

Jarilla–Coffea extract: a natural cosmetic product that improves eyelash and eyebrow growth in women

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Purpose: A combination of extracts, from two plant species, ie, *Coffea arabica* and *Larrea divaricata* (Jarilla) (ECOHAIR®), is being successfully used in Argentina as a cosmetic for hair recovery in androgenic and areata alopecia, and for eyelash and eyebrow growth. The objectives of this prospective study were to evaluate the capacity of Jarilla–Coffea extract gel of improving hair growth in relation to thickness, appearance of new hair, and hair length in comparison with a placebo in premenopausal and postmenopausal volunteers and to identify possible signs of ocular adverse local reactions related to the application of the gel.

Volunteers and methods: An open-label, placebo-controlled, prospective study was performed in healthy premenopausal and postmenopausal women during a daily administration period of 2 months (eyebrow growth) and 3 months (eyelash growth). The thickness of hair was determined using a video microscope MedicalScope®. The appearance of new hairs and total area with hair in eyebrow and eyelash length were quantified using a photographic record with Fotofinder® (Germany). The number of volunteers presenting variation in growth of new hair and length were also recorded.

Results: The product significantly increased the thickness of eyebrows (20% in 80% women) and eyelashes (19.44% in 100% of women). The gel also increased the appearance of new hairs, total area with hair, and length but there was no statistical difference between treatment and placebo.

Conclusion: The gel was capable of improving growth of eyelashes and eyebrows by inducing principally hair thickening without causing local adverse effects in a high percentage of volunteers.

Keywords: eyelash thickness, eyebrow thickness, plant extracts, menopause, adverse effects, cosmetic

Introduction

Both men and women suffer from hypotrichosis, which comprises either hair loss and/or thinning. This phenomenon, which affects not only the scalp but also eyelashes and eyebrows, is a result of an imbalance between cell proliferation and death. It is known that hair is involved in skin protection and thermal regulation. Especially, the eyelashes protect the eyes from light and dust, while the eyebrows protect the eyes from the sweat dripping down from the forehead. When hypotrichosis occurs, a deleterious effect on the patient's esthetic appearance also ensues. Hypotrichosis in eyelashes and eyebrows may be caused by ophthalmological infections and inflammatory process but also by dermatological diseases such as atopic dermatitis, contact dermatitis, psoriasis, lichen planus, and seborrheic dermatitis. Also hypotrichosis

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may be attributed to telogen effluvium caused by intake of drugs such as anticoagulants, lipid-lowering agents, anti-hypertensive agents (propranolol), valproate, barbiturates, bromocriptine, immunosuppressants, botulinum toxin, NSAIDs, and hormone imbalance.¹

It is known that premenopausal, perimenopausal, and postmenopausal women suffer from hypotrichosis as a consequence of hormone misbalance; under this situation, some changes in skin such as increased dryness and roughness, decreased elasticity, warts, freckles, increased wrinkles, loss and graying of head hair, and lengthening of eyebrows and eyelashes are observed.^{2,3}

Under these situations, extensions, masks of different types, eyelash bends, transplants, and permanent dyes are used on both eyelashes and eyebrows. One of the current products used to promote eyelash growth is a bimatoprost solution, which is a synthetic analog of prostaglandin F_{2α} used for the treatment of glaucoma. As side effect, bimatoprost is known to induce hair growth. The Food and Drug Administration (FDA) has approved it to be used in a 0.03% solution to stimulate the growth, thickness, and pigmentation of eyelashes in patients with hypotrichosis. However, the compound stimulates hair growth in some patients while in those with alopecia areata it is ineffective. On the other hand, when the product makes contact with the eye, serious adverse effects can occur. Analogs of prostaglandins (PGs) and prostanoids are associated with local and systemic effects not only when administered topically in the eye but also when placed in the eyelash insertion. These analogs can cause hyperpigmentation of the iris, as a consequence of an increase in melanogenesis, conjunctival hyperemia, eye dryness, erythema, itching, and uveitis. They have also been reported to cause headache, hirsutism, hepatic impairment, and to increase the sensitivity to herpes simplex virus (HSV) infection. In addition, it has been observed that both bimatoprost and latanoprost, other prostanoid, cause HSV dermatitis of the periocular skin.⁴ Taking these observations into account, the use of these prostanoids to promote the growth of the eyelashes is contraindicated in patients suffering from inflammatory diseases or from viral infections. Moreover, the commercially available products contain benzalkonium chloride as antibacterial, which has cytotoxic effect on the cornea and can cause erosion of the epithelium, irritation, and eye dryness.⁵

