Utilization of Green Tea Extract on Anti-aging Cream with Butylated Hydroxytoluene (BHT) and Tertiary Butylhydroquinone (TBHQ): Physical Stability Aspect

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Abstract. Green tea (*Camellia sinensis* (L.) Kuntze) is a potent natural ingredient with flavonoid content that can be used as an antioxidant and anti-aging for skincare products. The formula containing green tea extract is usually formulated as oil in water emulsion or cream. The active components of green tea are catechins which are characterized as less stable against oxidation. Therefore, it is needed to add other antioxidants such as ButylatedHydroxy Toluene (BHT) and Tertiary–Butyl Hydroquinone (TBHQ) to protect the product from degradation. The aim of this study was to obtain a physically stable antiaging cream formula. Each formula was tested for physical stability by measuring several variables including organoleptic, pH, relative density, viscosity, and flow properties, as well as droplet size. Accelerated stability testing is carried out for 3 mo at 40 °C and 75 % relative humidity. The results found that cream with the BHT formula is more stable than the TBHQ formula in terms of the parameters of density and droplet size. While the TBHQ formula only gave better stability in pH, the other variables from both formulas remain stable in 3 mo. It can be concluded that the green tea extract cream with BHT antioxidant is more stable than the TBHQ.

Keywords: *Camellia sinensis* (L.) Kuntze, Environmental friendly technology, flavonoid, natural antioxidant, prevent premature aging

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1 Introduction

Antioxidant compounds can be obtained from synthetic chemicals or from natural ingredients [1]. Antioxidant compounds isolated from natural sources usually come from plants. These natural plant antioxidant compounds are generally phenolic or polyphenolic compounds that can be present in the form of flavonoids including flavones, flavonols, isoflavones, catechins, and chalcones [2–4]. One of the natural ingredients that can be a source of natural antioxidants and rich in catechins is green tea (*Camellia sinensis* (L.) Kuntze) [5].

Antioxidants or anti-aging compounds are often used to prevent premature aging. Aging is a natural degeneration process that cannot be avoided and occurs constantly. When entering their forties, a person will experience aging shown by changes in their skin. Beginning with wrinkles and dryness followed by skin pigmentation, darkening the skin color. However, due to sun exposure and an unhealthy lifestyle, people may experience premature aging. Premature aging can take the form of wrinkles, dullness, and even the presence of melasma or blackish brown patches which, if not treated immediately, can cause black spots that generally occur in women [6]. Antioxidants can inhibit free radicals generated from sun exposure, especially UV A radiation with a wavelength of 320 nm to 400 nm, which is the main cause of premature aging (photoaging) [7]. Generally, antioxidants are defined as compounds that can delay, slow down and prevent the oxidation process.

To provide an anti-aging cream formula containing green tea extract, several formulas with catechin as the active ingredient has been produced [8], however because of the instability of a natural compound against oxidation, it is necessary to add antioxidants to protect the product from oxidation and provide better physical stability [9]. Two oil in water cream containing green tea extract formulations with the addition of Butylated Hydroxy Toluene (BHT) and Tertiary Butyl Hydroquinone (TBHQ) have been developed. The physical stability of cream containing green tea extract with BHT has increased compared to those without BHT, although the improvement was not very encouraging [10]. To further improve the physical stability, we have investigated the stability of green tea cream with another antioxidant that was reported to be a better antioxidant, namely Tertiary–Butyl Hydroquinone (TBHQ) [11]. In this study, the authors compared the physical stability of oil in water cream preparations containing green tea extract with TBHQ and BHT. The purpose of this study was to obtain a physically stable anti-aging cream containing green tea extracts that are used to treat premature aging.

The physical stability of cream preparations containing green tea extract was tested using the accelerated stability test method [12]. The variables observed and measured in this study are organoleptic, degree of acidity (pH), relative density, viscosity, and flow properties, as well as the droplet size of cream preparations. All formulated creams were stored in a climatic chamber with a temperature of 40 °C, 75 % relative humidity for 3 mo [13, 14]. Observations and measurements in triplicate were carried out immediately after preparations has been manufactured [t_0], and every 14 d for 3 mo.

