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### Photoprotective properties of Olea Europea by topical application versus UVB and Vitamin E acetate

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SUMMARY: Photoprotective properties of Olea Europea by topical application versus UVB and Vitamin E acetate.

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The aim of the present study was to assess the photoprotective, lenitive and antioxidant properties of Olea Europea (OE) versus UVB and Vitamin E acetate. The Olea Europea is a pool of functional highly specific and differentiated molecules, with antioxidant activities and "scavenger" mechanisms, since oxidative stress linked to the formation of free radicals exerts an important role in the UV-induced skin damage.

21 healthy subjects were studied during a 4-month study. They were subdivided into 3 groups (A, B and C), each formed by 7 patients. They all were irradiated with a source of UVB and evaluated by colorimeter.

In A group patients, following application of Olea Europea, we observed a 22% erythema decrease respect to MED; in B group parients, it was approximately 32%, in C group, it was about 18%. OE presented high efficacy, especially when it was applied both before and after photostimulation; results were more important when OE was combined with pure vitamin E acetate (reaching, respectively, 31%, 60% and 40% erythema decrease).

Olea Europea, when topically applied, present interesting capabilities of repairing UVB photodamage, therefore, OE integrates the peculiar and natural functional characteristics of skin photoprotection. RIASSUNTO: Proprietà fotoprotettive della Olea Europea mediante l'applicazione topica rispetto UVB e Vitamina E acetato.

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Lo scopo del presente studio era di valutare le proprietà foto protettive, lenitive et ant ossid nti della Olea Europea (OE) rispetto a UVB e Vitamina E a etato. L'Olea Europea è un pool di molecole funzionali alta neme specifiche e differenziate, con attività antiossidante e meccanismi "scavenger", dal momento che lo stress ossidativo legato alla formazione di radicali liberi esercita un ruolo importante nel danno cutaneo UV-indotto.

Sono stati studiati 21 soggetti sani durante uno studio di 4 mesi. Essi sono stati suddivisi in 3 gruppi (A, B e C), ciascuno formato da 7 pazienti. Tutti sono stati irradiati con una sorgente di raggi UVB e valutati da colorimetro.

Nei pazienti del gruppo A, a seguito di applicazione di Olea Europea, abbiamo osservato una diminuzione dell'eritema del 22% rispetto a volori della MED; nei pazienti del gruppo B, è stato circa del 32%, nel gruppo C, è stato circa del 18%. OE ha presentato una grande efficacia, soprattutto quando applicata prima e dopo fotostimolazione; i risultati sono stati più importanti quando OE è stato combinata con acetato di vitamina E pura (raggiungendo rispettivamente il 31%, 60% e 40% diminuzione eritema).

L'Olea Europea, quando applicata localmente, ha la capacità di riparare il foto danneggiamento UVB- indotto. Essa, pertanto, integra le peculiari caratteristiche fisiche e funzionali di fotoprotezione della pelle.

KEY WORDS: Antioxidants - Photoprotective properties - UVB. Antiossidanti - Proprietà fotoprotettive - UVB.

### Introduction

The kin has a wide range of antioxidant defence mechanisms, with very complex connections. This pool, however, can be easily destroyed by high doses of ultraviolet radiations (UV) (1). Since oxidative stress linked

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to the formation of free radicals exerts an important role in the UV-induced skin damage, supplementation of natural antioxidant system, with a shielding action of UV radiations and a lenitive capacity, would have a rational for use in photoprotection. The Olea Europea is a pool of highly specific and differentiated functional molecules, some of them increased in unsaturated components and therefore rich of double bonds with UV radiations absorbing activities, bioperoxides with antioxidant activities and "scavenger" effects of ROS and free radicals; then, contained green the polyphenols, soy isoflavones, vitamin C and E.

#### M. Nino e Coll.

The aim of the present study was to assess, on humans, the photoprotective, lenitive and antioxidant properties of Olea Europea versus UVB and Vitamin E acetate (2, 3). We also evaluated the Olea Europea properties when combined with vitamin E acetate.

### Materials and methods

21 healthy subjects were studied: they were 9 males and 11 females, aged between 18 and 52 years (average age: 29.4 years) enrolled at the Section of Dermatology of the University Federico II of Naples, during a 4month period. No patient had a history of photodermatosis or recent sun exposure. Subjects were selectionated as most phototype representatives of the population (phototypes 2 and 3 of Fitzpatrick).

Four preparations were tested:

- Olea Europea;
- Olea Europea not subjected to ozonation process;
- pure Vitamin E acetate;
- Olea Europea combined with pure vitamin E acetate.

For each volunteer, areas of treatment ( $2 \text{ cm}^2$  each area) were located on the medial surface of forearm; samples were applied with a slow 1-minute massage. Before proceeding with the test, each patient was given the MED (minimum erythematous doses) for UVB. The MED is defined as the minimum dose of UVB able to

induce a perceptible erythema with net limits. The measurement of MED was made 24 hours after irradiation with UVB by colorimeter.

The 21 patients were divided into 3 groups (each of 7 patients) and the different preparations, in the three groups, were so used:

- only before photostimulation (Protocol 1), A group;
- before and after photostimulation (Protocol 2), B group;
- only after photoostimulation (Protocol 3), C group.

# **Protocol 1:** application of the samples only before photostimulation.

Patients in the first group (A group) applied the preparations 24 hours before and immediately before photostimulation. This, amounted to MED-UVB individual, was conducted simultaneously on 5 test areas: one area (untreated area) for MED evaluation, the remaining 4 treated respectively with Olea Europea, Olea Europea not subjected to ozonation process, pure vitamin E acetate and Olea Europea combined with pure vitamin E acetate (Table 1).