The prostanoids cannot be used during pregnancy because it has been classified by the FDA as a class C drug, since it is likely to cause malformations in the fetus.⁶

To date, no innocuous effective products to promote eyelash or eyebrow growth have been developed. A combination of extracts obtained from two plant species, ie, *Coffea arabica*

and *Larrea divaricata* (Jarilla) (ECOHAIR[®]), is being successfully used in Argentina, by Garre-Guevara Laboratory, as cosmetic, for hair recovery in scalp in cases of androgenic and areata alopecia and for eyelash and eyebrow growth. The effectiveness of the product in scalp alopecia has been verified not only in in vivo alopecia models in C3H mice but also in a clinical trial.^{7,8}

To provide scientific support for its effect in eyelash and eyebrow growth, the product was tested in premenopausal and postmenopausal healthy women. The primary objective of this prospective study was to evaluate the capacity of Jarilla–Coffea extract gel for improving hair growth in relation to thickness, appearance of new hairs, and hair length in comparison with a placebo. The secondary objective was to identify possible signs of ocular adverse local reactions related to the application of the gel.

Volunteers and methods

Study design

An open-label, placebo-controlled, prospective study was performed to evaluate the efficacy of Jarilla–Coffea extract gel on eyelashes and eyebrows in premenopausal and postmenopausal healthy women during a daily administration period of 2 months, in the case of eyebrow growth, and 3 months, in the case of eyelash growth. These evaluation periods are in accordance with the product prospectus.

Ethical considerations and good clinical practices

This study was conducted in accordance with the intent and purpose of Good Clinical Practice regulations described in Title 21 of the US Code of Federal Regulations, the requirements of the Declaration of Helsinki (1964), Amendments Tokyo (1975), Venice (1983), and Hong Kong (1989), Seúl (2008), and for CLAIM Standard Operating Procedures and National Laws: “Guía de Buenas Prácticas de Investigación Clínica en Seres Humanos”, in accordance to Resolution 1490/2007, published in “Boletín Oficial de la República Argentina” 14/11/2007, page 6. The study was approved by a committee of independent ethics, which assessed the study protocol and informed consent. Once the protocol was approved by the independent Ethics Committee or institutional review board (CLAIM’s Institutional Ethics Committee), the acceptance of the informed consent of the volunteer was obtained, in which, all the aspects making the study, all subjects and related research on which it requires information, and what was done with a terminology was explained in a clearly understandable manner to the volunteer. Informed consent form: Eyelashes and Eyebrows: Nr 0167-16.

Products under study

The products used in this study were supplied by the Garre-Guevara Laboratory. The products were identified as Jarilla–Coffea extract eyebrows (PE928061A) and Jarilla–Coffea extract eyelashes (PE038061) fashioner gels and placebo. Both gels were viscous lotions slightly colored with penetrating odor. The placebo used was glycerin USP.

Volunteers

Volunteers were recruited during a period of 3 months (July 2016 to September 2016). The study sample comprised ten women volunteers aged between 44 and 60 years (mean: 51.2 ± 1.54). One volunteer was withdrawn from the study due to a lack of adherence, so the sample size was then reduced to nine volunteers.

Before enrolling in the trial, all volunteers were informed about the study and signed an informed consent. All volunteers were subjected to a thorough clinical revision. The following inclusion criteria were employed: 1) age: to be between 40 and 60 years; 2) volunteers with low density hair in eyelashes and eyebrows; 3) to have signed the informed consent form; and 4) volunteers who frequently used cosmetics and agreed to comply with the clinical trial requirements. Exclusion criteria were: 1) presence of any medical condition, or the ingestion or application of any medication by either the oral or the ocular route, such as steroidal and NSAIDs; 2) diagnosis of psoriasis, eczema, atopic dermatitis, folliculitis, lentigines, eye dryness, or ocular allergic diseases; 3) suffer from diabetes, asthma, or respiratory allergic diseases; 4) to have received treatments with immunosuppressant drugs as a consequence of organ transplantation; and 5) have a clinical record of cutaneous reactions after cosmetics application.

