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Article

Improper storage of sunscreen might decrease effectiveness

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SUMMARY

One of the leading causes of melanoma is overexposure to ultraviolet (UV), and each year over 100,000 cases of melanoma are diagnosed. Sunscreen, when used correctly, can reduce the risk of skin cancer. This study evaluated sunscreen efficacy under different storage temperatures. The FDA recommends that sunscreen be stored in a place away from excessive heat, but many consumers are unaware of this. We hypothesized that exposure to extreme temperatures can compromise sunscreen effectiveness, potentially increasing the risk of sunburn or sun damage. We tested 22 sunscreens by spreading sunscreen on plastic wrap placed on a UV light. The UV light that penetrated through the sunscreen was measured by a UV meter. Before adding to plastic wrap, sunscreens were stored at -10°C, 6°C, 22°C, 40°C, and 50°C for 18-24 hours before measuring UV absorption. We used repeated measures ANOVA to detect mean differences in the percentage of UV blocked between the different storage temperatures. Sunscreens stored at -10°C blocked 74% of UV light. Sunscreens stored at 50°C blocked only 64.8% of UV light. We also studied characteristics of sunscreen to see if specific qualities were associated with better temperature performance. Ten characteristics of sunscreen were studied, and none had statistically different effects on temperature partial inactivation. This study demonstrated sunscreen inefficiency when stored at 50°C to raise sunscreen users' awareness of proper storage temperature.

INTRODUCTION

Ultraviolet (UV) rays, both A and B, can damage the skin, but in different ways. Ultraviolet B *(*UVB) rays reach the outer layer of the skin, called the epidermis, and can cause sunburn (1). However, Ultraviolet A (UVA) rays reach the dermis (i.e., the inner layer of skin), causing wrinkles and premature aging (1). UV radiation is all electromagnetic energy emitted from the sun or artificial sources, while UV rays, such as UVA and UVB, are the specific types of radiation, classified by their wavelength (2). Exposure to UV radiation can damage skin cells by interacting with their DNA, proteins, and cellular structure (3).

There are many types of skin cancer, but almost all of them are caused by UV radiation, leading to abnormal cell growth (4). UV radiation can cause mutations in DNA, such as thymine dimers. When DNA becomes damaged, DNA replication, the process in the cell cycle where DNA doubles before mitosis, becomes unregulated. This allows the cell to replicate, even when cell-cycle checkpoints are not adequately passed. The DNA is damaged in such a way that cell-cycle regulation no longer works (5). Common types of skin cancer are melanoma, squamous cell carcinoma of the skin, basal cell carcinoma (BCC), and nonmelanoma skin cancer (4). BCC is caused by DNA damage in the basal cells of the epidermis (4). BCC mostly occurs areas that are exposed to the sun such as the head, neck, face, and arms (4). Melanoma is skin cancer from melanocytes, cells that produce pigment and give skin color (1). The risk of melanoma increases with age, but it is also common among younger people under the age of 40 (6). The median age at diagnosis is 66 (6). Squamous cell carcinoma in the keratinocyte cells of the outer epidermis and appears like scaly patches of red firm bumps (7).

Reducing the amount of UV exposure can decrease the risk of skin cancer (1). Sunscreens were made as a barrier to protect the skin from UV rays (8). Sunscreens are categorized into different levels of protection called sun protection factor (SPF) where the higher the SPF number is, the more it protects and prevents sunburn damage (8). The following are ways to reduce UV exposure: use a broad-spectrum sunscreen that is water resistant, reapply sunscreen every two hours, avoid sun exposure from 10 AM-2 PM when the UV exposure is the highest, and take caution near sand, snow, and water as it reflects radiation (9). Intentionally tanning is high risk, as a tan represents skin that has UV damage (9). However, one should be careful as sunscreens are only proven safe and effective for three years after the initial purchase (9). Sunscreens are composed of active ingredients, formula stabilizers, sensory enhancers, and added extra components (10). Some components of sunscreens, particularly the active ingredients, can break down over time (10). The quantity of active ingredients varies by type. The Food and Drug Administration (FDA) regulates a different maximum quantity of active ingredients, ranging from between avobenzone at 3% to zinc oxide at 25% in chemical vs. mineral sunscreens (11).

The primary types of sunscreens are mineral and chemical. Mineral sunscreens do not absorb into the skin. Rather, they create a barrier on your skin that reflects the UV light/rays (12). In mineral sunscreen the active ingredients are often titanium dioxide and zinc oxide (8). Zinc oxide and titanium dioxide act as physical barriers against sunlight, and they are a safer alternative to other chemicals because they reflect UV rays rather than absorbing them because the photon does not enter the skin (8). Recent changes in the approval of over-the-counter sunscreens, as part of the 2020 Coronavirus Aid, Relief, and Economic Security (CARES) Act, revealed that the FDA deems mineral sunscreens, containing up to 25% of either

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titanium dioxide or zinc oxide, to be generally recognized safe and effective (11).

Chemical sunscreen ingredients absorb UV radiation (10). Common active ingredients in chemical sunscreens include oxybenzone, octinoxate, octisalate, and avobenzone (12). Chemical sunscreens have a benzene ring, which absorbs a photon and dispenses light into heat (10). For people with sensitive skin, chemical sunscreens can cause redness or inflammation (8). The FDA regulation in the CARES Act of 2020 called for additional data on the safety and efficacy of chemical sunscreens with active ingredients such as avobenzone and oxybenzone, due to unanswered questions about consumer safety and environment harm (13).