2 Methods

2.1 Materials and instruments

The materials used in this study were green tea extract, stearic acid, cetyl alcohol, isopropyl palmitate, sepicid, HB, sorbitan monostearate, sorbitol solution (70 %), polysorbate 60,

oleum olivarum, glycerin, spiegel, squalene, TBHQ, BHT, and aqua demineralization. The instruments used in this study are climatic chamber KBF 240, Cyberscan 510 pH–meter, Brookfield Cone and Plate AT 71362 viscometer, picnometer, Sartorius analytical balance, glass instruments, 99017420002 series optical microscope, 40 Zeiss Axioscope photomicroscope, and water bath.

2.2 Formulation methods and stability testing

Cream preparations are manufactured according to the formula and composition as shown in Table 1. Each formula is made on a laboratory scale weighing 20 g in two replications and each replication is subjected to a physical stability test with five test parameters and three observations. Physical stability testing using the accelerated stability test method begins by inserting the cream in its container into a climatic chamber which has been set at 40 °C and 75 % relative humidity. Then the physical characteristics are tested at each point of observation for 3 mo. The physical properties of the tested preparations include organoleptic, pH, relative density, viscosity, and flow properties, as well as droplet size.

Table 1. Formula composition and weighing table of anti-aging cream preparation containing green tea extract and TBHQ or BHT

Materials	Amount	Formula A (TBHQ)	Formula B (BHT)
Green tea extract	0.25 %	50 mg	50 mg
Stearic acid	6 %	1.2 g	1.2 g
Cetyl alcohol	1 %	200 mg	200 mg
Isopropyl palmitate	1 %	200 mg	200 mg
Sepicid HB	0,15 %	30 mg	30 mg
Sorbitan monostearate	4 %	800 mg	800 mg
Sorbitol solution 70 %	3 %	600 mg	600 mg
Polysorbate 80	3 %	600 mg	600 mg
Oleum olivarum	6 %	1.2 g	1.2 g
Glycerol	1.5 %	300 mg	300 mg
Sepigel	1 %	200 mg	200 mg
Squalane	5 %	1 g	1 g
TBHQ	0.05 %	10 mg	–
BHT	0.05 %	–	10 mg
Aquadem	68.05 %	13.61 g	13.61

2.3 Data analysis

The data of 3 mo observation on the pH, relative density, viscosity, and droplet size of the preparations were then analyzed using the one–way ANOVA, while the organoleptic data and flow properties were analyzed descriptively [15, 16].

3 Results and discussion

An amount of 140 g of anti-aging cream containing green tea extract is prepared and then packed with a weight of 20 g each into seven plastic pots. Two types of formulas are distinguished in their antioxidant additives [8], the first formula with BHT and the second formula with TBHQ. Each formula was made in two replications. The specifications for the preparation of oil in water cream (emulsion), no phase separation, possess a light

moisturizing cream odor, white in color, pH of 4.85, and a 1.0 g mL^{-1} density.

3.1 Organoleptic

Organoleptic observations of anti-aging cream preparations containing green tea extract consisted of form, smell, and color. Visually, it appears that all anti-aging cream preparations both in the TBHQ and BHT formulas from week 0 wk to 12 wk do not change in form, that is, they remain in the form of cream or oil emulsion in water and there is no visible phase separation. After being observed through the sense of smell, it was found that all anti-aging cream preparations in the BHT and TBHQ formulas from 0 wk to 12 wk had no change in odor. Visually, there is a color change in the anti-aging cream preparations from 0 wk to 12 wk. The BHT formula at 0 wk to 6 wk is white, while at 8 wk to 12 wk it changes color to beige. Meanwhile, the color change was even faster in the TBHQ formula. During 4 wk to 12 wk the color changes to beige.