Study objectives and assessments

Volunteers were treated with Jarilla–Coffea extract eyebrow or eyelash gel on a daily basis, once a day, during 60 consecutive days with the eyebrow gel, or during 90 consecutive days with the eyelash gel. Two main variables were observed: 1) efficacy in relation to hair growth defined as thickness, appearance of new hair, area with hairs and hair length; and 2) safety and tolerability. The thickness of eyebrows and eyelashes was determined by a noninvasive methodology using a video microscope MedicalScope® with magnification of 200×. A calibrated micrometer scale having a least measurement of 0.01 mm was incorporated in the software of the video microscope and the diameter of at least ten hairs, from a same zone of the eyebrow or the eyelash, was measured always at the same position close to their bases using

the measuring eyepiece. An average of the measurements was obtained for each volunteer. Images were registered under CD protocol no. 0164-16-12E1 (eyebrow) and no. 0164-16-01E2 (for eyelash) on day 0 (T_0) and on day 60 (T_{60}) or on day 90 (T_{90}), respectively. The appearance of new hairs and total area with hair, and eyelash length were quantified using a photographic record with Fotofinder® (Fotofinder Systems GmbH, Bad Birnbach, Germany). The number of patients presenting variation in growth of new hair and length were also recorded.

Women were instructed to apply the product on the right eyebrows or eyelashes and to treat the left ones with the placebo, using the application instructions specified in the prospectus, that is: 1) daily application; 2) apply the product directly on the selective zone of eyebrows or eyelashes; 3) wash their hands between the application of placebo and treatment; 4) remove the excess of product; and 5) leave the product applied for 15–20 minutes. During the study period, women were forbidden to use other products or make up and to have sunbath.

Safety studies

Volunteers were subjected to ophthalmologic studies. First, a clinical history was obtained through examination of ocular surface with a slit lamp.

An evaluation of ocular surface was done after the treatment with the product. Daily appointments were made with each volunteer during the first and third week after treatment. The presence of two clinical signs and two symptoms were considered positive for side effects.

Scales of signs and symptoms that were used to evaluate side effects are shown in Table 1:

Table 1

Scales of signs and symptoms used to evaluate side effects	Description
Symptoms score	
0	Absent
1	Present without discomfort
2	Present with discomfort without interfering with daily activities
3	Present with discomfort and interfering with daily activities
Signs score	
0	Absent
1	Hardly visible
2	Visible but not severe
3	Visible and severe
Local tolerance	
0	Absent
1	Hardly visible
2	Visible but not severe
3	Visible and severe

To determine the presence of signs and symptoms, an instrumental analysis was done with a tearscope, which allowed evaluating the lachrymal film stability through the interference of colors produced by the instrument on the lachrymal film when the film is disrupted. A crystallization test was also done, which allows determining the quality of the lachrymal film. Four types of lachrymal crystallization are known:

Type I: like a tree with ramifications.

Type II: like type I but with less ramifications with a few small leaves.

Type III: partial crystallization without fern formation.

Type IV: no crystallization.

Statistical analysis

A level of significance of 0.05 was employed in the hypothesis test. Percent variations (PVs) were determined for each volunteer employing the following formula:

$$\text{Percent variation} = [(\text{effect } T_F / \text{effect } T_0) - 1] \times 100$$

The mean PV was calculated as the mean of individual PV ($\mu\text{PV} = [(\sum \text{PV}) / n]$), but not as PV of means $\text{PV}\mu = [(\mu T_F) / (\mu T_0) - 1] \times 100$, where T_0 : basal time, T_F : time at 90 or 60 days of application. The mathematical analysis was done with IBM SPSS Statistics 22 statistical package.

To determine if the increase in the mean thickness during the treatment was greater than that obtained with the placebo, the linear generalized model for repeated measures (LGMRM) was applied. Results obtained with the product between T_0 and T_{90} were analyzed with Student's *t*-test.

Results

Effects on eyelashes

Eyelash thickness

With the test product, the thickness means were 0.05 and 0.0594 at T_0 and T_{90} , respectively. This percent change was about 19.44% (Table 2 and Figure 1A). All women displayed increased eyelash thickness.

For the placebo, the thickness mean for eyelashes was 0.05 at T_0 and T_{90} ; the PV was 0% (Table 2 and Figure 1A). No women treated with placebo presented any change in eyelashes thickness. The results obtained with the active principle and the placebo are shown in a representative micrograph (Figure 2C).