Sunscreens can be further divided based on the level of protection they provide. Broad-spectrum sunscreen shields the skin from UVA and UVB rays. In 2010, scientists learned that UVA rays do not necessarily cause visible sunburn, but they do cause DNA damage (8). Before this finding, sunscreen was not designed to protect against UVA damage. In 2011, the FDA mandated that sunscreens advertised as broad-spectrum must protect against UVA and UVB (8). A study examined 32 people who used broad-spectrum sunscreen on the participants' faces for a year. All test subjects self-reported improvements in skin clarity and texture over the course of a year (14). Broad-spectrum sunscreens account for 25% of the sunscreen market (15).

Sunscreens should not be stored in direct sunlight (16). On a hot day with strong UV, sunscreen should be stored in a shady place (17). The ideal storage temperature for sunscreen is between 15 and 30°C (9). Sunscreens are frequently stored in cars where the internal temperature can be as cold as -3°C or as hot as 85°C (17). A pilot study tested the phase change of nine sunscreens exposed to extreme temperatures and found that four of the nine sunscreens experienced a phase change at 60°C (16). Toni Golen, the editor of Harvard Women›s Health Watch, explains that applying sunscreen that has been exposed to extreme temperatures can be a reason that people have sunburns even after applying sunscreen (18). Additionally, sunscreens that are expired or not stored correctly could break down in a way that produce skin irritants (10).

Sunscreen is essential to prevent aging and skin cancer; however, improper storage of sunscreen may reduce its effectiveness. While the FDA recommends storage temperatures for sunscreen, the exact consequences of improper storage are not part of the warning. It is unknown which storage temperatures reduce the effectiveness of sunscreen. We hypothesized that chemical and mineral sunscreens, when stored at extremely hot or cold temperatures, will lose their ability to block UV radiation. This research aims to characterize the efficacy of sunscreen stored at extremely low and high temperatures to bring awareness about proper sunscreen storage.

RESULTS

This study aimed to measure the effectiveness of sunscreens stored at extreme temperatures. We exposed 22 chemical and mineral sunscreens to -10°C, 6°C, 22°C, 40°C and 50°C temperatures for 18-24 hours (**Table S1**). We then spread the sunscreens on plastic-wrap, placed the plastic-wrap on a transilluminator, and measured the amount of UV light that was able to penetrate the sunscreen with a UV meter. The outcome variable was the percent of UV blocked with each sunscreen and each storage condition.

Sunscreens stored at -10°C significantly blocked more UV than sunscreens stored at 50°C (repeated-measures ANOVA, *F* (122) = 1.1073, *p* = .04284). While there were no other significant differences, sunscreens stored at -10°C and 6°C blocked a slightly higher percentage of UV light than sunscreens stored at room temperature (**Figure 1**).

We created a plot to show the temperature performance of each of the 22 sunscreen brands (**Figure 2**). Eucerin and COOLA sunscreens were less effective at hot temperatures. Aveeno Protect + Hydrate, CereVE, Native coconut and pineapple, and Native unscented all had worse performance at 6°C (**Figure 2**). Target Store brand had the best percentage of UV blocked through -10°C; 77% of UV was blocked.

After the result showing different temperature performance in different sunscreens, we were then interested in determining if any sunscreen characteristics were responsible for the difference in temperature stability. Our main goal was to determine if sunscreens have similar characteristics that impact their stability when stored in extreme temperatures. Therefore, we used repeated-measures ANOVA design to test the interaction between the storage temperature and sunscreen characteristics. The sunscreen characteristics were chemical/mineral, broad-spectrum, consistency, water resistance, avobenzone, tocopheryl acetate, PEG-100, phenoxyethanol, and cetyl alcohol.

The effect of the storage temperature on the percentage of UV blocked was not significantly different between chemical and mineral sunscreens (ANOVA interaction p=0.7158) as well as broad-spectrum and non-broad-spectrum sunscreens (ANOVA interaction, p=0.3337) (**Figure 3**). For sunscreen consistency, 10 sunscreens had a medium consistency, 4 had a thick consistency, and 8 had a thin consistency. The effect

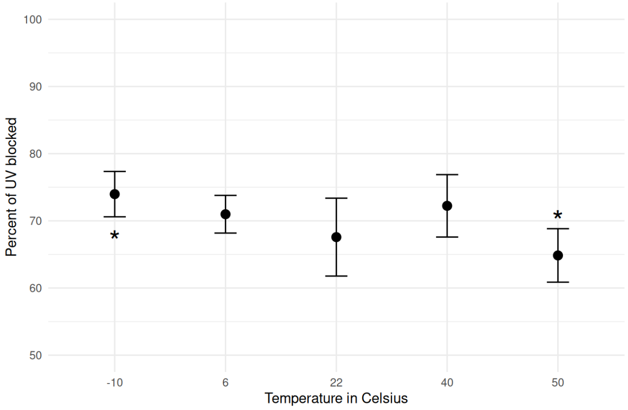

Figure 1: Sunscreen performance is related to temperature. Twenty-two sunscreens were stored at a variety of storage temperatures for 18-24 hours, then, the amount of light from a transilluminator that was blocked by the sunscreen spread onto plastic wrap, was recorded with a UV meter for two trials. The amount of UV read by the UV meter was compared to a no-sunscreen control, to derive percent of UV blocked. The bars represent 95% confidence intervals, and the dots represent the mean percentage of UV blocked. Each storage temperature was compared to each other storage temperature in an ANOVA test. This figure shows that sunscreens stored at 50°C block less UV light than those stored at -10°C (mixed-effects ANOVA for 50°C/-10°C, p=0.04284). The asterisks indicate a statistically significant difference (p<0.05) between groups.

