A Controlled Trial of Objective Measures of Sunscreen and Moisturizing Lotion

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Abstract

Taking an alcohol swab of a person's forearm and analyzing it using a spectrophotometer has been shown to be a reliable method for detecting the presence of sunscreen. The aims of this study were to determine if moisturizing lotions or other non-sunscreen products influence the absorbance readings from skin swabs in a controlled setting, and to establish the cutoff point in determining the presence or absence of sunscreen using a crystal cuvette instead of a plastic one. In a controlled trial of 30 volunteer office workers, absorbance readings from two popular brands of sunscreen with sun-protection factors (SPF) of 30 and 45 were compared with absorbance readings from two different moisturizing lotions, one with an SPF of 15 and another with no stated SPF. Moistur-

Background

Self-report data are most commonly used to collect information about sunscreen use (1). However, as with any self-reported behavioral information, these data are subject to social desirability and recall biases. Taking an alcohol swab of a person's forearm and analyzing it using a spectrophotometer was recently shown to be a reliable method for detecting the presence of sunscreen (2), and the feasibility of this method has been supported by subsequent studies (3, 4). In a controlled experiment in an indoor office setting, spectrophotometer absorbance readings showed very clear distinctions between the presence and the absence of sunscreen, and the UV absorption cutoff point for a positive sample was determined to be 0.147 (3).

In subsequent field studies where water activities, the amount of sunscreen used, and the sun-protection factor (SPF) value were not controlled, absorption readings still showed statistically significant differences between the

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izers with SPF 15 tested positive for sunscreen, with absorbance readings (mean, 3.77; min, 3.30) comparable to sunblock with SPF 30 or 45 (mean, 3.51; min, 2.02). Moisturizers with no stated SPF factor tested negative for the presence of sunscreen, with extremely low absorbance readings (mean, 0.06; max, 0.19) similar to control readings. The skin swabbing technique remains a valid and useful method for detecting the presence of sunscreen and does not result in false positives when moisturizers with no stated SPF are present. Using a conservative cutoff point of 0.30 with a crystal cuvette reduces any chance of false-positive readings and remains robust when sunscreen of SPF 15 or higher is present. (Cancer Epidemiol Biomarkers Prev 2009;18(5):1399–1402)

presence and the absence of sunscreen using the same cutoff point (0.147). However, these readings contained more "noise" or absorption readings, suggesting the presence of sunscreen or its active ingredients, which were noticeably higher than the cutoff point but not nearly as high as the readings normally associated with sunscreen use and detection (4). It was unclear whether these readings might be due to (*a*) the use of lower SPF sunscreen, (*b*) the use of non-sunscreen moisturizers, (*c*) the presence of residual sunscreen from a previous application, or (*d*) some other unknown factor.

It is also unclear where the cutoff point should be established in determining the presence or absence of sunscreen if, for example, the laboratory procedures or equipment differ from one research environment to another. The scientists who first reported this method of swabbing determined from their pilot studies and trials that a cutoff point of 0.147 at 320 nm wavelength was the most reliable indicator of the presence of sunscreen (2). Subsequent studies confirmed both the cutoff point and wavelength (3, 4). However, while preparing our laboratory for analysis of a large number of samples in the study reported in this article, our results indicated false-positive readings using the 0.147 cutoff point. We attributed these false-positive readings to the use of a reusable crystal cuvette instead of the disposable plastic cuvettes used in previous studies (3, 4) because the remainder of our methods and equipment were identical.

The aims of this study were to determine if moisturizing lotions or other non-sunscreen products might

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possibly be influencing the absorbance readings from skin swabs in a controlled setting, and to establish the cutoff point in determining the presence or absence of sunscreen from skin swabs using a crystal cuvette.

Context

During the summer of 2006, skin swabbing techniques were used to detect the presence of sunscreen on more than 500 lifeguards, parents, and children as part of a field study to compare self-reported sun protection behaviors with objective measures at 16 swimming pools participating in the Pool Cool sun-safety diffusion trial (5). In this very large study, with four samples per participant, laboratory methods were similar to those used in previous research (3, 4) with two exceptions: (a) a newer spectrophotometer was used (same brand and series) and (b) reusable crystal cuvettes were used instead of disposable plastic cuvettes to reduce cost and waste. The crystal cuvettes were thoroughly washed between samples, and control samples were used for quality assurance throughout processing. Whereas the detection of the presence of sunscreen was fairly obvious from the absorbance readings in most cases, we were concerned about the same noise issues identified in a previous field study using sunscreen swabbing (4). An additional office trial was therefore conducted to help examine possible sources of the variation in spectrophotometer readings and to establish the cutoff point for the presence of sunscreen.

