

Influence of Applied Quantity of Sunscreen Products on the Sun Protection Factor (SPF) – A Multi-Center Study organised by the DGK*Task Force Sun Protection

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Keywords

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Abstract:

It is often debated that the protection against solar induced erythema under real conditions is dependent upon the amount of sunscreen applied. When too little is applied it is believed that a lower sun protection than labelled will result. It was the aim of this study to quantify this effect. In this multi-center study the influence of three different amounts (0.5; 1.0; 2.0 mg/cm²) of three commercial sunscreen products in three reliable test centers following the test protocol of "The International Sun Protection Factor (SPF) Test Method" (IM 2003) has been investigated. The main result was a linear dependence of the SPF on the quantity applied. Taking into consideration the volunteer specific variations an exponential dependence of confidence interval of the *in vivo* SPF and amount applied is found. The highest amount applied (2.00 mg/cm²) is linked to the lowest confidence intervals. Thus, from the point of view of producing reliable and reproducible *in vivo* results under laboratory conditions the recommendation of this multi-center study is an application quantity of 2.0 mg/cm².

Introduction

Overexposure to sunlight, particularly to UV-rays, has harmful effects on human skin. Sunscreens have become widely used for the prevention of short- and long-time skin damage especially cutaneous erythema and premature skin ageing. The "Sun Protection Factor (SPF)" of a sunscreen product is widely recognized by the general public as a measure for the protection offered by a sunscreen preparation against sunburn.

The first known studies regarding the SPF have been published in the 1940's by H. Blum et al. [1] and in the 1950's by R. Schulze [2] and led to the definition of the concept of minimal erythema dose (MED) and SPF. The first formalised method for SPF determination and labelling was issued by the FDA in the USA in 1978 [3], followed in 1984 by the DIN 67501 Norm in Germany [4], which was applied mainly in Europe. These two standards differed in numerous ways, an important one being the amount of product applied to the skin (2.0 and 1.5 mg/cm² respectively), which lead to some discrepancies in the sun protection factors measured. All subsequently issued standards retained the application amount of 2.0 mg/cm². The European Cosmetic, Toiletry and Perfumery Association (COLIPA), in its 1994 SPF test method [5] introduced new techniques to characterise and specify the emission spectrum of the UV source and to colorimetrically select skin types. The Austrian ÖNORM [6] in 1998 and the new DIN standard [7] of 1999 were aligned to the COLIPA 1994 Method. In 2000, COLIPA, JCIA and ctfa-SA agreed on the harmonisation of the SPF measurement method. A joint agreement on the International SPF Test Method (IM 2003) was published in 2003 [8].

It is evident that all generally accepted methods used to determine the SPF of a sunscreen require that the product be applied in a thickness of 2.0 mg/cm² in order to get reproducible results. However under real conditions sun bathers tend to apply less than 2.0 mg/cm² of sunscreen product on exposed skin in an even layer.

Several studies have shown that consumers apply much less than this amount, typically between 0.5 and 1.5 mg/cm². Bech-Thomsen and H.C.Wulf [9] reported that the application of sunscreen by women, men and children was an average 0.5 mg/cm². Farr et al calculated the influence of the film thickness on the SPF on a theoretical basis, assuming the applicability of Beer-Lambert law. A 50% reduction of the layer thickness would thus reduce the SPF by the square root, e.g. SPF 25 to SPF 5 [10] However in a study with *in-vitro* and *in-vivo* experiments deviations from Beer-Lambert law have been reported [11]. In a previous study on excised human epidermis with different quantities R. Stokes and B. Diffey found that most users probably achieved a mean SPF of between 20-50% of that expected from the labelled SPF of the product [12].

C. Stenberg and O. Larkö [13] examined the SPF *in-vivo* with individuals who were asked to apply commercial sunscreens *ad libitum*. Under these conditions the SPF was only approximately 50% of the declared SPF.

It is unanimously accepted, that the SPF under real conditions is lower than the declared one, but there are discussions about its extent and the risk for consumers who are less protected than they expect from the labelled SPF.

This is the first multi-center study to investigate the influence of 3 different amounts (0.50; 1.00; 2.00 mg/cm²) of 3 commercial sunscreen products in 3 reliable test centers following the test protocol of “The International Sun Protection Factor (SPF) Test Method” (IM 2003) [8].

