution to protect the skin from free radicals lies in antioxidants. Therefore, efforts and treatments are needed to maintain skin health and beauty (Lestari et al., 2020). Gel-shaped moisturizers containing natural ingredients are a viable option because they can hydrate and repair skin exposed to free radicals, utilizing flavonoids that function as antioxidant compounds. The purpose of this literature review is to find out plants that have antioxidant activity in gel moisturizer preparations.

METHODS

The method used in this article is a literature review of original articles in Indonesian and English collected through databases such as Google Scholar and PubMed

published from 2014 to 2024 on the topic of the antioxidant activity of facial gel moisturizer formulas from various plants. This literature review used keywords such as antioxidants, cosmetics, moisturizers, and free radicals. There were nine articles that met the criteria out of 20 articles that were screened. The inclusion criteria comprised original research articles that focused on the formulation of gel moisturizer preparations containing plant extracts, provided quantitative antioxidant activity data, and were available in full text in either Indonesian or English. The exclusion criteria included articles that did not address gel moisturizer formulations, investigated only plant extracts without gel formulation, lacked antioxidant activity data, were review articles, were not available in full text, were published outside the 2014–2024 period, or were written in languages other than Indonesian or English. The initial search identified 20 articles, which were subsequently screened based on the inclusion and exclusion criteria, resulting in 9 articles that met all eligibility criteria.

RESULT AND DISCUSSION

Nowadays, along with the exploration of plants that have pharmacological effects, the development of cosmetic preparations containing natural ingredients is increasing rapidly. Moisturizer gel is an emerging and widely used cosmetic. Moisturizer gel is a product that aims to improve skin hydration. There are many moisturizing products commercially available today, but it should be noted that the selection of moisturizers depends on each person's skin needs and suitability. Trans Epidermal Water Loss (TEWL) is the ability of the facial skin to draw water to hydrate the subcutaneous and epidermis of the skin. Some ingredients that are humectants and can attract water to the skin include glycerin, sorbitol, propylene glycol, hyaluronic acid, sodium, and protein, petroleum, paraffin, dimethicone, cyclomethicone, and mineral oil are some ingredients that can reduce Trans Epidermal Water Loss (TEWL) occlusive (Adianingsih et al., 2022).

The extraction process to obtain thick extracts in the review of plants using the maceration method. Maceration is one method of separating compounds by immersing the sample using an organic solvent at a certain temperature. In the maceration method, there are two ways, namely maceration with one type of polar solvent, fractionated to obtain polar, semi-polar, and non-polar fractions and a multistage maceration method starting with maceration using non-polar solvents. The non-polar filtrate from maceration is separated and the macerated residue is re-macerated using a polar solvent, thus obtaining two non-polar and polar extracts without the fractionation process. But from the other hand, the maceration method can also avoid the risk of damage to compounds in plants that are thermolabile (Tetti, 2014).

Electron-donor compounds, such as metal-binding enzymes and proteins, have the ability to dampen the negative effects of oxidants. The 2,2-diphenyl-1-picrylhydrazyl (DPPH) method can be used to test antioxidants. Measuring the decrease in DPPH absorbance at the highest wavelength by adding DPPH reagent solution so that it is proportional to the concentration of free radical inhibitors is the procedure of the DPPH method. The principle of this method is the interaction of antioxidants with DPPH either by electron transfer or hydrogen radicals on DPPH will neutralize the free radical character of DPPH, if all electrons on DPPH free radicals become paired then the color of the solution changes from dark purple to bright yellow (Raudhotul et al., 2018). The Inhibitory Concentration (IC) value, also known as IC_{50} , indicates antioxidant activity through the effective concentration of the extract that can inhibit DPPH activity by 50%.

Analysis of antioxidant compounds in plants can be done using several methods, namely the FRAP, DPPH, ABTS, CUPRAC, and ORAC methods. Ferric Reducing Antioxidant Power (FRAP) method is used to measure antioxidants in aqueous solution. This method is simple, fast, and efficient, allowing one to determine the total antioxidant content of a substance based on its ability to convert Fe^{3+} to Fe^{2+} , comparing the antioxidant activity of a substance with its ability to do so. However, some problems arise in this method, such as slow reactions with certain antioxidants like glutathione (Lalitha & Jayanthi 2011).