To determine whether the increase in the mean thickness during the treatment was greater than that obtained with the placebo LGMRM was applied. The differences between

Table 2 Eyelash thickness: statistical descriptors

	N	Minimum	Maximum	Mean	SD
Active T_0	9	0.04	0.06	0.0500	0.00707
Active T_F	9	0.05	0.07	0.0594	0.00635
PV T_F/T_0	9	8.33%	25.00%	19.44%	4.93077%
Placebo T_0	9	0.04	0.07	0.0506	0.00950
Placebo T_F	9	0.04	0.07	0.0506	0.00950
PV T_F/T_0	9	0.00%	0.00%	0.00%	0.00000%

Abbreviation: PV, percent variation.

treatment and placebo were highly significant ($P < 0.001$). Besides, the mean thickness values obtained with the product between T_0 and T_{90} were significantly different ($P < 0.001$, Student's *t*-test).

Eyelash maximal length

For the test product, the maximal eyelashes length means were 34.37 mm and 35.54 mm at T_0 and T_{90} , respectively. The percent change was 3.59% (Table 3 and Figure 1B). About 55.6% of volunteers presented an increase in the maximal eyelashes length.

For the placebo, the maximal eyelash length means were 36.09 mm and 35.54 mm, at T_0 and T_{90} , respectively. The PV was -1.09% (Table 3 and Figure 1B). The effects obtained with the active principle and the placebo are shown in a representative micrograph taken with a Fotofinder® camera (Figure 2A, B).

To determine whether the increase in the mean maximal length obtained after treatment with the product was higher than that obtained with the placebo LGMRM was applied. There was no difference between treatment and placebo ($P = 0.11$).

Effects on eyebrows

Eyebrow thickness

The active principle induced an increase in the eyebrow thickness of 20% in 80% of volunteers (Table 4 and Figure 3A, B, E, F). The eyebrow thickness means were 0.055 and 0.06 at T_0 and T_{60} , respectively. A PV mean of 9.16% was observed (Table 4).

For the placebo, the mean thickness was 0.058 at T_0 and T_{60} . No PV was assigned in 100% of the volunteers (Table 4 and Figure 3A, B, E, F).

To determine whether the increase in the mean thickness after the treatment was higher than that obtained with the placebo LGMRM was applied. There was difference between mean values after treatment with the product and the placebo ($P < 0.001$).

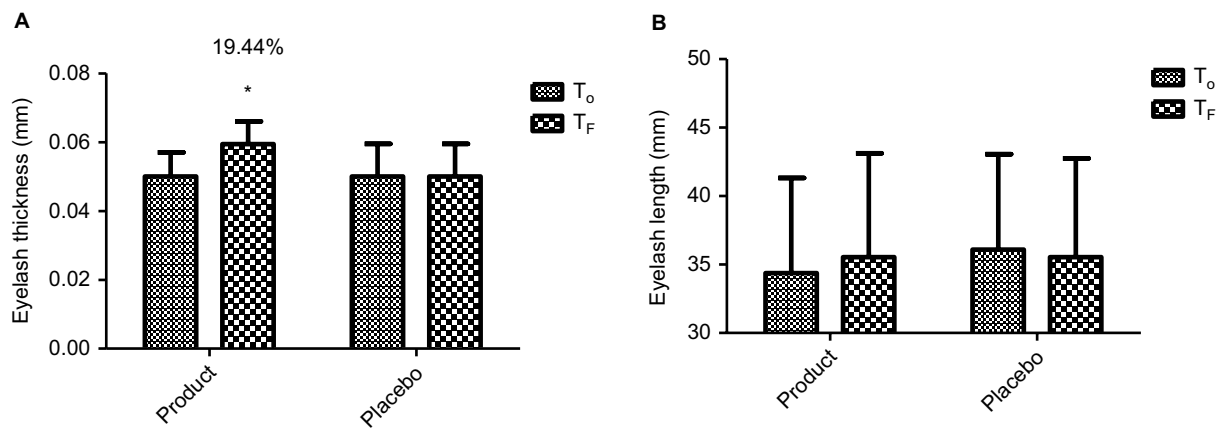


Figure 1 Effect of eyelash gel on (A) eyelash thickness and (B) length.

Notes: The product effect was compared with a placebo. Results represent the mean \pm SD of nine determinations. * $P < 0.05$: significant differences between T₀ and T₉₀ (Student's *t*-test).