Materials and methods

Test Materials:

Three commercial sunscreen products representing a variety of UV-filter combinations and a variety of cosmetic formulations (spray, w/o-emulsion and o/w-emulsion) were chosen. In order to reduce a possible influence of product application variability products “easy” to apply were selected. The labelled SPF's ranged from 20 to 25.

Product Code “A”:

Commercial European brand sun care lotion with labelled SPF 20; o/w-emulsion with the following UV-filter combination:

- 4-Methylbenzylidene Camphor
- Polysilicone-15
- Butyl Methoxydibenzoylmethane
- Sodium Phenylbenzimidazole Sulfonate

Product Code “B”:

Commercial European brand sun care spray with labelled SPF 20; hydrodispersion with the following UV-filter combination:

- Octocrylene
- Butyl Methoxydibenzoylmethane
- Titanium Dioxide
- Drometrizole Trisiloxane
- Ethylhexyl Triazone
- Terephthalylidene Dicamphor Sulfonic Acid

Product Code “C”:

Commercial European brand sun care lotion with labelled SPF 25; w/o-emulsion with the following UV-Filter combination:

- Ethylhexyl Triazone
- Titanium Dioxide
- Bis-Ethylhexyloxyphenol Methoxyphenyl Triazine

Methods:

In a multi-center study three test centers assessed the influence of 3 different amounts (0.50; 1.00; 2.00 mg/cm²) of the 3 sunscreen products described above on the SPF following the test protocol of “The International Sun Protection Factor (SPF) Test Method” (IM 2003) [8]. The test centers were located in Germany (Witten/Herdecke [TC III], Holzminden [TC II] and Schenefeld / Hamburg [TC I]). All practical testing was performed from October 2004 to December 2004.

In each test center three groups of at least 10 volunteers were recruited following the inclusion and exclusion criteria of the “IM 2003” [8]. The three application amounts of each test product were tested in the same group of volunteers to enable intra-individual comparison of data. All SPF readings were blind. All test centers used test materials of the same batch. Table 1 shows the assignment of test products to the three different test groups and the coding of the test products. The application amounts are given in the right column of Table 1.

Table 1: Assignment of test products to the groups of volunteers

Test Group	Code	Product	Application-Amount
Group 1	P3	COLIPA High Standard P3 SPF 14-17	2.00 mg/cm ²
	A	o/w-emulsion SPF 20-30	2.00 mg/cm ²
	A	o/w-emulsion SPF 20-30	1.00 mg/cm ²
	A	o/w-emulsion SPF 20-30	0.50 mg/cm ²
Group 2	P3	COLIPA High Standard P3 SPF 14-17	2.00 mg/cm ²
	C	w/o-emulsion SPF 20-30	2.00 mg/cm ²
	C	w/o-emulsion SPF 20-30	1.00 mg/cm ²
	C	w/o-emulsion SPF 20-30	0.50 mg/cm ²
Group 3	P3	COLIPA High Standard P3 SPF 14-17	2.00 mg/cm ²
	B	hydrodispersion SPF 20-30	2.00 mg/cm ²
	B	hydrodispersion SPF 20-30	1.00 mg/cm ²
	B	hydrodispersion SPF 20-30	0.50 mg/cm ²

In each group of volunteers the COLIPA Standard P3, SPF range 14 to 17 [8] was included with an application amount of 2.0 mg/cm².

The three test panels per test center were recruited from the general population of the neighbouring community of the study centers. Phototypes I, II, and III were included in accordance with the approximated distribution in the population of Central Europe and in all three test panels in a comparable frequency. Panellists were included on the basis of an informed consent after checking all inclusion and exclusion criteria of “IM 2003” [8].

Test Procedure

Selection of the volunteers, application of test products, irradiation and readings were in accordance with the “IM 2003” in all details except for the application amount. At each test center, the solar simulator spectrum was proven to be in the range of the method as described in “IM 2003”, Appendix III [8].

In a pre-test the minimal erythral dose of unprotected skin (MED_u) was detected for all volunteers by irradiation of 6 spots with an increment of 25 % (day 1). To estimate correct irradiation times for the MED-determination colorimetric measurements of the skin were carried out by using the $L^*a^*b^*$ colour space in accordance with “IM 2003”, Appendix IV. After reading of the MED_u on day 2, product applications and irradiations were performed. The “IM 2003” requires a geometric progression of irradiation doses of 1.12 for SPF of >25. Since all products had expected protection factors of about SPF 25, all products and all doses were investigated by use of the progression 1.12. Visual readings were performed 16 – 24 hours after irradiation on day 3.