3.2 Emulsion Type

The success of patchouli oil extraction from *Pogostemon cablin* Benth. leaves based on patchouli alcohol (PA) quality is influenced by fertilization factors and cultivation conditions, harvest time, fermentation, drying and scaling down, and extraction methods. The water–bubble distillation (WBD) and microwave-assisted hydrodistillation (MHD) are efficient methods for extracting good quality patchouli oil from *Pogostemon cablin* Benth. leaves.

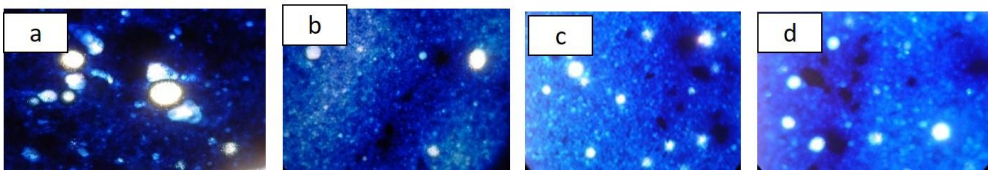

Fig. 1. Emulsion type of anti–aging cream preparation with TBHQ formula during week 0 (a), 4, (b), 8 (c), and 12 (d)

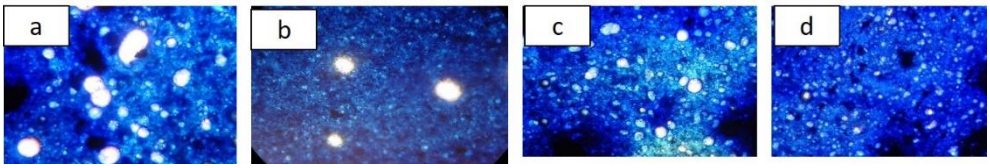

Fig. 2. Emulsion type of anti–aging cream preparation with BHT formula during week 0 (a), 4, (b), 8 (c), and 12 (d)

3.3 pH

The pH measurement of anti-aging cream preparations containing green tea extract from 0 to 12 wk carried out using a Cyberscan 510 pH meter. Resulting in a profile as shown in Figure 3, which indicates that the pH of both creams are relatively stable during 12 wk of storage.

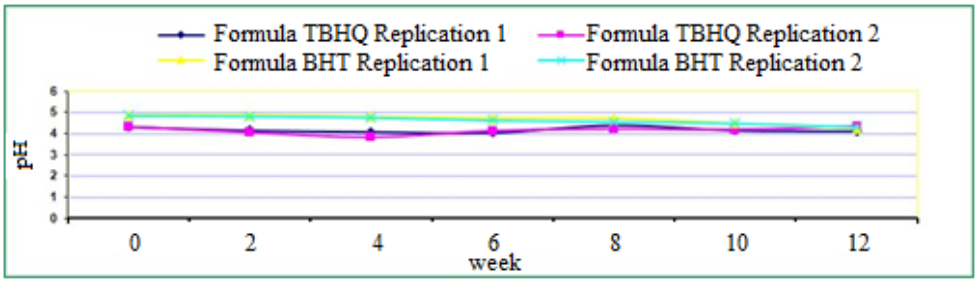

Fig. 3. pH value vs storage time profile

From an ANOVA test for the BHT and TBHQ formulas, a significance value of 0.000 was obtained, after being compared with the α value of 0.05. It can be concluded that the significance is < α, meaning H_1 was accepted. Therefore, there was at least one different variant in the seven samples (observation time), hence the Tamhane test was carried out. The results of the analysis of the significance of the pH data on the formula with BHT showed that there was a significant difference in the pH of the cream preparations when first made with the cream after being stored for the specified observation time. The same result was observed for the TBHQ formula, it was found that there was a significant difference in pH at 0 wk and 12 wk. The decrease in pH was possibly due to the hydrolysis action that occurs to the cream materials which caused an increase in the concentration of H^+ ions [17].

3.4 Relative density

The results of relative density observation from cream preparations carried out at 0 wk to 12 wk were reported as the storage time vs relative density profile of the cream preparations that can be seen in Figure 4.

