



ASSESSMENT OF THE CLINICAL EFFICACY AND SAFETY PROFILE OF AN UNDER-EYE CREAM FORMULATION CONTAINING LANACHRYST™ 2B PF, ALDAVINE™ 5X, AND TYROSTAT™-09 IN HEALTHY ADULT SUBJECTS

Rushabh Dharamshi¹, Poorna Pai¹, Kartiki Jadhav¹, Priyanka Sharma^{1*}

¹Amvigor Organics Private. Limited, 1311-1317, 1401, 1403, 1405, 1407, 1411-1420, 13th and 14th Floor, The Gateway Atmosphere, Mulund Goregaon Link Road, Mumbai, Mumbai Suburban, Maharashtra, Pincode-400080.

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*Corresponding author:

Priyanka Sharma

Amvigor Organics Private.
Limited, 1311-1317, 1401, 1403,
1405, 1407, 1411-1420, 13th and
14th Floor, The Gateway
Atmosphere, Mulund Goregaon
Link Road, Mumbai, Mumbai
Suburban, Maharashtra, Pincode-
400080.

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ABSTRACT

One of the primary locations wherein visible symptoms and symptoms of ageing start to emerge is across the eyes. Dark circles below the lower eyelids are an issue which has numerous reasons like aging, genetics, loss of sleep, stress, vessel fragility and poor circulation under eyes. There are an extensive variety of processes and products that could successfully deal with the pores and skin around the eyes. The aim of clinical research was to check the efficacy of an under-eye cream having a combination of 3 actives LANACHRYST™ 2B PF, ALDAVINE™ 5X, AND TYROSTAT™-09 from Lucas Meyer Cosmetics. 36 female subjects were selected and asked to apply Under eye cream around the eye area once a day for 56 days. Safety and efficacy were assessed by the dermatologist according to the Bazin's atlas. Melanin content of the skin was measured by a Mexameter. The cream was well appreciated for making under eye skin supple, for reducing under eye dark circles, wrinkles & fine lines around the eye. In 56 days, there was a reduction in melanin content by 22.9%, under eye dark circles by 13.8%, intensity of crow's feet wrinkles reduced by 10.2%. To conclude, dermatological evaluation for efficacy shows significant reduction in wrinkles & fine lines and reduction in under eye dark circles after 56 days of application of the under-eye cream containing LANACHRYST™ 2B PF, ALDAVINE™ 5X, AND TYROSTAT™-09.

KEYWORDS: Aldavine™ 5X, Lanachrys™ 2B PF, Tyrostat™-09, Under eye dark circle, Under eye puffiness, Melanin reduction.

1 INTRODUCTION

One of the primary locations wherein visible symptoms and symptoms of ageing start to emerge is across the eyes. The sensitive pores and skin on this area is extraordinarily thin^[1] and consists of little subcutaneous fats tissue.^[2] During aging, the pores and skin cells divide slowly, and the internal layer, known as the dermis, starts to thin. Networks of the elastin and collagen fibers loosen and unravel, leading to depressions at the surface.^[3] The pores and skin additionally lose its elasticity, is much less capable to hold moisture, sebaceous glands are much less efficient, and the skin

heals slowly. All of these contribute in formation of wrinkles.^[4]

Dark circles below the lower eyelids are another issue which can have numerous reasons like aging, genetics, loss of sleep, and stress,^[5] however, few other pathological reasons are:

- Vessel fragility: Dark under-eye circles start inside the capillaries. If the blood vessels are fragile, they break down easily and can lead to leak of hemoglobin into the skin that surrounds the eye. As the hemoglobin breaks down, pigments, which are

the degradation products, are formed and collected below the dermis and epidermis. This causes a dark and pigmented discoloration around but majorly below the eye.^[6]

- Poor circulation under eyes: Poor circulation results in less oxygenated blood, and hence, causes a bluish appearance.

Nowadays, there is an extensive variety of processes and products that could successfully deal with the pores and skin around the eyes. Fine wrinkles may be treated with dermal treatments that bring about the contraction of collagen or through treatments that plump up the epidermis and dermis.^[7]

The objective of this clinical research is to check the efficacy of an under-eye cream having a combination of 3 actives from Lucas Meyer Cosmetics viz, LANACHRYS™ 2B PF, ALDAVINE™ 5X, AND TYROSTAT™-09, in a single, non-randomized, blind, non-comparative study.

2 MATERIALS AND METHODS

The actives LANACHRYS™ 2B PF, ALDAVINE™ 5X, AND TYROSTAT™-09 are proprietary molecules manufactured by Lucas Meyer Cosmetics and solely distributed by Amvigor Organics Pvt. Ltd. in India.

