

The Formulation of a Serum Containing Tomato (*Solanum Lycopersicum L.*) Extract for a Green Anti-Aging Skincare Product

Van Quy Nguyen¹, Thieu Phu Lau², Thi Kim Chi Huynh³, Hoang Phuc Nguyen³, Thi Cam Thu Nguyen³, Thi Hong An Nguyen³, Xuan Ngoc Nguyen³, Thi Kim Dung Hoang³, Anh Khoa Ton^{3*}

¹Ho Chi Minh City University of Technology and Education, Vietnam

²Ton Duc Thang University, Vietnam

³Institute of Chemical Technology, Vietnam Academy of Science and Technology, Vietnam

*Corresponding author. Email: kevinton150691@gmail.com

ARTICLE INFO

Received: 11/10/2024
Revised: 13/11/2024
Accepted: 28/11/2024
Published: 28/12/2024

KEYWORDS

Anti-aging;
Tomato extract;
Polyphenol;
Flavonoid;
Green cosmetics.

ABSTRACT

Sunlight exposure can lead to photoaging that reduces the quality of life, from inside to outside. Tomato contains a lot of nutrients and valuable compounds which are powerful antioxidants. Hence, the utilization of safe and effective natural compounds for the creation of phytocosmetic is undoubtedly attractive for sustainable development. Herein, the small pieces of tomato were dried at 60°C for 6 hours to remove the water. The chloroform solvent was used for extraction by a 1:10 (w/v) ratio for 24 hours without light exposure. The yield was 0.273% compared with the dry tomato. Besides, the total polyphenol and flavonoid contents were 111.49 ± 0.31 µg gallic acid equivalents/mg extract and 150.15 ± 2.17 µg quercetin equivalents/mg extract, respectively. The anti-aging serum containing 0.10% tomato extract was an ideal topically phytocosmetic that was stable after physical evaluations, had no irritation by voluntary assessment as well, and had a uniform diameter of O/W droplet around 40 µm with a potential antioxidant for daily application. Therefore, we have formulated an anti-aging serum containing tomato extract for nourishing the outside beauty and taking care of inside health.

Doi: <https://doi.org/10.54644/jte.2024.1673>

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1. Introduction

Undoubtedly, aging is a complex process that is integrated into the development of life. In the aging process, the excessive reactive oxygen species will be liberated from the living cells and implicated in some accompanied aging diseases such as Alzheimer's and Parkinson's, schizophrenia, and so on [1]. Sunlight exposure is a means of aging that is induced by UV rays, one of the expressions is the signs presented on our skin such as black pots, wrinkles, acne, and so on if we don't take any preventive actions, it will result in more serious damage to our health, the protection from outside will bring the beauty and healthy for inside. Therefore, it is necessary to discover phytocosmetics that utilize natural compounds for the safety and sustainability trends in the future.

Tomato (*Solanum lycopersicum L.*) is a daily and essential food source for humans because it has a lot of valuable nutrients. Recently, the utilization of natural extraction has attracted intensive investigations, especially phytocosmetics because the customers believe in the health of these products, not only for humans but also for our environment. Tomato contains polyphenolics, flavonoids, saponins, carotenoids, monoterpenoids, and sesquiterpene. Lycopene is a component of a carotenoid compound that owns 11 conjugated double bonds to express the potential antioxidant capacity by a high quenching rate for singlet oxygen [2], [3]. Regarding the importance of lycopene, it is reported that lycopene can protect the skin from the photoaging induced by UV rays [4], prevent cardiovascular disorders [5], and have protective effects in anti-inflammation and anticancer [2], [6].

In this research, we developed an anti-aging serum containing tomato extract to exploit potential products for a green skincare era. The total phenolic and flavonoid compounds will be determined, and the antioxidant of the extract will reflect the ability of the bioactive in the phytocosmetic, besides, the

physical characteristics of the anti-aging serums will be assessed to evaluate and go to the conclusion for the final formulation.

2. Materials and Methods

2.1. Materials

Tomatoes were sourced from the market between April and June 2024. These tomatoes were red, fresh, and had no rotten behaviors. Sodium carbonate anhydrous, Potassium acetate 99%, and Aluminium chloride were purchased from Xilong, China. Gallic acid 98.5 % was purchased from Shyuanye, China. Quercetin dihydrate 95% was from Merck, Germany. Folin-Ciocalteu phenol reagent originated from Sigma Aldrich. L-ascorbic acid with a purity of 99% was from Fischer and DPPH with 97% purity was from Cool Chemistry Science and Technology, Beijing, China. Chloroform, Methanol, and Isopropanol were from Xilong, China. Double-distilled water was produced at the Institute of Chemical Technology with no further purification.

