# CORRELATION OF SUN PROTECTION FACTOR OF SUNSCREEN WITH ABSORBANCE AND TRANSMITTANCE IN THE ULTRAVIOLET RANGE OF RADIATION

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## ABSTRACT

Introduction: Ultraviolet solar radiation (UV) is harmful to people both because of sunburn and because of much more serious health problems, among which skin cancer is one of the most serious consequences of exposure to UVA and UVB radiation. Sunscreens protect the skin from harmful solar radiation, by absorbing or blocking UV radiation. The aim of the paper is to determine the correlation between the sun protection factor (SPF) and the absorbance of UV radiation of a domestic brand of sunscreen, and to compare the effectiveness of the domestic brand of sunscreen with other commercially available brands.

Method: The UV-Vis spectrophotometric method was used to determine the correlation of the sun protection factor (SPF) with the absorbance and transmittance of UVA and UVB radiation.

Results: The results showed a very good correlation and linear dependence of SPF with absorbance in the UVA ($R^2$ = 0.993) and in the UVB area ($R^2$ = 0.998). A statistically significant difference ($P < 0.0001$) was observed in the absorbance of UVA and UVB radiation of different brands of sun protection creams with the same protection factor.

Conclusion: This study shows a direct correlation between SPF sunscreen and absorbance, which assesses the effectiveness of sunscreens in blocking UVA and UVB radiation. However, it is important to point out that all the researched creams, although they block different amounts of UV radiation, all absorb both UVA and UVB rays.

**Keywords:** Ultraviolet radiation, sun protection factor, absorbance, transmittance

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## INTRODUCTION

The sun is a necessity for life, but there is more and more scientific evidence about the harmful effect of solar radiation, especially ultraviolet A (UVA) and ultraviolet B (UVB) on the skin, and therefore on overall human health. The ultraviolet (UV) spectrum of solar radiation has a range of wavelengths from 200-400 nm, which is divided into UVC (200-280 nm), UVB (280-315 nm) and (UVA (320-400 nm)) range.

However, since UVC radiation has the shortest wavelength and the highest energy, it is blocked by the ozone layer and cannot reach the Earth, so it does not have any harmful effects on humans. UVA and UVB rays can penetrate through the ozone layer and reach the earth's surface, especially in the last few decades due to damage to the ozone layer, the planet has been even more intensely exposed to UV radiation (1). The extent to which UV radiation would have negative effects on the skin depends on the exposure to UV radiation and the body's tolerance limit (2). Chronic exposure to UV radiation leads to immunosuppression, photoaging, and carcinogenesis. It involves modulation of the immune system, accumulation of genetic changes and ultimately leads to cancer. Research shows that 90 % of all skin cancers are related to exposure to harmful UV radiation from the sun. UVB radiation is more cytotoxic than UVA radiation because it causes direct DNA damage through absorption of photons by cyclobutane pyrimidine dimers that ultimately induce mutagenesis and skin cancer.

It has been described how UV radiation in addition stimulates the synthesis of reactive oxygen species (ROS). These reactive species damage mitochondrial enzymes and plasma membranes, thereby reducing the concentration of antioxidant substances in the skin. Furthermore, oxidative stress, by damaging the macromolecules of the skin, leads to the loss of cellular function. UVA radiation causes increased oxidative stress compared to UVB radiation,due to is deeper penetrating spectral range (3-12). It is particularly important to point out that exposure to UV radiation during childhood and adolescence is the main etiological cause of skin cancer (13).

In order to protect the skin from UV radiation, different formulations were originally developed such as sunscreens, lotions, etc. Later, these formulations were refined to provide protection against other harmful effects of UV radiation (skin aging, pigmentation, loss of collagen, skin cancer) (14,15). The sun protection factor (SPF) labeled on sunscreen products determines the level of protection against erythema caused by UV radiation.

Research shows that the use of sunscreens with a certain SPF has a wide spectrum of protection against UV radiation, such as protection against burns, photoaging, dyspigmentation, DNA damage, prevention of immunosuppression, photocarcinogenesis, and antioxidant protection (16-18). Therefore, sunscreens have become widely used for the prevention of short-term and long-term skin damage, and for this reason consumers are offered a wide selection of cosmetic products with protection against UV radiation.