The 2,2-azinobis-(3-Ethylbenzothiazoline-6- Sulfonic Acid (ABTS) method is a method for detecting anti-oxidative activity by using 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) as a basic reagent. It is a stable substance of the peroxidase enzyme that can be oxidized by H_2O_2 to form carbon radicals. The principle of this method is to stabilize carbon radicals by adding protons to them, which are then analyzed by light microscopy to determine the presence of carbon radicals. Detection of the presence of carbon radicals can be done using a spectrophotometer at a wavelength of 734 nm. The ABTS method is effective for both airborne and organic systems, providing faster, more consistent reactions, and is suitable for high pH environments. However, this method is body sensitive, requiring 12-16 hours of exposure under cold conditions. The FRAP method, which enables antioxidation by converting ferri-triptyridyl-triazine (TPTZ) into ferrous complexes (Fe^{2+}), is particularly useful for reducing the formation of single ferrous complexes at a wavelength of 593 nm (Maryam et al., 2015).

The Cupric Reducing Antioxidant Capacity (CUPRAC) method is based on the reaction between antioxidants and base radicals, which can be converted to cuprous (Cu^{+}) by using Cu(II)-neocuproine ($\text{Cu}^{2+}-(\text{Nc})_2$) as an oxidizer. Antioxidant activity is qualitatively affected by the color change of the sample. The moderately selective CUPRAC method has a high, fast, and stable reaction potential and can be used for antioxidants that are hydrophilic or lipophilic in pH, easy, safe, and cost-effective, and can be used in conventional laboratories with standardized standards. (Pekal et al., 2012) conducted a study using CUPRAC and Trolox as a screening method on three types of tea (black, green, and fruit) and found that high-quality tea had the highest TE/g content with a total phenolic content of 513.4 mg GAE/g (Choirunnisa et al., 2016).

The principle of the Oxygen Radical Absorbance Capacity (ORAC) method is to evaluate the effectiveness of antioxidant compounds by determining their ability to donate hydrogen atoms during reactions with peroxide radicals. This effectiveness is reflected by changes in fluorescence intensity throughout the reaction. In this assay, the bis-azide/AAPH initiator acts as a radical generator, producing peroxy radicals that degrade the fluorescent probe. The reduction in fluorescence serves as an indicator of antioxidant capacity against oxidative stress. The gradual decline in fluorescence intensity represents the oxidative degradation of the fluorescent molecule, which typically begins shortly after AAPH application and can be accurately monitored at emission wavelengths of 520 nm and 480 nm. Antioxidant molecules found in green tea can inhibit or delay this oxidative degradation, while partially active antioxidant components help stabilize fluorescence changes and enhance analytical sensitivity. Overall, the ORAC method is recognized as a rapid, reliable, and cost-effective analytical technique applicable to both hydrophilic and hydrophobic antioxidant compounds. It provides results with strong physiological relevance and wide applicability in antioxidant research. Additionally, the method offers high sensitivity, making it suitable for detecting subtle differences in antioxidant strength among various samples. Its reproducibility has also established ORAC as a widely adopted standard in comparative antioxidant evaluation studies (Ácsová et al., 2019).

Table 1. Antioxidant Activity of Plant Extract and Gel Moisturizer Formulations

| Plants extract | Solvent | IC ₅₀ Gel | IC ₅₀ extract | Extraction Method |
|--|-------------|----------------------|--------------------------|-------------------|
| Breadfruit (<i>Artocarpus altilis</i>) leaf extract | 96% ethanol | 66.69 ppm | - | Maceration |
| Onion skin extract (<i>Allium cepa</i> L.) | 96% ethanol | 146.40 ppm | 56.25 ppm | Maceration |
| Watermelon mesocarp extract [<i>Citrullus lanatus</i> (Thunb.) Matsum. & Nakai] | 70% ethanol | 138.05 ppm | 110.90 ppm | Maceration |
| Green tea leaf extract (<i>Camellia sinensis</i>) | 70% ethanol | 5.62 ppm | - | Maceration |
| Telang flower extract (<i>Clitoria Ternatea</i> L.) | 96% ethanol | 92.867 ppm | 71.125 ppm | Maceration |
| Black mulberry fruit extract (<i>Morus nigra</i> L.) | 96% ethanol | 104.65 ppm | 146.731 ppm | Maceration |
| Star fruit leaf extract (<i>Averrhoa bilimbi</i> L.) | 96% ethanol | 89.12 ppm | 24.78 ppm | Maceration |
| Red cabbage flowers (<i>Brassica oleracea</i> L.) | 96% ethanol | 57.48 ppm | 47.10 ppm | Maceration |
| Mulberry leaf extract (<i>Morus alba</i> L.) | 70% ethanol | 56.122 ppm | - | Maceration |

