การศึกษาประสิทธิผลของกลีเซอลิล ออกทิล แอสคอเบท และ ทรี โอ แอสคอบิก แอซิด
ที่มีผลต่อความกระจ่างใสและความยืดหยุ่นของผิวหนัง

# The Effect of Glyceryl Octyl Ascorbate and 3-0-Ethyl Ascorbic Acid
# on Skin Whitening and Skin Elasticity

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## Abstract

Skin whitening and anti-aging is desire in Asian consumer cosmetic because whiten the skin boost the confident and youthful look. Vitamin c is the common use in cosmetic and has new derivative but less study. We hypothesized that new vitamin c derivative can be used to skin whitening and improve skin elasticity.

Objective: To study the effect glyceryl octyl ascorbate and 3-0-ethyl ascorbic acid on skin whitening and skin elasticity.

Method: 30 volunteers, aged 20-35 were double blind randomly the sample. The sample formulation was applied on face. Used 2 pumps (approximately 0.5 g) on clearly face twice a day; in the morning and before bed, for 8 weeks. Mexameter and cutometer were used to measure and record.

Result: Both of vitamin c was shown statistic significantly ($p<0.001$) decreased melanin index on cheek but on forehead shown no shown significantly decreased. Comparing the reduction of melanin index between 3 groups shown statistically significantly ($p<0.05$) different when compare group 1,2 with group 3. The elasticity (R0, R2, R6 and R7) of both vitamin c shown statistically significant ($p<0.001$) increase elasticity excepted R0 value. Comparing the change of elasticity between 3 groups shown significantly ($p<0.001$) different when compare group 1,2 with group 3 excepted R0 value.

**Keywords:** glyceryl octyl ascorbate / 3-0-ethyl ascorbic acid / whitening / elasticity

**บทคัดย่อ**

      จากผลสำรวจผู้บริโภคสินค้าเครื่องสำอางในอาเซียนพบว่า ผู้บริโภคส่วนใหญ่เลือกซื้อสินค้าเครื่องสำอางประเภทเพิ่มความกระจ่างใสและลดเลือนริ้วรอย เนื่องจากการผิวที่กระจ่างใสเป็นที่ยอมรับกันในภูมิภาคนี้และการคงความอ่อนเยาว์ยังช่วยเสริมความมั่นใจได้อีกด้วย ผลิตภัณฑ์เครื่องสำอางส่วนใหญ่จึงเลือกใช้ไวตามินซี ในการเพิ่มความกระจ่างใสและลดเลือนริ้วรอย ปัจจุบันมีการพัฒนาไวตามินซีออกมามากในหลายอนุภัณฑ์ แต่ยังไม่มีผลการศึกษาวิจัยถึงประสิทธิผลในการเพิ่มความกระจ่างใสและลดเลือนริ้วรอยมากนัก

      วัตถุประสงค์ เพื่อศึกษาประสิทธิผลของกลีเซอลิล ออกทิล แอสคอแบท และ ทรี โอ เอททิล แอสคอบิก แอซิด ในความกระจ่างใสและความยืดหยุ่นของผิว

      วิธีการศึกษา ใช้อาสาสมัครจำนวน 30 คน ช่วงอายุ 20 – 35 ปี ถูกแบ่งเป็น 3 กลุ่ม โดยให้ทาตัวยาเป็นเวลา 8 สัปดาห์ ใช้ทาบนใบหน้าที่สะอาดเช้าและก่อนนอน กดจำนวน 2 ครั้ง ติดตามผลทุก 2 สัปดาห์โดยใช้เครื่อง Mexameter วัดจำนวนเมลานินในผิวและ Cutometer วัดความยืดหยุ่นของผิวโดยใช้ค่า R0 คือความกระชับ,R2 คือ ความยืดหยุ่นสุทธิ, R6 คือ เวลาที่ผิวใช้ในการหดกลับหลังจากถูกดึก และ R7 คือคความสามารถในการหดกลับของผิวสู่ภาพเดิม

      ผลการวิจัย พบว่าไวตามิน ซีทั้ง 2 ชนิดสามารถลดได้จำนวนของเมลานินได้เหมือนกันที่บริเวณหน้าแก้ม โดยพบการลดลงอย่างมีนัยสำคัญทางสถิติ (p<0.001) ตั้งแต่สัปดาห์ที่ 2 ของการศึกษา แต่จำนวนของเมลานินที่หน้าผาก มีแนวโน้มลดลงเช่นกัน แต่ไม่พบความแตกต่างอย่างมีนัยสำคัญทางสถิติ เมื่อค่าของเมลนานินที่ลดลงของทั้ง 3 กลุ่มมาเปรียบเทียบกันพบว่า การลดลงของไวตามินซีทั้ง 2 ชนิด พบความแตกต่างอย่างมีนัยสำคัญทางสถิติ (p<0.05) เมื่อเทียบกับกลุ่มควบคุม ความยืดหยุ่นของผิวพบว่า ไวตามินซี ทั้ง 2 ชนิดสามารถเพิ่มความยืดหยุ่นของผิวได้ทุกตัวแปร ยกเว้นค่า R0 หรือ ความกระชับของผิว โดยพบความแตกต่างอย่างมีนัยสำคัญทางสถิติ (p<0.001) เมื่อนำค่าที่ลดลงของทุกตัวแปรมาเปรียบเทียบกับพบว่า ไวตามินซีทั้ง 2 ชนิด พบความแตกต่างอย่างมีนัยสำคัญทางสถิติ (p<0.001) เมื่อเทียบกับกลุ่มควบคุม ยกเว้นค่า R0 หรือ ความกระชับของผิว