Ingredients in the serum formulation: Glycerin, Xanthan gum, Olivem 1000, Almond oil, Vitamin E, and Geogard ECT were from local Vietnam manufacturers.

2.2. Tomato extraction

The tomatoes were washed with tap water, removed from the seed, and chopped into small pieces. The sliced tomato was spread onto an aluminum foil and dried at 60°C in an oven. After 6 hours, weigh the remaining tomatoes until constant mass, and calculate the water loss and volatile content. Next, the dried tomato was added into chloroform by a 1:10 (w/v) ratio and stirred in the closed glass vessel at 100 rpm for 24 hours. Afterward, the mixture would be filtered to remove the sediment, and the filtrate was recollected by a separating funnel to get rid of the water layer. Finally, the red tomato extract was removed from chloroform by a Buchi Rotavapor at 50 °C, 400 mbar, 60 rpm for 30 minutes to collect the red tomato extract and store it at -20°C in the dark until use.

2.3. Total Phenolics evaluation

The standard curve was prepared by a solution of gallic acid at the concentration of 100 µg/mL in distilled water. Next, keep diluting to obtain a range of standard concentrations at 10, 20, 30, 40, and 50 µg/mL of gallic acid. Then, 1 mL of these solutions was added 5 mL of 10% Folin-Ciocalteu reagent, mixed, and left in the dark for 10 minutes. Finally, 4 mL of Na₂CO₃ 7.5% solution was added to each of the mixtures and shaken to obtain a homogenous solution. Let them in the dark for 60 minutes and measure the absorbance at 765 nm by Jasco V-770 UV-Vis spectrophotometry [7].

Regarding the tomato extract, weigh 1 mg and dissolve it in 4 mL of distilled water. Next, 1 mL of this solution was added 5 mL of 10% Folin-Ciocalteu phenol reagent and conducted similarly as described above [7]. The experiment was triplicate, and the total polyphenol content was calculated by using the following calculation [8]:

$$\text{TPC} = C \times K \times V / m \quad (1)$$

Herein,

- TPC: Total Polyphenol content (µg GAE (Gallic acid equivalents)/mg extract),
- C: the concentration obtained from the standard curve (µg/mL),
- V: the volume of extract solution (mL),
- K: the dilution factor,
- m: the mass of extract in volume V (mg).

2.4. Total Flavonoids determination

A standard calibration curve was constructed by a range of quercetin concentrations of 20, 40, 60, 80, and 100 µg/mL in methanol. Next, 0.5 mL of these concentrations were mixed homogeneously with 1.5 mL of methanol. After 5 minutes, 0.1 mL of AlCl₃ 10% was added and left free for 6 minutes. Finally, 0.1 mL of CH₃COOK 1M solution was inserted and 45 minutes later, these solutions were

measured for their absorbance at the wavelength 415 nm by Jasco V-770 UV-Vis spectrophotometry [9].

On the other hand, 1 mg of tomato extract was dissolved in 4 mL methanol. Next, 0.5 mL of this solution was added 1.5 mL methanol and shaken homogeneously. The subsequent steps were conducted as described above. The experiment was triplicate, and the total flavonoid content was calculated by using the following formula [8]:

$$\text{TFC} = C \times K \times V / m \quad (2)$$

Herein:

- TFC: the total flavonoid content, (μg QE (Quercetin equivalents)/mg extract),
- C: the value X from the standard curve ($\mu\text{g}/\text{mL}$),
- V: the volume of extract solution (mL),
- K: the dilution factor,
- m: the mass of extract in volume V (mg).

2.5. The DPPH antioxidant activity

Weigh 8 mg tomato extract to dissolve in the mixture of chloroform and methanol (1:3, v/v) to obtain the 2000 $\mu\text{g}/\text{mL}$ solution. Next, the standard concentrations were diluted and became 31.25, 62.5, 125, 250, 500, 1000 and 2000 $\mu\text{g}/\text{mL}$. Then, 100 μL of these tomato extracts were added to 3 mL DPPH 0.1 mM solution. The negative control contains only MeOH and DPPH. Incubate the samples in the dark room for 30 minutes and measure the absorbance at 517 nm by Jasco V-770 UV-Vis spectrophotometry [7], [8].