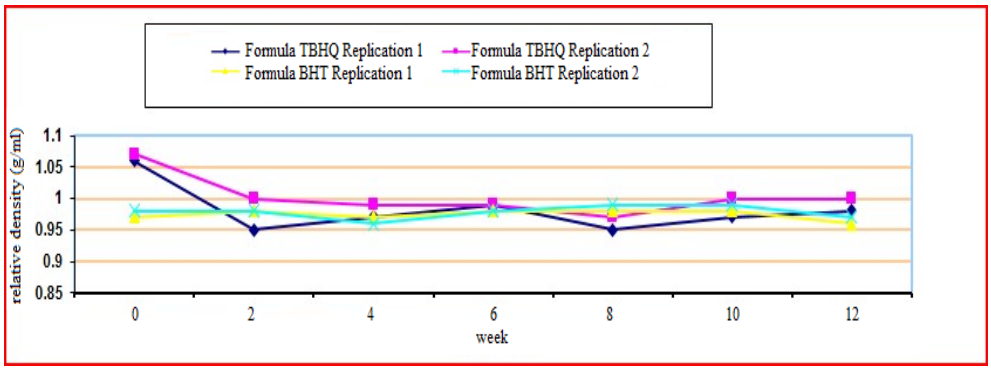

Fig. 4. Relative density vs storage time profile

From the ANOVA test for the BHT and TBHQ formulas, a significance value of 0.000 was obtained, after being compared with the α value of 0.05. It was concluded that the significance < α, meaning H1was accepted. Therefore, there is at least one different variant in the seven samples (observation time), hence the Tamhane test is carried out. From the data significance result of the BHT formula's relative density, we found that there was a significant difference of relative density between 4 wk and 6 wk; 4 wk and 8 wk; 6 wk and 12 wk.

Likewise, from the data significance result of the TBHQ formula's relative density,

there was a significant difference in the cream preparations when they were initially prepared with the ones that had been stored until the designated observation time. The reduction in relative density is presumed due to the release of oil during initial weighing or it could also be from a decrease in the concentration of surfactants or inaccurate selection of surfactants that have not been able to function to reduce the interfacial tension properly, causing the separation of the oil and water phases in the preparation. Another possibility that could occur is water evaporation caused by the storage processing at relatively high temperatures [18].

3.5 Viscosity and flow properties

The viscosity and flow properties of anti-aging cream preparations containing green tea extract were observed at 0 wk to 12 wk using Brookfield Cone and Plate Digital Viscometer AT 71362. CPE 41 spindles were used at a speed of 0.3 rpm. Table 2. showed the viscosity of the BHT formula and Table 3. shows the viscosity of TBHQ formula. The storage time vs viscosity profile of the preparation can be seen in Figure 5.

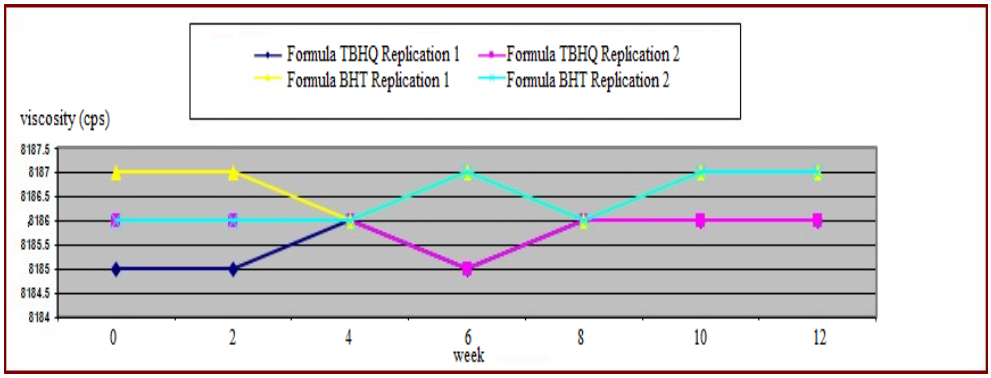

Fig. 5. Flow properties profile of anti-aging cream (viscosity vs storage time)

