A NEW *IN VITRO* METHOD TO PREDICT *IN VIVO* PHOTOPROTECTION AGAINST VISIBLE LIGHT-INDUCED PIGMENTATION AND A NEW VISIBLE LIGHT PHOTOPROTECTION FACTOR

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INTRODUCTION

Ultraviolet radiation is the main cause of skin pigmentation, but more recently visible light (VL) (400-700 nm) has been shown to be an important contributor especially in melano-competent subjects. To evaluate sunscreen efficacy on VL photoprotection, an assessment method has recently been proposed based on *in vivo* pigmentation, leading to the calculation of the visible light photoprotection factor (VL-PF). However, even if *in vivo* methods remain the most representative of real life, *in vitro* methods are more suited to screening sunscreen formulations. The aim of this study was to evaluate the correlation between an *in vivo* and an *in vitro* method in assessing protection against VL-induced pigmentation.

MATERIALS & METHODS

First the *in vitro* protective properties of 10 sunscreens with very high photoprotection (≥ SPF50+) were analyzed using transmission measurements in the VL spectrum. Then, a monocentric, double-blind, randomized controlled study with intra-individual comparisons in 20 healthy subjects was performed to measure the VL-PF *in vivo* of those sunscreens. This VL-PF was reinterpreted as a percentage using the formula (1-(1/VL-PF))×100 and named the pVL-PF (0% corresponds to an untreated exposed area and 100% corresponds to theoretical complete protection against the VL, equivalent to an unexposed area). The correlation between the pVL-PF and the percentage of blocked light was evaluated using the coefficient of determination R, for each test area, for each wavelength from 400 to 700 nm, and for every wavelength range. The statistical analysis was performed using the Pearson correlation.

RESULTS

➢ *In vitro*, the transmission spectra of the tinted sunscreens showed that the best protection was obtained in the high energy VL (**Fig. 1**).

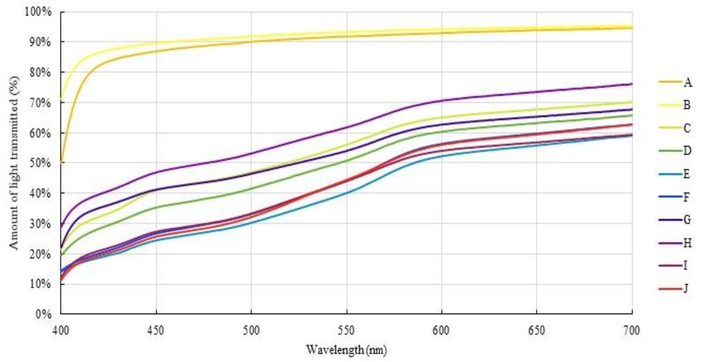

Figure 1: *Transmission spectra of the ten tested sunscreens (A to J)*

➢ *In vivo*, twenty subjects with a mean age of 34.9 years (range: 18-49 years) were included in the *in vivo* study. The pVL-PF obtained ranged from 9.7% (for the non-tinted sunscreen) to 66.4% (for one of the tinted sunscreens) (**Fig. 2**).