**Introduction**

From skincare survey, skin whitening and anti-aging product is the most desire in Asian cosmetic consumer. They use skin whitening and anti-aging because it can boost the confident, culture conception (darker skin tones denoting lower social class and fair skin associated to youthful look. (Ramaa C., 2014)

Skin whitening and anti-aging agent that are common use in cosmetic is vitamin C or ascorbic acid. Nowadays, a new ascorbic acid derivative is glyceryl octylascorbate; otcyl and glyceryl group is added in to an ascorbic acid. It shows the humectant properties from glycerin, anti-bacterial from octanol, fibroblast activation, melanogenesis suppressing and wrinkle improving effect (Takeshi N., et al., 2013). Another new one is 3-0-ethyl ascorbic acid. Its show inhibits melanogenesis properties, stimulate collagen synthesis and anti-inflammation (Corum, 2011).

Melanogenesis is a biosynthesis pathway for produce of the pigment melanin in the human skin. Melanin plays an important role to protect the skin from free radicals that generate within the cytoplasm in shielding the host from various types of ionizing, including UV light. Melanin is produced from a process called melanogenesis through a combination of enzymatically catalyzed and chemical reaction. The melanin synthesis pathway occurred in melanosomes, consist of eumelanin and pheomelanin.

Skin aging is a complex biological process associated with change of skin physical properties. Skin aging has 2 factors; intrinsic and extrinsic aging. Intrinsic aging or innate aging is occurring by natural of an accumulative of irreversible age-associated degenerative change. Free radical or oxidative stress theory of aging proposes that the accumulation of oxidative cellular damage is a major contribute to the aging process and a key determination of longevity of the species. Aging is associated with change of DNA structure, proteins, the lipids, and prostaglandins. The accumulation of these molecular changes, in particular proteins, forms the basis of cellular aging. During skin aging the effectiveness of endogenous antioxidant system is diminished and ROS formation causes mainly DNA damage. Oxidative stress due to aging process not only induces DNA damage, but also, intracellular lipid peroxidation, abnormal protein oxidation reactions, all of which result in cell damage, inflammation, immune suppression, oxidative stress, hyperplastic responses in skin as

well as perturbed hormonal balances, and premature skin aging (Alfredo G. *et al*, 2014). Extrinsic aging is superimposed in intrinsic skin aging process primarily to UVR (solar ultraviolet radiation) and partly by other factor such as infrared light smoking and air pollution (Masamitsu I, 2009). At the microscopic level, sun damage is characterized by accumulate of abnormal elatotic material, process called solar elastotic. At the damage advance, pleiomorphic elastotic structure predominant. Although these structures are compose of elastotic material, do not have a normal fibrous appearance. Confocal laser scaning microscopy using elastin-specific antibodies show how the normal morphology of dermal elastic fiber changes to one of globular elastotic structure. Solar elastotic structure is shown by confocal vicroscopy to replace almost all of the dermal collagen, which dominant component in healthy skin

## Objectives

1. To study the effect of glyceryl octyl ascorbate and 3-O ethyl ascorbic acid on skin whitening
2. To study the effect of glyceryl octyl ascorbate and 3-O ethyl ascorbic acid on skin elasticity

## Materials and Methods

This study was a Randomized, Double-blind, Placebo-Controlled Trail. 30 subjects age 20-35 years old, who wanted to whiten their facial skin at Mae Fah Luang University Hospital, Bangkok and matched with all inclusion were recruited. The formulation no.1 or glyceryl octyl ascorbate, no.2 or 3-0-ethyl ascorbic acid and no.3 or placebo were applied on face in each group. They used 2 pumps (approximately 0.5 g) on clearly face twice a day; in the morning and before bed, for 8 weeks. The measurement was followed at week 0, 2, 4, 6 and 8 while using the samples. The melanin index and elasticity value (R0, R2, R6 and R7) were recorded. Repeated measurement ANOVA were used to analyze the different amount of melanin index and skin elasticity value in each group and one-way ANOVA to analyze the different amount of melanin index and skin elasticity value between groups.

**Result**

     This study divided subjects in to 3 groups, each group contained 11 subjects, group 1 as glyceryl octyl ascorbate, group 2 as 3-0-ethyl ascorbic acid and group 3 as placebo. The study was measured at Mea Fah Luang Hospital Bangkok, treatment for 2 months from May to July 2016. During the study, 3 subjects were quit while studying; 2 subjects from group 1, 1 subject from group 2.