Table 2. The viscosity of anti-aging cream preparation containing green tea extract and BHT

Storage Week	Formula BHT							
	Replication 1				Replication 2			
	1	2	3	Mean	1	2	3	Mean
0	8 183	8 187	8 186	8 185	8 184	8 186	8 186	8 186
2	8 183	8 186	8 187	8 185	8 185	8 186	8 186	8 186
4	8 186	8 186	8 187	8 186	8 185	8 185	8 186	8 186
6	8 183	8 185	8 186	8 185	8 183	8 186	8 187	8 185
8	8 187	8 186	8 186	8 186	8 187	8 186	8 186	8 186
10	8 187	8 187	8 185	8 186	8 186	8 186	8 187	8 186
12	8 187	8 186	8 187	8 186	8 186	8 186	8 186	8 186

Table 3. The viscosity of anti-aging cream preparation containing green tea extract and TBHQ

Storage Week	Formula TBHQ							
	Replication 1				Replication 2			
	1	2	3	Mean	1	2	3	Mean
0	8 187	8 187	8 187	8 187	8 186	8 186	8 186	8 186

(Continued on next page)

Table 3. Continued

Storage Week	Formula TBHQ							
	Replication 1				Replication 2			
	1	2	3	Mean	1	2	3	Mean
2	8 187	8 187	8 187	8 187	8 185	8 186	8 186	8 186
4	8 186	8 186	8 186	8 186	8 186	8 186	8 186	8 186
6	8 187	8 187	8 187	8 187	8 188	8 187	8 186	8 187
8	8 186	8 186	8 187	8 186	8 186	8 187	8 186	8 186
10	8 187	8 187	8 187	8 187	8 186	8 187	8 187	8 187
12	8 187	8 187	8 187	8 187	8 187	8 186	8 187	8 187

The observation result for the flow properties of cream preparation scarried out at 0 wk to 12 wk, based on the viscosity measurement data yielded a stable viscosity as shown in Figure 5. Assumed from data analysis with the addition of the Tamhane test, viscosity data for both BHT and TBHQ formulas hold no significant difference for each observed time, from 0 wk to 12 wk. The flow properties of the cream preparations are observed as pseudoplastic [18].

3.6 Droplet size

The droplet size (dvs) observation results of cream preparations based on micromeritic data can be seen in Table 4 for the BHT formula and Table 5 for the TBHQ formula whereas the storage time vs droplet size profile can be seen in Figure 6.

Table 4. The droplet size of anti-aging cream preparation containing green tea extract and BHT during 0 wk to 12 wk

Storage Week	Formula BHT							
	Replication 1				Replication 2			
	1	2	3	Mean	1	2	3	Mean
0	6.75	3.05	4.38	4.73	2.17	2.98	2.92	2.69
2	4.03	2.16	2.54	2.91	1.9	2.17	2.1	2.06
4	2	1.67	1.88	1.85	1.2	2.16	2.07	1.81
6	2.43	4.76	4.09	3.76	1.57	2.07	1.97	1.87
8	2.99	2.31	2.64	2.65	1.3	2.07	1.82	1.73
10	3.59	1.67	3.54	2.93	1.6	2.09	1.68	1.79
12	2.63	2.21	1.95	2.26	1.71	2	1.84	1.85

Table 5. The droplet size of anti–aging cream preparation containing green tea extract and TBHQ during 0 wk to 12 wk

Storage Week	Formula TBHQ							
	Replication 1				Replication 2			
	1	2	3	Mean	1	2	3	Mean
0	3.33	3.07	3.18	3.19	3.77	2.52	2.9	3.06
2	2.09	2.39	2.53	2.34	2.54	2.41	2.57	2.51
4	2.66	2.65	2.88	2.73	3.09	2.85	2.95	2.96
6	2.85	2.84	2.87	2.85	2.8	2.99	3.07	2.95
8	2.54	2.49	2.71	2.58	3.22	2.52	3.3	3.01
10	3.62	3.31	3.18	3.37	2.68	2.03	2.03	2.25
12	2.85	2.78	2.82	2.82	3.38	2.3	2.7	2.79