Although SPF was recognized by the FDA (Food and Drug Administration) as a standard for measuring sun protection as far back as 1978, new research is still

necessary in order to synthesize photoprotective ingredients in sunscreens (19). Understanding the chemical properties and pharmacology of sunscreens is necessary for the development of better formulations of UV protection agents (20,21).

Namely, it is important that photoprotective sunscreens must be photostable, chemically inert, non-irritating and non-toxic, and must also ensure the removal of singlet oxygen and other ROS in order to provide complete protection against solar UV radiation (22-25). Photoprotection provided by topical sunscreen against exposure to solar UV radiation can be determined *in vivo* and/or *in vitro* (26,27).

Although these methods correlate in some cases, it should be noted that for a number of products the SPF determined *in vivo* is much higher than the SPF *in vitro*. (28). However, although the *in vivo* method is useful and precise, it is a time-consuming, complex (exposure of volunteers to UV radiation) and expensive process, especially when information on protection from long wavelengths (UVA) is needed. In vitro methods are faster, more economical and simpler. Therefore, various analytical methods (UV spectroscopy, HPLC methods, etc.) were investigated in order to accurately and precisely determine the protection factor and SPF values. (29-32).

## RESEARCH AIM

This preliminary research has two goals. The first is to determine the correlation between the SPF marked on the product and the absorbance/transmittance in the UV area and the second is to determine a statistically significant difference in the absorbance of UV radiation of domestic brand sunscreens and other commercially available sunscreens on the market of Bosnia and Herzegovina. According to the knowledge and the available literature, this is the first research that has been conducted on commercially available sunscreens in Bosnia and Herzegovina, as well as in the region.

## MATERIAL AND METHODS

### Chemicals and samples

- Ethanol of analytical purity (p.a) - Fluka (96 %).

- Isopropanol of analytical purity (p.a) – Fluka

Domestic brand sun creams with different SPF (6, 15, 30, 50) and creams of different brands with the same SPF (50) were purchased in pharmacies and other stores in the territory of Bosnia and Herzegovina.

### Instruments and accessories

- VWR UV-1600 PC (China) spectrophotometer (single beam).Wavelength range 190-1100 nm.

- Hellma Analytics quartz cuvettes 10 mm (6030-UV 6030)

- Ultrasonic water bath (WUC – AO3H) – Witeg (Germany)

- Analytical balance - Adam Equipment PW 184 (accuracy 0.001 g).

Microsoft Excel 2019, Microsoft Office software package (Microsoft, USA) and MedCalc software (MedCalcStatistical Software Version 14.8.1) were used for statistical processing of the obtained results.

### Preparation of samples for absorbance measurement

For analysis, 0.02 g of the standard as well as a sample of sunscreen factor 6, 15, 30 and 50 were prepared. All samples were analyzed in triplicate. The prepared samples were dissolved in 100.00 mL of 70% ethanol. In order to improve the dissolution of the samples, the solutions were transferred to an ultrasonic water bath and subjected to ultrasound for the duration of 10 minutes.

The solutions were filtered through quantitative filter paper. The absorption spectrum in the ultra-violet region was measured with the prepared samples within 20 minutes of preparation. 70% ethanol was used as a blank. A blank sample is poured into the quartz cuvette up to the mark, the sample is poured into the second quartz cuvette and the absorbance is measured in the range of wavelengths from 200 to 400 nm with a shift of 5 nm. In this way, $\lambda_{max}$ is determined, i.e. the wavelength at which a solution of a certain SPF shows maximum absorbance.

The obtained absorbance was recorded and corrected to the same mass of 0.0200 g. The corrected absorbance is calculated according to the formula:

$$A = Measured\ A \cdot \frac{0{,}0200}{sample\ mass}$$

Finally, the mean value of the corrected absorbance of three measurements for each sample is calculated. The transmittance (T) of the sunscreen solution was calculated using the equation: $A = -\log T$. In the same way, 20 sunscreens of the same SP factor (50), but of different brands (B1-B20), were analyzed, and the absorbance at λmax in the UVA and UVB radiation range was determined.