Analysis of Data

The analysis of data was performed in accordance with the IM Appendix VI. Mean values for the SPF for a certain product were calculated in each test center(i) according to equation (1)

$$\overline{SPF}_i = \frac{\sum_{k=1}^{n_i} SPF_k}{n_i} \quad (1),$$

Where SPF_k is the SPF value measured with subject k, and n_i is the number of subjects for the product in test center i.

Confidence intervals were calculated according to equation (2)

$$CI_{95}(i) = t \cdot \frac{s_i}{\sqrt{n_i}} \quad (2),$$

Where

$CI_{95}(i)$ = confidence interval for the mean based on 95% significance level,

t = t-value from paired t-test,

s_i = standard deviation in test center i,

n_i = number of subjects in test center i,

Evaluations of the overall arithmetical mean SPF values for all three test centers for a certain product and application amount were performed according to equation (3):

$$\overline{SPF} = \frac{\sum_{i=1}^m n_i \cdot \overline{SPF}_i}{\sum_{i=1}^m n_i} \quad (3)$$

where n_i are the numbers of subjects in the respective test center leading to the mean values of the SPF in that test center, and m is the number of test centers,.

The standard deviations s of the overall means SPF values for a certain product and application amount of all three testing facilities were calculated using equation (4):

$$s = \sqrt{\frac{1}{N-1} \cdot \left[\sum_{i=1}^m (n_i - 1) \cdot s_i^2 + \sum_{i=1}^m n_i \cdot (\overline{SPF}_i - \overline{SPF})^2 \right]} \quad (4)$$

Where N is the total number of subjects from all test centers for a certain product. The respective confidence intervals were calculated according to equation (5):

$$CI_{95} = t \cdot \frac{s}{\sqrt{N}} \quad (5)$$

Results and Discussion

The average SPF data and the statistical parameters obtained in the three test centers are listed in Table 2.

Table 2: Summary of results

Application amount (mg/cm ²)		TC I			TC II			TC III			All test centers			
		2.0	1.0	0.5	2.0	1.0	0.5	2.0	1.0	0.5		2.0	1.0	0.5
Product A	n _i	11	13	11	12	12	12	10	10	10	N	33	35	33
	SPF	26.3	9.5	5.1	20.3	8.2	5.1	22.1	11.6	8.0		22.8	9.7	6.0
	St. dev (s _i)	5.5	1.8	0.8	3.3	1.5	0.8	1.4	1.0	0.8	(s)	4.5	2.0	1.6
	t	2.23	2.18	2.23	2.20	2.20	2.20	2.26	2.26	2.26		2.04	2.03	2.04
	CI ₉₅ (i)	3.7	1.1	0.5	2.1	1.0	0.5	1.0	0.7	0.6	CI ₉₅	1.6	0.7	0.6
	CI ₉₅ (i), %	14.0	11.2	10.0	10.5	11.2	10.0	4.4	5.8	7.0	CI ₉₅ , %	7.0	7.1	9.3
Product B	n _i	10	14	11	12	12	12	10	10	10	N	32	36	33
	SPF	24.0	10.1	4.4	26.9	13.8	8.6	21.1	11.5	7.8		24.2	11.7	6.9
	St. dev (s _i)	4.2	2.9	0.7	5.6	3.0	1.7	1.2	1.2	0.8	(s)	4.8	3.0	2.2
	t	2.26	2.16	2.23	2.20	2.20	2.20	2.26	2.26	2.26		2.04	2.03	2.04
	CI ₉₅ (i)	3.0	1.7	0.5	3.6	1.9	1.1	0.9	0.9	0.6	CI ₉₅	1.7	1.0	0.8
	CI ₉₅ (i), %	12.5	16.6	11.4	13.3	13.8	12.7	4.1	7.7	7.4	CI ₉₅ , %	7.1	8.6	11.2
Product C	n _i	11	10	11	12	12	12	10	10	10	N	33	32	33
	SPF	30.3	19.7	7.0	34.6	18.9	12.4	26.5	17.0	10.9		30.7	18.5	10.1
	St. dev (s _i)	7.0	2.3	1.5	7.2	3.8	2.7	1.3	1.6	1.2	(s)	6.7	2.9	3.0
	t	2.23	2.26	2.23	2.20	2.20	2.20	2.26	2.26	2.26		2.04	2.04	2.04
	CI ₉₅ (i)	4.7	4.6	1.0	4.6	2.4	1.7	0.9	1.1	0.8	CI ₉₅	2.4	1.0	1.1
	CI ₉₅ (i), %	15.5	8.3	14.1	13.2	12.6	13.7	3.4	6.6	7.6	CI ₉₅ , %	7.8	5.6	10.5
P3 Standard	Appl. am. (mg/cm ²)	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0		2.0		
	n _i	11	11	11	12	12	12	10	10	10	N	99		
	SPF	15.9	15.2	14.9	15.6	15.5	15.5	13.9	14.3	14.4		15.1		
	St. dev (s _i)	2.4	2.8	2.4	3.7	2.3	3.5	1.0	1.3	1.0	(s)	2.5		
	t	2.23	2.23	2.23	2.20	2.20	2.20	2.26	2.26	2.26		1.99		
	CI ₉₅ (i)	1.6	1.9	1.6	2.3	1.5	2.2	0.7	0.9	0.7	CI ₉₅	0.5		
	CI ₉₅ (i), %	10.1	12.3	10.8	14.9	9.5	14.3	5.2	6.6	5.2	CI ₉₅ , %	3.3		