### Protocol 2: application of the samples

before and after photostimulation.

Patients in the second group (B group) applied the preparations 24 hours before and immediately before pho-



Table 1 - PM®O erythema reduction values respect to MED.



Table 2 - Erythema reduction values when samples are applied only before photostimulation (protocol 1).

tostimulation, such as patients with Protocol 1; then, products were applied again, on these areas, shortly after photostimulation and after a further 1-hour interval (Table 2).

## **Protocol 3:** application of the samples only after photostimulation.

Patients in the third group (C group) applied products (2 applications, one every hour) immediately after photostimulation with UVB on the 4 areas not treated previously (Table 4).

Readings of phototest were made after 24 hours by 3 different operators clinically estimating the 4 irradiated areas:

- a) erythema in the area corresponding to MED (area 1);
- b) the effect produced on the erythema by applying: Olea Europea, Olea Europea not subjected to ozonation process, pure vitamin E acetate and Olea Europea combined with pure vitamin E acetate according to the different ways in the 3 different protocols (areas 2, 3, 4 and 5).

As objective clinical evaluation, on these areas was also performed colorimetric assessment, taking as the index of erythema  $\Delta$  a \*. This was calculated as follows:

a) a \*  $\Delta$  for protocol 1 = a \* only irradiated skin (MED-UVB) - a \* each of the areas of skin treated respectively with Olea Europea, Olea Europea not subjected to ozonation process, pure vitamin E acetate and Olea Europea combined with pure vitamin E acetate, applied only before irradiation;

- b)  $\Delta$  a \* for protocol 2 = a \* only irradiated skin (MED-UVB) a \* each of the areas of skin treated with the same products before and after irradiation respectively;
- c) a \*  $\Delta$  for protocol 3 = a \* only irradiated skin (MED-UVB) a \* each of the areas of skin treated with the same products only after irradiation respectively.

The reduction of the degree of erythema MED UVBinduced, was calculated as follows: the percentage reduction of erythema = ( $\Delta$  a sample x \* / a \* MED-UVB individual) \* 100.

The following equipment assessments were used:

- a) as source of UVB irradiation: the TL12 Lamp Philips equipped with 6 tubes of 40 W, with irradiance of 1.4 mW/cm<sup>2</sup> and spectrum emission between 290 and 320 nm each;
- b) as colorimeter: Spectrocolorimeter type X-Rite 968, measured light reflected in the visible range (400-780 nm) and translated into digital systems, to determine quantitatively the degree of UV-induced erythema. The measurement system used, was called L\* a\* b\*: it is based on a system of three-dimensional Cartesian axes in which the axis L\* identify the variations between white and black, a \* axis between red and green and b \* axis between yellow and blue.





Table 3 - Erythema reduction values when samples are applied before and after photostimulation.

### Results

Results showed that in A group, following application of Olea Europea, there was a 22% decreased incidence of erythema respect to MED; in B group incidence decrease was about 32%; in C Group it was about 18% (Table 1). Tables 2, 3 and 4 show the different results obtained among Olea Europea, Olea Europea not subjected to ozonation process, pure vitamin E acetate and Olea Europea combined with pure vitamin E acetate in the three different protocols.

### Discussion

Free radicals play an important role in UV-induced skin damage.

Recent studies showed that skin supplementation with antioxidants reduces the oxidative stress and subsequent photodamage (4). In vitro studies on human keratinocytes have also documented a protective role of antioxidants in UV-induced immunosuppression (5). Therefore, it can be considered of practical use in photoprotection.

The effectiveness of photoprotection by topical application is confirmed by the demonstration that the keratinocytes of the most differentiated layers have more antioxidant capacity than germinal layers ones. In vitro studies have shown that the antioxidant capacity of keratinocytes increases during the process of cell differentiation (6).

Furthermore, the antioxidants are at significantly higher concentrations in epidermis than the dermis, according with the fact that the human epidermis is the first barrier against oxidative damage.

The colorimetric measures obtained confirms that Olea Europea, when topically applied, has high preventive and repairing properties against UVB photodamage through synergies of action of its active ingredients. Nevertheless, Olea Europea presented more efficacy respect to pure vitamin E acetate, especially when it was applied both before and after photostimulation: it was, infact, found a rate of 32% erythema decrease compared to MED, most important of pure vitamin E acetate (20% decrease). We further observed that results were more important when Olea Europea was combined with pure vitamin E acetate (reaching, respectively, 31%, 60% and 40% erythema decrease).

The vehiculation of ozone in a complex matrix, as that of Olea Europea, induces the formation of highly enriched of "unsaturated components" small molecules with absorbing of harmful UV radiations with ROS and free radicals "scavenger" action as enhancement of the antioxidant action.

Olea Europea, when topically applied, can be able to prevent and repair UV-induced skin photodamage because integrates and enhances the natural and functio-



Table 4 - Erythema reduction values when samples are applied only after photostimulation.

nal characteristics of skin photoprotection. It can therefore be used both for UVB prevention and post-irradiation.

### Conclusions

Olea Europea, when topically applied, presents in-

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teresting capabilities of repairing UVB photodamage. These abilities are greater when it was applied before and after photostimulation, confirming its photoprotective and lenitive properties. The results are further enhanced when using Olea Europea combined with pure vitamin E acetate. The topical application of Olea Europea can, therefore, integrate the peculiar and natural functional characteristics of skin photoprotection.

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