



Noninvasive *In-Vivo* Determination of Sunscreen-UVA Protection Factors

Introduction

The development and evaluation of UVA (320–400 nm) sunscreens is important because UVA sunlight can penetrate deep into human skin and cause severe internal damage, as well as erythema and photoaging. Human phototesting is usually used for determining the sunscreen protection factor (SPF), defined as the ability of a sunscreen to prevent a threshold erythema from solar radiation. The SPF definition is now considered as the tentative final FDA standard for measuring SPF. The SPF measured with this method, however, results primarily from UVB (280–320 nm) and not from UVA. Phototesting is unsuitable for the evaluation of UVA protection factors (PFA) because of the extremely long exposure time necessary to generate erythema from UVA radiation and the concerns of reciprocity. Recently, several *in vivo* and *in vitro* methods using animals have been proposed. However, none of the methods is capable of measuring the efficacy of UVA sunscreens *in vivo* on human skin.

A HORIBA Jobin Yvon spectrofluorometer with double grating monochromators for both excitation and emission can be used for noninvasive *in-vivo* determination of sunscreen PFA via diffuse-reflectance spectroscopy. Because of the extremely low level of stray light that these monochromators allow, our spectrofluorometers can measure highly scattering samples, such as human skin, with excellent sensitivity and precision. Diffuse-reflectance (DR) spectra of human skin both with sunscreen and without can be directly

measured *in vivo* and differentiated within minutes. The ratio of spectra with and without sunscreen results in transmittance spectra of sunscreens, which allows calculation of PFAs. This noninvasive method provides an alternative diagnostic tool for quick and accurate determination of PFA of sunscreen products.

Experimental method

DR spectra with and without sunscreen were measured by synchronously scanning both excitation and emission monochromators at the same wavelength. Light from a xenon lamp was passed through a double-grating monochromator and then was focused into one arm of a randomized, bifurcated fiber-optic bundle. This light then was transmitted to the common end of the bundle to irradiate the skin. Backscattered light was collected by the emission bundle, passed through another double-grating monochromator, and sent to a photomultiplier-tube detector.

A 5-cm × 5-cm area on the arm was initially marked and checked under a Wood's lamp (UV light). DR spectra without the sunscreen product were measured on at least four different spots. The same area was then applied with 50 mg of product (2 mg cm⁻²) and viewed under a Wood's lamp for uniformity of coverage. After air-drying for 20 minutes, the DR spectra were measured again at the same spots. The average of four spectra for each measurement was used to calculate PFAs. The active ingredients in the sunscreens were oxybenzone and Parsol®.

Results and discussion

PFA value of a sunscreen product is defined in the following equation¹:

$$PFA = \frac{\sum_{320 \text{ nm}}^{400 \text{ nm}} S(\lambda)A(\lambda)}{\sum_{320 \text{ nm}}^{400 \text{ nm}} S(\lambda)A(\lambda)T(\lambda)}$$

where $S(\lambda)$ is the source spectrum², $A(\lambda)$ is the reference action spectrum³ for UV-induced erythema in human skin, and $T(\lambda)$ is the calculated transmittance spectrum of the sunscreen product obtained on human *in vivo*. $T(\lambda)$ is

$$T(\lambda) = \sqrt{\frac{I_{\text{with}}(\lambda)}{I_{\text{without}}(\lambda)}}$$

where $I_{\text{with}}(\lambda)$ and $I_{\text{without}}(\lambda)$ are the re-emitted intensities at wavelength λ from the skin, with and without sunscreen, respectively. Software was specially developed to calculate the PFAs from these parameters.

The typical transmittance spectra of sunscreen products are shown in Fig. 1. Table 1 summarizes PFAs calculated for three sunscreen products¹, and also lists PFAs reported by the manufacturers via the phototesting method.³ Calculated PFAs using the DR method show

good agreement with the stated values for all three products. Discrepancies at the higher PFA arise from the inverse relation of the measured transmittance to the calculated SPF value. The accuracy of PFAs can be improved for high-PFA products by using thinner films and correcting for the standard film thickness.⁴

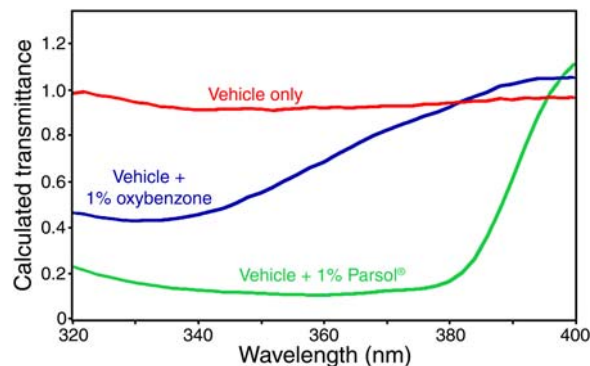


Fig. 1. Sunscreen transmittance spectra for human skin with vehicle, but no active ingredient, with 1% oxybenzone, and with 1% Parsol®.

Sunscreen	PFA (by DR)	PFA (by manufacturer)
Shade 15	3.1	2.9
Shade 45	4.5	5.3
Photoplex	6.5	8.3

Table 1. PFAs for several sunscreens measured by DR compared to values reported by the manufacturers.

Parsol® is a registered trademark of Givaudan.

¹ Kollias, N., Gillies, R., and Anderson, R.R. "The non-invasive determination of UVA sunscreen effectiveness *in vivo*". In *Biological Responses to UVA Radiation*; Urback, F., ed., Valdenmar Publishing: Overland Park, KS, 1992, p. 371.

² Sayre, R.M. and Agin, P.P. *J. Am. Acad. Dermatol.* **1984**, *23*, 429.

³ McKinlay, A.F. and Diffey, B. L. *CIE Journal* **1987**, *6*, 17.

⁴ Gillies, R. and Kollias, N. "Noninvasive *In Vivo* Determination of Sunscreen Ultraviolet A Protection Factors Using Diffuse Reflectance Spectroscopy". In *Sunscreens: Development, Evaluation, and Regulatory Aspects*, N.J. Lowe, N.A. Shaath, and M.A. Pathak, eds., New York, Marcel Dekker; Vol. 10, 1997, pp. 601–610.