**Table 1** The General information of each group

|  | Number of subject | Average ages | Std. Deviation | Sex Male | Female |
|---|---|---|---|---|---|
| Group 1 | 9 | 30.11 | 2.66 | 3 | 6 |
| Group 2 | 10 | 28.00 | 3.01 | 1 | 10 |
| Group 3 | 11 | 29.45 | 3.14 | 0 | 11 |

     From table 4.1, Group 1 contained 9 subjects, group 2 contained 10 subjects and group 3 contained 11 subjects. The average ages of each group were $30.11\pm2.66$, $28.00\pm3.01$ and $29.45\pm3.14$ respectively.
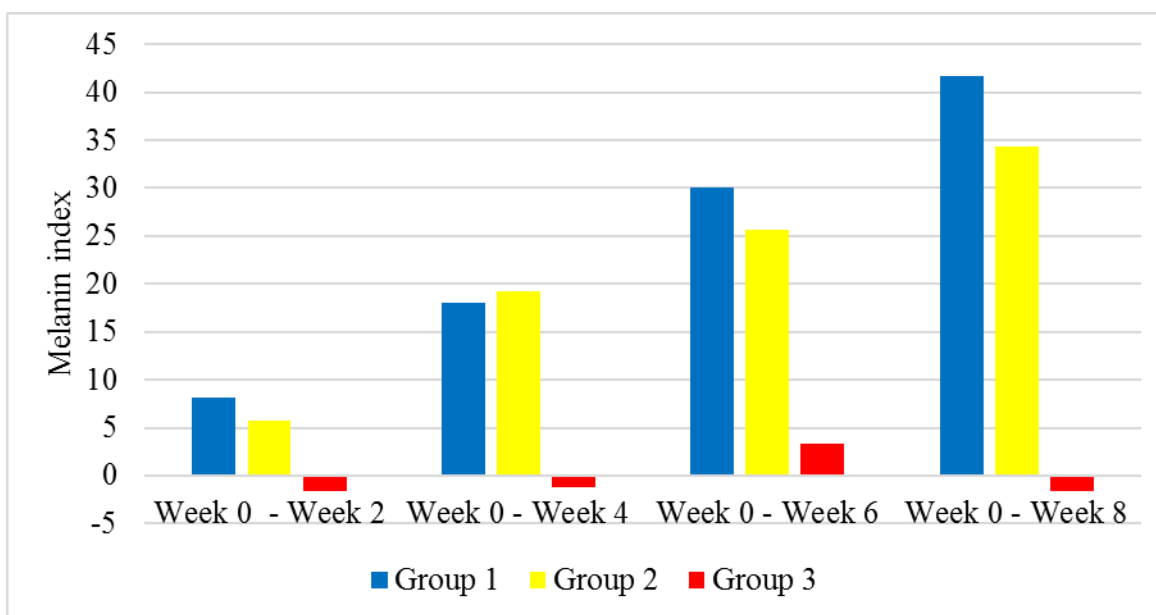
Result of Melanin Index

**Table 2** The comparison of melanin index on cheek between 3 groups

|  | Week 0 | Week 2 | Week 4 | Week 6 | Week 8 | p-value |
|---|---|---|---|---|---|---|
| Group 1 | 265.778 | 257.667 | 247.759 | 235.648 | 224.148[b] | <0.001 |
| SD | 54.116 | 53.536 | 54.022 | 57.274 | 55.591 |  |
| Group 2 | 254.333 | 248.567 | 235.15[a] | 228.65[a] | 220.033[a] | <0.001 |
| SD | 32.63 | 33.7 | 33.846 | 34.058 | 34.606 |  |
| Group 3 | 287.727 | 289.379 | 288.909[a] | 284.364[a] | 289.394[a,b] | 0.38 |
| SD | 53.486 | 53.278 | 49.359 | 54.418 | 50.741 |  |
| p-value | 0.281 | 0.139 | 0.033 | 0.031 | 0.004 |  |

Note; a is statistically significant between group 2 and 3

    b is statistically significant between group 1 and 3

**Figure 1** The reduction of melanin index on cheek between 3 groups



From table 2 and figure 1, the average of melanin index on cheek of group 1 (glyceryl octyl ascorbate) on week 2, 4, 6 and 8 were 257.67±53.54, 247.76±54.02, 235.65±57.27 and 224.15±55.59 respectively. When used mean difference of melanin index to plot a graph, show the reduction of melanin trend to be increase or melanin index was decrease. Repeated measurement ANOVA used to analyze the mean difference of melanin index among 5 weeks of measurement, shown statistically significant (p<0.001).

The average of melanin index of group 2 (3-0-ethyl ascorbic acid) on week 2, 4, 6 and 8 were 248.567±33.7, 235.15±33.846, 228.65±34.058 and 220.033±34.606 respectively. When used mean difference of melanin index to plot a graph, show the reduction of melanin trend to be increase or melanin index was decrease. Repeated measurement ANOVA used to analyzed the mean difference of melanin index among 5 weeks of measurement, shown statistically significant (p<0.001) as table 1.
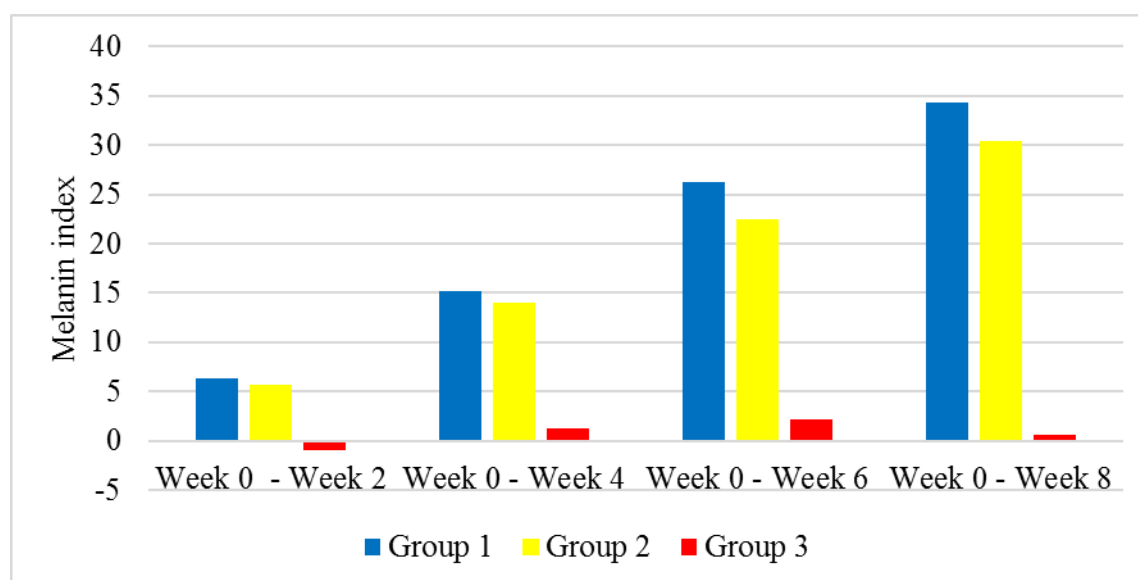
The average of melanin index of placebo on week 2, 4, 6 and 8 were 289.379±53.278, 288.909±49.359, 284.364±54.418 and 289.394±50.741 respectively. When used repeated measurement ANOVA to analyze the mean difference of melanin index among 5 weeks of measurement, shown no statistically significant as table 1.