n_i = number of subjects in test site i,
 N = overall number of subjects in all test sites,
 SPF = average of the individual values (SPF_k) referring to the n_i or N,
 St. dev = standard deviation of the respective mean SPF value,
 t = t-value from paired t-test,
 CI₉₅ (i), CI₉₅ = confidence intervals calculated from equation (2) or (5), respectively

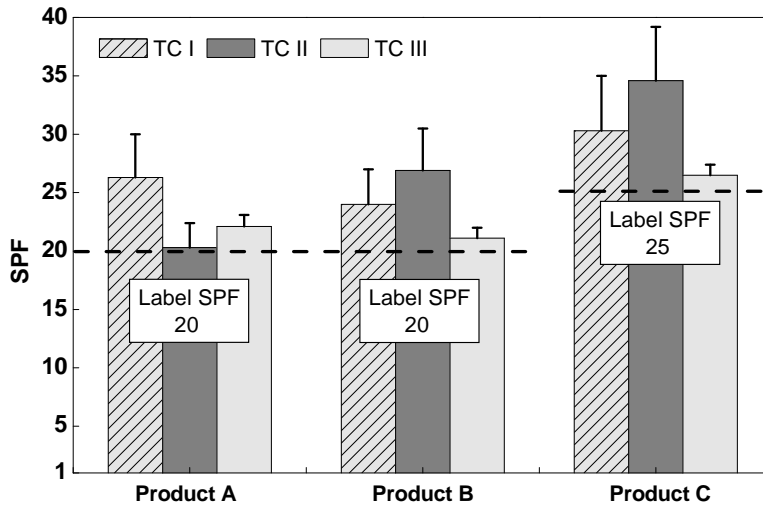


Figure 1 shows the SPFs of the three products determined with an application amount of 2.0 mg/cm², according to the “IM 2003”.

Figure 1: SPFs of products A, B and C determined in three test centers at an application amount of 2.00 mg/cm²; experimental errors indicated as confidence intervals; labelled SPF values are drawn as horizontal lines (labelled SPF = 20 for products A and B, and 25 for product C)

All test centers determined for all 3 sunscreens SPFs slightly higher than the labelled SPFs. In accordance with the IM 2003 confidence intervals of these measurements were all less than 17% in any single measurement.

Following the idea of this multi-center study further investigations of sample A, B and C at reduced product application amounts were carried out. As described in Table 1 50% and 25% of the generally applied amount in the “IM 2003” (2.00 mg/cm²), thus 1.00 mg/cm² and 0.5 mg/cm² were used for *in vivo* SPF testing.