Antioxidant activity (%) is calculated as:

$$\text{Antioxidant activity (\%)} = \frac{(A_0 - A_t)}{A_0} \times 100 \quad (3)$$

Herein:

- A_0 : Absorbance value of the standard DPPH sample
- A_t : Absorbance value of the test sample

2.6. Serum containing tomato extract preparation

2.6.1. The formulation of serum

Table 1. The formulation of serum base and serum containing tomato extract.

Phase	%w/w	Ingredient
A	To 100%	Water
B	2.5	Glycerin
	0.3	Xanthan gum
C	3.0	Olivem 1000
	5.0	Almond oil
D	2.5	Glycerin
	0.2; 0.1; 0.05 or 0.0167	Tomato extract
E	0.2	Vitamin E
	0.9	Geogard ECT
pH adjustment		

Table 1 shows the composition of the serum with and without tomato extract. First, mix glycerin and xanthan gum in phase B to form a paste. Add phase B to phase A, stirring vigorously to swell the xanthan

gum. Let it stand while proceeding to the next step. Mix phases C and D. Heat phase A/B to 75°C. When bubbles appear at the bottom of the glass beaker of phase A/B, heat phase C/D to the same temperature, 75°C. Stir phase C/D to create a homogeneous mixture. Quickly add phase C/D to the hot phase A/B, stirring vigorously until a smooth, shiny white or orange emulsion is formed. Allow the product to cool to below 40°C, then add phase E and stir to mix into the product. Adjust the pH to between 5.3 and 5.8.

2.6.2. Physicochemical characteristics

Physical observations by color, odor, skin irritation, appearance, and phase separation were made against the base serum and serums containing tomato extract.

Skin irritation test: the experiment was conducted by a group of 20 volunteers at the Institute of Chemical Technology in Ho Chi Minh City. Approximately, 0.5g of the serum was applied on hand 2 times per day to evaluate the swelling, itchiness, or redness [10].

Determination of water content and volatile substances: approximately 1 g of sample was weighed and placed in a vial. Then, they were heated in the oven at 100 ± 5 °C to a constant mass. The water content and volatile substances are calculated by using this equation [11]:

$$\text{Water content and volatile substances} = \frac{(m_1 - m_2)}{m_1} \times 100 \quad (4)$$

Herein: m_1 : mass of the sample before drying, (g)

m_2 : mass of the sample after drying, (g)

Homogeneity: take 2-3 g of serum at different concentrations and place them in centrifuge tubes. Centrifuge at 4,000 rpm for 30 minutes for 3 cycles [12].

Physical stability (thermal stress) test: prepare 5 small glass jars containing the base serum and 4 serums containing different extract amounts. Place them in a freezer at 4°C for 24 hours, then transfer them to an oven at 40°C for 24 hours. Conducting this experiment for 6 cycles. Finally, observe whether the phase separation of the serum occurred [10], [13].

Optical microscopy: a small amount of serum samples was spread onto a glass slide. Their size and shape were visualized by an optical microscope (IM-5/Optika, Italy) at 4X magnification [11].

2.6.3. The DPPH antioxidant activity

Weigh 4 g of serum with 0.10% extract and dissolve it in 5 mL of a chloroform and isopropanol mixture (1:1 ratio). Filter to remove any insoluble residues. Then, dilute the sample into 7 different concentrations by using methanol.

Add 100 μ L of the test solution to a test tube containing 3 mL DPPH 0.1mM. The negative control contains only MeOH and DPPH. Incubate the samples in the dark at room temperature for 30 minutes, then measure the absorbance at 517 nm by UV-Vis spectrophotometry [12].

Antioxidant activity (%) is calculated as:

$$\text{Antioxidant activity (\%)} = \frac{(A_0 - A_t)}{A_0} \times 100 \quad (5)$$

Herein:

- A_0 : Absorbance value of the standard DPPH sample
- A_t : Absorbance value of the test sample

3. Results and Discussion

Tomatoes are a kind of tropical fruit that contains a lot of potentially useful components for health benefits such as minerals, vitamins, proteins, amino acids, monounsaturated fatty acids (linoleic and linolenic acids), flavonoids (flavones, flavanols, flavonols, flavanones, isoflavones and anthocyanins), phenolic acids (quercetin, kaempferol, naringenin, caffeic acid and lutein) along with carotenoids (lycopene, β -carotenoids) [14], [15]. Herein, polyphenols are classified into principal classes: flavonoids, stilbenes, phenolic acids, and lignans [16]. Sunlight contains UVA (315-400 nm), UVB (280-315 nm), and UVC (100-280 nm) which are key factors in producing the reactive oxygen species