Based on the Table 1, breadfruit (*Artocarpus altilis*) leaf extract was extracted using maceration technique and 96% ethanol solvent. In this article, the antioxidant activity test was not conducted in the form of extract, but the antioxidant test was conducted in the form of moisturizer gel preparation with the best preparation in formulation 3 with IC₅₀ of 66.69 ppm which is classified as a strong antioxidant. Flavonoid compounds contained in breadfruit leaves (*Artocarpus altilis*) are natural antioxidants in plants that function to neutralize free radicals, inhibit cell damage in the body, and prevent disease (Asni Setiani et al., 2018). The antioxidant mechanism of breadfruit leaf extract (*Artocarpus altilis*) is through proton donors from samples that act as antioxidants by reducing DPPH to DPPH-H which forms 2,2-diphenyl-1-picrylhydrazyl compounds (Okzelia and Nurdaini, 2019). The formulation used in making this preparation is 20% breadfruit leaf extract (*Artocarpus altilis*); 2% carbopol; 2% TEA; 10% glycerin; 5% propylene glycol; 0.2% methylparaben; 0.2% propylparaben; and 100 mL of distilled water to produce a moisturizer that meets the SNI standards for topical preparations. Breadfruit leaf extract gel is semi-solid, greenish-yellow in color has a distinctive aroma of breadfruit leaves, and all preparations are homogeneous. pH value is in accordance with the pH of topical preparations, has good distribution, does not irritate the skin, and is able to moisturize the skin (Shofiah, 2024).

Onion skin extract (*Allium cepa* L.) shows an IC₅₀ value of 56.25 ppm which includes strong antioxidants and IC₅₀ in formula preparations of 146.40 ppm including the medium category with maceration extraction method and 96% ethanol solvent. Onion skin (*Allium cepa* L.) has flavonoid content that functions as antioxidants that can prevent the development of free radicals in free and repair damaged body cells. The formulation used is 10% shallot skin extract; carbopol 1%; glycerin 5%; propylene glycol 5%; TEA 1%;

methylparaben 0.1%; aquadest 100 mL produces a good preparation based on the physical stability test (Feladita et al., 2021).

Watermelon mesocarp extract [*Citrullus lanatus* (Thunb.) Matsum. & Nakai] extracted using maceration method and 70% ethanol solvent has antioxidant activity with IC₅₀ values of 110.90 ppm and 138.05 ppm for moisturizer preparations where both are classified as moderate antioxidants. This extract has flavonoid compounds where there are free hydroxyl groups that have the ability to ward off free radicals by breaking chain reactions and turning them more stable compounds (Hasanah et al., 2020). The antioxidant mechanism of watermelon mesocarp extract [*Citrullus lanatus* (Thunb.) Matsum. & Nakai] has a mechanism that is through proton donors from samples that have antioxidant activity so that DPPH can be reduced to DPPH-H which forms the compound 2,2- diphenylpicrylhydrazine. The maximum wavelength of DPPH was first scanned at 800-400 nm because DPPH is a mauve-colored solution. With the DPPH reduction process, the solution changes color from pale mauve to yellow. The formula used to make this preparation is watermelon mesocarp extract [*Citrullus lanatus* (Thunb.) Matsum. & Nakai] as much as 15%; carbopol 1.2%; TEA 3.5%; propylene glycol 15%; Phenoxyethanol 0.5%; mica powder q.s; fragrance q.s; and distilled water 100 mL to produce a moisturizing gel that meets all the standard parameters of a good gel preparation, can increase the percentage of skin moisture (Okzelia & Mardiyyah, 2023).

