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## ***"Efficacy Study of A New Composition With The Effect of Improving The Appearance of Scars And Stretch Marks"***

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### **Abstract**

The purpose of this study was to investigate whether a body oil with a combination of *Carthamus Tinctorius* (Safflower) seed oil, Tocopheryl Acetate, and Retinyl Palmitate are effective in improving the appearance of scars and stretch marks. The investigators recruited 60 participants with visible skin scars and stretch marks that had developed no more than 3 years to conduct the efficacy evaluation tests and consumer use tests. Changes in skin pigmentation, skin color ITA°, stretch mark length, scar elevation, skin elasticity, skin moisture content of the stratum corneum and transdermal water loss at the test sites were measured before and after 21 days of continuous use of the body oils under normal conditions in both the test and control groups. The results of the test showed that there was a significant improvement in all the indicators before and after sample application in the test group compared with the control group ( $P < 0.050$ ). This suggests that the new combination of essential oils and vitamin derivatives has the efficacy to reduce the appearance of scars and stretch marks.

**Key words:** Scars, stretch marks, Body oil, Human Efficacy Evaluation tests, User Trial

### **1. Introduction**

Skin scarring is an inevitable product of skin injury in which the organism is unable to fully achieve histological regeneration, which in turn initiates the process of tissue repair, replacing it with connective tissue, and thus skin scarring is a fibroproliferative disorder with abnormal morphologic appearance and altered function [1-3]. Stretch marks are mainly pink or purplish wavy patterns formed when the abdominal skin suffers varying degrees of damage or breakage due to external pulling forces. The occurrence of stretch marks is very common among women, with 60% to 90% of pregnant women suffering from stretch marks [4-6]. Both skin scarring and stretch marks can have negative physical, psychological and mental effects on people, thus affecting their quality of life, and thus there is an increasing interest in the prevention and treatment of both. It has been suggested [7-8] that the production of hyperplastic scarring is closely related to large number of collagen fibers arranged in a spiral pattern. Clinical treatment measures for patients with hyperplastic scarring mainly include local

drug injection, topical medication, surgical excision, laser therapy, and pressure therapy. During the treatment process, the improvement effect is evaluated from the dimensions of clinical symptoms such as improvement of skin elasticity, skin color, stretch mark condition score, percentage of scar flat area, pigmentation, itching or pain sensation [9-10,14]. In recent years, the use of skin imaging techniques in the field of scarring has increased significantly [11]. Lately, there have been many clinical studies of plant essential oils applied to cosmetics to improve scarring and stretch marks, but not many combined with vitamin derivatives. [12-13]. This paper explores the efficacy of a body oil combining vitamin A palmitate, vitamin E derivative (vitamin E acetate), and safflower seed oil to improve the appearance of skin scars and stretch marks through human efficacy evaluations and consumer use tests.

## 2. Materials and Methods

### I. Test Group Formulation

Containing about 50% Ethylhexyl Palmitate, about 30% Carthamus Tinctorius (Safflower) seed oil, about 10% Tridecyl Trimellitate, and less than 0.6% of Tocopheryl Acetate, Retinyl Palmitate, Bisabolol, Rosmarinus Officinalis (Rosemary) Leaf Oil and Lavandula Angustifolia (Lavender) Oil). Of these, Carthamus Tinctorius (Safflower) seed oil, Tocopheryl Acetate and Retinyl Palmitate [15-23] are the key ingredients of this formula to improve the appearance of scars and stretch marks. Carthamus Tinctorius (Safflower) seed oil is rich in linoleic acid (omega-6), which replenishes skin lipids, promotes ceramide synthesis, strengthens the barrier function and reduces water loss [24-26]. And linoleic acid regulates prostaglandin synthesis pathway, inhibits pro-inflammatory factors (e.g. IL-6), and relieves acne and redness (for oily acne skin) [27-28]. Tocopheryl Acetate is hydrolyzed in the skin to free vitamin E ( $\alpha$ -tocopherol), which traps lipid peroxidation free radicals and protects cell membranes from oxidative damage, and can be regarded as a precursor antioxidant. At the same time, it cannot only synergize with vitamin C to enhance the photoprotective effect, reduce UVB-induced erythema and DNA damage, and play a photoprotective role, but also strengthen the antioxidant capacity of sebaceous membranes, reduce the transcutaneous water loss (TEWL), and assist in the repair of barrier function [29]. Retinyl Palmitate, as a derivative of Vitamin A, is first enzymatically cleaved in the skin to Retinol, which is then gradually converted to Retinoic Acid. This process slows down the irritation of using A-acid directly, making it suitable for sensitive skin. Secondly, it accelerates keratinocyte differentiation by activating Retinoic Acid Receptors (RAR/RXR) in the nucleus of skin cells, improving roughness and dullness and enhancing epidermal metabolism. Further, it neutralizes free radicals, reduces UV-induced oxidative damage, and indirectly inhibits collagen degradation. Finally, it increases type I collagen production by regulating fibroblast activity, improving wrinkles and sagging [30].