(ROS), leading to oxidative stress and directly or indirectly damaging the DNA. Hence, after long-term exposure to sunlight, the skin will appear deep wrinkles, hyperpigmented spots, and even the progression of precancerous lesions [17]. Antioxidant flavonoids and phenolic acids can counteract the production of ROS, maintain stable lipid peroxidation levels, and inhibit collagen degradation and keratinocyte apoptosis, promoting the expression of key factors in cellular repair [17]. Another research has revealed the molecular mechanisms of polyphenols such as autophagy and senescence, and DNA repair system in aged dermal fibroblasts, for instance, quercetin stimulated NRF2 and enhanced the expression of HO-1 and catalase or, interestingly, quercetin/curcuminoid at a ratio of 3:1 can activate the migration of fibroblasts which strongly encourages the trend of using natural plant extracts in cosmetics and functional foods [16].

It is believed that there are many factors influencing the yield of tomato extracts such as drying temperature, solvent, raw material, solvent ratio, the source of plants, and so on. Using a suitable conventional organic solvent is the primary step for obtaining the expected compounds such as hexane, acetone, ethanol, ethyl acetate, chloroform, and petroleum ether. Tomatoes have a high number of carotenoids which are soluble in nonpolar solvents such as chloroform. After extraction, it was removed by a rotavapor to recollect the extract without bringing any concerns about safety risks in health. However, to strengthen this statement, future clinical study or *in vitro/in vivo* evaluation should be taken in the future. Besides, modern extraction techniques have presented some methods to isolate the natural ingredients such as the extraction with the assistance of ultrasound, microwave, enzyme, pressure, and supercritical fluid [18]. Herein, the tomato was dried at 60°C for 6 hours until it became an unchanged mass and extracted by using chloroform with a 1:10 (w/v) ratio. Consequently, we obtained 0.273% of tomato extract over the dried raw sample. **Figure 1** depicts gallic acid and quercetin calibration curves by measuring the absorbances over a range of respective standard solutions. Herein, the gallic acid calibration curve is $y = 0.0109x + 0.1312$ ($R^2 = 0.9965$) while quercetin is $y = 0.0072x - 0.0227$ ($R^2 = 0.9970$). Hence, the total polyphenol and flavonoid were $111.49 \pm 0.31 \mu\text{g GAE/mg extract}$ and $150.15 \pm 2.17 \mu\text{g QE/mg extract}$. Moreover, **Figure 2** exhibits the DPPH antioxidant of the tomato extract. In this experiment, we use the DPPH free radical scavenging method to determine the antioxidant capacity of the tomato extract sample. We can find that at the highest concentration of 2000 $\mu\text{g/mL}$, the antioxidant percentage was only $42.05 \pm 0.014\%$, this value was relatively close to the elsewhere research which said that the antioxidant activity of tomato extract against DPPH which were prepared in methanol was about 37.68% [19].