## RESULTS

To determine the correlation between absorbance and transmittance depending on the SPF value, sunscreens of the same brand (standard brand) with different SPF values (6, 15, 30 50) were analyzed.

Figure 1. shows the UV absorption spectra of the tested sunscreens in the wavelength range of 220-400 nm. The three creams SPF 15, 30 and 50 have three significantly pronounced peaks in the wavelength range of 220-400 nm, indicating that they absorb UVC, UVB and UVA radiation. However, since UVC radiation does not reach the earth's surface, it was not further.considered. The protection factor six cream has the lowest pronounced peaks in both areas of UV radiation. The first peak of all samples is centered around 240 nm (UVC region). The second peak was centered around 300 nm (UVB region) for SPF 50, 30 and 15 and 310 nm for SPF six. The third peak for all samples is centered around 350 nm (UVA region). The UV spectra of four sun creams of different factors show that all creams absorb both UVB and UVA radiation, but with different intensities. As can be seen from the graph, the cream with protection factor 50 has the highest absorbance.

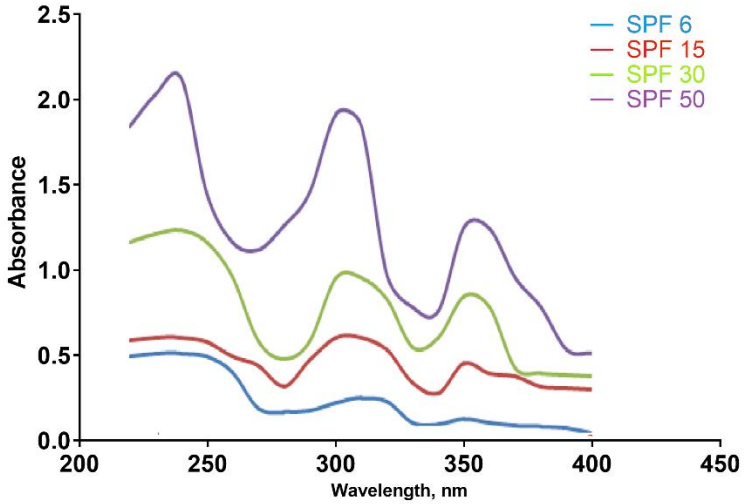


**Figure 1.** *UV absorption spectrum Absorbance versus wavelength 220-400 nm for SPF 6,15,30 and 50 standard brand (SB) creams*

The results of absorbance dependence on SPF (6, 15, 30 and 50) are shown in Figure 2. and Figure 3. The results show that there is a very good correlation and linear dependence of the absorption of sunscreen and SPF ($R^2$ = 0.998 and $R^2$ = 0.993). It is also clear from the graph that the researched standard brand has better absorption in UVB than in the UVA radiation range.