### II. Control Group Formulation

Containing about 50% Ethylhexyl Palmitate and about 10% Tridecyl Trimellitate.

### III. Test equipment

Antera 3D ® multifunctional skin imaging analyzer, skin immediate elasticity (ISE) test probe (ElastiMeter), Skin Moisture Content Test Probe (Corneometer CM 825), Skin Transdermal Moisture Loss Meter (VaPoMeter).

### IV. Basic principles of human testing

The cosmetic efficacy evaluation trial in this paper adhered to the ethical principles of the International Declaration of Helsinki by requiring participants to sign an informed consent form,

which explained the nature of the study, its purpose, and the potential risks of participating in the study, and emphasized that participation in the test was voluntary, and that participants could withdraw from the study at any time and for any reason. All participants were allowed to ask questions about the research test and were given full event consideration before signing. All informed consent signatures were made prior to the start of the study. The investigator took the necessary medical precautions to maximize the protection of the participants during the trial. Necessary product safety evaluations were completed and passed prior to the conduct of the human efficacy evaluations.

## V. Experimental design

Double-blind control principle

## VI. Participants inclusion criteria

This test ultimately included 60 healthy Asian participants (excluding pregnant and lactating women) aged 20-40 years [9], of which 10 were male and 50 were female, with a mean age of  $34.5 \pm 2.1$  years. Sixty of the participants had scars, of which 18 had both scarring and stretch marks. Participants were asked to read and understand all the contents of the informed consent form and sign it voluntarily; had at least one skin scar or stretch mark that was more obvious and had been produced for no more than 3 years [9]; had no obvious skin lesions, hairs, moles, capillary dilatation, or lesions on the skin of the subject site; had no serious systemic diseases, no immunodeficiency or autoimmune diseases; had no active allergic Diseases; not sensitive to commonly used cosmetics, general light exposure or would not have had a serious reaction; not highly sensitive; not received skin treatment, skin peeling, medical cosmetic treatment at the test site within 3 months prior to participating in the assessment; not used hormone drugs and immunosuppressants within 1 month prior to participating in the assessment; not sunbathing or over-exposure to ultraviolet rays (e.g., hiking, phototherapy, used of tanning salons) within 1 week prior to participating in the assessment.

## VII. Procedures

On the first visit, the investigators explained the test to the participants and signed the informed consent form. Sixty participants were randomly divided into two groups, test group and control group. Before starting the test, participants sat still for at least 30 minutes in a test room with a temperature of  $21 \pm 1^\circ\text{C}$  and a relative humidity of  $50\% \pm 10\%$ . The investigators identified the areas of the participants' body with scars and stretch marks as test areas and recorded the test areas of each participant for the next return visit. Finally, the investigators used the equipment to collect the skin pigmentation, skin color ITA°, stretch mark length, scar elevation height, immediate skin elasticity (ISE), skin stratum corneum moisture content and skin transdermal water loss from the test sites before using the product.

**Sample Use:** The test group used the test group formulation, gently massaged until fully absorbed, twice a day for 21 days. The control group used the control formulation in the same way as the test group.

**Test return visit:** After the participants had used the product for 21 days, the investigators used the equipment to collect test parameters of the test areas. Before starting the test, participants sat still for at least 30 minutes in a test room at a temperature of  $21 \pm 1^\circ\text{C}$  and a relative humidity of  $50\% \pm 10\%$ . A skin marker was used to fix the measurement area during the test to avoid errors due to positional shifts. At the same time, the investigators distributed questionnaires to investigate the participants' satisfaction with the effect of the product and the degree of itchiness of the skin before and after the use of the product.

Post-test follow-up: 4 weeks after discontinuation of the product to observe the persistence of the effect.

### VIII. Data processing

Calculate the mean, standard deviation, maximum and minimum values of each parameter for the test and control groups at each time point, and calculate the difference between the test and control groups after using the product at each time point, respectively.

Normal distribution test: The above difference values were tested for normal distribution, and if the value of asymptotic significance (two-sided) of the normality test is  $>0.050$ , then the series of data obeys normal distribution.