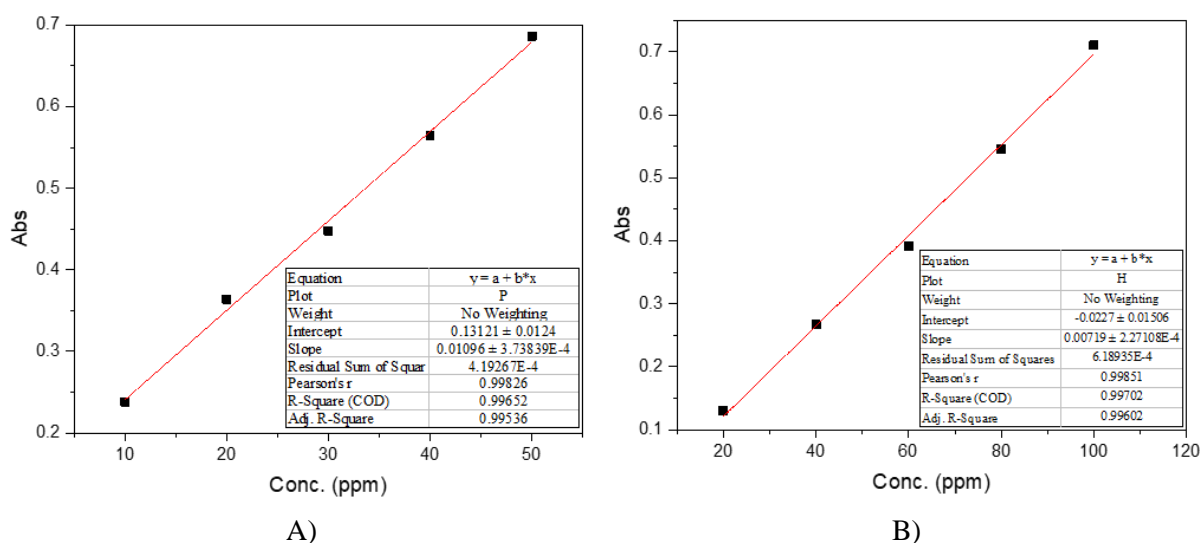


Figure 1. The calibration curves of A) gallic acid and B) quercetin for determination of total polyphenol and flavonoid, respectively.

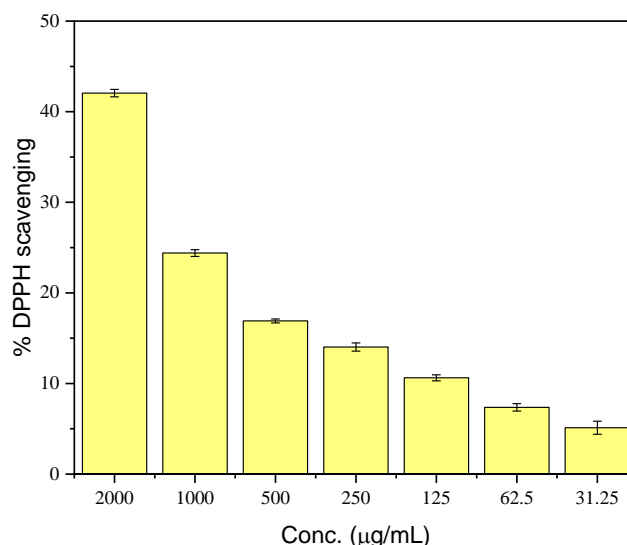


Figure 2. The DPPH antioxidant of tomato extract.

After determining the concentration of polyphenolics and flavonoids, we proceeded to transfer this extract to the serum for expression of its potential efficacy. The amount of tomato extract was chosen at different weight percentages such as 0.20%, 0.10%, 0.05%, and 0.0167%, and compared with the serum base. **Table 2** depicts the related parameters of formed serums. In the process of formulating serums, the composition of each phase, amount of emulsifier, content of viscosity modifier, mixing speed, temperature, especially, the input of extract is all important for a skincare product. It is shown that all the serums have reached the appropriate pH for skin adaptability. On the other hand, except for the base serum that had a milky formulation, the other ones had different orange colors depending on the concentration of added tomato extracts.

Besides, the irritation test shows us evidence of no redness, swelling, or itching which is examined by a group of 20 volunteers at the Institute of Chemical Technology. Although the serum containing tomato extract had an orange color, it left no remaining color or any residues after applying on the skin because its absorption was good owing to the appropriate size to penetrate through the skin, and the mixing, as well as the temperature, were all well integrated to create a homogenous, stable and silky emulsion. However, the amount of the extract presented in the emulsion is probably influenced by the physical parameters resulting in dissimilarity in stability, storage, and treatment efficacy.

Table 2. The appearance of the serum base and serum containing different amounts of tomato extracts.

Parameter	Serum base	0.20%	0.10%	0.05%	0.0167%
Organoleptic	Semi-solid, milky form, odorless	Semi-solid, orange, odorless	Semi-solid, orange, odorless	Semi-solid, orange, odorless	Semi-solid, orange, odorless
Homogeneity	Homogenous	Homogenous	Homogenous	Homogenous	Homogenous
pH	5.48	5.36	5.43	5.56	5.55
Type	Oil-in-water	Oil-in-water	Oil-in-water	Oil-in-water	Oil-in-water
Stability	Stable	Stable	Stable	Stable	Stable
Skin irritation	No	No	No	No	No