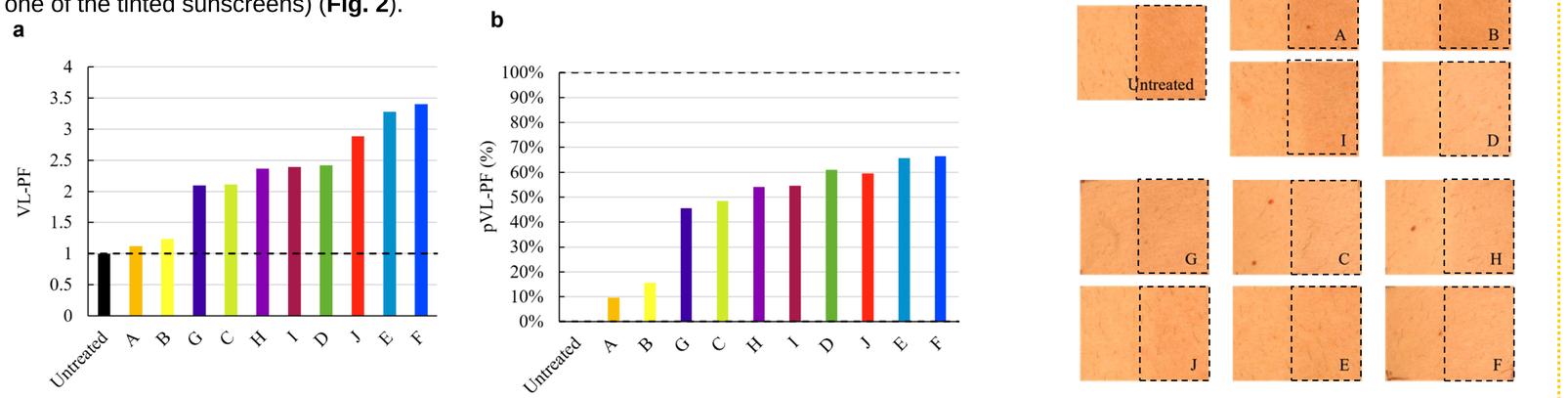

Figure 2: *In vivo evaluation of the protection against VL-induced pigmentation of ten tested sunscreens (A to J). VL-PF (**a**) pVL-PF=(1-(1/VL-PF))×100 (**b**) and photos (**c**)*

➢ A highly significant correlation was demonstrated between *in vivo* VL protection factor and *in vitro* transmittance measurements (**Fig. 3**). The correlations were the highest at 420 nm (R^2 = 0.9910) for separate wavelengths (**Fig. 3a**), and between 400 and 469 nm (R2 = 0.9904) for wavelength ranges (**Fig. 3b**).

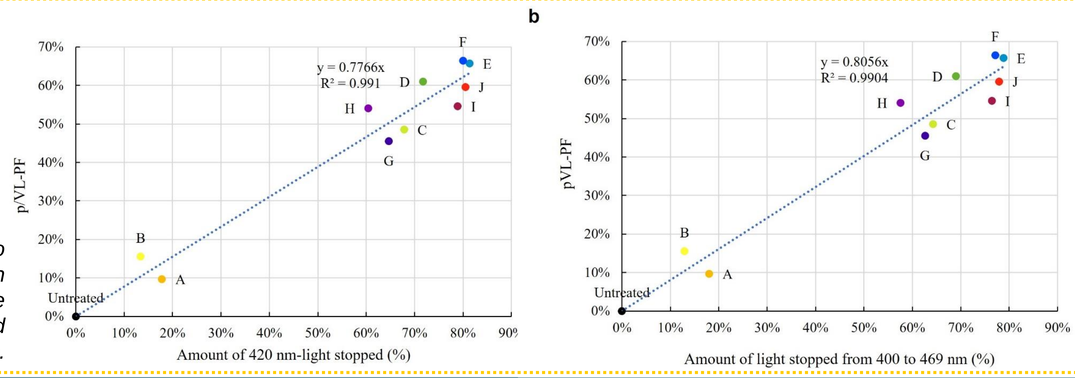

Figure 3: *Correlation between the in vivo protection against VL-induced pigmentation and the amount of VL blocked in vitro by the ten tested sunscreens at 420 nm (**a**) and between 400 and 469 nm (**b**).*

➢ The results indicated a strong linear relationship between *in vivo* pVL-PF measurement and the percentage of VL blocked *in vitro*.

DISCUSSION

The pVL-PF is a new interpretation of the original VL-PF to compare more intuitively from 0% to 100% the performance of different formulations on VL-induced pigmentation. It makes it also easier to understand for dermatologists and consumers who are looking for high VL photoprotection. Interestingly, the best correlation between *in vivo* pigmentation and the *in vitro* transmittance was observed from 400 to 469 nm, which corresponds to the absorption spectrum of opsin-3. Indeed, melanocytes directly sense blue light through direct stimulation of the opsin-3 receptor. In conclusion, the *in vitro* method using transmittance measurement from 400 to 469 nm is a good predictive tool to evaluate sunscreen VL photoprotection efficacy and could be used to select formulations before final *in vivo* evaluation.

EADV 2022, Milan