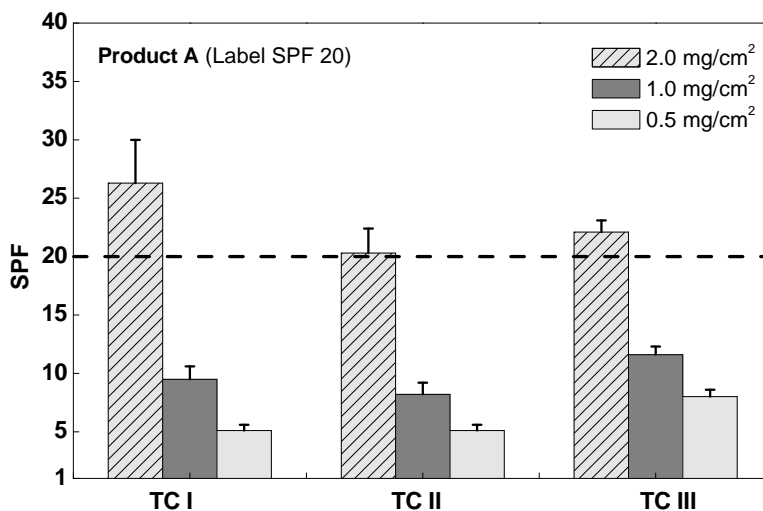


Figure 2: SPF values of product A with different application amounts measured in three test centers; (TC I, TC II and TC III); statistical experimental errors indicated as confidence intervals.

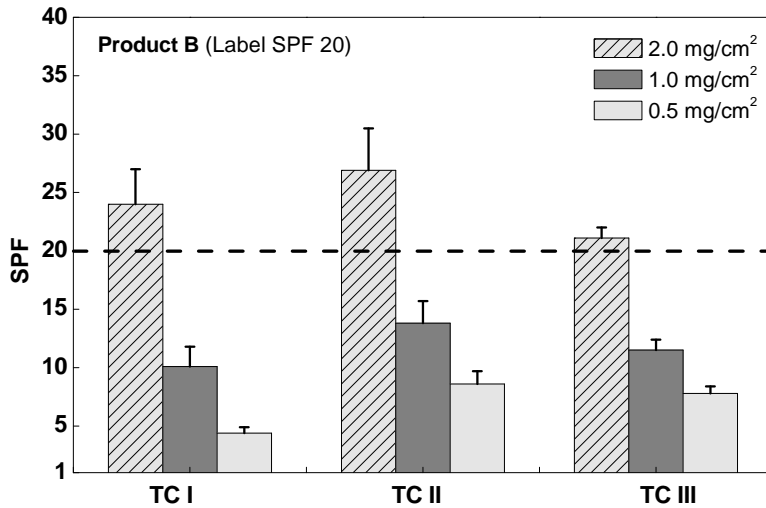


Figure 3: SPF values of product B with different application amounts measured in three test centers, (TC I, TC II and TC III); statistical experimental errors indicated as confidence intervals.

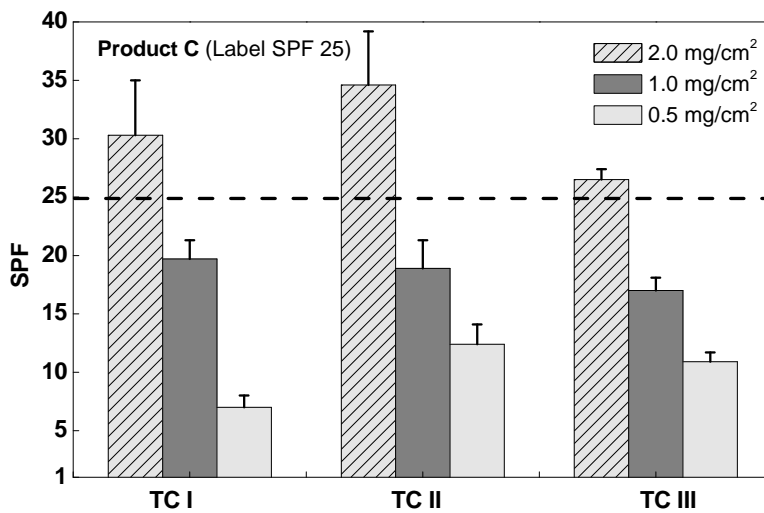


Figure 4: SPF values of product C with different application amounts measured in three test centers, (TC I, TC II and TC III); statistical errors indicated as confidence intervals.