Table 3 gives us data about water content and volatile substances. All the samples have a water loss of over 85% which was relevant compared to the input water. **Figure 3** expressed the homogeneity of all samples after 3 cycles of centrifugation. The serum base had a homogeneous mixture and there was no phase separation after 24h of preparation. However, the serums containing 0.20% and 0.0167% tomato extract didn't express a stable emulsion because of phase separation formation. Especially, the

serum with 0.10% tomato extract was stable after 3 cycles of 4,000 rpm for 30 min centrifugation, it could be stable for up to 10 months, approximately [4]. In **Figure 4**, after 12 days of freeze-thaw (6 cycles), the homogeneity as well as the odor and color of all samples were still stable. The free-thaw cycle is the process the checking the segregation of an emulsion against the change of temperature, it is noted that there is an increase in the size of the globular after free-thaw cycles compared with room temperature resulting in the flocculation or phase separation of the skincare product [4]. **Figure 5** shows the globule size of the emulsions at 4X magnification. The serum base had a spherical shape and was around 60 μm in diameter. Concerning the 0.20% tomato extract-contained serum, it had a spherical shape, but the size of the globule was around 110 μm which was the largest diameter compared to the other ones. The smallest size distribution was the serum containing 0.10% tomato extract with a size of approximately 40 μm . However, the more the added extract, the bigger the size was which was visualized at 0.05% and 0.0167%, respectively. The particle size of the O/W droplet plays an important role in the stability of the emulsion. The smaller the diameter, the better the shelf life and the more stable because it cannot flocculate or be separated by gravity [13]. This could be the reason for the phase separation of serum containing 0.20% and 0.0167% tomato extract after centrifugation as depicted in **Figure 3**. The serum containing 0.10% of tomato extract is superior to the others owing to its stability, and the size of the formed droplet, so we will investigate the potential radical scavenging of the formulation. Lastly, in **Figure 6**, we have evaluated the antioxidant of serum containing 0.10% tomato extract and seen that the higher the used amount of serum, the better the antioxidant is, however, the data is not as significantly high as we expected but still expressed the ability of the antioxidant serum for daily application. Thus, this phytocosmetic is an ideal agent for promisingly combating aging.

Table 3. The water content and volatile substances of serum base and serum containing different amounts of tomato extracts.

Name	m ₁ (g)	m ₂ (g)	% weight loss
Serum base	1,0105	0,1358	87,47
Serum 0,20% extract	1,003	0,1412	86.18
Serum 0,10% extract	1,005	0,1517	85,33
Serum 0,05% extract	1,007	0,1431	86.39
Serum 0,0167% extract	1,002	0,1057	89,63

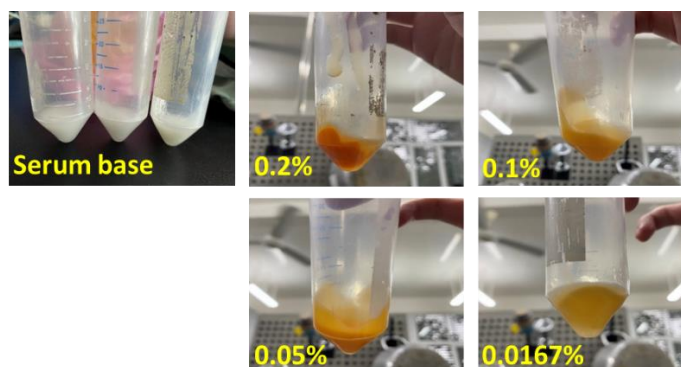


Figure 3. The homogeneity of serum base and serum containing different amounts of tomato extracts after 3 cycles of centrifugation.

Table 4. The result of centrifugation test.

Centrifuge speed	Serum base	0.20% extract	0.10% extract	0.05% extract	0.0167% extract
4,000 rpm; 30 min	-	+	-	-	+

(3 cycles)					
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(-): not separate into two phases,

(+): split into two phases.

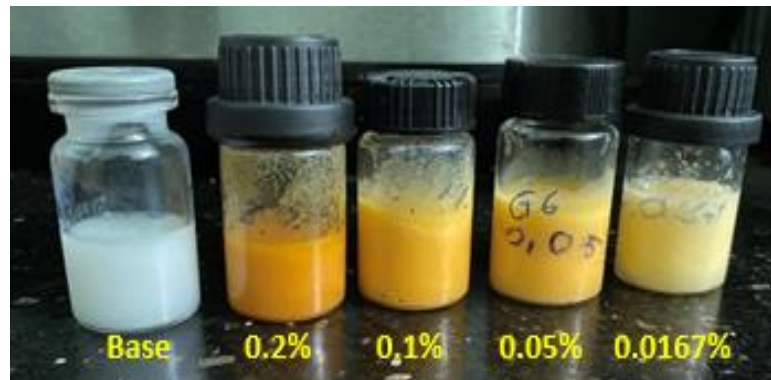


Figure 4. The serum base and serum containing different amounts of tomato extracts after 6 cycles of freeze/thaw.