Analysis of variance: the above differences were subjected to analysis of variance, and when the two sets of data were normally distributed at the same time, the paired *t*-test or independent samples *t*-test was used to analyze the variance between the two sets of data; when the two sets of data were not normally distributed at the same time, the nonparametric test was used to analyze the variance between the two sets of data.

Participants' self-assessment results were analyzed using a 4-point Likert scale (1. very satisfied, 2. satisfied, 3. dissatisfied, 4. very dissatisfied), which was combined into a dichotomous variable divided into a positive answer group = 1+2 and a negative answer group = 3+4). The percentage of the combined "positive answer group" was the total percentage of participants who provided consent. A one-sample test of proportions (binomial test) was used, combined with a confidence interval analysis, with the objective of verifying that the proportion of positive answers was significantly higher than 65.1%.

## 3. Results

### 1.1. Descriptive statistics

**Table 1.** Descriptive statistics of raw data

Statistic Test items		Mean value		Standard devi- ation		Maximum value		Minimum value	
		Test grou- p	Con- trol group	Test group	Con- trol group	Test group	Con- trol group	Test group	Con- trol group
Skin immedi- ate elasticity	T <sub>0d</sub>	67.0	65.3	25.77 0	22.60 3	128.0	126.0	27.0	30.0
	T <sub>21d</sub>	78.1	68.3	32.17 7	22.34 7	181.0	128.0	30.0	34.0
Skin Color ITA°	T <sub>0d</sub>	36.6	36.7	7.401	7.264	51.5	51.3	14.6	15.0
	T <sub>21d</sub>	38.5	37.6	7.786	7.385	53.1	52.0	15.0	14.9

Pigmentation	T <sub>0d</sub>	42.7	42.7	5.664	5.463	57.0	56.8	30.3	30.6
	T <sub>21d</sub>	41.3	42.3	5.942	5.398	55.7	56.7	30.0	30.5
Skin stretch mark length	T <sub>0d</sub>	21.9	22.9	6.860	4.042	34.1	28.6	14.9	18.0
	T <sub>21d</sub>	19.3	22.9	6.326	3.990	32.1	28.7	12.7	18.1
Scar elevation	T <sub>0d</sub>	1.4	1.4	0.331	0.271	2.0	1.9	0.6	0.7
	T <sub>21d</sub>	1.3	1.4	0.300	0.237	1.9	2.0	0.5	1.0
Skin transdermal water loss	T <sub>0d</sub>	14.5	14.3	2.859	2.875	20.9	20.8	10.4	10.2
	T <sub>21d</sub>	10.8	12.2	2.529	2.613	17.8	19.0	6.7	7.9
Skin stratum corneum moisture content	T <sub>0d</sub>	31.25	31.44	6.90	6.90	43.50	43.53	19.10	19.73
	T <sub>21d</sub>	40.15	33.16	7.74	8.41	58.75	48.90	25.97	19.05

Note 1: The larger the value of skin immediate elasticity ISE (unit: N/m), the better the elasticity; the larger the value of skin color ITA°, the whiter the skin; the smaller the value of pigmentation, the less pigmentation; the length of skin stretch marks (unit: mm); the height of scar elevation (unit: mm); the smaller the value of skin transcutaneous water loss (unit: g/m<sup>2</sup>h), the more intact the skin barrier function; and the greater the skin moisture content of the stratum corneum, the better the skin moisturization. The larger value of stratum corneum moisture content indicates better skin moisturization.

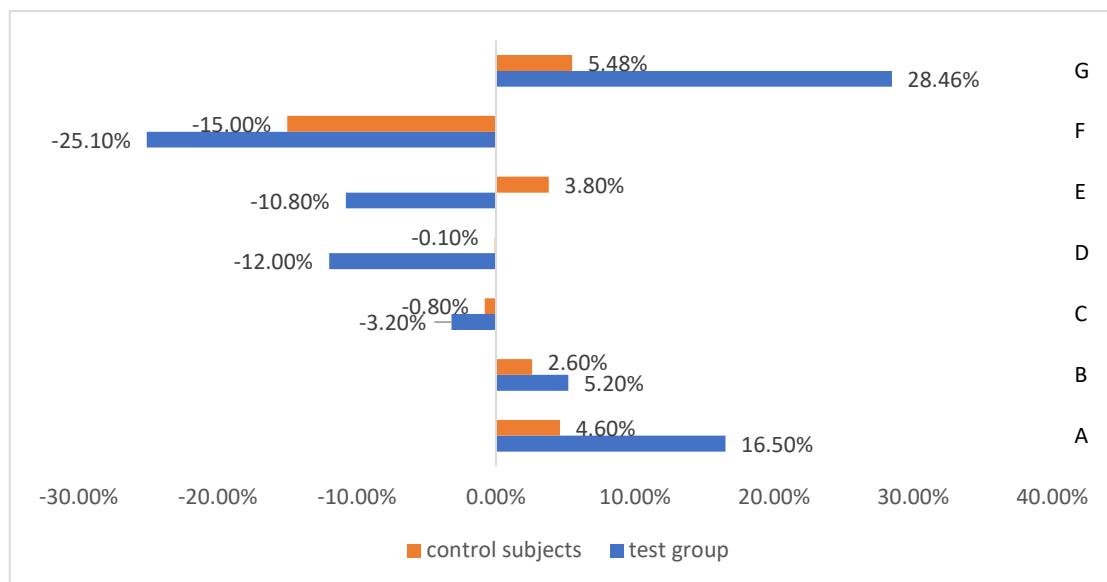
### 1.2. Difference-in-difference analysis results