Table 3 Eyelash maximal length: statistical descriptors

Statistical descriptors	N	Minimum	Maximum	Mean	SD
Active T ₀	9	21.5720	44.3710	34.3730	6.9594393
Active T _F	9	22.7930	46.4040	35.5494	7.5886040
PV T _F /T ₀	9	-5.13	23.92	3.59%	9.04619
Placebo T ₀	9	22.7930	43.9620	36.0910	6.9738635
Placebo T _F	9	24.8320	47.2160	35.5494	7.2176550
PV T _F /T ₀	9	-13.67	8.95	-1.09%	8.65175

Abbreviation: PV, percent variation.

Eyebrow new hairs

A 60% of women treated with the gel presented an increase in the number of hairs in the eyebrows; the mean new hair values were 119 and 130 at T₀ and T₆₀, respectively, with a mean increment value of 12.57% and a maximal increment of 56% (Table 5 and Figure 3A–D). Moreover, the mean values of new hairs obtained with the placebo were 118 and 123, at T₀ and T₆₀, respectively, with a percent increase of 3.94% and a maximal increment of 62%. In this case, 50% of volunteers presented a change.

To determine whether the increase in the mean value of new hairs after the treatment was higher than that obtained with the placebo LGMRM was applied. There was no difference between treatment and placebo ($P=0.749$).

Total area with hair in eyebrows

The mean total areas with hair obtained after treatment with the product were 0.435 and 0.444, for T₀ and T₆₀, respectively. The mean percent increase was 15.53%, with a maximum value of 132% (Table 6). This change was observed in 50% of volunteers (Figure 3C, D).

For the placebo, the total areas with hair means were 0.400 and 0.418 for T₀ and T₆₀, respectively. The mean percent

increase was 5.69%, with a maximum value of 84%. This change was observed in 50% of volunteers (Figure 3C, D).

To determine whether the increase in the mean total area with hair after the treatment with the product was greater than that obtained with the placebo LGMRM was applied. There was no difference between the total area with hair between treatment and placebo ($P=0.907$).

Safety studies

When applied on the eyelashes, the product did not cause any ocular adverse reactions. The lachrymal film in all volunteers did not present any alteration. The crystallization was type I in all cases. In addition, there was no difference – in the effects obtained on the eyelids and the orbital ledge skin – of product treated and placebo.

Discussion

In this work, the efficacy of Jarilla–Coffea extract eyelashes and eyebrow gels was demonstrated. In all healthy premenopausal and postmenopausal women with scant eyelashes and eyebrows, both these gels were capable of increasing the eyelash and eyebrow growth in relation to thickness in all women treated, improving the hair appearance by this way. Moreover, other parameters such as the eyelash maximal length (that increased in 55.6% of the volunteers) and the eyebrow new hairs (in 60% of the volunteers) related to growth were modified by this gel. However, there was no statistical difference with the placebo ($P=0.11$ and 0.749 , respectively).

It could be possible that the product had increased thickness of eyelash and eyebrow hair by promoting hair from telogen to anagen phase. Previously, it was demonstrated that the plant extracts, present in the gel, were capable to induce hair