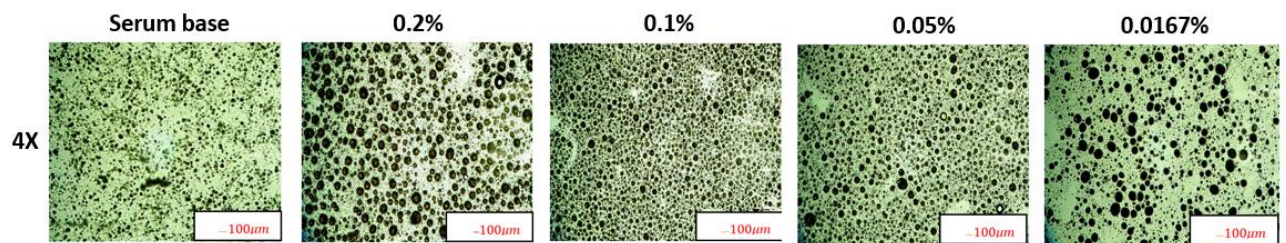


Figure 5. The optical microscope of serum base and serum containing different amounts of tomato extracts at 4X magnification.

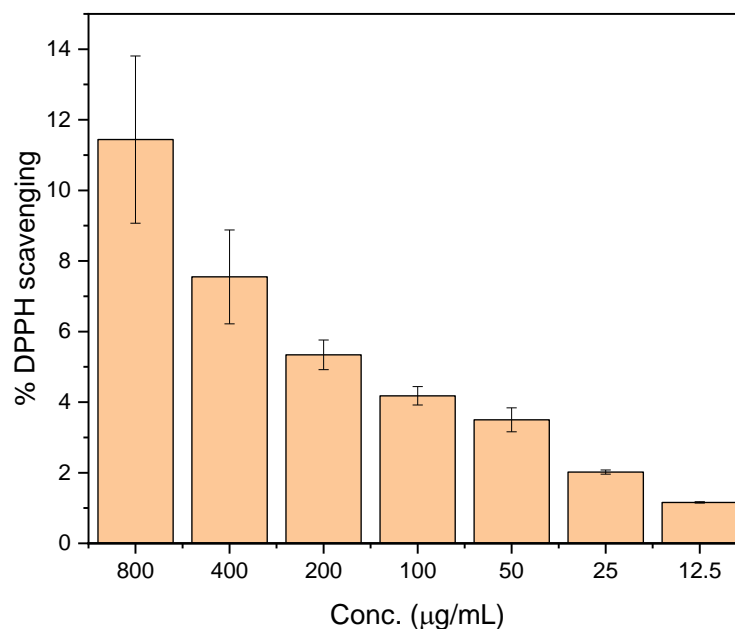


Figure 6. The DPPH antioxidant of serum containing 0.10% tomato extract.

4. Conclusions

This study investigated the ability of tomato extract to formulate an anti-aging serum. The extract had $111.49 \pm 0.31 \mu\text{g GAE/g}$ and $150.15 \pm 2.17 \mu\text{g QE/g}$ and showed antioxidant activity by DPPH radical quenching. The serum containing 0.10% tomato extract was optimum for stability, compatibility, and potential human skin topical application. We suggest doing more investigation into the stability of

the serum for several weeks as well as choosing a suitable active delivery system for the integration into the product. This hopes to bring the best treatment efficacy by deeper penetrating through the skin.

Acknowledgments

This research was completed successfully with the participation of members from the Vietnam Academy of Science and Technology, Ho Chi Minh University of Technical Education, Ton Duc Thang University. Many thanks for our contributions and efforts.

Conflict of Interest

The authors declare no conflict of interest

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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First author

Dr. Nguyen Van Quy is currently a lecturer at Ho Chi Minh City University of Technology and Education. He received a PhD degree from the School of Chemical Engineering, Sungkyunkwan University (SKKU), South Korea in 2022. He has published more than 20 research articles. His research interests are in composites and biomaterials.

Email: quynv@hcmute.edu.vn. ORCID:  <https://orcid.org/0000-0002-5952-0700>



Lau Thieu Phu obtained his bachelor's degree at the Faculty of Applied Science, Ton Duc Thang University in 2024. His major is Organic Synthesis.

