CLINICAL EFFICACY OF PHOTOPROTECTIVE FORMULATIONS CONTAINING OR NOT THE ASSOCIATION OF VITAMIN C AND E DERIVATIVES

Vitoria TONINI[1,2], Céline PICARD[2], Patrícia MAIA CAMPOS*[1].

[1] *School of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, Ribeirão Preto, Brazil.*
[2]*Université Le Havre Normandie, Normandie Univ, URCOM UR 3221, F-76600 Le Havre, France.*

ABSTRACT

This study aimed to evaluate the physico-mechanical properties and clinical efficacy of photoprotective formulations containing (F2) or not (F1) vitamin derivatives as a complementary strategy to photoprotection[1,2]. Photoprotective formulations, containing (F2) or not (F1) the association of ascorbyl tetraisopalmitate and dl-alpha-tocopherol acetate, were characterized in terms of its flow properties and viscoelastic behaviour. Clinical efficacy was evaluated by means of non-invasive biophysical and skin imaging measurements obtained from the skin of the anterior and posterior regions of the forearms of 10 healthy research subjects, before and after a 28-day period. F1 and F2 exhibited shear thinning and a predominant elastic behaviour (G' > G'') within the linear viscoelastic region, with a higher strain value at the G'– G'' crossover and lower values of G' and G'' observed for F2. Characterization of basal conditions of the forearm skin revealed a significant reduction in dermis echogenicity for the posterior region. After 28 days of use of both formulations, it was observed significant improvements in skin hydration parameters. For F2, a subtle improvement in the Corner Density parameter, related to density of primary lines, was observed as well as an increase of higher magnitude in dermis echogenicity ratio, both in photoexposed skin regions. F2 showed more sustained elastic behaviour and higher resistance to imposed oscillatory strain. Comparative evaluation of basal conditions verified a significantly lower dermal density for the photoexposed skin. Both F1 and F2 were effective increasing skin hydration and dermis echogenicity in both evaluated regions. For F2, was also observed an improvement in density of primary lines, as higher values of the Corner Density parameter are correlated with younger ages and lower levels of photoexposure.

OBJECTIVES

- To evaluate the effect of the addition of vitamin derivatives on the physico-mechanical properties and clinical efficacy of photoprotective formulations.

METHODS

Flow properties and viscoelastic behavior were evaluated using the Discovery HR-2 Hybrid Rheometer (TA Instruments). A randomised double-blind clinical study was performed under the CEP/FCFRP Protocol nº 490 - CAAE: 00669818.7.0000.5403[3,4] to assess long-term photoprotective efficacy[5]. 10 healthy subjects, aged 28-31 years, phototype II or III[6], were recruited and subjected to a 7-day washout period. Measurements were obtained from the skin of the anterior (photoprotected, PP) and posterior (photoexposed, PE) regions of the forearms, before and after a 28-day period. Non-invasive biophysical and skin imaging techniques were employed to evaluate clinical efficacy under real use conditions.

- MoistureMap® MM 100 (Courage & Khazaka)
- Corneometer® CM 825 (Courage & Khazaka)
- Tewameter® TM 210 (Courage & Khazaka)
- Visioscan® VC 98 (Courage & Khazaka)
- Dermascan® C ultrasound (Cortex Technology)

RESULTS

F1 and F2 exhibited shear thinning and predominant elastic behaviour (G' > G'') within the linear viscoelastic region. F2 showed a slight higher strain value at the G'– G'' crossover, lower values of moduli with weaker strains overshoot of G'' (Fig. 1).

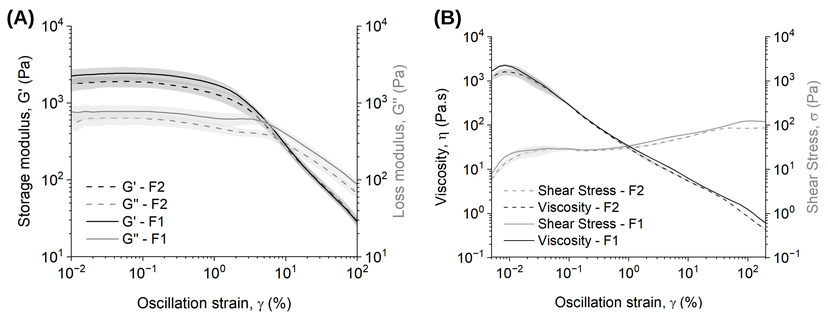

Fig. 1. Dynamic strain sweep test, 1 Hz, 25ºC, G' and G'', as a function of applied strain (A) and steady state shear test, Viscosity, η, and Shear Stress, σ, as a function of shear rate (B). Comparison between photoprotective formulations containing (F2) or not (F1) the association of vitamin derivatives. Mean values of triplicates ± SD.