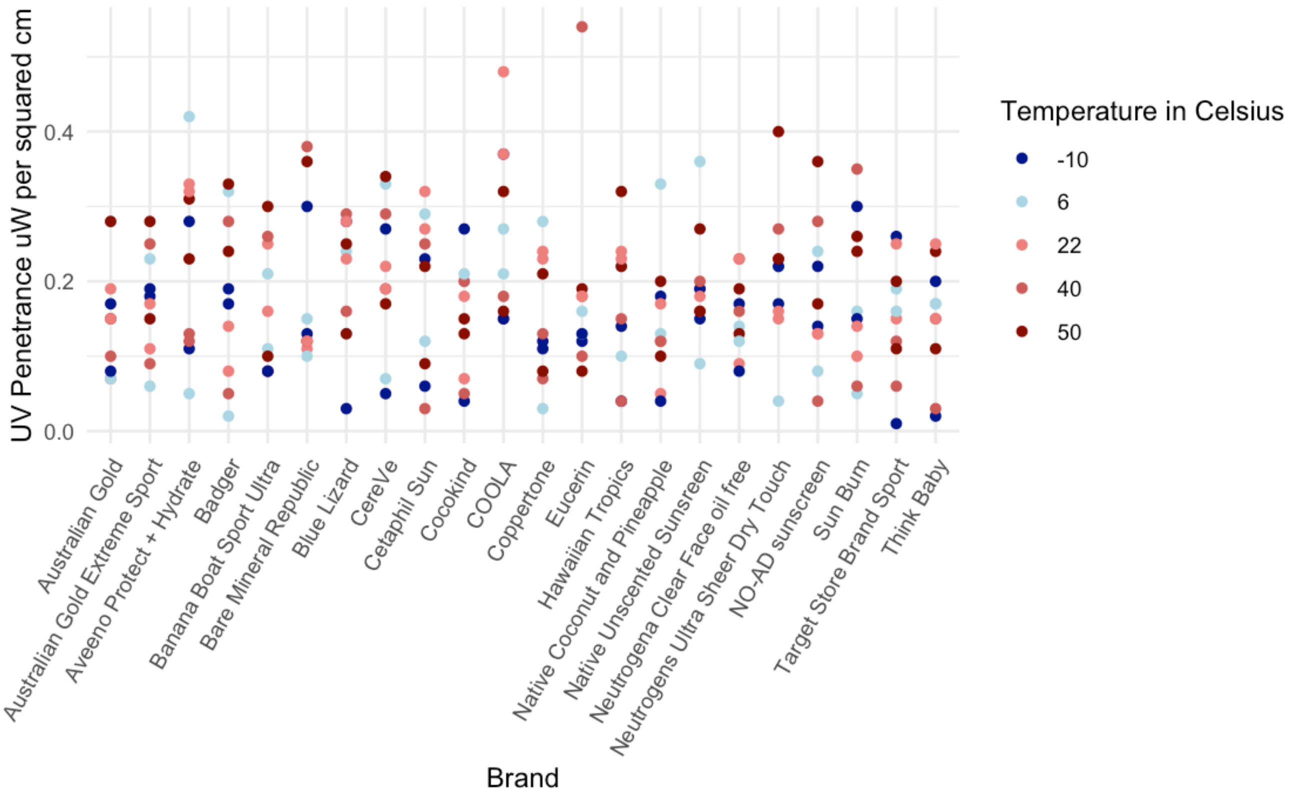

Figure 2: Sunscreen UV efficacy by temperature. A scatter plot shows the difference in UV reading between all sunscreens and tested temperatures (-10, 6, 22, 40, and 50°C). The y-axis shows UV which is the raw reading of the UV meter to display how much light was passing through the sunscreen. A low reading shows that a sunscreen blocked a larger percentage of UV, whereas a higher rating shows less sunscreen efficacy. Dark blue represents -10°C, light blue represents 6°C, light red represents 22°C, red represents 40°C and dark red represents 50°C. Data are shown for both replicates of this study. Each dot represents one replicate (total n=220).

of the storage temperature on the percentage of UV blocked was not significantly different when comparing consistency (ANOVA interaction, *F* (20, 174) = 0.8273, *p* = .856) (**Figure 3**). Also, there was no significant effect of the storage temperature between water resistant and non-water-resistant sunscreens (ANOVA interaction, p=0.2823) (**Figure 3**).

Avobenzone is an active ingredient in chemical sunscreens, and we found that it was the most common out of the 22 sunscreens. The effect of the storage temperature on the percentage of UV blocked was not significantly different between sunscreens with and without avobenzone (ANOVA interaction, *F* (4, 190) = 0.9178, *p* = .4353) (**Figure 4**). Phenoxyethanol is a preservative in many types of sunscreens and the effect of the storage temperature on the percentage of UV blocked was not significantly different between sunscreens with and without phenoxyethanol (ANOVA interaction, *F* (4, 190) = 0.5368, *p* = .6945) (**Figure 5**). Tocopheryl acetate is an antioxidant with vitamin E, it is also a preservative of the active ingredient. The effect of the storage temperature on the percentage of UV blocked was not significantly different between sunscreens with and without tocopheryl acetate (ANOVA interaction *F* (4, 210) = 1.4326, *p* = .205) (**Figure 5**). Cetyl alcohol is a common ingredient for moisturizers as an emulsifier and thickening agent. The effect of the storage temperature on the percentage of UV blocked was not significantly different between sunscreens with and without cetyl alcohol (ANOVA interaction, *F* (4,190) = 0.6790, *p* =