Email: lauthieuphu@gmail.com. ORCID: <https://orcid.org/0009-0006-5607-8300>



Dr. Huynh Thi-Kim-Chi received the Ph.D. degree in Organic Chemistry from Graduate University of Sciences and Technology, Vietnam, in 2022. She has worked as an active researcher of the Laboratory of Organic Chemistry and Polymer Technology in ICT, VAST.

She has published more than 10 peer-reviewed papers related to organic synthesis, medicinal chemistry, and environmental chemistry, and *in silico* studies for bioactivity.

Besides, her research has also focused on drug delivery systems, optical and chemical sensors, and materials for environmental detection and treatment. Email: maihuynh1224@gmail.com. ORCID: <https://orcid.org/0000-0002-6214-8022>



Nguyen Hoang-Phuc is currently working as a senior engineer of the Laboratory of Organic Chemistry and Polymer Technology in ICT, VAST. He received the master's degree in chemical engineering from Ton Duc Thang University, Vietnam, in 2021.

His is interested in organic synthesis, SAR, *in silico* studies for bioactivities, environmental treatment and biosensor, and chemical process equipment design.

Email: nguyenhoangphuc377@gmail.com. ORCID: <https://orcid.org/0000-0003-0140-7112>



Nguyen Thi-Cam-Thu completed the master's degree in chemical engineering from Ton Duc Thang University, Vietnam, in 2022. She is currently working as a staff of the Laboratory of Organic Chemistry and Polymer Technology in Institute of Chemical Technology (ICT), Vietnam Academy of Science and Technology (VAST).

She is also in charge of synthesis and application of chemical and optical sensors along with Assoc. Prof. Thi-Kim-Dung Hoang and Dr. Thi-Kim-Chi Huynh. Moreover, her research interests include organic synthesis, structure-activity relationship (SAR), and sensors practical applications for environmental detection and treatment.

Email: nguyenthicamthu1108@gmail.com. ORCID: <https://orcid.org/0000-0001-5458-8012>



Nguyen Thi-Hong-An received a degree in Chemical Engineering from Ton Duc Thang University in 2017. She is currently working as a staff of the Laboratory of Organic Chemistry and Polymer Technology in ICT, VAST.

Her research focuses on organic synthesis, SAR, bioactivities, polysaccharide extraction, and the application of polysaccharide materials for environmental treatment and sensing toxins in water.

Email: nguyenan2093@gmail.com. ORCID: <https://orcid.org/0000-0001-7652-0019>



Nguyen Xuan Ngoc hold a bachelor's degree in biotechnology from Ton Duc Thang University in 2024. She is currently employed as a staff member in the Laboratory of Organic Chemistry and Polymer Technology in Institute of Chemical Technology (ICT), Vietnam Academy of Science and Technology (VAST).

Email: nguyexuannhocbtr@gmail.com. ORCID: <https://orcid.org/0009-0009-5179-6620>



Assoc. Prof. Hoang Thi-Kim-Dung received the Ph.D. degree in Organic Chemistry from Ministry of Education and Training, Vietnam, in 2010. She is currently Head of the Laboratory of Organic Chemistry and Polymer Technology and Director of ICT, VAST. She has had active studies with over 50 peer-reviewed articles in high-index journals and applied for 5 more patents. She has also been the project supervisors of more 8 projects which were funded by VAST, Department of Science and Technology of Hochiminh City, National Foundation for Science & Technology Development (Vietnam), and so on. Her research interests include organic synthesis, medicinal chemistry, *in silico* studies for bioactivities, optical and chemical sensors, polymers and materials for environmental detection and treatment.

Email: hoangthikimdung@gmail.com. ORCID: <https://orcid.org/0000-0002-9369-8051>



Dr. Ton Anh Khoa received his Ph.D. degree at the Department of Chemical Engineering from National Taiwan University of Science and Technology, Taiwan in 2019. After graduating, he worked as a senior scientist at a Nanotechnology Fabrication company for 2 years and a postdoc researcher for 1.5 years at Academia Sinica, Taiwan which focused on targeted nanomedicine for cancer treatment, both diagnostics and therapeutics.

Currently, he has worked as an active and enthusiastic researcher of the Laboratory of Organic Chemistry and Polymer Technology at Ho Chi Minh Institute of Chemistry, Vietnam Academy of Science and Technology.

His interests are nanomedicine for cancer treatment, drug delivery systems, nano-based sensors for biomedical and environmental detection as well as green, sustainable cosmetics.

Email: kevinton150691@gmail.com. ORCID: <https://orcid.org/0009-0001-0121-5869>. Phone: +84-98.998.9994

Corresponding author