Report on the interlaboratory ring test

Heiner Gers-Barlag
Beiersdorf AG, Hamburg, Germany

Executive Summary

Five sunscreens which were formerly investigated in a DGK ring study were supplied to two Australian institutes and measured according to the proposed UVA index and UVA balance method. As expected the determination of the $\mathrm{SPF}_{\text{in vitro}}$ resulted in very high standard deviations. But the relative UVA parameters were detected by all labs very reproducibly. No relevant differences were found between data from the DGK study and from the Australian institutes.

The method exhibits a high reproducibility and is considered as potential candidate for future harmonized UVA measurements.

Aim of the present study

Due to the increasing knowledge about the harmful effects of the long wavelength UVA radiation authorities, companies and experts intensely debated during the past years how the UVA protection in sunscreens should be determined. A finally resulting and agreed method needs to cover relevantly the effects on UVA exposed skin and it needs to be highly reproducible to differentiate and to compare validly the UVA protection efficacy of sunscreens.

The DGK task force "sun protection" published recently the results of a ring study [1] and demonstrated that a high agreement between several labs were achieved when they determined *in vitro* relative UVA protection parameters like the UVA index. The aim of the present study was to perform these measurements in two Australian institutes and to compare the results with the data from the DGK study.

Method

Product application and transmission measurements were performed according to the descriptions in [1]. Each single measurement was controlled to meet the requirements set by Klette et al. [2] in order to avoid any saturation or overloading of the instrumentations.

Substrate
Roughened Plexiglas® (**s**upplied by Schönberg GmbH & Co KG, D-22457 Hamburg, Germany) plates for single use from one batch were chosen. A roughened PMMA plate with a

Samples
The five sunscreens already used in the DGK study were also chosen for this ring study. The selection represent the majority of all sun products with regard to product type, UV filter composition and SPF. For more details see Table I. All samples came from the same batches as used in the DGK study. In order to analyse possible alterations in the absorbance properties due to storage or aging all samples were remeasured by LAB 1 of the DGK paper (= Beiersdorf AG).

Sample	SPF	Type of emulsion	UV-filter system
Sample A	12	O/W emulsion	Ethylhexyl Methoxycinnamate Butyl Methoxydibenzoylmethane
Sample B	15	W/O emulsion	Methylbenzylidene Camphor Ethylhexyl salicylate Butyl Methoxydibenzoylmethane Octocrylene
Sample C	15	O/W emulsion	Ethylhexyl Methoxycinnamate Zinc Oxide Phenylbenzimidazole Sulfonic Acid
Sample D	24	Hydro-dispersion	Methylbenzyldine Camphor Ethylhexyl Triazone Butyl Methoxydibenzoylmethane Titanium Dioxide
Sample E	30	O/W emulsion	Octocrylene Titanium Dioxide Butyl Methoxydibenzoylmethane Terephthalylidene dicamphorsulfonic acid

Tab. I: List of sunscreens

Calculation
$SPF_{in\ vitro}$, $PPD_{in\ vitro}$ and the UVA index were calculated according to [1]. Additionally we calculated also a modified UVA index the UVA balance [3].

Institutes
Two Australian institutes participated in the ring study:

Mr John Staton
Dermatest Pty Ltd.
22 King Street
Rockdale NSW 2216
Australia

Mr Gavin Greenoak
Australian Photobiology Testing Facility
Suite 204 - 205 Ross Street Building A03
University of Sydney NSW 2006
Australia

Results

Each samples was applied to 3 or 4 plates. The resulting absorbance curves are given in the Appendix. The remeasurement in LAB1 was performed on one single plate. The absorbance of the sunscreens did not exceed the value of 2 indicating that the chosen product rate of 0.75 mg/cm^2 have been appropriate.

When the SPF$_{\text{in vitro}}$ is determined the big variation of this parameter becomes obvious. Even in the same lab the data from one to the other measurement were extremely deviating (see particularly LAB 1, samples B and D). The results from the Australian institutes underline the non homogeneity (see table II).

A key step in the UVA method is the adjustment of the absorbance curve to the SPF$_{\text{in vivo}}$. By introducing this step the non-reproducible data turn into a set of reproducible data. The PPD$_{\text{in vitro}}$ factor as well as the UVA index and the UVA balance were reproducibly found within a small standard deviation. The data of both Australian institutes supplemented the DGK data excellently.

