Determination of Titanium Dioxide in Commercial Sunscreens by Inductively Coupled Plasma–Optical Emission Spectrometry

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ABSTRACT

A variety of sunscreen products have been developed to afford the consumer protection against some of the deleterious effects, for example erythema, caused by solar ultraviolet radiation. The requirement that suncare products offer broad-spectrum protection has resulted in the use of inorganic oxides, such as *micronized* titanium dioxide (TiO_2), in their formulation. However, there are now concerns about the photocatalytic effects of the TiO_2 in these products and its potential phototoxicity to the skin through the generation of reactive oxygen species such as hydroxyl and superoxide anion radicals. It is important, therefore, that the amounts of TiO_2 in suncare products be closely monitored and maintained within stipulated ranges. A simple, fast and reliable analytical method for the determination of TiO_2 in commercial sunscreen products by inductively coupled plasma–optical emission spectrometry (ICP-OES) was developed and validated. The limits of detection and quantitation were found to be 0.018 and 0.062 μg mL^{-1}, respectively. The average percentage recovery of TiO_2 was 102.32 ± 2.87 % with a RSD of 2.81 %. The method was applied to determine the concentration of TiO_2 in 22 commercial suncare samples of which 14 contained TiO_2. The amounts of TiO_2 measured in these sunscreens ranged from 0.05 to 3.21 %. To our knowledge this is the first study that reports the amounts of TiO_2 in sunscreen products available on the South African market.

1. Introduction

Solar ultraviolet radiation (UVR) is indispensable for life on earth and yet it is responsible for a number of adverse effects on human skin. These include the chronic effects of photoaging and skin cancer, and the acute effects of sunburn in fair-skinned people, photosensitivity rashes and immunosuppresion.[1] The many harmful effects of sunlight have been the impetus behind the continuing search by scientists for an ideal photoprotectant.

Sunscreens were primarily designed for the prevention of sunburn but are now recognized as an important strategy in the prevention of squamous cell carcinoma, photoaging and UV-induced immunosuppression.[2–4] They contain organic and inorganic substances that absorb, reflect and scatter UVR, thus attenuating the nature and amount of UVR reaching viable cells in the skin. Sunscreen products are formulated in a variety of strengths termed sun protection factors (SPF). The SPF is the ratio of the energy required to produce minimal sunburn (skin reddening) in the presence of a sunscreen to the energy required to produce the same effect in the absence of a sunscreen.

Terrestrial solar ultraviolet radiation can be divided into two regions: the ultraviolet-B (UVB) region from 290 to 320 nm and the ultraviolet-A (UVA) region from 320 to 400 nm. The SPF is essentially a measure of the effect of UVB radiation on the skin since sunburn is primarily caused by UVB radiation. It is now appreciated that UVA radiation plays an important part in the induction of skin cancer, chronic photoaging, photo-immuno-suppression, and wrinkling of the skin.[5] Therefore, modern sunscreens must offer broad-spectrum protection against both the UVB and UVA wavelengths. In order to achieve broad-spectrum protection and boost the SPF of a formulation,[6] cosmetic manufacturers have turned to metal oxides such as *micronized* titanium dioxide (TiO_2), *ultrafine* zinc oxide (ZnO) and iron oxide (Fe_2O_3); with TiO_2 being the most commonly used. These oxides scatter, reflect and absorb UV radiation and are often termed 'physical blockers'.
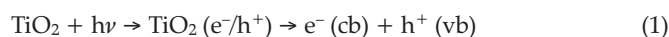
Titanium dioxide is effective in blocking out both the UVA and UVB wavelengths, and because of the micron size of TiO_2 it remains invisible on the skin. Rayleigh's theory best explains the optical behaviour of ultrafine TiO_2. It states that the intensity of light scattered is inversely proportional to the fourth power of the wavelength. Therefore, TiO_2 particles of size ranging from 20 to 50 nm effectively scatter UVA and absorb UVB radiation, and wavelengths approaching the visible region are transmitted through the material, making it virtually transparent to the naked eye.[7]

However, the two most common crystalline forms of TiO_2, *anatase* and *rutile,* are both photocatalysts and semiconductors with band-gap energies of about 3.23 eV and 3.06 eV, respectively, corresponding to light of 385 and 400 nm. Light at or below these wavelengths can excite electrons from the valence band (vb) to the conduction band (cb) generating single electrons (e$^-$) and positively charged spaces called holes (h$^+$). After formation, electrons and holes either recombine or migrate rapidly to the particle surface, where they now react with adsorbed species.