Green tea leaf extract (*Camellia sinensis*) in this article is not tested for antioxidant activity in the form of extracts, but antioxidant tests are carried out in the form of moisturizer gel preparations with an IC₅₀ value of 5.62 ppm which is classified as a strong antioxidant extracted using maceration method and 70% ethanol solvent. The greater the concentration of the active substance in the formulation, the smaller the IC₅₀ value so that the antioxidant activity is higher. Green tea leaves (*Camellia sinensis*) are an example of natural ingredients that contain antioxidants, this is because green tea contains catechin components that function as antioxidants (Aris Purwanto and Irfan Zamzami, 2020). The formula used to make this preparation is green tea leaf extract (*Camellia sinensis*) 0.01%; methylcellulose 0.037%; carbopol 0.555%; propylene glycol 0.124%; methylparaben 0.00025%; green tea oil 0.005%; aquadest 20 mL. Tea leaf extract gel (*Camellia sinensis*) has different antioxidant activity based on variations in methylcellulose concentration with carbopol (Purwanto & Zamzani, 2020).

Telang flower extract (*Clitoria Ternatea* L.) has strong antioxidant activity based on the IC₅₀ value of 71.125 ppm and for the preparation of moisturizer gel of 92.867 ppm extracted using maceration method and 96% ethanol solvent. The difference in IC₅₀ values between gel moisturizer preparations and extracts is due to the addition of tragakan in the formulation process which results in lower IC₅₀ values in gel moisturizer preparations than extract preparations. Telang flower (*Clitoria Ternatea* L.) contains active compounds flavonoids, saponins, terpenoids, and tannins which act as radical antidotes, inhibitors of lipid peroxidation, and other free radical mediation processes (Riswanto et al., 2022). The formulation used in making this preparation is telang flower extract (*Clitoria Ternatea* L.) 0.01%; tragacanth 0.05%; glycerin 0.1%; propylene glycol 0.05%; methylparaben 0.0015%; distilled water 100 mL (Nurwaini et al., 2024).

Black mulberry fruit extract (*Morus nigra* L.) has antioxidant activity with moderate IC₅₀ values of 146.731 ppm and 104.65 ppm for gel moisturizer preparations extracted using maceration method and 96% ethanol solvent. The difference in IC₅₀ values in extracts and preparations is because the formulation uses DPPH solution (2:3) so the IC₅₀ value in the moisturizer gel preparation is higher than the extract. Black mulberry (*Morus nigra* L.) contains the most phenolic and flavonoid compounds called anthocyanins. These

anthocyanin compounds are cyanidin-3-glucoside and cyanidin-3-rutinoside. Previous studies have shown that mulberry anthocyanins, quercetin 3-(6-malonyl glucoside), and rutin from mulberry (*Morus nigra* L.) leaves are excellent antioxidant agents. The formula used in this preparation is customized gel base; glycerin 10%; methylparaben 0.18%; propylparaben 0.02%; fragrance q.s; aquadest 100 mL (Budiman et al., 2019).

Star fruit leaf extract (*Averrhoa bilimbi* L.) has secondary metabolite compounds, namely flavonoids, tannins, saponins, and steroids. Where flavonoids and polyphenol derivatives are compounds that function as antioxidants because these compounds are phenol compounds, which are compounds with an -OH group attached to an aromatic carbon ring. The free radical products of these compounds are resonance stabilized and therefore unreactive compared to most other free radicals so they can function as effective antioxidants. Star fruit leaf extract (*Averrhoa bilimbi* L.) extracted by maceration method and 96% ethanol solvent has very strong antioxidant properties based on the IC_{50} value of 24.78 ppm and gel moisturizer preparation of 89.12 ppm which is included in the strong category. This is supported by research conducted by (Andriani et al., 2019) which states that 96% ethanol extract of star fruit leaves has very strong antioxidant activity, namely 25.74 μ g/ml and research conducted by (Hasim et al., 2019). The formulation of this preparation includes star fruit leaf extract (*Averrhoa bilimbi* L.) 15%; HPMC 5%; glycerin 10%; methylparaben 0.3%; propylparaben 0.3%; aquadest 100 mL (Zaky et al., 2021).