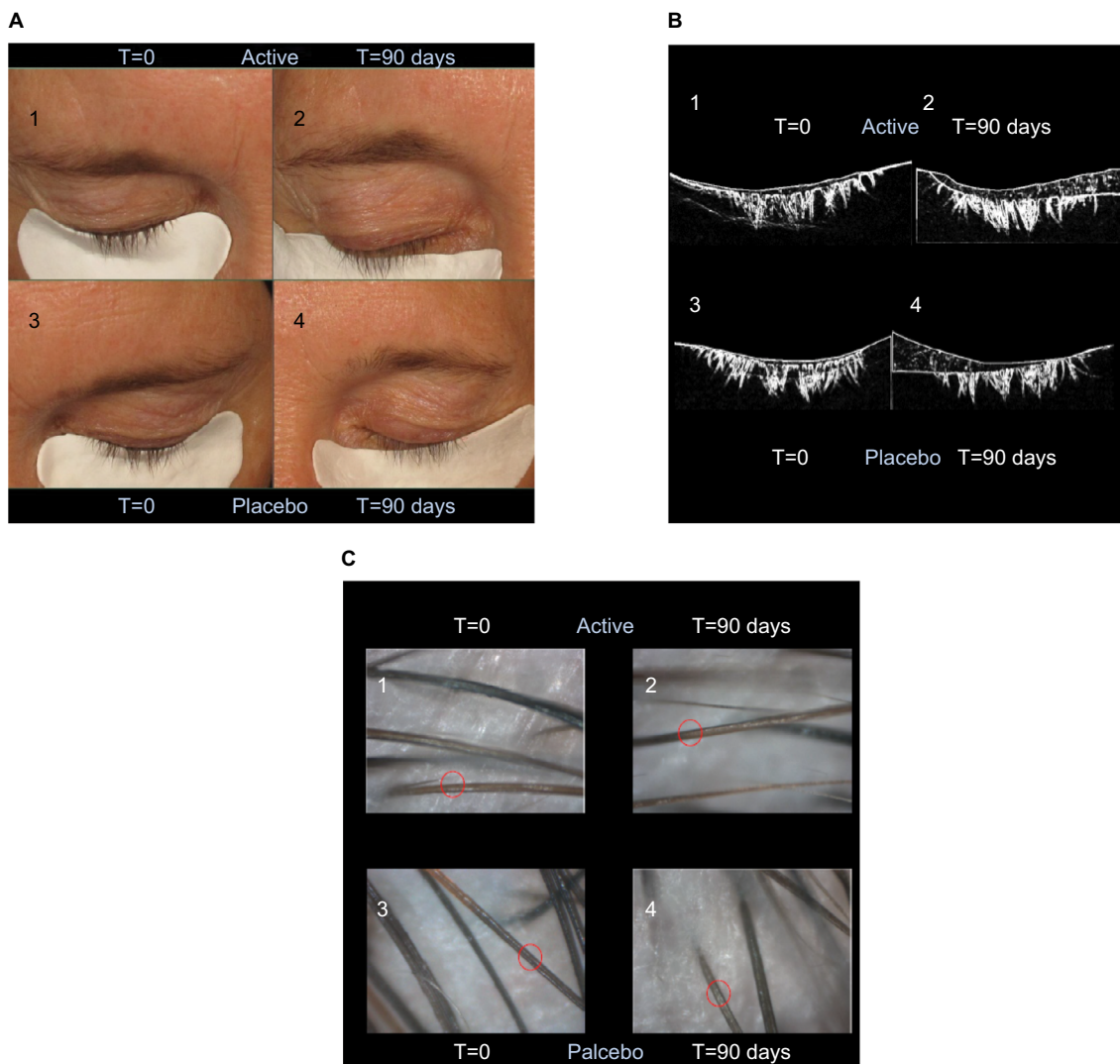


Figure 2 Effect of eyelash gel on (A, B) eyelash length and (C) thickness.

Notes: (A) Representative micrographs of eyelashes treated either with gel (upper) or with placebo (lower). Micrographs were taken with Fotofinder®. (B) Analysis of the micrographs shown in (A). (C) Representative micrographs of eyelashes treated with either gel (upper) or with placebo (lower). Analysis of thickness was done with video microscope. 1: T₀; 2: T₉₀ treated with gel; 3: T₀; 4: T₉₀ treated with placebo. T₀: initial time, T₉₀: final time.

Table 4 Eyebrow thickness: statistical descriptors

Statistical descriptors	N	Minimum	Maximum	Mean	SD
Active T ₀	10	0.04	0.07	0.0550	0.00850
Active T ₆₀	10	0.04	0.07	0.0600	0.009129
Active PV	10	0.00%	20.00%	9.1667%	6.19886%
Placebo T ₀	10	0.04	0.07	0.0580	0.01033
Placebo T ₆₀	10	0.04	0.07	0.0580	0.01033
Placebo PV	10	0.00%	0.00%	0.0000%	0.00000%

Abbreviation: PV, percent variation.

from telogen to anagen phase in C3H mice.⁷ Moreover, the capacity of a lotion, made with the two extracts, in stimulating hair growth in humans has previously been determined. Such study was conducted in patients with noncicatricial alopecia

in a randomized, prospective, open-label, single-branch, phase IV cohort study. The clinical study revealed that the lotion improves the appearance, volume and appearance of new hair due to an induction of hair growth, and a decrease of hair loss (from the moment of its application) in 84.6% of patients (total patients 52), after 90 days of treatment. The decrease in hair loss was higher in women, and the best results were obtained in grades I and II androgenic alopecia and in alopecia areata.⁸

The mechanism of the extracts involved in promoting anagen phase could be related to a modulation of PGs. It was demonstrated that unlike PGD₂, which has an inhibitory effect, prostaglandin F_{2α} and PGF₂ induce hair growth. Therefore, PGs may be considered a pharmacological target for the treat-