When compare the change of melanin index on cheek between 3 groups at week 0, mean of melanin index showed no statistically significant as table 4.8 but mean of melanin index at week 4 showed statistically significant (p-value < 0.05) between group 2 and 3. At the end of the study, the change of melanin index all group showed statistically significant (p-value < 0.05) when analyzed by one way-ANOVA.

**Table 3** The comparison of melanin index on forehead between 3 groups

|         | Week 0  | Week 2  | Week 4  | Week 6  | Week 8  | p-value |
|---------|---------|---------|---------|---------|---------|---------|
| Group 1 | 308.204 | 301.852 | 293.056 | 281.907 | 273.852 | <0.001  |
| SD      | 45.793  | 46.538  | 44.675  | 50.026  | 44.443  |         |
| Group 2 | 326.833 | 321.167 | 312.767 | 304.383 | 296.417 | <0.001  |
| SD      | 53.977  | 55.172  | 55.177  | 54.859  | 53.129  |         |
| Group 3 | 316.121 | 317.076 | 314.848 | 313.909 | 315.47  | 0.293   |
| SD      | 57.357  | 56.891  | 54.756  | 55.756  | 54.51   |         |
| p-value | 0.746   | 0.714   | 0.608   | 0.417   | 0.214   |         |

**Figure 2** The reduction of melanin index on forehead among 3 groups



From table 3 and figure 2, the average of melanin index on forehead of glyceryl octyl ascorbate on week 2, 4, 6 and 8 were 301.852±46.538, 293.056±44.675, 281.907±50.026 and 273.852±44.443 respectively. Repeated

measurement ANOVA was used to analyzed the mean difference of melanin index among 5 weeks of measurement, shown statistically significant (p<0.001) as table 3.

Table 3 and figure 2, the average of melanin index of 3-0-ethyl ascorbic acid on week 2, 4, 6 and 8 were 321.167±55.172, 312.767.15±55.177, 304.383±54.859 and 296.417±53.129 respectively. When used mean difference of melanin index to plot a graph, show the reduction of melanin trend to increase or melanin index was decrease. Repeated measurement ANOVA shown statistically significant (p<0.001) difference of melanin index at 5 weeks of measurement as table 3

The average of melanin index of placebo on week 2, 4, 6 and 8 were 317.076±56.891, 314.848±54.637, 313.909±55.756 and 315.470±54.510 respectively. The mean difference of melanin index among 5 weeks showed no statistically significant as table 3 (when used repeated measurement ANOVA to analyze).

When compare the change of melanin index on forehead between 3 groups shown after 8 weeks was shown mean of melanin index does not difference by no statistically significant when analyzed by one way-ANOVA as table 3.

Result of Elasticity (R0, R2, R6 and R7)

**Table 4** The comparison of R0 value between 3 groups

|  | Week 0 | Week 2 | Week 4 | Week 6 | Week 8 | p-value |
|---|---|---|---|---|---|---|
| Group 1 | 0.096 | 0.076 | 0.16 | 0.184 | 0.216 | 0.1 |
| SD | 0.097 | 0.03 | 0.215 | 0.076 | 0.069 | |
| Group 2 | 0.079 | 0.139 | 0.17 | 0.442[a] | 0.507 | 0.068 |
| SD | 0.02 | 0.085 | 0.102 | 0.493 | 0.661 | |
| Group 3 | 0.078 | 0.094 | 0.093 | 0.076[a] | 0.104 | 0.477 |
| SD | 0.015 | 0.063 | 0.063 | 0.015 | 0.066 | |
| p-value | 0.719 | 0.111 | 0.387 | 0.022 | 0.067 | |