Tyrostat™ - 09: Tyrostat™ – 09 contains *Rumex occidentalis* extract with water & glycerin. It has strong inhibitory effect on the tyrosinase enzyme. It helps to reduce formation of unwanted pigmentation, restore uneven skin tone.

Tyrostat™ - 09 safely reduces skin pigmentation and erythema for a more uniformed and lighter skin complexion.

Aldavine™ 5X: Aldavine™ 5X mainly targets and inhibits the biochemical mediators like VEGF cytokine, PGE2 activity and MMP-2, which are up-regulated in the skin upon aging, UV exposure, and environmental stress. It maintains microcapillary integrity and soothes sensitive skin (reduces tingling sensation and increases skin comfort). It increases skin elasticity and resiliency, reduces the appearance of dark circles and reduces the eye puffiness.

Lanachrys™ 2B PF: It is an extract of *Chrysanthellum indicum*, which can effectively restore, activate and regulate the microcirculation. It helps in reduction of the appearance of cellulite and puffiness under the eye by stimulating limiting oedema. It has a draining effect and reduces eye puffiness.

2.1 Preparation of formulation

The finished formulation was formulated by the Formulation team at Amvigor Organics Pvt. Ltd. Oil phase & water phase were prepared separately. Both the phases were heated at 70-80°C. The thickener was added

into the water phase at the same temperature and was mixed until it gets dispersed completely. Oil phase was added into water phase at same temperature & was stirred continuously to form an emulsion. The emulsion was cooled below 40°C. At this stage all the actives including LANACHRYS™ 2B PF, ALDAVINE™ 5X, AND TYROSTAT™-09 and preservatives were added.

2.2 Under eye cream efficacy

The study was performed at Mascot Spincontrol. The selection was based on a preliminary interview, that explains the study's modalities, its practical considerations, and all possible cosmetic benefits, inconveniences or potential risks. For the study a selection of 36 female subjects was made. The consent form was read, approved, and signed by all the subjects to substantiate the fact that they freely accept the conditions of the study which were described to them. The most recent recommendations of the World Medical Association (64th WMA Declaration of Helsinki, Fortaleza, Brazil, October 2013) and ICH-E6 - Good Clinical Practice (GCP) guidelines were all adhered to in this investigation.

2.2.1 Inclusion criteria

- Healthy subjects (no infectious and evolutive pathology which could make the subject vulnerable and stop the study)
- Between 25 and 55 years of age
- Skin is healthy on the studied anatomic unit (free of eczema, wounds, inflammatory scars etc)
- Having visible under eye dark circles
- Having Crows feet wrinkle of grade 2 and above according to the Bazin's atlas

2.2.2 Exclusion criteria

- Being pregnant or breastfeeding or having stopped to breastfeed in the past three months
- Having a progressive asthma
- Being epileptic
- Having a diagnosed or highly probable allergy to one or several compounds of the cosmetic products
- Following a chronic medicinal treatment comprising any of the following products: aspirin-based products, anti-inflammatories, anti-histamines, corticotherapy, taken by general or local routes (the only medication permitted is paracetamol)
- Having undergone a surgery requiring a general anaesthetic of more than one hour in the past 6 months
- Having changed her cosmetic habits in the 14 days preceding the start of the study on the studied anatomic unit
- Having applied a cosmetic product (included make-up) or skin care product on the studied areas the day of the study (only face cleaned with water is accepted)

2.2.3 Product application

The application is carried out by the subjects themselves at home; except the first application of test products will be carried out at Mascot Spincontrol under the guidance of the CRA:

Table I: Product Application for the trials.

Products	Application area	Frequency of application	Application duration	Conservation
Cream	Around the eyes	Twice daily	56 days	At ambient temperature

Modalities of application

Cleanse the face with water. Take approximately 4-5 drops of the product on fingertip. Apply dot wise around both eyes. Then gently massage using fingertips till the product is absorbed into the skin. Apply the test product twice daily i.e. morning & evening for the period of 56 days.