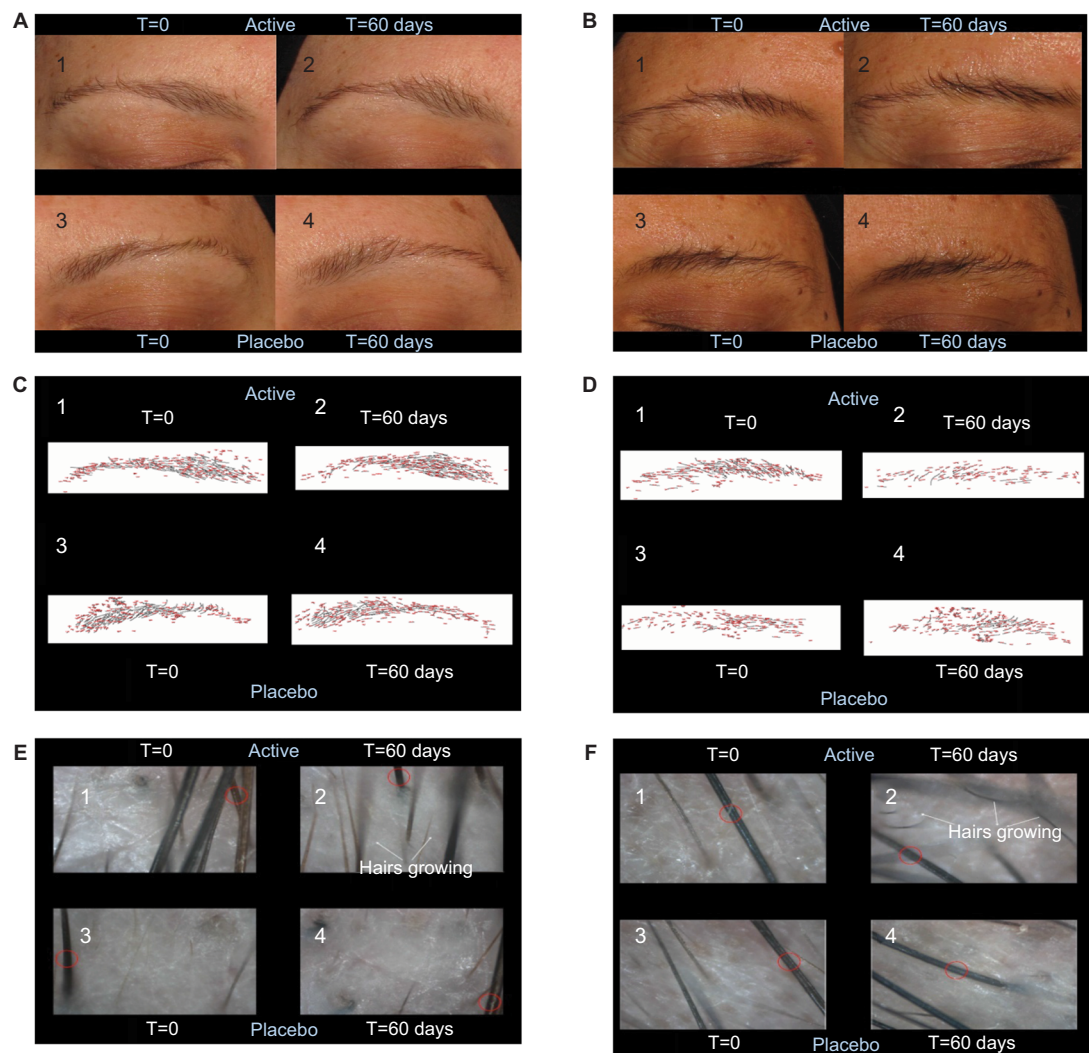


Figure 3 Effect of eyebrow gel on (A–D) eyebrow growth and (E, F) thickness.

Notes: (A, B) Representative micrographs of eyebrows treated either with gel (upper) or with placebo (lower). Micrographs were taken with Fotofinder®. (C, D) Analysis of the micrographs shown in (A, B). (E, F) Representative micrographs of eyebrows treated either with gel (upper) or with placebo (lower). Analysis of thickness was done with video microscope. 1: T_0 ; 2: T_{60} treated with gel; 3: T_0 ; 4: T_{60} treated with placebo. T_0 : initial time, T_{60} : final time.