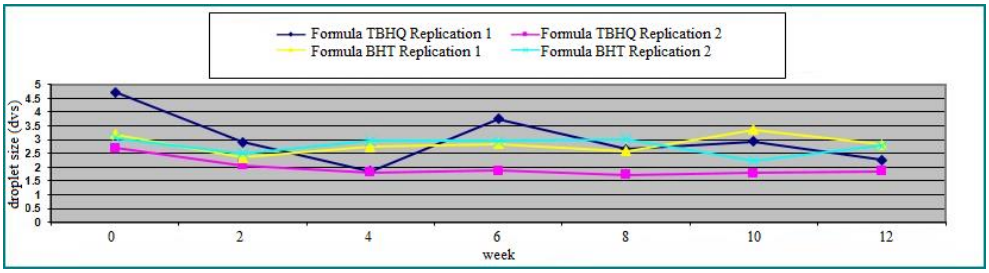

Fig. 6. Droplet size vs storage time profile

A significance value of 0.030 was obtained through the one–Sample Kolmogorov–Smirnov Test carried out on the BHT formula. That value was then compared with the α value of 0.05, which showed that the test result's significance was < α, meaning H_1 was accepted. Therefore the droplet size was not normally distributed. Where as for the TBHQ formula, a significance value of 0.939 was obtained, after being compared with the α value of 0.05, the result was that the significance is > α, meaning H_0 was accepted, showing that the droplet size was normally distributed. The histogram chart for the droplet size distribution is shown in Figure 7a and 7b for BHT and TBHQ formula respectively.

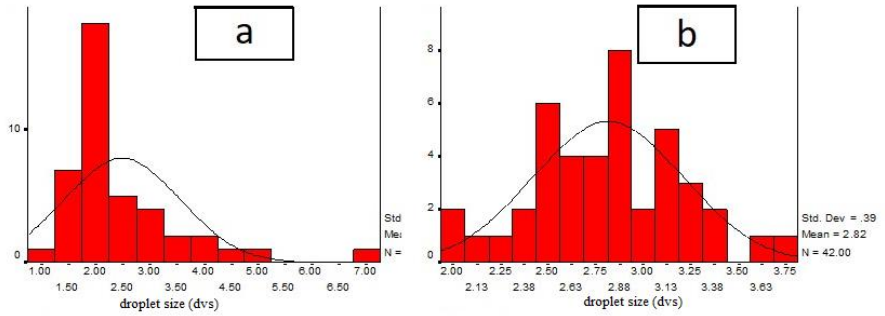

Fig. 7. Droplet Size Histogram of Anti–aging Cream for Formula BHT (a) and Formula TBHQ (b)

The droplet size data significance analysis result of the BHT formula showed that there was no significant difference at any time. As for the data on the droplet size of the TBHQ formula, it was found that there were significant differences between (2 wk and 4 wk) and (2 wk and 6 wk). Based on the results of statistical data analysis using the one–way ANOVA test, it was found that there were differences in particle size for the TBHQ formula during 0 wk and 12 wk, which is probably due to the increased solubility during the storage of the preparation in the climatic chamber [18].

4 Conclusion and suggestions

Based on the results of the study, green tea anti-aging cream with the BHT formula is more stable than the TBHQ formula in terms of relative density and droplet size. While the TBHQ formula is better in terms of pH stability. For other parameters, both formulas show stable results. It can be concluded that anti-aging cream preparations containing a combination of green tea extract and BHT are more stable than anti-aging creams containing a combination of green tea extract and TBHQ. For future studies, it is suggested to use a combination of BHT and TBHQ to obtain anti-aging creams that are more stable against the oxidation process.

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