Methods

Thirty office workers volunteered for this trial. Participants were randomly assigned to two equal groups of 15. Group 1 received doses of Banana Boat Ultra Sunblock Lotion (SPF 30). Group 2 received Coppertone Oil Free Sunscreen Lotion (SPF 45). The sunscreen brands and SPF values were chosen based on the most commonly mentioned brands and SPF values reported by participants in our previous skin cancer prevention research. Both sunscreens claimed to provide UVA and UVB protection, although specific ingredients and wavelength were not stated on the labels. Within the two groups, participants were further randomly assigned to receive one of two different daily moisturizing lotions: (*a*) Lubriderm Daily Moisture (SPF 15) and (*a*) Eucerin Daily Replenishing Lotion (no stated SPF).

Participants were contacted the day before the trial and reminded not to apply any sunblock or lotions on either forearm the following morning. They were asked again on the morning of the trial whether they had applied any sunscreen or lotions to their skin that day, and any who answered "yes" were excluded from the study.

The morning of the trial, two research staff members began applying sunscreen or lotion at 10:00 a.m. A cardboard cutout and a ballpoint pen were used to mark off three sections (2.5×4 cm each) on the dorsal surface of each participant's right forearm, following methods from previous trials ([3, 5]). The sections were labeled A, B, and C and then randomly assigned to one of the three treatment applications: (a) sunscreen (0.02 mL =

 2 mg/cm^2), (b) lotion (0.02 mL = 2 mg/cm^2), or (c) nothing (control section). Sunscreen or lotion was applied using a needle-free syringe for measurement accuracy and rubbed evenly with a disposable gloved finger. Participants were blinded to their study conditions. Two different (and blinded) research assistants returned ~1 hour later to take swabbing samples.

After explaining the procedure, research assistants wearing disposable gloves swabbed each participant's right forearm with disposable alcohol wipes (70% isopropyl alcohol). Each 2.5×4 cm section was swabbed twice with the same wipe, using light and even pressure, reversing the side of the wipe each time to ensure that a thorough sample was collected. Used wipes were sealed in prelabeled glass vials containing 4 mL of 100% ethyl alcohol.

All procedures for this study were approved by the Institutional Review Board at Emory University.

Analysis. Before analysis, vials of 100 percent ethyl alcohol were prepared for use as calibration references. Reference standards were made with clean alcohol wipes placed directly into 4 mL of 100% ethyl alcohol. Samples with absorbance readings within 1 SD were kept as reference standards (controls). After the initial calibration of the spectrophotometer, swabbing samples from participants were analyzed with a calibration reference standard rerun after every three participants to confirm that the spectrophotometer calibration had not changed. Readings for participant samples and controls were taken in separate cuvettes to prevent cross-contamination.

Swabs were analyzed for the presence of sunscreen based on UV absorption following previous research methods (3). After shaking the vial to mix its contents, 1.5 mL of sample were transferred via disposable pipette into a 10-mm UV, open-top crystal cuvette (Fisher Scientific) and placed in a UV-Vis spectrophotometer (Beckman DU 530). Absorbance was measured across the UV spectrum from 280 to 400 nm at 5-nm intervals. The cuvette was thoroughly washed with alcohol (100% ethanol) and dried before testing of the next sample. The pipette tips were discarded after each sample.

In accordance with previous studies (2-4), absorbance readings at 320 nm were used to establish the presence or absence of sunscreen. The UVA spectrum (320-400 nm) and the UVB spectrum (280-320 nm) meet at 320 nm, so it is considered the most reliable indicator of the presence of sunscreen (2). The absorbance readings were compared with readings of the control sample swabs taken from participants where neither sunscreen nor moisturizing lotion had been applied. Descriptive analyses and graphical analyses were done on all absorbance readings, and one-way ANOVA was conducted with lotion type as the between-subjects factor.

Results

After data collection was completed, one participant indicated that he had used moisturizing lotion the morning of the study, and his results were excluded from analysis, leaving data from 29 participants. A summary of all readings is shown in Table 1, and a graphical representation of the data in Fig. 1. All swabbing samples known to contain sunscreen (SPF

30/45) recorded very high absorbance readings at 320 nm, with an average reading of 3.513 and a minimum of 2.019. The SPF 15 moisturizing lotion recorded similarly high absorbance readings (mean, 3.767; min, 3.301), whereas the SPF 0 lotion recorded extremely low readings (mean, 0.061; max, 0.193). ANOVA showed a statistically significant difference [F(3) = 667.58, P <0.001, $\omega^2 = 0.96$] among the four groups (control, lotion without SPF, lotion with SPF 15, and sunscreen). A Tukey post hoc analysis revealed mean differences between the control or lotion-no-SPF samples and the lotion-SPF or sunscreen samples. The visual inspection reaffirms the statistical analyses. Based on the above data, an absorbance reading of 0.300 is an appropriate cutoff point, any value above which is considered a positive indicator of the presence of sunscreen using this method. Calculations with these data using the 0.147 cutoff reveal a specificity of 93%, with three false-positive readings, whereas specificity is 100% at the 0.300 cutoff. Sensitivity is 100% for both cutoff points.