2.2.4 Dermatological Evaluation (Cosmetic Acceptability)

Safety of the product is assessed by the dermatologist, through the grading on the test area of defined clinical signs (observed by the dermatologist) and functional signs (felt by the subjects and reported to the dermatologist), at T0, T+14 days, T+28 and T+56 days visit, as follows:

Clinical signs include:

- Erythema
- Oedema
- Dryness
- Scaling
- Peeling

Functional signs include:

- Itching
- Tingling

The evaluation is performed using the following scale:

- 0: None
- 1: Slight
- 2: Moderate
- 3: Severe

2.2.5 Dermatological Evaluation by a Dermatologist (Efficacy)

The overall product efficacy, is assessed by the dermatologist, through the grading at T0, T+14, T+28 and T+56 days visit for the following parameters:

- Reduction in under eye dark circles (Scale: 1 to 10; where 1=No dark circle, 5=Moderate dark circle with smaller area & 10=severe dark circle with larger area)
- Intensity of crow's feet wrinkles and fine lines using Page n° 40 & 41 Crow's Feet Wrinkles of Skin Ageing Atlas (Scale: 0 to 6)^[8]

2.2.5.1 For Under Eye Dark Circles

The result will be given in terms of a score from 1 to 10 for under eye dark circle. The significant decrease in the score for under eye dark circle parameter shows an effect

of the product in terms reduction in under eye dark circle.

2.2.5.2 For Intensity of Crow's Feet Wrinkles and Fine Lines

The result was given in terms of a score from 0 to 6 for intensity of crow's feet wrinkles. The decrease in the grade of the intensity of wrinkles and fine lines shows an effect of the product in terms of reduction in wrinkles and fine lines.

2.2.6 Mexametry

Mexametry is the technique used to determine the melanin content of the skin. This method is based on absorption/reflection. The probe of the Mexameter® MX 18 emits 3 specific light wavelengths. A receiver measures the light reflected by the skin. The positions of emitter and receiver guarantee that only diffuse and scattered light is measured.^[9] As the quantity of emitted lights defined, the quantity of light absorbed by the skin can be calculated. The apparatus used is a MX 18 (Courage and Khazaka, Germany).

Three measurements were carried out on the pigmented under eye area, for each subject and each examination time on T0, T+14 days, T+28 days and T+56 days.

3 RESULT AND DISCUSSION

3.1 Subject's Self Evaluation

The cream was well appreciated for making under eye skin supple, for reducing wrinkles & fine lines around the eye and reducing under eye dark circles with 100% agreement, for helping to look under eye skin younger & healthier with 86%, 86% to 100% agreement, for helping to improve overall appearance of eye area with 94%, 94% to 100% agreement respectively from 14 days, 28 days to 56 days of application.

The cream was well appreciated for its appealing fragrance, for not making the skin sticky with 94% agreements, for quick penetration in skin with 100% agreement at the given time point.

The cream showed no itching, irritation and burning sensation on the skin with 100% agreement from 14 days, 28 days to 56 days of application.

Table II: Summary of Self-assessment.

Number of subjects	T + 14 days		T + 28 days		T + 56 days	
	36		36		35	
	% agreement	Significant? (5%)	% agreement	Significant? (5%)	% agreement	Significant? (5%)
The test product helps to make my under-eye skin supple.	100%	Yes	100%	Yes	100%	Yes
The test product helps in reduction wrinkles and fine lines around the	100%	Yes	100%	Yes	100%	Yes
The test product helps in reduction of under eye dark circle.	100%	Yes	100%	Yes	100%	Yes
The test product helps to look my under-eye skin younger and	86%	Yes	86%	Yes	100%	Yes
The test product helps to improve overall appearance of eye area.	94%	Yes	94%	Yes	100%	Yes
The fragrance of the product is pleasant.	94%	Yes	-	-	-	-
The test product penetrates quickly into skin.	100%	Yes	-	-	-	-
The product does not make my skin sticky.	94%	Yes	-	-	-	-
The test product does not cause itching on the skin.	100%	Yes	100%	Yes	100%	Yes
The test product does not cause irritation on the skin.	100%	Yes	100%	Yes	100%	Yes
The test product does not give burning sensation to the skin	100%	Yes	100%	Yes	100%	Yes

3.2 Dermatological Evaluation: Cosmetic Acceptability

On the basis of Dermatological Evaluation for Cosmetic Acceptability, it has been observed that there is no

Table summarises the means and standard deviations of the grades of the studied parameters observed at T0, T+14 days, T+28 days, T+56 days as well as the

occurrence of erythema, oedema, dryness, scaling, peeling, itching and tingling after 56 days of application.

3.3 Dermatological Evaluation: Efficacy

The following corresponding statistical results for the evolution in time (Student test or Wilcoxon test, two-tailed for paired groups at 5%, after checking the normality of the distributions by a Shapiro-Wilk test at 1%).

Table III: Raw values for under eye dark circle and crow's feet wrinkles.