In an air-saturated aqueous environment, electrons react with

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oxygen, and h^+ with hydroxyl ions or water, forming superoxide or hydroxyl radicals:

$$TiO_2 + h\nu \rightarrow TiO_2 (e^-/h^+) \rightarrow e^- (cb) + h^+ (vb) \qquad (1)$$

$$e^- (cb) + O_2 \rightarrow O_2^{\cdot -} \qquad (2)$$

$$h^+ (vb) + OH^- \rightarrow {}^{\cdot}OH \qquad (3)$$

$$h^+ (vb) + H_2O \rightarrow {}^{\cdot}OH + H^+ \qquad (4)$$

The superoxide and, in particular, the hydroxyl radicals formed are the active agents for the degradation of organic compounds:

$${}^{\cdot}OH \text{ or } O_2^{\cdot -} + \text{substrate} \rightarrow \text{photo-oxidized products} \qquad (5)$$

Anatase is more photoactive than rutile.[8] This greater photoactivity has been ascribed to a lower rate of electron-hole recombination and a greater efficiency of oxygen adsorption. Therefore, rutile should be the preferred form of TiO_2 for use in sunscreen products. However, over-the-counter sunscreens were found to contain either anatase or rutile or a mixture of the two,[9] and Ricci *et al.*[10] state that anatase is the form used in sunscreen and cosmetic products. Brezová *et al.*[11] found that nine commercial sunscreen preparations containing TiO_2 did indeed generate reactive radical species, such as superoxide anion radical and hydroxyl radical, when irradiated with wavelengths of light longer than 300 nm.

Most sunscreen formulations employ a combination of physical and chemical (organic) UV-filters to provide broad-spectrum protection. Therefore, the possibility exists that TiO_2 can photocatalyze the degradation of the organic filters. Indeed it has been shown that TiO_2 can cause a significant loss of the most commonly used absorbers, namely avobenzone,[12] 2-ethyl-hexyl-*para*-methoxycinnamate,[10,13] oxybenzone,[10,14] octocrylene[10] and octyl salicylate.[10] Ricci *et al.*[10] further found that the process occurs more rapidly in the presence of a surfactant, which is often present in the formulation as an emulsifying agent. These interactions between components of the formulation reduce the efficacy of the product and lead to a loss of photoprotection and consequently more potential damage to the skin.

The ability of photo-excited TiO_2 to generate free radicals also accounts for its photocytotoxic effects.[9,15–23] Investigations by Dunford *et al.*[9] have shown that TiO_2 particles isolated from commercial sunscreen products induced DNA strand-breaks and other lesions in DNA plasmids and in human cells. Consequently, the presence of TiO_2 in sunscreen formulations can initiate or lead to photo-oxidative damage of the skin.

Moreover, because of the small size of these physical blockers it is possible that they may penetrate the skin *via* intercellular routes, or through hair follicles, pores and sweat glands. A number of studies have reported on the percutaneous absorption of microfine titanium dioxide[24–30] and although they reach different conclusions because of the different protocols used, titanium dioxide has been detected no further than the *stratum granulosum*.[30–34] The fact that microfine titanium dioxide does not appear to penetrate viable skin tissue is fortunate, since it has been found to be incorporated into cells[16,17] and once inside the cells it can trigger a chain of reactions (involving reactive oxygen species such as the hydroxyl radical) that can lead to DNA mutations and cell death.[35,36] Van der Molen *et al.*[37] showed that some forms of TiO_2 changed the integrity of the intercellular spaces of the *stratum corneum* severely. Such a scenario is highly undesirable. In addition, titanium dioxide has been found to be genotoxic.[38,39]

In order to prevent the deleterious effects enumerated above, the surface of the TiO_2 must be deactivated. Consequently,

coating of ultrafine TiO_2 is a topic of active research. The coating has the added advantage that it ensures good dispersability of the very fine particles. However, it has been noted that surface treatment of TiO_2 particles may or may not prevent photo-oxidative reactions, and that some coatings work better than others.[40–43] Doping of the crystal lattice with transition metal ions can also alter the photoactivity of TiO_2. However, the choice of dopant is critical as it can either enhance or reduce the photo-activity. Wakefield *et al.*[44] have shown that doping micronized titanium dioxide with 1 % manganese has a number of beneficial effects including enhanced UVA absorption, less degradation of other organic constituents of the formulation and a reduction in free radical generation.