Figures 2 to 4 clearly show an expected decrease of the protection factor, when using smaller amounts of sunscreen products. In order to investigate this decrease of protection mean values per product were calculated. Figure 5a-c presents these mean SPF values for product A, B, and C averaged from the results of all three laboratories as a function of application amount

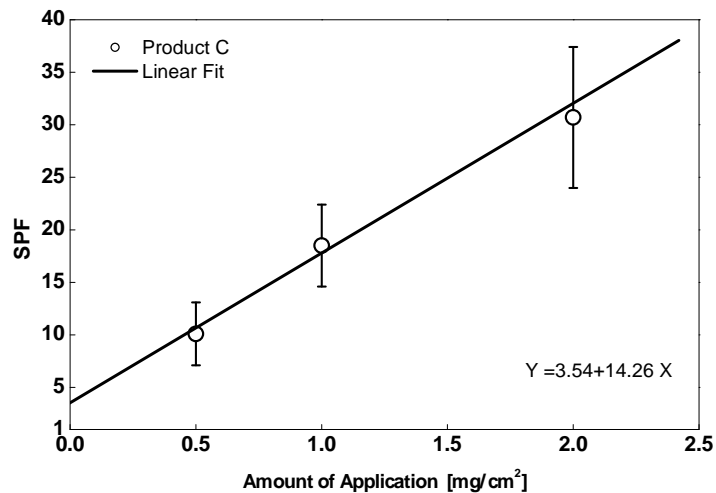
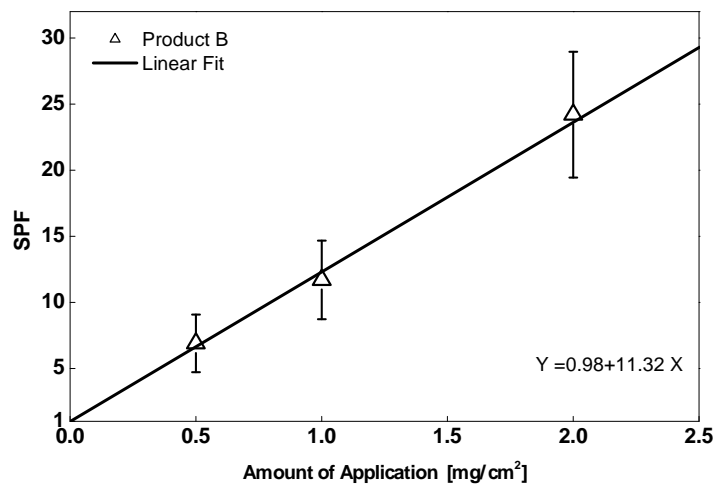
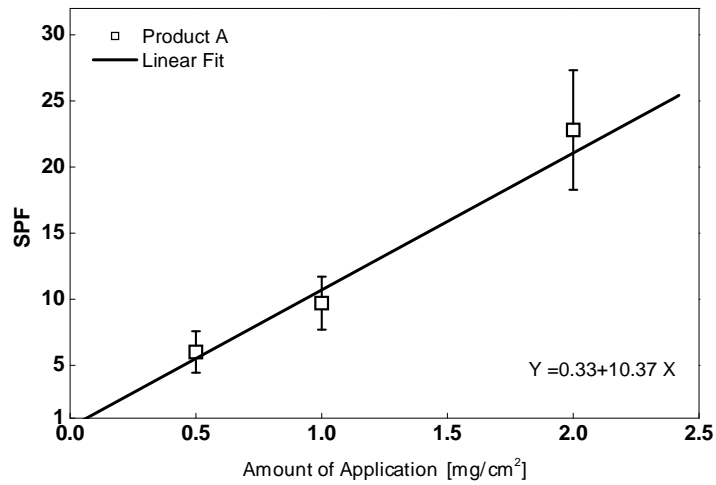


Figure 5a-c: SPF values of product A, B, and C averaged from the results of all three test centers as function of application amount; statistical errors indicated as confidence intervals; mathematical correlation calculated on the basis of a linear regression ($Y=b+mx$, individual parameter given in the graph).

At a first glance, the dependence of the SPF and application amount appears to be linear for all three products. For a closer examination, linear regression lines were calculated, which are shown with the respective experimental data in Figure 5a, 5b and 5c. For all different types of product a correlation coefficient of $R > 0.98$ is calculated (Product A: $R=0.981$, Product B: $R=0.997$ and Product C: $R=0.995$). In all cases the mathematical model of a linear correlation is within the confidence intervals of the measured individual *in vivo* SPF values.

With a linear relationship of the SPF on application amount one would expect an intercept of the regression on the SPF axis of 1.0. This is perfectly given for product B (intercept = 0.98). For products A and C the intercept is slightly different from 1. While for product A a value of 0.33 is calculated which might be accepted as very close to 1 for product C an intercept of 3.54 results from the linear model.