Note; a is statistically significant between group 2 and 3

**Figure 3** The reduction of R0 value between 3 groups
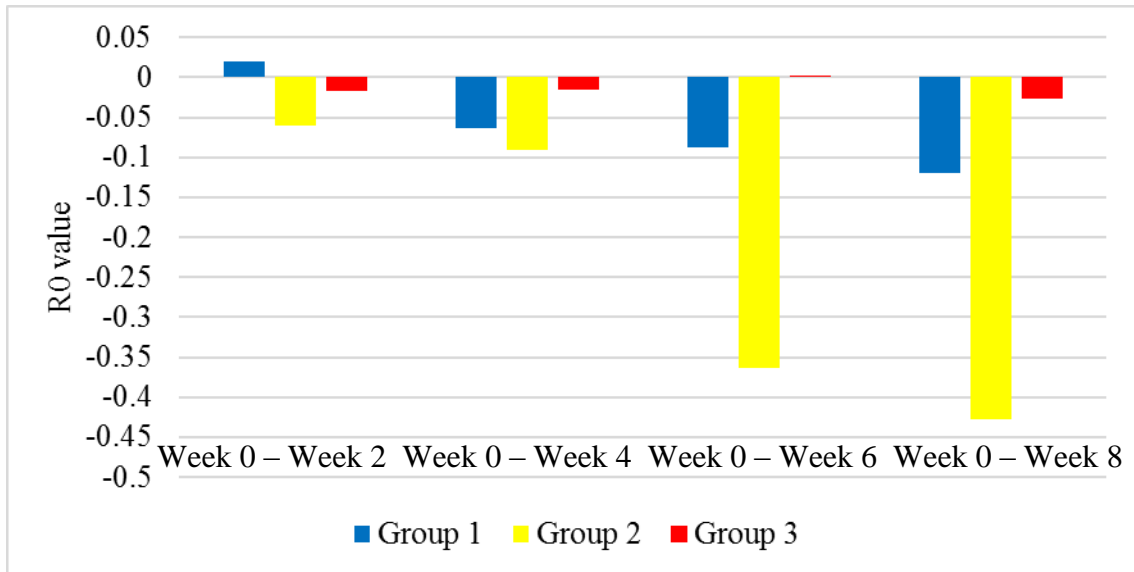


From table 4.4 and figure 4.3, the average of R0 value of group 1 (glyceryl octyl ascorbate) on week 2, 4, 6 and 8 were 0.076±0.03, 0.160±0.325, 0.184±0.076 and 0.216±0.069 respectively. When using repeated measurement ANOVA to analyze the mean difference of R0 value showed no statistically significant as table 4.

The average of R0 value of group 2 (3-0-ethyl ascorbic acid) on week 2, 4, 6 and 8 were 0.139±0.085, 0.17±0.102, 0.442±0.093 and 0.507±0.661 respectively. The mean difference of R0 value showed no statistically significant as table 4 (when used repeated measurement ANOVA to analyzed).

The average of R0 value of group 3 (placebo) on week 2, 4, 6 and 8 were 0.094±0.063, 0.093±0.063, 0.076±0.015 and 0.104±0.066 respectively. The mean difference of R0 value showed no statistically significant as table 4 (when used repeated measurement ANOVA to analyzed).

When compare the change of R0 value between 3 groups from week 0 to week 8 was shown no statistically significant by used one-way ANOVA to analyze.

**Table 5** The comparison of R2 value between 3 groups

| | Week 0 | Week 2 | Week 4 | Week 6 | Week 8 | p-value |
|---|---|---|---|---|---|---|
| Group 1 | 0.496 | 0.512 | $0.612^b$ | $0.667^b$ | $0.74^b$ | <0.001 |
| SD | 0.056 | 0.062 | 0.074 | 0.085 | 0.109 | |
| Group 2 | 0.541 | 0.572 | $0.627^a$ | $0.693^a$ | $0.691^a$ | <0.001 |
| SD | 0.069 | 0.085 | 0.104 | 0.11 | 0.144 | |
| Group 3 | 0.517 | 0.514 | $0.511^{a,b}$ | $0.51^{a,b}$ | $0.508^{a,b}$ | 0.039 |
| SD | 0.049 | 0.051 | 0.047 | 0.052 | 0.052 | |
| p-value | 0.259 | 0.098 | 0.004 | <0.001 | <0.001 | |