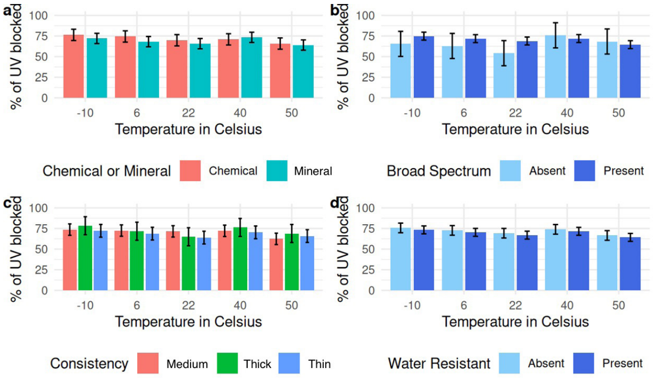

Figure 3: Comparison of partial inactivation of chemical and mineral, broad-spectrum, consistency and water-resistant sunscreens by temperature. Bar graphs show the estimated marginal mean of percentage of UV blocked for sunscreens by a) chemical or mineral, b) broad spectrum, c) consistency, or d) water resistance. These are ANOVA interaction plots. There was no significant interaction between temperature and any of the conditions studied. The bars represent 95% confidence intervals and the mean percentage of UV blocked. Each sunscreen is tested at each temperature, with two replicates n=12 mineral sunscreens (3A blue bar), n=10 chemical (3A red bar), n=20 broad-spectrum (3B blue bar), n=2 not broad-spectrum (3B light blue bar), n=8 thin consistency (3C blue bar), n=10 medium consistency (3C red bar), n=4 thick consistency (3C green bar), n=12 water resistant(3D blue bar) and n=10 not water resistant (3D light blue bar).

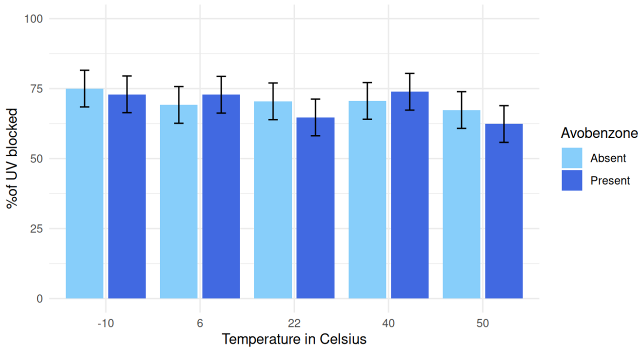

Figure 4: Sunscreens with and without avobenzone are equally temperature sensitive. A bar graph shows the estimated marginal mean of percentage of UV blocked. This is an ANOVA interaction plot. There was no significant interaction between temperature and avobenzone. The bars represent 95% confidence intervals and the mean percentage of UV blocked. 11 sunscreens had avobenzone (dark blue bar), 11 did not (light blue bar).

.5983) (**Figure 6**). PEG100 is also an emulsifier which creates a creamy texture. The effect of temperature on the percentage of UV blocked was not significantly different between sunscreens with and without PEG100 (ANOVA interaction *F* (4, 190) = 0.1524, *p* = .9593) (**Figure 6**).

DISCUSSION

While the FDA recommends sunscreen storage temperature, there is no information regarding what happens if sunscreen is stored outside of its range (9). Sunscreen is beneficial in preventing the risk of skin cancer. Therefore, it is vital for sunscreen users to know the consequences of storing sunscreen at extreme temperatures. However, there is very little research on the consequences of improper sunscreen storage. This study examined the consequences of 18-24 hours extreme temperatures for a wide variety of sunscreens. We examined the percent of UV blocked by sunscreens stored at various temperatures. The major finding is that sunscreens stored at 50°C blocked significantly less UV than sunscreens stored at -10°C. There were no observed significant interactions between temperature and any of the sunscreen characteristics. This reflected that while there was a difference in sunscreen efficacy between storage at -10°C and 50°C, none of the studied sunscreen characteristics are responsible for this difference.

In 2010, a study examined the efficacy of sunscreen with an SPF of 8 (19). The sunscreen in its original packaging was stored at 3 different temperatures (25°C, 29°C, and 40°C) over the course of 15 days, and they found no significant changes in efficacy (19). These results are similar to our own study; however, this study did not test sunscreen stored at the extreme temperatures -10°C and 50°C, as at 50°C our study found significant change compared to -10°C. Additionally, our study used 22 sunscreens at SPF 30 compared to the one sunscreen at SPF 8 that was used in the 2010 study.

In our study, we observed that the following sunscreens had a phase change after being stored at 50°C: Cetaphil Sun, Blue Lizard, Badger, Hawaiian Tropics, Coppertone, Cocokind, and Thinkbaby. Sunscreens stored at room temperature were smooth and formed a creamy texture when spread out. When these seven sunscreens were stored at 50°C, they had inconsistent texture and would not spread evenly on the plastic wrap due to the grainy, non-homogenous, texture. We believe that the phase change observed with some sunscreens at 50°C could be a major cause of the lower effectiveness of sunscreens at this temperature. The largest composition of sunscreen is usually formulation stabilizers, preventing the sunscreen consistency from being too thick (10). Another previous study that looked at the temperature stability of sunscreens examined the physical characteristics of nine sunscreen stored at extreme temperatures (16). This study saw a similar result as ours, where two of the nine sunscreens had a phase change after being stored at 60°C and there was no observed difference in sunscreens that had been frozen (16). However, in the 2010 Rego study they homogenized the sunscreen after temperature exposure and before analysis (19). If a phase change did occur, then homogenization could have recombined the separated sunscreen.