**Table 2.** Difference-in-difference analysis results

Test items	Nick-names	Test group	Control subjects	Comparison between groups
Skin immediate elasticity	A	16.50%*	4.60%*	*

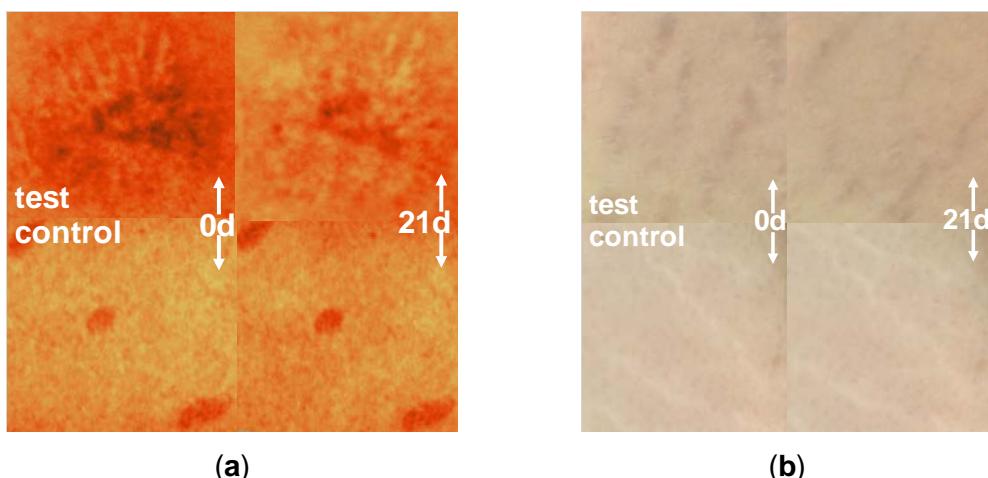
<b>Skin Color ITA°</b>	B	5.20%*	2.60%*	*
<b>Pigmentation</b>	C	-3.20%*	-0.80%*	*
<b>Skin stretch mark length</b>	D	-12.00%*	-0.10%	*
<b>Scar elevation</b>	E	-10.80%*	3.80%*	*
<b>Skin transdermal water loss</b>	F	-25.10%*	-15.00%*	*
<b>Skin stratum corneum moisture content</b>	G	28.46%*	5.48%	*

Note 2: \*Indicates a significant  $p$ -value  $<0.050$ , indicating a significant difference between after use of the samples and before use of the samples or comparison of the test group with the control group.



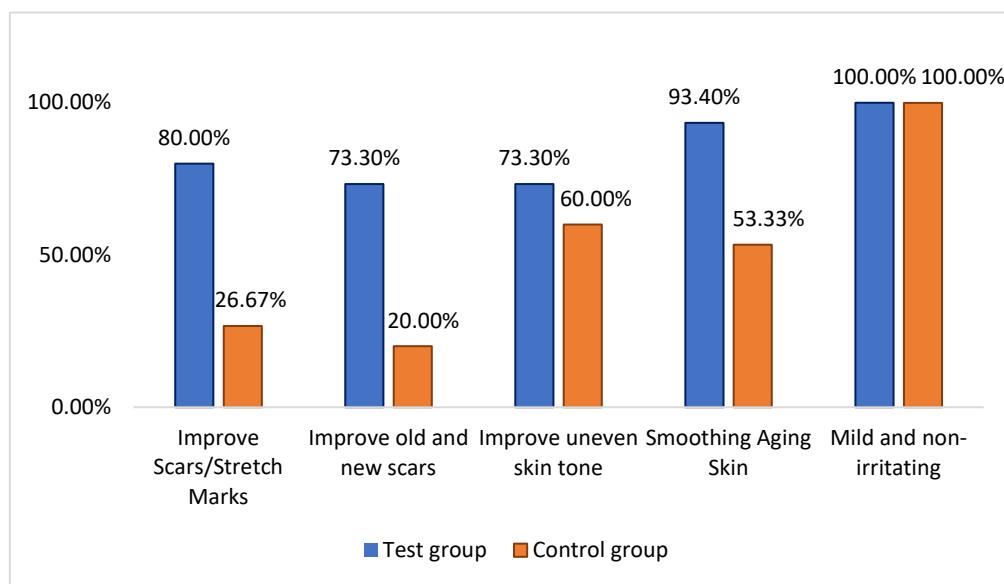
**Figure 1.** Comparison of the rate of change of each test indicator between the test and control groups