Note; a is statistically significant between group 2 and 3

b is statistically significant between group 1 and 3

**Figure 4** The reduction of R2 value between 3 groups
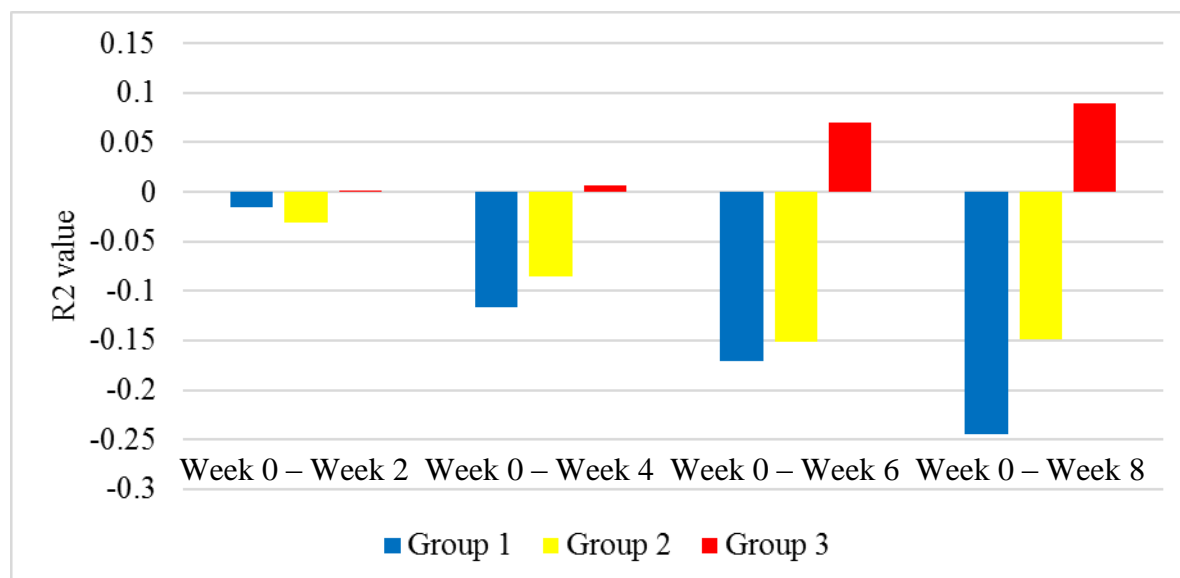


From table 5 and figure 4, the average of R2 value of group 2 (glyceryl octyl ascorbate) on week 2, 4, 6 and 8 were 0.512±0.062, 0.612±0.074, 0.667±0.085 and 0.74±0.109 respectively. The mean difference of R2 value showed statistically significant ($p < 0.05$) (when used repeated measurement ANOVA to analyzed).

The average of R2 value of group 2 (3-0-ethyl ascorbic acid) on week 2, 4, 6 and 8 were 0.572±0.085, 0.627±0.104, 0.693±0.110 and 0.691±0.144 respectively.

When used repeated measurement ANOVA to analyzed the mean difference of R2 value shown statistically significant ($p < 0.05$) as table 5.

The average of R2 value of group 3 (placebo) on week 2, 4, 6 and 8 were $0.514\pm0.051$, $0.511\pm0.047$, $0.510\pm0.052$ and $0.508\pm0.052$ respectively. When used repeated measurement ANOVA to analyzed the mean difference of R2 value shown statistically significant ($p < 0.05$) as table 5.

When compare the change R2 value between 3 groups showed statistically significant difference since week 4 to the ends of the study by used one-way ANOVA to analyze.

**Table 4.6** The comparison of R6 value between 3 groups

|  | Week 0 | Week 2 | Week 4 | Week 6 | Week 8 | p-value |
|---|---|---|---|---|---|---|
| Group 1 | 1.307 | 1.187 | $1.034^b$ | $0.814^b$ | $0.752^b$ | <0.001 |
| SD | 0.242 | 0.205 | 0.234 | 0.202 | 0.16 | |
| Group 2 | 1.165 | $1.043^a$ | $0.895^a$ | $0.726^a$ | $0.606^a$ | <0.001 |
| SD | 0.251 | 0.23 | 0.258 | 0.263 | 0.178 | |
| Group 3 | 1.308 | $1.296^a$ | $1.321^{a,b}$ | $0.1334^{a,b}$ | $1.363^{a.b}$ | 0.004 |
| SD | 0.158 | 0.166 | 0.163 | 0.164 | 0.17 | |
| p-value | 0.258 | 0.026 | <0.001 | <0.001 | <0.001 | |

Note; a is statistically significant between group 2 and 3

b is statistically significant between group 1 and 3

**Figure 5** The reduction of R6 value between 3 groups
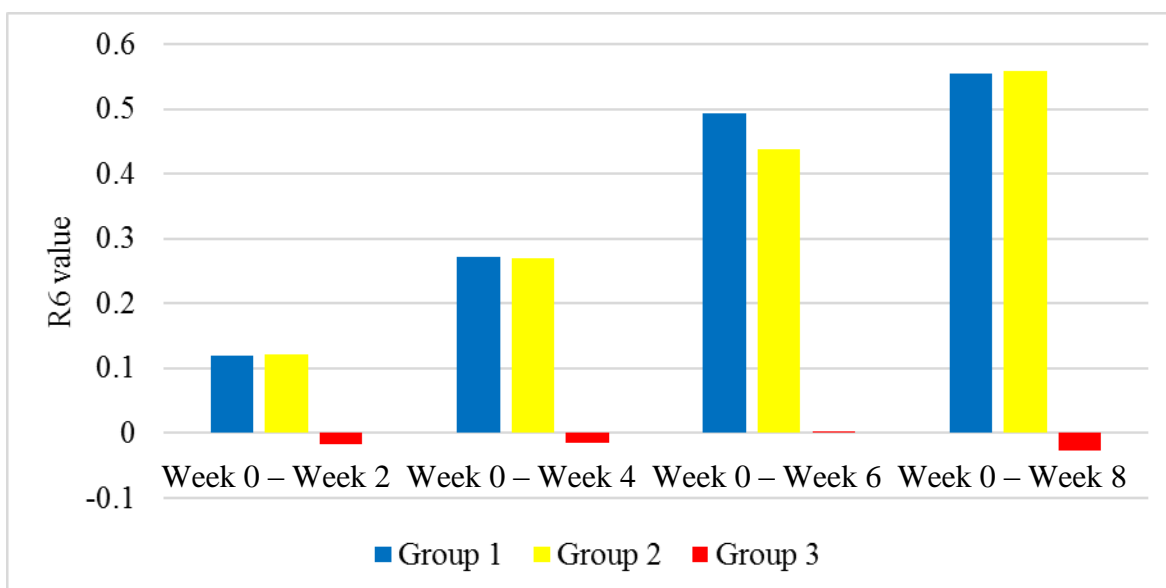


From table 4.6 and figure 4.5, the average of R6 value of group 1 (glyceryl octyl ascorbate) on week 2, 4, 6 and 8 were $1.187\pm0.205$, $1.034\pm0.234$, $0.814\pm0.202$ and $0.752\pm0.16$ respectively. When used repeated measurement ANOVA to analyzed the mean difference of R6 value shown statistically significant ($p < 0.05$).