Emulsifiers allow oily and watery substances to be mixed to form a creamy texture (10). The melting temperature of some emulsifiers, such as PEG-100, is 47°C (10). Therefore, the observed temperature partial inactivation at 50°C could be related to melting emulsifiers. Although, we did not see a significant interaction between the percentage of UV blocked in sunscreens with PEG-100 and without PEG-100. Therefore, it is possible that another emulsifier could be causing the phase change. Future studies could test for a change in sunscreen effectiveness of the same sunscreen before and after a heat related phase-change.

There were several limitations of this study that need to be considered when interpreting the results. First, we used a previously published method of spreading sunscreen on plastic wrap. Overall, this method worked, but it was challenging to keep the sunscreen thickness consistent because it would slide around the plastic wrap. Second, the UV meter was only capable of measuring light from 290-390 nanometers (nm); therefore, UVB light between 280-290 nm and UVA light between 390-400 nm could not be detected by this UV meter. While we did not have access to a spectrophotometer

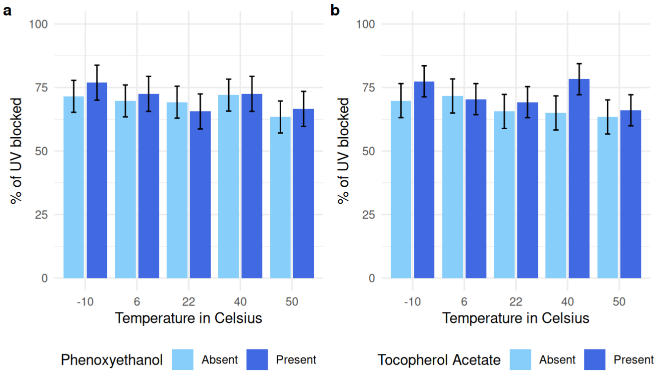

Figure 5: Temperature partial inactivation with and without phenoxyethanol and tocopheryl acetate. A bar graph shows the estimated marginal mean of percentage of UV blocked. These are ANOVA interaction plots. There was no significant interaction between temperature and a) phenoxyethanol or b) tocopheryl acetate. The bars represent 95% confidence intervals and the mean percentage of UV blocked. 15 sunscreens had tocopherol acetate (dark blue bar is tocopherol acetate present, light blue bar is tocopherol acetate absent), 2 had phenoxyethanol (dark blue bar is phenoxyethanol present, light blue bar is phenoxyethanol absent).

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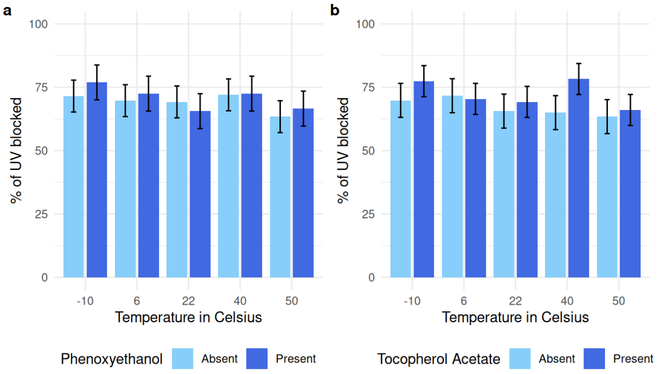

Figure 6: Temperature partial inactivation of sunscreen with and without cetyl alcohol and PEG-100. A bar graph shows the estimated marginal mean of percentage of UV blocked. These are ANOVA interaction plots. There was no significant interaction between temperature and a) cetyl alcohol or b) PEG-100. The bars represent 95% confidence intervals and the mean percentage of UV blocked.10 sunscreens had PEG-100, 12 did not (dark blue bar is PEG100 present, light blue bar is PEG100 absent). 10 sunscreens had cetyl alcohol and 12 did not (dark blue bar is cetyl alcohol present, light blue bar is cetyl alcohol absent).

in this study, we acknowledge that the UV meter is a much more limited tool than a spectrophotometer. Ideally, this study should be replicated with a spectrophotometer to measure the sunscreen performance in the full UV spectrum.

Third, the sunscreens froze completely solid in the freezer, and we had to allow extra time for them to thaw thoroughly before testing them. It is possible that this thawing period changed the sunscreen, and not the freezing period. Also, we used small aliquots of sunscreen. It is possible that the results may have been impacted due to a small aliquot in a 15 mL Falcon tube being tested instead of the original quantity and packaging. These smaller aliquots could freeze faster than a large bottle of sunscreen. Additionally, light from a transilluminator and light from the sun are different, and some sunscreens may perform better with solar UV.

Finally, and most importantly, this experiment spread sunscreens on plastic, which is obviously different from human skin. A sunscreen's performance on a plastic film might differ dramatically from its performance on the skin. An *in vivo* experiment would have practical and ethical hurdles. Skin has a different texture from plastic, more layers, and a more structured foundation. For all these reasons, we believe that an experiment in an *in vitro* skin model might have different results. For future experiments, there are new materials that are being used to mimic human skin. These *in vitro* models have features that replicate all layers of the skin, blood flow, and sweat glands (20).