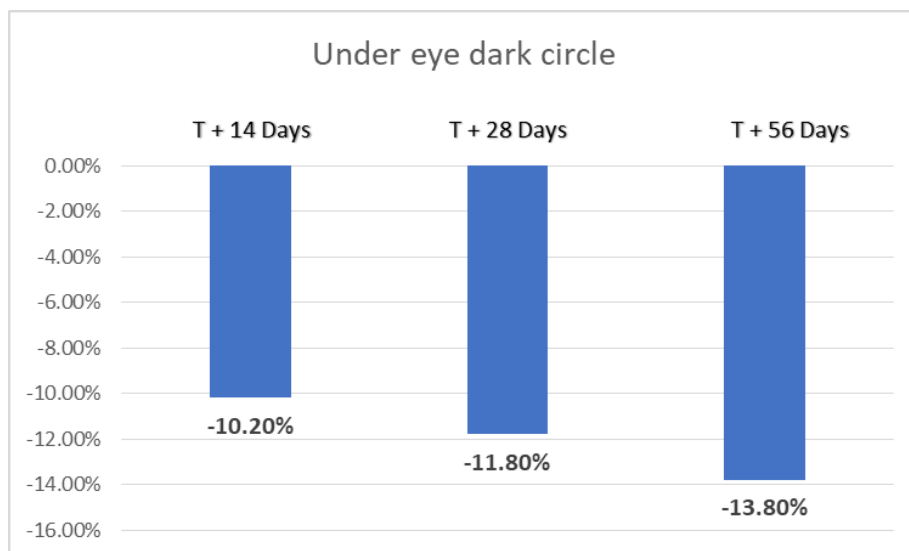
		T0	T+14 days	T+ 28 days	T + 56 days
Under eye dark circle	Mean	5.19	4.67	4.58	4.43
	Std Dev	0.75	0.76	0.69	0.65
	Significance at 5%		Yes	Yes	Yes
Crow's Feet wrinkles	Mean	3.76	3.76	3.72	3.39
	Std Dev	1	1	0.96	0.83
	Significance at 5%		No	No	Yes

3.3.1 Under Eye Dark Circles

The statistical analysis shows a significant decrease in under eye dark circles by 10.2%, 11.8% & 13.8% at T+14 days, T+28 days & T+56 days respectively.

Table IV: Reduction of under eye dark circles.

Parameters	Reduction		
	T+14 days	T+ 28 days	T + 56 days
Under eye dark circle	10.20%	11.80%	13.80%

**Figure 1: Reduction in the under eye dark circles.**

3.3.2 Intensity of Crow's Feet Wrinkles and Fine Lines

The statistical analysis showed a significant decrease in intensity of crow's feet wrinkles by 10.2% after 56 days of application.



Figure 2: Reduction in Puffy eyes.

Table V: Reduction of wrinkles.

Parameters	Reduction	
	T+ 28 days	T + 56 days
Crow's Feet wrinkles	1.10%	10.20%

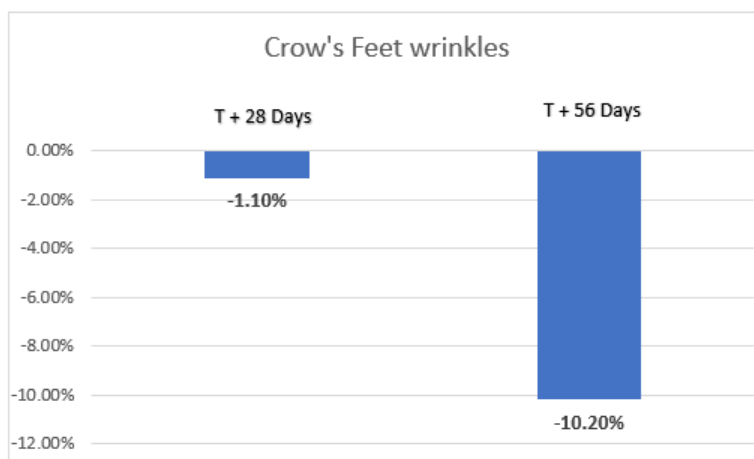


Figure 3: Reduction in Crow's Feet Wrinkles

3.4 Mexametry

The following Table summarises the means and standard deviations of the raw values of the studied parameter, observed at T0, T+14 days, T+28 days, T+56 days, as well as the corresponding statistical results for the

evolution in time (Student t-test or Wilcoxon test, two-tailed for paired groups at 5%. after checking the normality of the distributions by a Shapiro-Wilk test at 1%).