The average of R6 value of group 2 (3-0-ethyl ascorbic acid) on week 2, 4, 6 and 8 were $1.043\pm0.23$, $0.895\pm0.258$, $0.726\pm0.263$ and $0.606\pm0.178$ respectively. The mean difference of R6 value shown statistically significant ($p < 0.05$) as table 6 (when used repeated measurement ANOVA to analyzed).
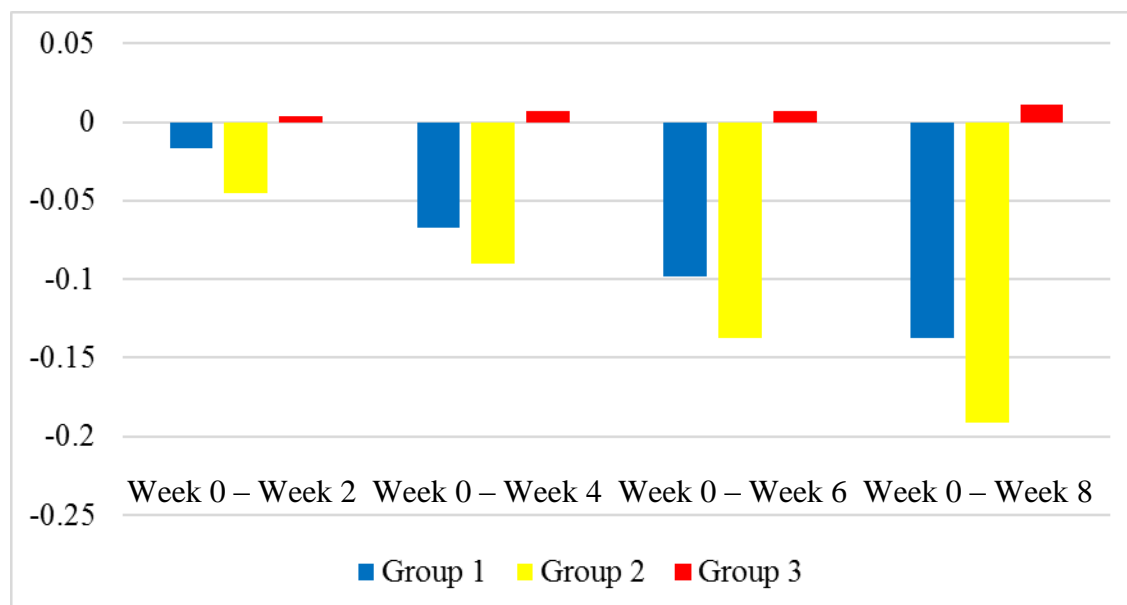
The average of R6 value of group 3 (placebo) on week 2, 4, 6 and 8 were $1.296\pm0.166$, $1.321\pm0.163$, $1.334\pm0.164$ and $1.363\pm0.17$ respectively. The mean difference of R6 value shown statistically significant ($p < 0.05$) as table 6 (when used repeated measurement ANOVA to analyzed).

When compare the change R6 value between 3 groups shown statistically significant difference since week 2 between group 2 and 3. Since week 4 to week 8, the data showed statistically significant ($p < 0.05$) between group 1, 3 and 2, 3 (when used one-way ANOVA to analyzed).

**Table 7** The comparison of R7 value between 3 groups

| | Week 0 | Week 2 | Week 4 | Week 6 | Week 8 | p-value |
|---|---|---|---|---|---|---|
| Group 1 | 0.458 | 0.475 | 0.525 | 0.556 | 0.595 | <0.001 |
| SD | 0.078 | 0.06 | 0.073 | 0.067 | 0.061 | |
| Group 2 | 0.473 | 0.518 | 0.563 | 0.61[a] | 0.664 | <0.001 |
| SD | 0.07 | 0.062 | 0.072 | 0.074 | 0.095 | |
| Group 3 | 0.542 | 0.537 | 0.535 | 0.535[a] | 0.531 | 0.288 |
| SD | 0.107 | 0.107 | 0.106 | 0.107 | 0.103 | |
| p-value | 0.088 | 0.244 | 0.61 | 0.141 | 0.008 | |

Note; a is statistically significant between group 2 and 3

**Figure 6** The reduction of R7 value between 3 groups



From table 7 and figure 6, the average of R7 value of group 1 (glyceryl octyl ascorbate) on week 2, 4, 6 and 8 were 0.475±0.060, 0.525±0.073, 0.556±0.067 and 0.595±0.061 respectively. The mean difference of R7 value shown statistically significant ($p < 0.05$), when used repeated measurement ANOVA to analyzed.

The average of R7 value of group 2 (3-0-ethyl ascorbic acid) on week 2, 4, 6 and 8 were 0.518±0.062, 0.563±0.072, 0.610±0.074 and 0.664±0.095 respectively.