While this experiment did use 22 sunscreens, there are many future directions to study this effect in a larger variety of sunscreens. For instance, both block sunscreens and spray-sunscreens were not used in this study but could be included in future experiments. This might be especially interesting considering these sunscreens are not a creamy texture and therefore use very different emulsifiers, which might lead to different temperature stability. Other sunscreen SPFs that are less commonly used could be tested to see the effect under the same conditions. We saw a different result between 40°C and 50°C, but the exact partial inactivation temperature was not discovered. Therefore, we could test smaller increments of temperatures in this range to find the exact partial inactivation temperature of each sunscreen. We were not expecting to see the result of the phase change at 50°C, therefore, we did not closely examine the entire aliquot of sunscreen. Future experiments should look carefully for this phase change, as was done in the Jung study (16). Finally, the 2010 Rego study exposed sunscreen to heat while it was in its original packaging, which might have influenced the results (19). Therefore, future studies could use original packaging instead of aliquots.

While the FDA recommends that sunscreens are stored at room temperature, it does not report what happens when sunscreens are not stored in ideal conditions. We examined the efficacy of sunscreen stored at extreme temperatures and found that sunscreen stored at 50°C for 18-24 hours was less effective than other conditions. This experiment raises awareness of which extreme temperatures inactivate sunscreen and why it is so essential to keep sunscreen in proper storage conditions. We hope that this study will raise awareness of the dangers of leaving sunscreen in extremely hot temperatures.

MATERIALS AND METHODS

Sunscreen preparation

We obtained 12 mineral and 10 chemical sunscreens with SPF 30 in lotion form (**Table S1**). Sunscreens were stored at roughly room temperature (22°C) before being brought into the testing environment. 10 mL of each sunscreen were put into 15 mL Falcon tubes (Fisher Scientific) at room temperature (22°C) for the control group, and for the experimental groups 10 mL of each sunscreen was added to different falcon tubes and incubated at -10°C, 4°C, 40°C, and 50°C. The lower temperatures were selected to mimic what temperatures could be expected for sunscreen use in winter sports. The higher temperatures were selected based on modeling the expected temperatures outside or inside of a car on a hot day. The control tubes were stored at room temperature at 22°C. The experimental tubes were placed in a -10°C freezer, a 4°C refrigerator, and in a water bath at 40°C and 50°C for 18-24 hours. After treatment, all sunscreens were left at room temperature for two hours.

UV meter detection

A clean square 12'' x 12'' piece of plastic wrap was placed on the Transilluminator (Benchmark Scientific, Compact). While wearing protective gear, the Transilluminator was turned on and we measured the UV light passing through the plastic bag with a UV detector meter as an initial reading. The transilluminator created light with wavelength of 312 nm. After the initial reading was taken, the transilluminator was turned off. Approximately 10 mL of each sunscreen was measured and spread evenly on one side of the plastic wrap. Sunscreen consistency was measured by observing characteristics of sunscreen spread on plastic wrap. A thin sunscreen spread on plastic wrap without manipulation. A medium sunscreen was able to be spread with manipulation. A thick sunscreen did not spread at all on plastic wrap. The transilluminator was turned on and the UV light passing through the sunscreen was measured using a UV detector meter until there was a stable reading for at least 15 seconds. The experiment was repeated two times. The results were analyzed using Rstudio (21).

UV percentage calculation

The outcome variable was the percentage of UV blocked. The maximum amount of UV from the transilluminator was 0.6. The percentage of UV blocked was calculated as:

$$percent\ UV\ blocked = \frac{1 - UV\ penetrance}{0.6} \times 100\%$$

Statistical analysis

A repeated measures ANOVA model was used to run the comparisons between all the different means. The estimated marginal means of the percentage of UV blocked were output for the ANOVA. Confidence intervals (CI) were set to 95%. Repeated measures were used to allow for multiple trials of the sunscreens. There was more than one of the same type of sunscreen in each model, therefore the random effect of sunscreen was used. A random effect is used in repeated measures ANOVA to account for the fact that one subject (sunscreen) was measured multiple times, and therefore it's not independently contributing new information to the model each measurement (22). The primary test calculated the estimated marginal mean (95% CI) of UV blocked for each of the five temperatures. Secondary tests were interested in what characteristics influenced sunscreen partial inactivation at different storage temperatures. A repeated measures ANOVA model was used where the interaction between characteristics and temperature was reported (22). The estimated marginal means for each temperature and sunscreen characteristic was recorded. The characteristics included broad-spectrum, chemical or mineral, water resistance, avobenzone, phenoxyethanol, tocopheryl acetate, PEG-100, consistency, and cetyl alcohol. After the test was run, an interaction p-value was recorded. The lmer function in R software was used for all ANOVA models, version 4.5.2 (21).