Since coating or doping of TiO_2 nanoparticles cannot guarantee that they become photoinactive and hence ensure their safety, it is imperative that the levels of physical blockers incorporated in sunscreen formulations be closely monitored. In view of this, this study was undertaken to quantify the amounts of titanium dioxide in commercial sunscreens available on the South African market. The technique chosen to analyze the samples was inductively coupled plasma-optical emission spectrometry (ICP-OES), since ICP has a high sensitivity for refractory elements such as titanium and a wide dynamic range. To the best of our knowledge, this is the first such study to be carried out on suncare products commercially available in South Africa.

2. Experimental

2.1. Instrumentation

A JobinYvonn JY24, inductively coupled plasma–optical emission spectrophotometer equipped with a cross-flow nebulizer, was used. The experimental conditions were: wavelength, 337.280 nm; generator power, 1000 W; plasma gas flow rate, 12 L min^{-1}; auxiliary gas flow rate, 0.00 L min^{-1}; sheath gas flow rate, 0.2 L min^{-1}; nebulizer pressure, 3 bars; sample flow rate, 1 mL min^{-1}. The data were analyzed by using JYESS software.

2.2. Reagents and Samples

Titanium dioxide (99.8 % – Analytical Reagent Grade) from Riedel-de Haën A.G., Seelze-Hannover, was used for the preparation of standard solutions. Sulphuric acid (98.0 % – AnalaR) was supplied by Associated Chemical Enterprises (Pty) Ltd, Johannesburg, South Africa, and BDH Chemicals Ltd, Poole, England. Potassium hydrogen sulphate ($KHSO_4$) was supplied by BDH Chemicals Ltd, Poole, England.

Twenty-two commercial sunscreen products were purchased from retail shops in Durban, South Africa, and the selection included most of the popular brands on the market. The suncare products were labelled SA1-22.

2.3. Preparation of Standard Solutions

A 300 mg mL^{-1} stock solution of Ti^{4+} was prepared by dissolving 0.050 g of pure TiO_2 in 100 mL of hot, concentrated sulphuric acid. Working standard solutions were then prepared by serial dilution from a 9.5 mg mL^{-1} solution.

2.4. Sample Analyses

A sample mass of 0.150 g of each of the commercial sunscreen products was weighed into fused silica crucibles. The samples were placed in an electrical furnace with the temperature set at 600 °C. The samples were left in the furnace for three hours to give a carbon-free ash. After the three hours, the samples were allowed to cool before being taken for fusion in which a mass of

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0.50 g of KHSO$_4$ was added to the crucible. The crucible was heated over a Bunsen flame for approximately 10 minutes to fuse the mixture. The molten product was then dissolved in hot, concentrated sulphuric acid and the solution transferred to a beaker. Further heating of the solution was done to ensure complete solubilization of the TiO$_2$. Sample solutions were diluted with deionised water to 50 mL. Further dilution was done to 10 % (v/v) and the solutions were subjected to ICP-OES analysis.

2.5. Recovery Analysis

In order to validate the method used for the analysis of samples, a known amount of TiO$_2$ was spiked into a TiO$_2$-free commercial product. A mass of 0.150 g of the spiked sample was accurately weighed into a fused silica crucible. The crucible was then put in an electrical furnace at 600 °C for three hours after which it was taken out and left to cool. After the sample had cooled, 0.50 g of potassium hydrogen sulphate (KHSO$_4$) was added to the carbonless ash product for fusion over a Bunsen burner. The sample was heated slowly and then left on the burner for approximately ten minutes. The molten product was dissolved in hot, concentrated sulphuric acid and then diluted as required with deionized water. The diluted sample was subjected to ICP-OES analysis. The commercial sunscreen product chosen for the recovery analysis was initially analyzed to confirm the absence of TiO$_2$ in the same way as described in Section 2.4.

3. Results and Discussion

We developed an ICP-OES method for the determination of TiO$_2$ in commercial sunscreen products in which the number of steps involved in the dissolution of TiO$_2$ were minimised to avoid losses and increase sample throughput. In addition, the use of potentially hazardous hydrofluoric acid was avoided.

Standard solutions ranging from 0.475 to 9.512 μg mL^{-1} titanium were used for the calibration. The calibration graph was linear over this concentration range and the coefficient of determination (R^2) was >0.998 in all cases. Table 1 displays typical linear regression analysis results. The standard error of the slope of the calibration line was found to be 6.303. The sensitivity of the instrument (estimated as the mean of the slopes of calibration curves) was of the order of 10^3 μg mL^{-1}. The limit of detection (LOD) was calculated to be 0.018 μg mL^{-1}, determined as $3S_{y|x}b^{-1}$ ($S_{y|x}$ is the standard error of the slope of the calibration curve, and b is the slope). The limit of quantitation (LOQ) was calculated as 3.33 × LOD,[45] which was approximately 0.062 μg mL^{-1}. The within-day and between-day precisions,

Table 1 Typical results of the linear regression analysis of TiO$_2$ calibration data.