Both SC water content and surface hydration distribution parameters showed significant increase in skin hydration after 28 days, compared to baseline (Fig. 2 and 3). A significant increase in *CornerDensity* parameter, related to primary line density, was observed for F2 on PE skin (Fig. 1E)[5,7]. Indeed, higher *CornerDensity* values have been correlated to younger ages and lower levels of photoexposure[8].

Fig. 2. *Gray Index* (a.u) (A), *Gray Index T* (a.u) (B), *MeanGL* (a.u.) (C) and *MeanGL20* (a.u) (D), *CornerDensity* (a.u.) (E) parameters, and SC water content (a.u.) (F) obtained from the anterior (photoprotected, PP) and posterior (photoexposed, PE) regions of the forearms, before (T0) and after (T28) 28 days of applying formulations containing (F2) or not (F1) the association of vitamin derivatives. * T-test ($p < 0.05$), ** t-test ($p = 0.0235$) and *** t-test ($p = 0.0111$) of normal data (Shapiro Wilk, $p > 0.05$). ● Mann-Whitney test ($p < 0.05$) and ● ● Wilcoxon test ($p < 0.05$) of non-normal data.

Significant reductions ($p < 0.05$) in skin microrelief parameter SER (A), related to roughness, SESC (B), related to scaling, and SEW, related to the number and width of wrinkles (Fig. 4), were also observed.

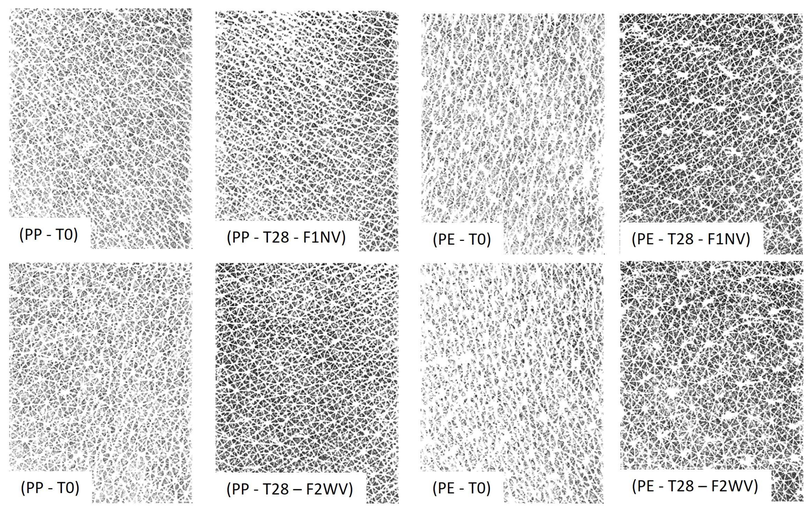

Fig. 3. Representative micro-topography images of surface hydration obtained from the anterior (photoprotected, PP) and posterior (photoexposed, PE) regions of the forearms, before (T0) and after 28 days (T28) of use of photoprotective formulations containing (F2) or not (F1) the association of vitamin derivatives.

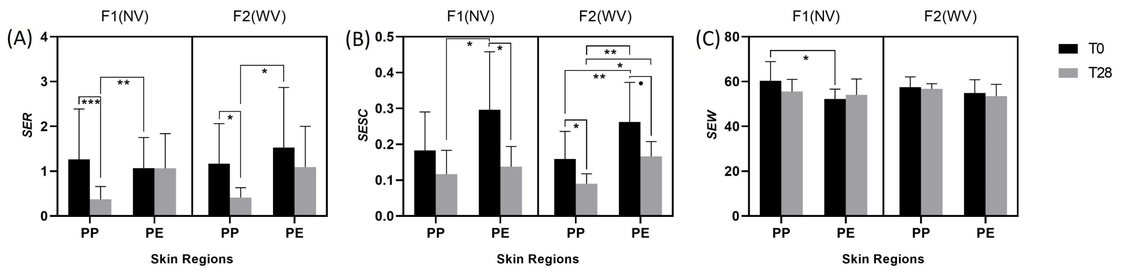

Fig. 4. SESM (A), SER (B), SESC (C) and SEW (D) parameters obtained from the anterior (photoprotected, PP) and posterior (photoexposed, PE) regions of the forearms, before (T0) and after (T28) 28 days of applying the photoprotective formulations containing (F2) or not (F1) the association of vitamins derivatives. * T test, ($p < 0.05$) of normal data (Shapiro Wilk, $p > 0.05$). ** Mann-Whitney test, ($p < 0.05$) and *** Wilcoxon test ($p = 0.0039$) of non-normal data.