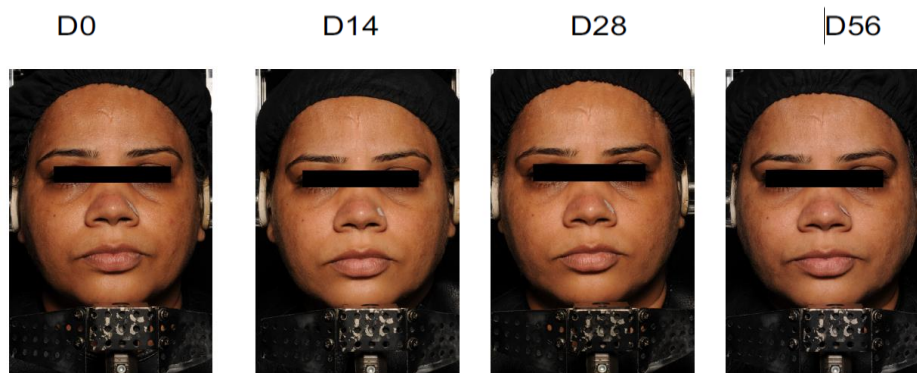


Figure 3: Reduction in melanin content under eye area.

Table VI: Raw values for Melanin reduction.

		T0	T+14 days	T+ 28 days	T + 56 days
Melanin	Mean	461.34	401.31	385.87	353.86
	Std Dev	73.43	75.36	71.35	71.89
	Significance at 5%		Yes	Yes	Yes

The statistical analysis shows a significant decrease in melanin content of the under eye dark circles by 13.0%, 16.4% & 22.9% at T+14 days, T+28 days & T+56 days, respectively.

Table VII: Reduction of Melanin.

Parameters	Reduction		
	T+14 days	T+ 28 days	T + 56 days
Melanin	13.0%	16.4%	22.9%

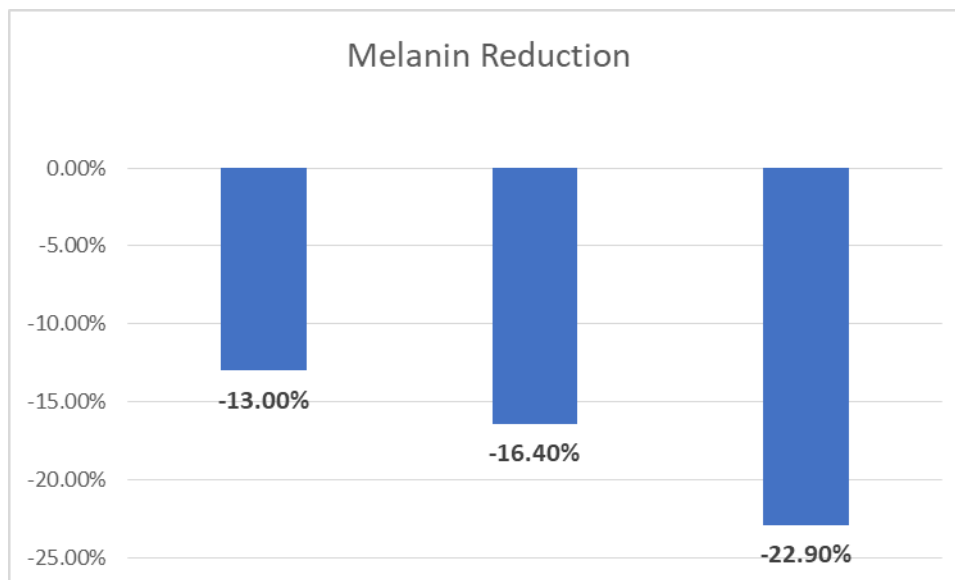


Figure 4: Reduction in Melanin.

4 CONCLUSION

The under-eye cream containing 3 actives from Lucas Meyer Cosmetics was applied twice a day on a panel of 36 Indian female subjects, aged between 36 and 55 years old, having under eye dark circles & crow's feet wrinkles. The statistical analysis shows a significant decrease in the melanin content of the under eye dark circles by 13.0%, 16.4% & 22.9%, respectively after T+14 days, T+28 days & T+56 days of product application shows reduction in under eye dark circles.

The statistical analysis showed a significant decrease in intensity of crow's feet wrinkles grade by 10.2% after 56 days of application, significant decrease in under eye dark circles by 10.2%, 11.8% & 13.8% at T+14 days, T+28 days & T+56 days, respectively.

To conclude, Dermatological Evaluation for efficacy shows significant reduction in wrinkles & fine lines and reduction in under eye dark circles after 56 days of application of the under-eye cream. Significant decrease

in melanin content of under eye dark circles through Mexametry shows reduction in under eye dark circles. Appreciation from the panel is obtained for the product through for product efficacy, product characteristics & product acceptability. Dermatological Evaluation for cosmetic acceptability showed no occurrence of clinical and functional signs for the cream till 56 days of application.

5 CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

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