The mean difference of R6 value shown statistically significant ($p < 0.05$) as table 7 (when used repeated measurement ANOVA to analyzed).

The average of R7 value of group 3 (placebo) on week 2, 4, 6 and 8 were 0.537±0.107, 0.535±0.106, 0.535±0.107 and 0.531±0.103 respectively. The mean difference of R7 value shown no statistically significant as table 7 (When used repeated measurement ANOVA to analyzed).

When compare the change R7 value between 3 groups shown statistically significant difference at week 8 between group 2 and 3 by used one-way ANOVA to analyze.

**Conclusion**

Melanin index

At the end of the study, glyceryl octyl ascorbate, 3-0-ethyl ascorbic acid and placebo reduced melanin index on cheek as 41.63, 34.30 and -1.667 respectively. In additional, glyceryl octyl ascorbate, 3-0-ethyl ascorbic acid and placebo reduced melanin index on forehead as 34.352, 30.417 and 0.652 respectively. Both of vitamin c shown same result but glyceryl octyl ascorbate could reduced melanin index more than 3-0-ethyl ascorbic acid. From the comparing the use of 10% L-ascorbic acid and 10% zinc sulfate solution in the treatment of melisma reported 10% L-ascorbic acid reduced MASI (the melasma area and severity index) score (Wahyu L., et al (2012). It's implied that 10% L-ascorbic acid reduced melanin. From our study, both of glyceryl octyl ascorbate and 3-0-ethly ascorbic acid showed ability to suppress melanin synthesis. The glyceryl octyl ascorbate decreased melanin index to 41.63 on cheek and 34.35 on forehead at day 56. Moreover, 3-0-ethyl ascorbic acid decreased melanin index to 34.30 on cheek and 30.42 on forehead at day 56. From the mean of melanin index, glyceryl octyl ascorbate decreased melanin index more than 3-0-ethyl ascorbic acid, at lower percentage than 3-0-ethyl ascorbic acid because glyceryl octyl ascorbate is an amphiphic molecule (Takeshi N., et al (2013). It's can be dissolve in water (hydrophilic) and oil (lipophilic). Lipids involve in stratum corneum are 50% ceramide, 25% cholesterol, 15% of free fatty acid and small amount of other lipid (Darlenski R., et al (2011). So, glyceryl octyl ascorbate has lipophilic properties same to the skin, so it easy penetrate into the skin.

R0 value

After treatment for 8 weeks, glyceryl octyl ascorbate, 3-0-ethyl ascorbic acid and placebo were increased R0 value as 0.12, 0.428 and 0.026. When used repeated measurement ANOVA to analyze in each group shown no statistically significant but both of vitamin c shown ability to increase R0 value. In the result of 3 groups, when using one-way ANOVA to analyze the data shown no statistically significant. It's mean the change of R0 value shown no different between 3 groups.

R2 value

After treatment for 8 weeks, glyceryl octyl ascorbate, 3-0-ethyl ascorbic acid and placebo were increased R2 value as 0.244, 0.149 and 0.026. When using repeated measurement ANOVA to analyze, group 1 shown statistically significant ($p < 0.05$) since week 4, group 2 shown statistically significant ($p < 0.001$) at week 6. In the result of 3 groups, when used one-way ANOVA to analyze the data shown both vitamin c shown statistically significant ($p < 0.05$) since week 4. It's mean the change of R2 value shown different between 3 groups.

R6 value

After treatment for 8 weeks, glyceryl octyl ascorbate, 3-0-ethyl ascorbic acid and placebo decreased R6 value as 0.555, 0.558 and -0.055. When used repeated measurement ANOVA to analyze, group 1 showed statistically significant ($p < 0.05$) since week 2, group 2 showed statistically significant ($p < 0.001$) at week 2. In the result of 3 groups, when used one-way ANOVA to analyze the data showed 3-0-ethyl ascorbic acid showed statistically significant ($p < 0.05$) since week 2. Moreover, both of vitamin c showed statistically significant ($p < 0.001$) at week 4. It's mean the change of R6 value shown different between 3 groups.

R7 value

After treatment for 8 weeks, glyceryl octyl ascorbate, 3-0-ethyl ascorbic acid and placebo increased R7 value as 0.137, 0.191 and -0.026. When used repeated measurement ANOVA to analyze, group 1 showed statistically significant ($p < 0.001$) since week 6, group 2 showed statistically significant ($p < 0.001$) at week 2. In the

result of 3 groups, when used one-way ANOVA to analyze the data shown 3-0-ethyl ascorbic acid showed statistically significant ($p < 0.05$) at week 2.

From study of ascorbic acid and the formation of collagen reported number of collagen was increase when supplied 100 mg of vitamin c (William B., et al (1952). It's implied that collagen in the skin increase. From our study, the trend of elasticity value was increase. Both of glyceryl octyl ascorbate and 3-0-ethyl ascorbic acid showed statistically significant in all parameter excepted R0 (skin firmness). Although, most skin made up of collagen (70%) but has another component such as elastin. Elastin has an important role to maintain normal skin elasticity and flexibility (Takanori I., et al (2005), so elastin has an important to increase skin firmness too.

**Suggestion**
1. More clinical research with hyperpigmentation such as melasma, acne spot or age spot.
2. More clinical research with age people, who has wrinkle and loss skin elasticity.
3. The longer period of the experiment may give better result.

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