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APPENDIX

Supplementary Table

Name	Active Ingredients (% by weight)	Chemical /Mineral	Broad Spectrum	Water Resistant	Consistency	PEG 100	Cetyl Alcohol	Tocopheryl Acetate	Phenoxy ethanol
Australian Gold	Titanium Dioxide; Zinc Oxide	Mineral	Broad spectrum	Water resistant	Medium	Present	Absent	Present	Absent
Australian Gold Extreme Sport	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Thick	Absent	Present	Present	Present
Aveeno Protect + Hydrate	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Thin	Absent	Absent	Absent	Present
Badger	Zinc Oxide	Mineral	Broad spectrum	Water resistant	Thin	Absent	Absent	Present	Absent
Banana Boat Sport Ultra	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Thick	Present	Present	Present	Present
Bare Mineral Republic	Titanium Dioxide; Zinc Oxide	Mineral	Broad spectrum	Water resistant	Medium	Absent	Absent	Absent	Absent
Blue Lizard	Titanium Dioxide; Zinc Oxide	Mineral	Broad spectrum	Not water resistant	Medium	Absent	Absent	Absent	Present
COOLA	Avobenzone; Homosalate; Octisalate	Chemical	Not broad spectrum	Water resistant	Thin	Absent	Absent	Absent	Absent
CereVe	Homosalate; Octisalate; Octocrylene	Chemical	Broad spectrum	Water resistant	Thin	Absent	Absent	Absent	Absent
Cetaphil Sun	Titanium Dioxide; Zinc Oxide	Mineral	Broad spectrum	Water resistant	Thick	Present	Absent	Present	Absent
Cocokind	Zinc Oxide	Mineral	Not broad spectrum	Not water resistant	Thin	Absent	Absent	Absent	Absent
Coppertone	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Medium	Present	Absent	Present	Present
Eucerin	Ensulizole; Octinoxate; Octisalate	Chemical	Broad spectrum	Not water resistant	Thin	Absent	Present	Absent	Present
Hawaiian Tropics	Avobenzone; Homosalate; Octocrylene	Chemical	Broad spectrum	Water resistant	Medium	Absent	Absent	Present	Present
NO-AD Sunscreen	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Medium	Absent	Absent	Present	Present
Native Coconut and Pineapple	Zinc Oxide	Mineral	Broad spectrum	Not water resistant	Medium	Absent	Present	Present	Absent
Native Unscented Sunscreen	Zinc Oxide	Mineral	Broad spectrum	Not water resistant	Medium	Absent	Present	Present	Absent
Neutrogena Clear Face Oil Free	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Thin	Absent	Absent	Absent	Present
Neutrogena Ultra Sheer Dry Touch	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Medium	Present	Absent	Absent	Absent
Sun Bum	Avobenzone; Homosalate; Octisalate	Chemical	Broad spectrum	Water resistant	Medium	Present	Absent	Absent	Absent

Table S1: A summary of the sunscreens studied. This table shows the characteristics of each of the 22 sunscreens studied. The table includes the sunscreen name, active ingredients, chemical or mineral formulation, thickness, and whether it was broad spectrum, water resistance, and of the following ingredients were present: tocopherol acetate, PEG-100, cetyl alcohol and phenoxyethanol.

R Studio Code

```
#install.packages("purrr")
library(purrr)
library(dplyr)
library(ggplot2)
library(readr)
#code to load data files
Sunscreen_Trial_1 <- read_csv("/Users/sophiaspencer/Desktop/SCIENCE/LACSEF 2024-2025/Sunscreen - Trial 1.csv")
View(Sunscreen_Trial_1)
Sunscreen_Trial_2 <- read_csv("/Users/sophiaspencer/Desktop/SCIENCE/LACSEF 2024-2025/Sunscreen - Trial 2.csv")
Ingredient <- read_csv("/Users/sophiaspencer/Desktop/SCIENCE/LACSEF 2024-2025/Sunscreen Ingredient Data.csv")
merged_data <- reduce(list(Sunscreen_Trial_1, Sunscreen_Trial_2), full_join, by = "Number")
summary(merged_data2)
# Rename the columns
Sunscreen_long <- Sunscreen_Trial_1 %>%
rename (
 Freezer="Freezer T1",
 Fridge="Fridge T1",
 Temp_50="Temp 50 T1",
 Temp_40="Temp 40 T1",
Impute_Control="Impute Control T1",
Control="Control T1",
Time_in_fridge="Time in fridge T1",
Time_out_Fridge="Time out Fridge T1",
Fridge_Temp="Fridge Temp T1",
Time_in_freezer="Time in freezer T1",
Time_out_freezer="Time out freezer T1",
Freezer_Temp="Freezer Temp T1",
Time_in_50="Time in 50 T1",
Time_out_50="Time out 50 T1",
Time_in_40="Tme in 40 T1",
Time_out_40="Time out 40 T1"
)
summary(Sunscreen_Trial_1)
# Show all variable names in the data frame
variable_names <- names(Sunscreen_Trial_1)

# Print the variable names
print(variable_names)

Sunscreen_long_T2 <- Sunscreen_Trial_2 %>%
rename (
 Freezer="Freezer T2",
 Fridge="Fridge T2",
 Temp_50="Temp 50 T2",
 Temp_40="Temp 40 T2",
Impute_Control="Impute Control T2",
Control="Control T2",
Time_in_fridge="Time in fridge T2",
Time_out_Fridge="Time out Fridge T2",
Fridge_Temp="Fridge Temp T2",
Time_in_freezer="Time in freezer T2",
Time_out_freezer="Time out freezer T2",
Freezer_Temp="Freezer Temp T2",
Time_in_50="Time put in50 T2",
Time_out_50="Time out 50 T2",
Time_in_40="Time in 40 T2",
Time_out_40="Time out 40 T2"
)

sunscreen_append <- bind_rows(Sunscreen_long, Sunscreen_long_T2)
sunscreen_append <- reduce(list(sunscreen_append, Ingredient), full_join, by = "Number")
```