Product A and B both classical types of sunscreen emulsions (product A: W/O and product B: O/W) fit excellently to the linear model while product C as hydrodispersion seems to follow a saturation process with higher values at lower amount of application than expected by a linear model. From a galenic point of view a better spreadability of a more fluid sunscreen might be the reason of this non-linearity. Nevertheless taking into account the high correlation coefficient of all three test products a linear model of the correlation of SPF and amount of product applied seems to satisfy a wide range of different product types quite excellent.

Each of the generally accepted SPF methods requires the product to be applied to a thickness of 2.00 mg/cm^2 . Compared to consumer practice 2.00 mg/cm^2 seems to be a relatively large amount. Thus, justification cannot be consumer daily usage, but justification is clearly driven by the aim of producing reliable and reproducible results in an *in vivo* laboratory SPF test. An experimental proof of this assumption has not yet been provided, but is now possible on the basis of the experimental data of the present study.

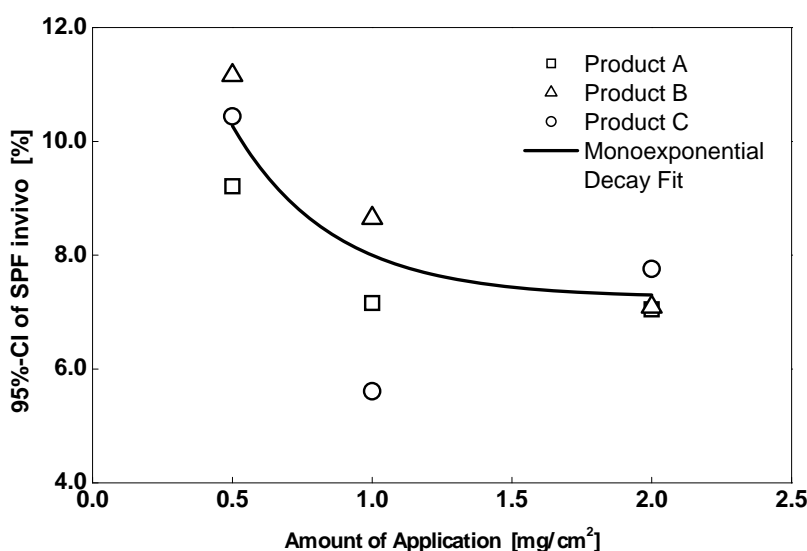


Figure 6: Standard deviation as function of application amount; correlation function calculated on the basis of a mono exponential decay fit

Staying in the way of presenting statistical criterions of the IM 2003 the impact of the application amount on the 95%-CI of the *in vivo* SPF is shown in Figure 6. The 95%-CI's shown are based on the complete data sets of all testing laboratories as calculated with equation [5]. There is a clear tendency of an increasing confidence interval with smaller amounts of applied sunscreen. A correlation is calculated on the basis of a mono-exponential model as shown in Figure 6. Following this correlation the most reliable results were obtained at an application rate of 2.00 mg/cm². Taking into account that products were chosen that present beside a wide range of UV-filters, products which were so called "easy" to apply the correlation calculated and shown in Figure 6 is a clear and powerful argument of using 2.00 mg/cm² as a standard amount of product applied in an *in vivo* laboratory SPF test.

In this study a correlation (close to linearity) of sun protection factor and amount applied was obtained. This, linear dependence found in the *in vivo* investigations demonstrates that the Beer-Lambert law is not applicable for the *in vivo* situation thus a clear difference of *in vivo* and *in vitro* is demonstrated (an *in vitro* study with the same sunscreen products A, B, and C is in preparation in order to investigate the UVA-Balance and further *in vitro* parameter at different amounts of product applied on a PMMA plate [14]). It has been shown that the extinction of an inhomogeneous film depends in a non-linear way on that of the homogeneous "parent film" and is always smaller [15]. By changing the product amount a so called mechanical parameter of the SPF testing procedure is changed. The characteristic of the absorption spectrum will not change at different amounts of product applied. Having this in mind a reasonable explanation of the obtained linearity of SPF and product amount might be an effective absorbing product thickness that might be not linear linked to the amount of product applied by weight. Obviously amount of product is bound to a defined film thickness of the sunscreen on the skin, or may be a defined saturation of the *stratum corneum* with the absorbing filters. Nevertheless the skin surface is no plane surface. It is a surface with heights and valleys. An induced erythema reaction is beside the overlay of absorption spectrum of the used sunscreens and the erythema effectiveness curve as well a result of a homogeneous film of product protecting the skin. It is conceivable that "more" product is applied in the valleys than on the heights. Changing the amount of product as investigated might be different for a valley or a height. Heights might be covered as before while less product is left for the valley on a relative comparison when product quantity is reduced. By this the expected exponential link of amount of product and SPF might be compensated and a linear correlation as experimentally found is being obtained.