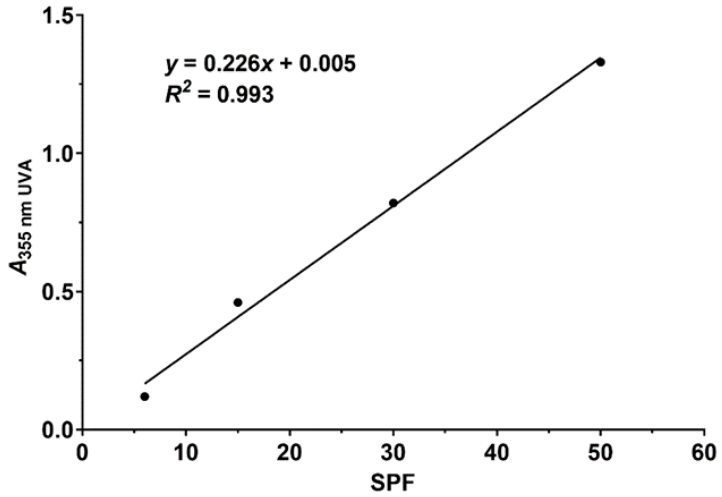


**Figure 2.** *Dependence of absorbance on SPF at (λ max ) in the UVA area*

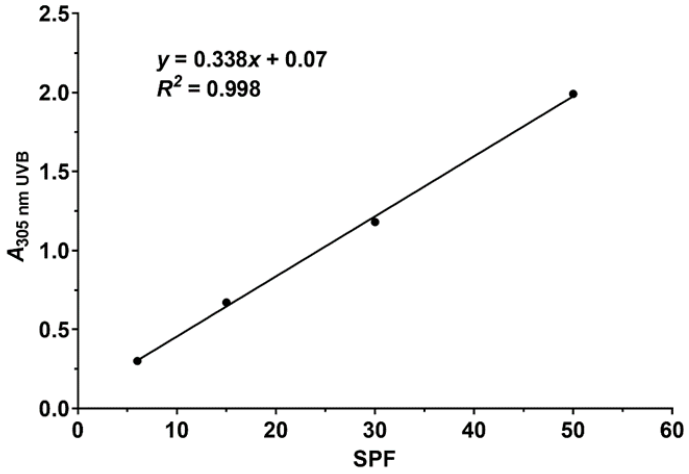


**Figure 3.** *Dependence of absorbance on SPF at (λ max) in the UVB area*

The transmittance results obtained (shown in Figures 4. and 5.) indicate a good correlation between absorbed and transmitted electromagnetic radiation (EMR). Namely, the higher the absorbance, the greater the protective effect of the sunscreen, which further indicates the fact that the transmittance is lower, that is, that less EMR penetrates the skin. The correlation coefficient obtained from the equation of direction is slightly better ($R^2$ = 0.9982) in the UVB area compared to the UVA ($R^2$ = 0.9829) area.
Since absorbance shows a linear dependence on SPF, and transmittance a logarithmic dependence, absorbance is the chosen quantity in evaluating the effectiveness of sunscreen. However, although transmittance is not linearly proportional to SPF, it can provide useful quantitative information as to what percentage of EMR is transmitted by a sunscreen with a specific SPF.

For example, from Figure 5. it can be determined that a cream with a protection factor of 6 transmits as much as 75.8% of UVA radiation, in contrast to a cream with a factor of 50, which transmits only 4.7% of UVA radiation. Or if we compare the same factor with different UV radiation, a cream with a protection factor of 50 only lets in 1% of UVB radiation, while the same cream lets in 4.7% of UVA radiation. These results indicate a good correlation between absorbance and transmittance, and the percentage of absorbed radiation can be unambiguously determined with this method. The higher the percentage of absorbed radiation, the greater the protective effect of the cream. So the SPF 50 cream absorbs as much as 99 % of UVB radiation.