### 1.3. Image Analysis Comparison Examples



**Figure 2.** (a): Appearance of scars before (0d) and after (21d) using the product; (b): Appearance of stretch marks before (0d) and after (21d) using the product

#### 1.4. Participants self-assessment



**Figure 3.** Bar graph of participants' satisfaction with product effectiveness

#### 4. Discussion

The results of the test showed that before and after the use of the product, when comparing the test group to the control group, the mean ISE value of the test group significantly improved from 67.0 at T0d to 78.1 at T21d (+16.5%), while the control group only increased from 65.3 to 68.3 (+4.6%). The significantly greater improvement in the test group suggests it is effective in enhancing immediate skin elasticity. ITA° increased from 36.6 to 38.5 (+5.2%) in the test group and from 36.7 to 37.6 (+2.6%) in the control group. The increase in skin tone brightness was more pronounced in the test group and may be related to moisturisation or barrier repair. The mean value of hyperpigmentation decreased from 42.7 to 41.3 (-3.2%) in the test group and from 42.7 to 42.3 (-0.8%) in the control group. The improvement in the test group was slightly better than the control group, but the effect was relatively limited. Stretch mark length decreased from 21.9 to 19.3 (-12.0%) in the test group, while there was no significant change in the control group (-0.1%). The data from the test group suggest that

the treatment may have a definite effect on stretch mark repair. Scar height decreased from 1.4 to 1.3 (-10.8%) in the test group compared to a slight increase in the control group (+3.8%). The decreasing trend in the test group may reflect the scar levelling effect. The moisture content of the test group increased significantly from 31.25 to 40.15 (+28.46%), while the control group only increased from 31.44 to 33.16 (+5.48%). The significant enhancement in the test group indicates that the treatment was effective in enhancing the moisturising capacity of the skin. Moisture loss decreased from 14.5 to 10.8 (-25.1%) in the test group and from 14.3 to 12.2 (-15.0%) in the control group.

Participants were all statistically satisfied with the product's efficacy in helping to improve and lighten the appearance of scars and stretch marks, even skin tone, smooth and tone the body's aging skin, retain moisture, and be gentle and non-irritating after use. There were no adverse skin reactions and no aggravation of the skin at the 4-week follow-up after 21 days of the trial. This indicates that the body oil consisting of *Carthamus Tinctorius* (Safflower) seed oil, Tocopheryl Acetate, and Retinyl Palmitate have the efficacy to lighten scars and stretch marks.

In the future, we are planning to study the absorption and penetration, and investigate if the ingredient combination offer a better solution and prevention to hyperplastic scars and stretch marks. The number of this trial participants, the test period, the number of return visits and the assessment indicators were small. In the future, we are planning to improve the trial protocol and increase the assessment indicators, such as increasing the assessment of scar hardness and inflammation indicators.

## 5. Conclusion

The test group significantly outperformed the control group in most of the test items, especially in skin elasticity, skin moisture content, stretch mark repair and scar smoothing, which demonstrated that the body oil composed of *Carthamus Tinctorius* (Safflower) seed oil, Tocopheryl Acetate and Retinyl Palmitate not only had a synergistic effect on improving the appearance of scars and stretch marks, but also had a significant effect on improving the skin condition. At the same time, none of the participants experienced any adverse skin reactions after long-term use, suggesting that the safety of the formulation is suitable for sensitive skin. Improvements in hyperpigmentation were relatively limited and may require more long-term or targeted interventions. The control group showed less change, which may reflect natural recovery or the influence of other external factors, but the trial group showed much greater improvement than the control group, which supported the validity of the trial group.

In terms of test design, the current study contains both instrumental measurement and subjective evaluation. By setting up a control group, adding post-trial follow-up and a number of objective assessment indicators, this provide a more comprehensive assessment in product efficacy on fading scars and stretch marks.

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