https://doi.org/10.59720/25-191

```
library(lme4)
library(dplyr)
#install.packages("lme4")
#install.packages("Matrix")
library(Matrix)
#building statistical model to test sunscreen inactivation
sunscreen_append$Consistency <- as.factor(sunscreen_append$Consistency)
sunscreen_append$ Chemical_Mineral <- (sunscreen_append$ Chemical_Mineral )
summary(sunscreen_append)
sunscreen_append$ Broad_Spectrum <- as.factor(sunscreen_append$ Broad_Spectrum )
sunscreen_append$ Active_1 <- as.factor(sunscreen_append$ Active_1)
sunscreen_append$ Brand <- as.factor(sunscreen_append$ Brand)
sunscreen_append <- sunscreen_append %>%
  mutate(diff40 = Temp_40 - Impute_Control)
#a positive value means more UV through the sunscreen than the control (inactivation)
#install.packages(tidyverse)
library(tidyverse)
# Assuming your dataset is named 'sunscreen_append'
# Convert the dataset to long format
long_sunscreen_append <- sunscreen_append %>%
  pivot_longer(cols = c(Fridge, Freezer, Temp_40, Temp_50, Impute_Control),
               names_to = "temperature",
               values_to = "UV") %>%
  mutate(temperature = case_when(
    temperature == "Impute_Control" ~ 22,
    temperature == "Freezer" ~ -10,
    temperature == "Fridge" ~ 6,
    temperature == "Temp_40" ~ 40,
    temperature == "Temp_50" ~ 50
  ))
long_sunscreen_append <- long_sunscreen_append %>%
  mutate(blockedUV = ((1 - (UV / 0.6)))*100)

# Add a variable for sunscreen id
long_sunscreen_append <- long_sunscreen_append %>%
  mutate(Sunscreen = row_number())

# View the transformed dataset
print(long_sunscreen_append)

long_sunscreen_append$temperature <- factor(long_sunscreen_append$temperature)
#install.packages("emmeans")
library(multcomp)
library(emmeans)
library(lme4)
# Fit the ANOVA model with random effects
long_sunscreen_append$temperature <- factor(long_sunscreen_append$temperature)
simplemodel1 <- lmer(blockedUV ~ temperature + (1|Number), data = long_sunscreen_append)
# Perform pairwise comparisons using emmeans
emm <- emmeans(simplemodel1, ~ temperature)
print(emm)
tukey_results <- pairs(emm, adjust = "tukey")
unadjusted_output <- anova(simplemodel1)
print(unadjusted_output)

# View the results
print(tukey_results)

emmeansresult <- emmeans(simplemodel1, "temperature")
print(emmeansresult)

#create mean graph, figure 1
```

```
resultdf <- as.data.frame((emmeansresult))
class(resultdf)
print(resultdf)
#install.packages("tidyverse")
library(tidyverse)
class(emmeansresult)
figure1 <- ggplot(resultdf, aes(x = temperature, y = emmean)) +
  geom_point(size = 3) +
  geom_errorbar(aes(ymin = lower.CL, ymax = upper.CL), width = 0.2) +
  labs(x = "Temperature in Celsius", y = "Percent of UV blocked") +
  ylim(50, 100)+
  theme_minimal()
figure1
model_int_bs <- lmer(blockedUV ~ temperature * Broad_Spectrum + (1|Number), data = Sunscreen_long_T2)
print(model_int_bs)
model_bs <- lmer(blockedUV ~ temperature + Broad_Spectrum + (1|Number), data = Sunscreen_long_T2)
print(model_bs)
anova(model_bs,model_int_bs)
bs_means <- emmeans(model_int_bs,~temperature *Broad_Spectrum)
print(bs_means)
# Convert emmeans object to a data frame
bs_df <- as.data.frame(bs_means)
bs_df_count <- Sunscreen_long_T2%>%
  group_by(temperature, Broad_Spectrum) %>%
  summarise(N = n())
print(bs_df_count)
figure2 <- ggplot(bs_df, aes(x = temperature, y = emmean, fill = factor(Broad_Spectrum))) +
  geom_bar(stat = "identity", position = position_dodge(width = 0.9), width = 0.8) +
  geom_errorbar(aes(ymin = lower.CL, ymax = upper.CL), position = position_dodge(width = 0.9), width = 0.2) +

  labs(
    x = "Temperature in Celsius",
    y = "% of UV blocked",
    fill = "Broad Spectrum") +
   scale_fill_manual(values = c("0" = "lightskyblue", "1" = "royalblue"),
                     labels = c("0" = "Absent", "1" = "Present")) +
  theme_minimal()+
  theme(legend.position = "bottom")
print(figure2)

library(tidyverse)
long_sunscreen_append <- long_sunscreen_append %>%
  mutate(Consistency = factor(tolower(as.character(Consistency ))))

model_int_C <- lmer(blockedUV ~ temperature * Consistency + (1|Number), data = long_sunscreen_append)
intC <- emmeans(model_int_C,~temperature * Consistency )
print(intC)
contrast(intC,interaction = "pairwise")
print(model_int_C)
model_C <- lmer(blockedUV ~ temperature + Consistency + (1|Number), data = long_sunscreen_append)
print(model_C)
modelC<-anova(model_C,model_int_C)
print(modelC)
C_means <- emmeans(model_int_C,~temperature * Consistency)
print(C_means)
# Convert emmeans object to a data frame
C_df <- as.data.frame(C_means)
#just out of curiosity are chemical and mineral sunscreen different ignoring temperature
#model_CM <- lmer(UV ~ Chemical_Mineral + (1|Number), data = Sunscreen_long_T2)
#emmeansresult <- emmeans(model_CM, "Chemical_Mineral")
#print(emmeansresult)
#print(model_CM)
```

```
# Load necessary libraries
library(lme4)
library(emmeans)
C_df_count <- long_sunscreen_append%>%
  group_by(temperature, Consistency) %>%
  summarise(N = n())
print(C_df_count)
figure4 <- ggplot(C_df, aes(x = temperature, y = emmean, fill = factor(Consistency))) +
  geom_bar(stat = "identity", position = position_dodge(width = 0.9), width = 0.8) +
  geom_errorbar(aes(ymin = lower.CL, ymax = upper.CL), position = position_dodge(width = 0.9), width = 0.2) +

  labs(
       x = "Temperature in Celsius",
       y = "% of UV blocked",
       fill = "Consistency") +
  theme_minimal()+
  theme(legend.position = "bottom")
print(figure4)
```