Discussion

The cutoff point of 0.300 used in this study is slightly higher and more conservative than the 0.147 cutoff point used in past studies (3, 4), accounting for the use of a

reusable crystal cuvette, which is believed to have slightly different light-absorbing properties than a disposable plastic cuvette (6), because all other methods and equipment used were identical to those used in previous studies (3, 4).

Moisturizers with SPF 15 tested positive for sunscreen, with absorbance levels comparable to sunblock with SPF 30 or 45. Moisturizers with no SPF factor tested negative. Thus, what may seem to be false-positive readings (where swabbing results show sunscreen but participants do not report using it) may be due to the use of self-report questions that do not ask about use of moisturizer with sun protection ingredients.

Noise in swabbing analyses that are noticeably higher than control samples but well below those of SPF 15+ may be due to (a) the use of very low SPF moisturizers (SPF 2, 4, or 8); (b) the presence of remaining traces of sunscreen from a previous application (perhaps even the day before) that was not thoroughly washed off; or (c) other factors such as dirt that may have found its way onto participants' arms or into the swabbing solution. Additional research is needed to examine these potential causes.

Previous studies have shown that people do not apply enough sunscreen to meet the recommended dose of 2 mg/cm² (7). However, studies using the swabbing technique have been able to detect the presence of sunscreen even when applied by subjects in unknown

n	Sunblock with SPF 30/45	Lotion with SPF 15	Lotion with no SPF
1	2.053		0.099
2	3.054	3.902	
3	3.361	3.607	
4	3.955		0.053
5	3.122		0.111
6	3.742	3.519	
7	3.217	3.301	
8	2.726		0.091
9	3.560	3.438	
10	4.000		0.032
11	4.000		0.022
12	4.000	4,000	0.022
13	4.000	1.000	0.035
14	3 711		0.023
15	3 419	4 000	0.020
16	2 840	1.000	0.080
17	3 957		0.022
18	4 000	3 856	0.022
10	3 188	3 507	
20	4 000	3.307	0.039
20	2 019	4 000	0.007
21	4 000	4.000	0.033
22	3 737	3 635	0.035
23	4.000	2 745	
24	4.000	3.743	0.018
25	2.424		0.010
20	2.434	4 000	0.195
27	4,000	4.000	
20*	4.000	4.000	
29	4,000	4 000	
30	4.000	4.000	
Mean	3.513	3.767	0.061
SD	0.878	0.971	0.050
Median	3.742	3.856	0.037
Min	2.019	3.301	0.018
Max	4.000	4.000	0.193

*Participant was removed for applying lotion that morning.



Figure 1. Spectrophotometer absorbance readings (± 1 SD) of alcohol solutions with and without sunscreen ingredients. Absorbance readings at 320 nm wavelength, where the UVA spectrum (320-400 nm) and the UVB spectrum (280-320 nm) meet, show the detection of sunscreen in the SPF 15 group (mean, 3.767) and the SPF 30+ group (mean, 3.513) but show no sunscreen detection in the control group and the SPF 0 group (mean, 0.061), which are both well below the conservative cutoff point of 0.300.

quantities in nonlaboratory settings (4). Whether the amount of sunscreen applied in these settings would be considered adequate or not is unknown; however, this suggests that it is unlikely that inadequate application of SPF 15+ sunscreen is the cause of lower absorbance readings. Previous research using the 0.147 cutoff point also found that once applied, sunscreen could still be detected by the swabbing technique for at least 6 hours (3). This supports our inference that residual sunscreen from a previous application may account for some of the lower, nonzero absorbance readings found in studies using skin swabbing techniques.

Like the previous studies, this study indicates that the swabbing method is a reliable objective method for determining the presence of sunscreen, thus making it a very useful technique for evaluating behavioral interventions aimed at increasing sunscreen use. This method will detect all organic sunscreen ingredients, regardless of SPF level, and has been shown to detect lighter applications of sunscreen in field research (4). However, it does not detect sunscreens containing exclusively inorganic ingredients, nor does it measure features related to the quality of sunscreen (e.g., UVA coverage, specific SPF, waterproof ability) or adequacy of application. Whereas some intermediate absorbance readings may show up in laboratory analyses, the effectiveness of the swabbing technique in detecting the presence of any meaningful amount of SPF 15+ sunscreen remains very strong. Furthermore, it is clear that regular moisturizing lotion without SPF does not affect absorbance readings using this technique, and any absorbance readings higher than the cutoff point of 0.300 indicate the presence of sunscreen in some quantity.

This study is limited by the lack of specific testing for residual sunscreen from previous applications. Thus, it is still unclear whether very low SPF or residual sunscreen from previous applications may affect absorbance readings. Further research should address these questions.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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