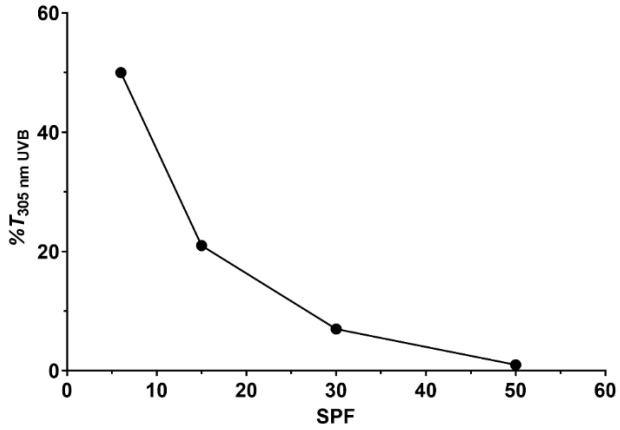


**Figure 4.** *Transmittance of sunscreens of the same brand at $\lambda_{max}$ in the UVB region*

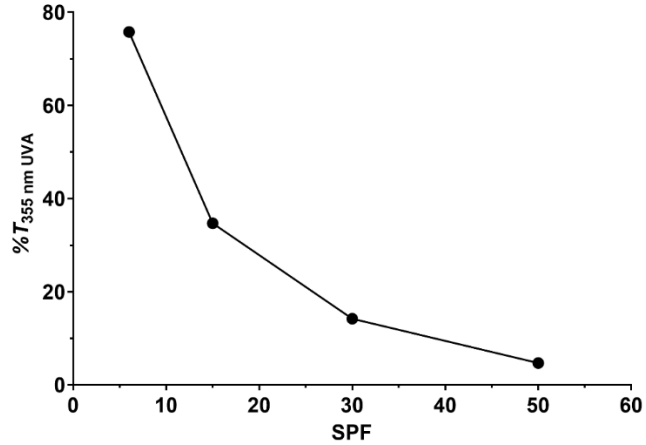


**Figure 5.** *Transmittance of sunscreens of the same brand at $\lambda_{max}$ in the UVA range*

Twenty different samples of sun protection creams (B1 to B20) with the same SPF 50 were analyzed using the described procedure, and were compared with the standard sun protection cream, i.e. the standard brand (SB). Figures 6. and 7. show the results of the obtained absorbance values in the UVB and UVA range of samples (B1-B20) SPF 50 compared to SB. The graphs show the mean values of three identical absorbance measurements with the associated standard deviations for different cream brands (SPF 50) in relation to the standard brand.

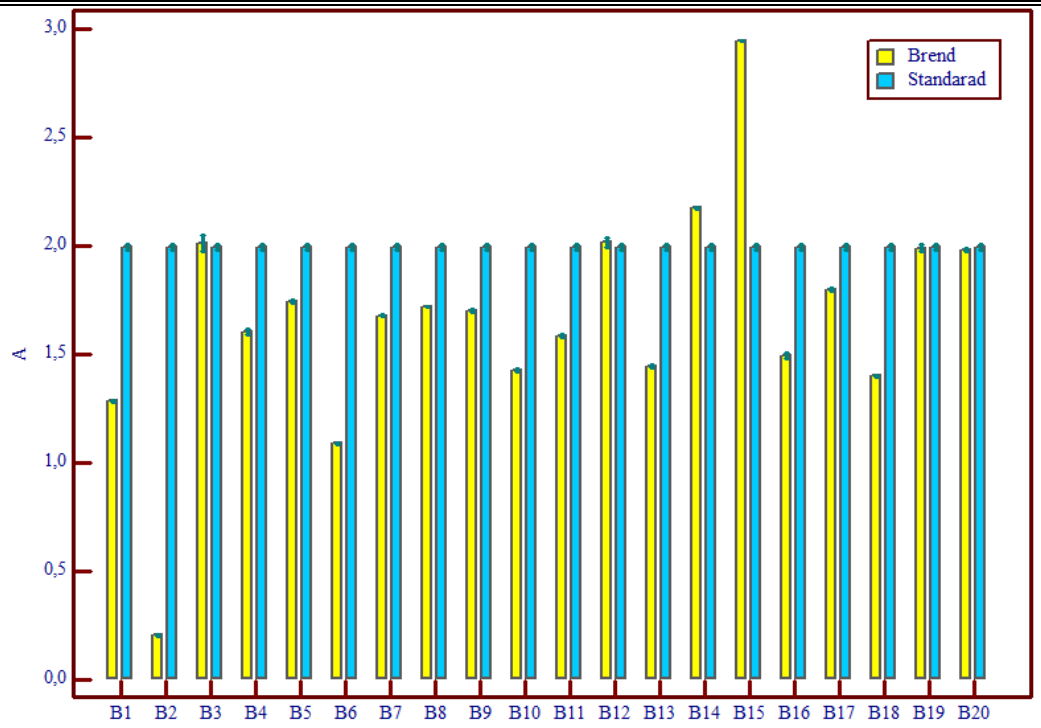


**Figure 6.** *Absorbance at $\lambda_{max}$ in the UVB area of different cream brands and SPF 50 standards*

The graph shown indicates the fact that there are evident differences in the absorbance of different samples in comparison with the selected standard in the UVB area. Namely, it can be observed that some standards (brands) have lower absorbances, among which B2 stands out the most, and on the other hand, sample 15 has a significantly higher absorbance in comparison to the standard. The results of the statistical analysis according to the SD (standard deviation) indicate that the measure of dispersion of the results is small, which further indicates the precision of the method. Statistical analysis revealed that four samples, brands (B3, B12, B19 and B20) do not show a statistically significant difference ($P>0.0001$), i.e. the protection in the UVB area of the mentioned brands is comparable to the standard brand, the other samples show statistical significance at level $P<0.0001$.