Summary

Consumers who are using sunscreen products need an orientation which protection they can expect from the product they bought. The SPF is such an orientation factor, though it is discussed, that the protection under real outdoor conditions may differ to the labelled protection.

A holistic approach was chosen to generate answers concerning the question: "How much protection is preserved if the consumer applies less than 2.0 mg/cm² sunscreen under real conditions?" To answer this question the influence of three different amounts of product applied (0.50; 1.0; 2.0 mg/cm²) on the *in vivo* SPF of three selected commercial sunscreen products was investigated in three experienced test centers.

These samples of the sun care market represent a broad range of technologies. Despite this variety of products the approach has shown a linear link of the level of protection and the amount of product applied. Thus reducing the applied amount less decrease of protection than previously assumed is obtained.

The linear way of consumers thinking can be overtaken as well for the SPF, independent of any physical laws which are still the basis of transmission and/or absorption measurements. This

result should be pointed out as an important fact for the consumer and also for the manufacturer of sunscreen products.

Beside this clear linear link of SPF and product amount applied the comparison of the 3 different application amounts justifies the usage of 2.00 mg/cm² for the SPF determination under laboratory conditions. A quantity of 2.00 mg/cm² is identified to generate the most reliable and reproducible SPF results at different test centers independent of the type of sunscreen formulation.

This multi-center study is a contribution to clarify the polarised discussion concerning the risk for consumers applying less than 2.00 mg/cm² of sunscreen as used under laboratory conditions in comparison to “real” conditions. Further investigations on a wider range of products are necessary to verify the results of this study.

References

1. Blum H, Eicher M, Terus W: Evaluation of protective measures against sunburn. *Am J Physiol.* 146, 118-125, 1945.
2. Schulze R: Einige Versuche und Bemerkungen zum Problem der handelsüblichen Lichtschutzmittel. *Parfümerie und Kosmetik.* 37,p 310-315, 1956.
3. Department of health, education and welfare, FDA, USA: Sunscreen drug products for over-the-counter human drugs; proposed safety, effective and labeling conditions. *Federal Register.* 43/166, 38206-38269, 25 August 1978.
4. Deutsches Institut für Normung: Experimentelle dermatologische Bewertung des Erythemschutzes von externen Sonnenschutzmitteln für die menschliche Haut. *DIN 67501.* 1984.
5. COLIPA: COLIPA SPF test method. *Ref 94/289.* 1994
6. ÖNORM: Sunscreen products - Laboratory testing of sun protection factors (SPF). *ÖNORN S 1130,* 1998.
7. Deutsches Institut für Normung (DIN): Experimental evaluation of erythema protection of external sunscreen products for the human skin. *Revision of DIN 67501,* 9/1999.
8. International Sun Protection Factor (SPF) Test Method. COLIPA, February 2003.
9. Bech-Thomsen N, Wulf HC. Sunbathers' application of sunscreen is probably inadequate to obtain the sun protection factor assigned to the preparation. *Photodermatol Photoimmunol Photomed* 1993; 9: 242-244.
10. Farr, P. M. and B. L. Diffey (1985) How reliable are sunscreen protection factors? *British Journal of Dermatology* 112,113-118.
11. Brown S, Diffey BL. The effect of applied thickness on sunscreen protection: in vivo and in vitro studies, *Photochem Photobiol* 1986; 44: 509-513.
12. Stokes RP, Diffey BL. How well are sunscreen users protected? *Photodermatol Photoimmunol Photomed* 1997;13:186-8.
13. Stenberg C, Larkö O. Sunscreen application and its importance for the sun protection factor. *Arch Dermatol* 1985; 121: 1400-1402.
14. Bimczok et al, manuscript in preparation
15. Ferrero L, Pissavini M, Marguerie S, Zastrow L. Efficiency of a continuous height distribution model of sunscreen film geometry to predict a realistic sun protection factor. *J. Cosmet. Sci.* 2003, 54: 463 -481