Basal conditions of PE skin presented significant lower dermal density, related to photoaging[6]. After a 28-day period, an increase in dermis echogenicity was observed on both PP and PE skin regions (Fig. 5 and 6), with an increase of higher magnitude for F2 on PE skin[9]. No significant changes were observed in dermis thickness and TEWL

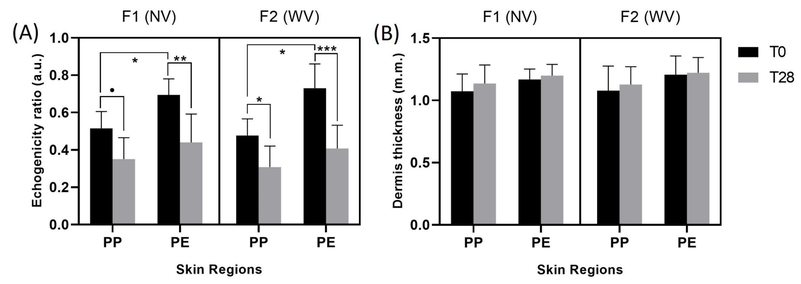

Fig. 5. Echogenicity ratio (dermis subechogenic pixels/dermis hyperechogenic pixels) (A) and dermis thickness (B) obtained from the anterior (photoprotected, PP) and posterior (photoexposed, PE) regions of the forearms, before (T0) and after (T28) 28 days of applying formulations containing (F2) or not (F1) the association of vitamin derivatives. * Test-t ($p < 0.05$), ** test-t ($p = 0.0010$) and *** test-t ($p < 0.0001$) of normal data (Shapiro Wilk, $p > 0.05$). ● Wilcoxon's test ($p < 0.05$) of non-normal data.

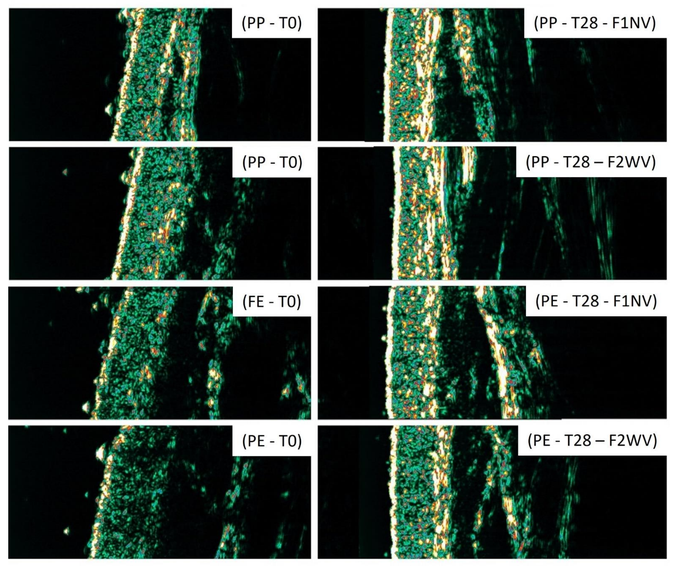

Fig. 6. Representative ultrasound images of a single participant obtained from the anterior (photoprotected, PP) and posterior (photoexposed, PE) regions of the forearms, regions before (T0) and after 28 days (T28) of applying the photoprotective formulations containing (F2) or not (F1) the association of vitamin derivatives.

CONCLUSION

F1 and F2 exhibited a close shear thinning and predominant elastic behaviour with a more sustained elasticity and a slight higher resistance to imposed oscillatory strain for F2. Basal conditions of PE skin showed significantly lower dermal density, yet both F1 and F2 were effective increasing skin hydration and dermis echogenicity on both evaluated regions, with an increase of higher magnitude for F2 on PE skin. F2 also showed improvement in density of primary lines on PE skin. Since the association did not significantly affect formulations' viscoelastic properties, these results suggest that differential clinical effects are likely related to the intrinsic properties of the vitamin derivatives.

REFERENCES

1. Mercurio DG, et al. *J Photochem Photobiol B.* 2015; 153: 121–126. 2. Maia Campos PMBG, et al. *Photochem Photobiol.* 2006; 82: 683-688. 3. WMA. Declaração de Helsinque. 18th WMA General Assembly, Finland, June 1964 and amended by the 64th WMA General Assembly, Brazil, 2013. 4. Brasil. Resolução nº 466, de 12 de dezembro de 2012. 5. Shirata MMF, Maia Campos PMBG. *Photochem Photobiol*. 2021; 97: 805–815. 6. Fitzpatrick TB. *Arch Dermatol*. 1988; 124(6):869–71. 7. Capitani RD, et al. *Biomed Bio-pharm Res*. 2012; 2: 215–224. 8. Bazin R, et al. *Skin Res Tech.* 2010;16: 23–29. 9. Infante VH, et al. *Photochem Photobiol*. 2022.

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