Table 5 Eyebrow new hairs: statistical descriptors

Statistical descriptors	N	Minimum	Maximum	Mean	SD
Active T_0	10	56	204	119.70	57.337
Active T_F	10	60	269	130.70	66.369
Active PV	10	-36%	56%	12.57%	29.776%
Placebo T_0	10	61	162	118.30	32.308
Placebo T_F	10	53	206	123.80	49.571
Placebo PV	10	-24%	62%	3.94%	27.362%

Abbreviation: PV, percent variation.

Table 6 Eyebrow total area with hair: statistical descriptors

Statistical descriptors	N	Minimum	Maximum	Mean	SD
Active T_0	10	0.15924	0.84720	0.4350210	0.23240888
Active T_F	10	0.17062	0.76764	0.4448500	0.20246945
Active PV	10	-51.13%	132.60%	15.5322%	51.52571%
Placebo T_0	10	0.14609	0.75613	0.4002340	0.16880262
Placebo T_F	10	0.12053	0.75707	0.4184350	0.19337064
Placebo PV	10	-24.83%	84.87%	5.6920%	33.34614%

Abbreviation: PV, percent variation.

ment of androgenetic alopecia.^{9,10} In line with these findings, it was found that inhibitors of PGs synthesis, such as aspirin, indomethacin, and ibuprofen, can inhibit hair growth.^{11,12} The extracts present in the gel have flavonoids (quercetin derivatives)

and nordihydroguaiaretic acid (NDGA) – compounds that can modulate endogen PGs. For example, NDGA is an inhibitor of lipooxygenase, therefore, this polyphenol could increase the PG synthesis, by activating cyclooxygenase (COX) pathway, thus

increasing eyelash growth. Moreover, and depending on its concentration, NDGA may also inhibit the synthesis of PGD2 through the inhibition of COX-1. In addition, it has also been demonstrated that the flavonoids present in Chinese herbs, such as amentoflavone, quercetin-3-*O*-rutinoside (rutin), and hinokiflavone, are PGD2 inhibitors with good pharmacokinetic properties and high efficacy in the treatment of hair loss.¹³

By other way, it is known that ROS can modulate the balance between the anagen and telogen phases, causing damage to DNA, cell proteins, and lipids leading to cell arrest and follicular cell death.¹⁴ These events are known to promote the hair follicle to shift from the anagen to the telogen phase, or hair loss phase. Normally, the ROS levels are controlled by the activity of antioxidant enzymes, such as superoxide dismutase (SOD), catalase (CAT), and peroxidase (Px); however, there are cases in which the ROS production exceeds their metabolism rate leading to different diseases. It is known that the superoxide anion is involved in the occurrence of alopecia, since this free radical interferes with the production of nitric oxide, which is an essential factor for hair growth.¹⁵

The extracts present in the gel are also known to have antioxidant activities, such as Px, CAT, and SOD-like activities.^{16,17} Therefore, the effects observed after the treatment with the gel could also be related to these biological activities exerted by the extracts.

It is thought that herb-based formulations may influence hair growth by stimulating or inhibiting a variety of growth factors, cytokines, hormones, enzymes, as well as through the modulation of signaling pathways. Some herbs and their active constituents allow maintaining the hair growth cycle in the anagen phase, while others inhibit hair apoptosis during the catagen phase.

The fact that the product increased hair length and the appearance of new hairs but not in a significant manner suggests that a longer treatment is necessary.

It is noteworthy that neither ocular nor dermal side effects were observed with Jarilla–Coffea extract gel in contrast with the products used nowadays to increase eyelash or eyebrow growth or for alopecia treatment.

Conclusion

This study concludes that the Jarilla–Coffea extract gel was capable of improving eyelash and eyebrow growth by inducing hair thickness, and also the appearance of new hairs and hair length were changed, without exerting local adverse effects.

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Author contributions

CA was involved in conception of the study and performed analysis and interpretation of data. Moreover, he contributed toward data analysis, drafting, and critically revising the manuscript and agrees to be accountable for all aspects of the work. MRA was involved in extract preparation and quality control of products and analysis and interpretation of data. SPD planned the trial performing visits and instrumental evaluation and protocol design. All authors contributed to data analysis, drafting or revising the article, gave final approval of the version to be published, and agree to be accountable for all aspects of the work.

Disclosure

The authors report no conflicts of interest in this work.

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