Red cabbage flowers (*Brassica oleracea* L.) contain natural antioxidants in the form of flavonoid compounds, saponins, and tannins. Red cabbage flower extract (*Brassica oleracea* L.) extracted using the maceration method and 96% ethanol solvent has very strong antioxidant activity with an IC_{50} value of 47.10 ppm and in gel moisturizer preparation of 57.48 ppm which is included in the strong category. The antioxidant activity of the extract is stronger than that of the preparation. The antioxidant activity after making the preparation becomes higher IC_{50} because of the possibility of a reaction between the extract and one of the components in the gel preparation, so the antioxidant activity of the preparation is smaller than the extract. The antioxidant activity of ethanol extract of red cabbage flower (*Brassica oleracea* L.) and vitamin C as a comparison compound has a very strong IC_{50} category of 47.10 ppm and 6.30 ppm. The formula used is ethanol extract of red cabbage flower (*Brassica oleracea* L.) 10%; HPMC 7%; propylene glycol 15%; methylparaben 0.03%; propylparaben 0.08%; aquadest 100 ml. The concentration of red cabbage flower extract in gel preparations has a different effect on the values of viscosity, pH, spreadability, organoleptic, and antioxidant activity (Effendi et al., 2019).

Mulberry leaf extract (*Morus alba* L.) in this article is not tested for antioxidant activity in the form of extracts, but antioxidant tests are carried out in the form of moisturizer gel preparations with an IC_{50} value of 56.122 ppm which is classified as a strong antioxidant extracted using maceration method and 70% ethanol solvent. The greater the concentration of active substances in the formulation, the smaller the IC_{50} value so that the antioxidant activity is higher. Many plants are efficacious as antioxidants, namely plants containing carotenoids and polyphenols, especially flavonoids, so many are formulated as natural antioxidants that can be made in oral dosage forms as vitamins and topically as skin care products. The formula of this preparation is mulberry leaf extract (*Morus alba* L.) 9%; carbopol 1%; glycerin 22%; TEA 1%; phenoxyethanol; DMDM Hydantoin 0.1%; aquadest 100 mL. The mulberry leaf extract (*Morus alba* L.) gel preparation fulfills all physical properties test parameters, namely homogeneity testing, pH, organoleptic, spreadability, and adhesiveness (Reinard et al., 2022).

The difference extraction process using 96% and 70% ethanol solvents aims to produce pure thick extracts of active compounds in a plant. 96% ethanol allows to attract

of active compounds more optimally, so based on the literature review. Although using the same extraction method with 96% ethanol produces a stronger IC_{50} value than 70% ethanol. The IC_{50} value in plant extracts is smaller than the IC_{50} of the preparation due to several factors such as the environment that can reduce antioxidant activity, as well as the addition of excipients and other substances in the formulation process of moisturizer gel can affect the concentration of antioxidants compared to thick extracts alone. Based on the literature review of the antioxidant activity of gel moisturizer preparations from various plants, antioxidant activity in 70% ethanol extract of green tea leaves (*Camellia sinensis*) in gel moisturizer preparations with maceration method with a very strong IC_{50} value of 5.62 ± 0.37 ppm and 70% ethanol extract of star fruit (*Averrhoa bilimbi* L.) with maceration method with IC_{50} value of 24.78 ppm.

CONCLUSION

Plants that have very strong antioxidant activity are 70% ethanol extract of star fruit (*Averrhoa bilimbi* L.) with maceration method where the IC_{50} value is 24.78 ppm and the antioxidant activity of moisturizer gel preparation is very strong, namely 70% ethanol of green tea leaves (*Camellia sinensis*) with maceration method with IC_{50} value of 5.62 ± 0.37 ppm.

AUTHOR CONTRIBUTION

Author Initial: Contribution.

AIMU: Concepts; design; literature search; manuscript preparation; revising manuscript based on reviewers comments.

KNH: Literature search; revising manuscript based on reviewers comments; initial article screening; design.

RR: Literature search; Initial article screening.

NS: Manuscript review; manuscript preparation; conducting final manuscript review.

CONFLICT OF INTEREST

None to declare

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