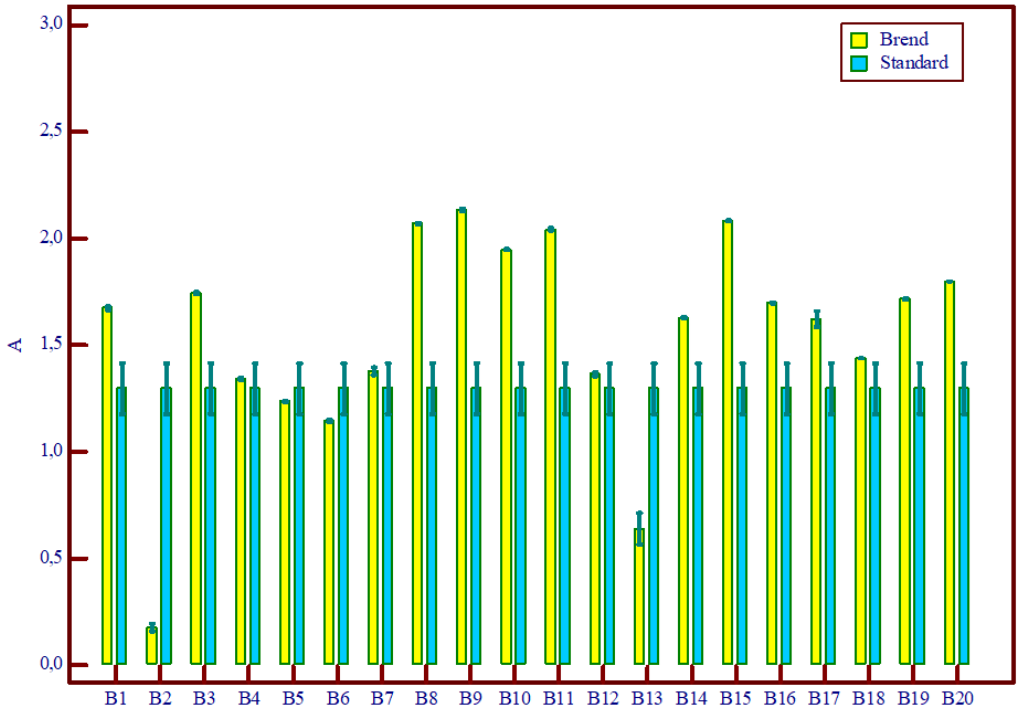


**Figure 7.** *Absorbance at $\lambda_{max}$ in the UVA range of different cream brands and SPF 50 standards*

Statistical analysis of 20 brands compared to the standard in the UVA area shows that six brands (B4; B5; B6; B7; B12; B18) do not show a statistically significant difference, i.e. the protection in the UVA area of the brands is comparable to the standard brand. The rest of the samples show statistical significance at the P<0.0001 level, the researched brands have a greater protective effect in the UVA area compared to the standard brand.

## DISCUSSION

The results of this research are in accordance with the research of Chou J, et al. (33). who also determined the linear dependence of absorbance and SPF, but the aforementioned authors conducted the research only in the UVB area and the correlation coefficient was $R^2$ = 0.998, which is in agreement with the results in this paper ($R^2$ = 0.9987). Furthermore, the difference between these two studies is in the use of different solvents, authors Chou J. et al. used isopropanol in their experiment. In this research, the spectrum was recorded with the solvent isopropanol (results not shown) and ethanol, but since there was no significant difference in absorbance depending on the solvent, ethanol was used in the experiment, because it was also used in other research, and on the other hand, it is financially more favorable. (34,35).

Authors Nalanda BR and Subhadip C. (36). determined the absorbance of sunscreens with different SPF values (15, 20, 24, 30, 50 and 60) using the spectrophotometric method, and also determined the correlation between absorbance and SPF ($R^2$ = 0.9908), which is slightly worse than that obtained in this research. Also, in the aforementioned research conducted on only six samples (of

different brands) that were compared with a standard sunscreen (Olay), it was determined that only two brands (33%) have similar absorbance to the standard sunscreen. In this work, four brands (20 %) did not have a statistically significant difference in absorbance in the UVB area, while in the UVA area six brands (30 %) did not have a statistically significant difference.