Sample	**A**	**B**	**C**	**D**	**E**
in-vivo SPF	**12**	**15**	**15**	**24**	**30**
Lab1, neu	**8,1**	**22,0**	**6,0**	**15,9**	**40,5**
Lab 1	8,3	9,1	7,4	23,8	17,5
Lab 2	7,1	14,2	5,3	14,4	17,6
Lab 3	6,2	14,5	3,6	10,4	6,1
Lab 4	8,4	13,4	5,2	14,1	16,8
Lab 5	9,1	7,7	3,5	15,4	13,8
Lab 6	8,2	14,2	4,5	14,9	15,1
Lab 7	8,2	13,9	5,7	6,9	26,5
Dermatest	17,8	32,5	15,7	32,5	30,9
APTF	8,8	24	7,7	13,3	45,4
Mean, DGK	7,9	12,4	5,0	14,3	16,2
SD, DGK	0,96	2,80	1,34	5,19	6,04
Mean, DGK with Australia	9,1	15,9	6,5	16,2	21,1
SD, DGK with Australia	3,37	7,69	3,74	7,60	11,58

Tab. II: Determination of the SPF$_{\text{in vitro}}$

Sample	**A**	**B**	**C**	**D**	**E**
in-vivo PPD factor	**1,8**	**4,5**	**n.d.**	**5**	**10,5**
Lab1, neu	2,1	5,4	2,3	6,0	11,3
Lab 1	2,2	6	1,9	6,2	10,3
Lab 2	2,2	6,1	2,4	6,7	12,2
Lab 3	2	5,8	2,2	5,1	10,4
Lab 4	2	5,8	2,2	5,1	10,4
Lab 5	2,1	5,1	1,5	6,5	11,9
Lab 6	2,1	6,1	2,1	5,8	9,5
Lab 7	2,4	6,3	2,3	7,4	13,5
Dermatest	2,2	5,7	2	7,2	10,9
APTF	2,2	5,7	2,6	7,6	13,8
Mean, DGK	2,1	5,9	2,1	6,1	11,2
SD, DGK	0,14	0,39	0,30	0,85	1,40
Mean, DGK with Australia	2,2	5,8	2,1	6,4	11,4
SD, DGK with Australia	0,12	0,35	0,32	0,93	1,50

Tab. III: Determination of the PPD$_{\text{in vitro}}$

Sample	A	B	C	D	E
Lab1, neu	**17,5**	**36,3**	**15,1**	**24,8**	**37,5**
Lab 1	19,0	40,0	13,0	26,0	34,0
Lab 2	19,0	40,0	16,0	28,0	41,0
Lab 3	17,0	38,0	15,0	21,0	35,0
Lab 4	16,0	38,0	15,0	21,0	35,0
Lab 5	18,0	34,0	13,0	27,0	40,0
Lab 6	17,0	41,0	14,0	24,0	32,0
Lab 7	20,0	42,0	15,0	31,0	45,0
Dermatest	18,4	38,1	13,6	30,0	36,3
APTF	18,7	37,8	17,4	31,9	46,0
Mean, DGK	18,0	39,0	14,4	25,4	37,4
SD, DGK	1,41	2,65	1,13	3,69	4,65
Mean, DGK with Australia	18,1	38,8	14,7	26,7	38,3
SD, DGK with Australia	1,25	2,34	1,45	4,05	4,98

Tab. IV: Calculation of the UVA index

Sample	A	B	C	D	E
Lab1, neu	**10,0**	**31,7**	**9,0**	**21,5**	**35,4**
Lab 1	10,9	35,7	6,4	22,6	32,1
Lab 2	10,9	36,4	10,0	24,8	38,6
Lab 3	9,1	34,3	8,6	17,8	32,4
Lab 4	9,1	34,3	8,6	17,8	32,4
Lab 5	10,0	29,3	3,6	23,9	37,6
Lab 6	10,0	36,4	7,9	20,9	29,3
Lab 7	12,7	37,9	9,3	27,8	43,1
Dermatest	10,9	33,6	7,1	27,0	34,1
APTF	10,9	33,6	11,4	28,7	44,1
Mean, DGK	10,4	34,9	7,8	22,2	35,1
SD, DGK	1,27	2,78	2,16	3,68	4,83
Mean, DGK with Australia	10,5	34,6	8,1	23,5	36,0
SD, DGK with Australia	1,12	2,48	2,26	4,06	5,19

Tab. V: Calculation of the UVA balance (according to [3])

Conclusion

The data demonstrate convincingly that the in vitro determination of the UVA protection of sunscreens can be performed in a very reproducible manner. In this study the PMMA substrate as well as the application procedure were chosen to be equal to the conditions of the DGK study. Each measurement run in the valid range and the data for the relative UVA parameters, the UVA index and the UVA balance were in good accordance. On this level, no differences were detectable between the data from the two Australian institutes and the DGK data.

The method is valid and reproducible and should be considered as potential candidate in the ongoing debate about harmonized and standardized UVA methodologies.

Literature

1. Gers-Barlag H, Wendel V, Klette E, Bimczok R, Springob C, Finkel P, Rudolph T, Gonzenbach HU, Westenfelder H, Schneider P, Kockott D, Heinrich U, Tronnier H, Johncock W, Langner R, Driller H, Pflücker F, Wünsch T; *The reproducibility of an in vitro determination of the UVA INDEX describing the relative UVA protection of sun care products;* IFSCC Magazine **5** (3), 161 - 166 (2001)
2. Klette E, Wendel V, Wittern KP, Gers-Barlag H; A quick, practical test procedure to evaluate the performance of instruments used for in vitro UV protection measurements, Int. J. Cos. Sci. **24** (2002), 1-7.
3. Gers-Barlag H, In vitro determination of the UVA balance; Sun Conference June 2003, London.

Appendix

Absorbance curves for Sample A to E