The results of this research show that the domestic brand (standard brand) has better efficiency in UVB compared to the UVA area, while the other investigated samples have better prection in the UVA area. According to the results of Miyamura Y, et al. (37) who found that UVB radiation is more cytotoxic than UVA radiation because it causes direct DNA damage through photon absorption by cyclobutane pyrimidine dimers that ultimately induce mutagenesis and skin cancer, the domestic brand in this research has a better protection effect compared to other commercially available sunscreens.
Observed differences in absorbance in both the UVA and UVB areas between different brands are probably due to the different chemical composition of the analyzed samples. Namely, most sunscreens contain a mixture of several active chemical substances, each of these absorbs light in different parts of UVA or UVB range. (38-41). Although there is a good correlation between SPF and absorbance, shown by the results presented in this paper as well as research by other authors, there may still be deviations from the indicated factor of a specific sample and the actual value.

Namely, the authors Fonseca AP and Rafaela N. (42) made a conclusion based on their research that all tested on their research concluded that all tested samples had a lower actual SPF compared to the indicated SPF values, especially for SPF 50 with a much larger difference, which is comparable to the results obtained in this paper. Namely, it is important to point out that, regardless of the different absorbance values between the domestic brand and other investigated brands, the results showed that only one sample (B2) has significantly lower absorbance in both UVA and UVB areas.

## CONCLUSION

This study shows a direct correlation between the absorbance, which evaluates the effectiveness of sun protection in blocking UVA and UVB radiation, and the SPF of the sunscreen. All analyzed creams with the same protection factor (50), even though they block different amounts of UV rays, all protect the skin from the penetration of harmful UVB and UVA rays into the skin. The applied spectrophotometric method is simple, fast, robust, sensitive, cheap and suitable for in vitro determination of the effectiveness of different sunscreens. Therefore, the developed method could provide a quick and useful alternative method for measuring the effectiveness of preparations for protecting the skin from UV radiation.

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# KORELACIJA FAKTORA ZAŠTITE OD SUNCA KREME ZA SUNČANJE S APSORPCIJOM I PROPUSNOSTI U ULTRAVIOLETNOM PODRUČJU ZRAČENJA

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## SAŽETAK

Uvod: Ultravioletno sunčevo zračenje (UV) je štetno za ljude kako zbog opeklina tako i zbog znatno ozbiljnijih zdravstvenih problema, među kojima je karcinom kože jedan od najozbiljnijih posljedica izlaganja UVA i UVB zračenju. Kreme za sunčanje štite kožu od štetnog sunčevog zračenja, jer apsorbiraju ili blokiraju UV zračenje.

Cilj rada je utvrditi korelaciju između zaštitnog faktora od sunca (SPF) i apsorbancije UV zračenja domaćeg brenda kreme za sunčanje, te usporediti učinkovitost kreme za sunčanje domaćeg brenda s drugim komercijalno dostupnim brendovima.

Metoda: UV-Vis spektrofotometrijska metoda je korištena za određivanje korelacie zaštitnog faktora (SPF) kreme za sunčanje s apsorbancijom i propusnosti UVA i UVB zračenja.

Rezultati: Rezultati su pokazali vrlo dobru korelaciju i linearnu ovisnost SPF s apsorbancijom u UVA ($R^2$ = 0,993) i u UVB području ($R^2$ = 0,998). Uočena je statistički značajna razlika (P< 0,0001) u apsorbanciji UVA i UVB zračenja različitih brendova krema za zaštitu od sunca s istim zaštitnim faktorom,

Zaključak: Ova studija pokazuje izravnu korelaciju između SPF kreme za sunčanje i apsorbancije koja procjenjuje učinkovitost krema za sunčanje u blokiranju UVA i UVB zračenja. No, važno je istaći kako sve istraživane kreme iako blokiraju različite količine UV zračenja, sve apsorbiraju i UVA i UVB zrake.

**Ključne riječi:** Ultra violetno zračenje, zaštitni faktor od sunca, apsorbancija, transmitancija

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