Parameters	Value
Calibration range/μg mL^{-1}	0.475–9.512
Slope (b)/10^3	1.026
Standard error of slope ($S_{y|x}$)	6.303
Correlation coefficient	0.999
LOD[a]/μg mL^{-1}	0.018
LOQ[b]/μg mL^{-1}	0.062

[a] LOD calculated as $3S_{y|x}b^{-1}$ ($S_{y|x}$ is the standard error of the slope and b is the slope of the calibration line.)
[b] LOQ calculated as 3.33 × LOD.

expressed in terms of the relative standard deviations of five replicates of a TiO$_2$ standard solution (9.5 μg mL^{-1}), were 1.60 and 7.2 % respectively, which are satisfactory. The mean recovery (of three replicate samples) for TiO$_2$ was 102.32 ± 2.87 % with an RSD of 2.81 %.

Some of the pertinent parameters of the method used in this work are compared in Table 2 with those of other techniques reported previously for the determination of TiO$_2$ in sunscreen products. Our method compares well overall with the other methods.

Of the 22 sunscreens analyzed, 14 contained TiO$_2$ in their formulation. Table 3 shows the results of the TiO$_2$ content in these 14 South African commercial sunscreens. The amounts of TiO$_2$ in all the samples analyzed are less than 5 %, with a minimum of 0.05 % and a maximum of 3.21 %. The low relative standard deviations (0.79 to 8.20 %) and the percentage recoveries achieved are good indications of the robustness of the analytical method. The concentration of TiO$_2$ in the suncare products was far below the European Cosmetics and Personal Care Association (COLIPA) limit of 25 %.[53] Unfortunately, the product labels did not contain any quantitative information that could be used for comparative purposes.

4. Conclusion

We have reported an improved method for the determination of TiO$_2$ in commercial suncare products. This method is simple, efficient and exhibits good precision and recovery, and the detection technique (ICP-OES) is selective for titanium. It was applied to the analysis of 22 commercial suncare products available on the South African market, of which 14 contained

Table 2 A comparison of different techniques reported for the determination of TiO$_2$ in sunscreen preparations.

Technique	Linear range	% Recovery	Limit of detection	Precision expressed as %RSD	Reference
ICP-OES	0.475–9.512 μg mL^{-1}	102.3 ± 2.9	0.018 μg mL^{-1}	1.6 (within-day) 7.2 (between-day)	This work
Volumetric	1–25 %	96–105			45
Spectrophotometry	0.1–6.0 μg mL^{-1}	97.2–98.5	0.1 %	0.62–1.85	46
Atomic absorption spectrophotometry	0–120 ppm				47
X-ray fluorescence spectrometry		>99.0	0.02 % by mass	<0.4	48
Energy dispersive X-ray fluorescence spectrometry	1–10 %		0.5 ± 0.06 %	2	49
ICP-OES	0.2–2 μg mL^{-1}	101 ± 4	0.035 μg mL^{-1}	0.6–5	50
ICP-OES		95.0 ± 2.6 %	0.2 μg g^{-1}	5.1	51

Table 3 Mean % (m/m) concentrations of TiO$_2$ measured in 14 sunscreens sold in South Africa.

Product	SPF No.	Country of manufacture	% TiO$_2$[a]	95 % confidence interval	% RSD
SA4	40	South Africa	3.21	±0.06	0.79
SA5	40	South Africa	1.41	±0.12	3.39
SA6	8	Greece	0.33	±0.05	5.78
SA7	15	Greece	1.14	±0.12	4.12
SA8	30	Greece	1.58	±0.08	2.03
SA9	40	Greece	1.09	±0.14	5.07
SA10	15	South Africa	1.42	±0.10	2.95
SA11	30	South Africa	1.28	±0.15	4.65
SA13	15	Germany	1.61	±0.17	4.25
SA14	30	Germany	1.79	±0.13	2.82
SA15	4	EU	0.05	±0.00	1.23
SA19	25	Germany	1.26	±0.26	8.20
SA21	40	South Africa	1.03	±0.11	5.78
SA22	30	South Africa	0.82	±0.06	3.12

[a] Each value is the mean of three replicates.

TiO$_2$. The amounts of TiO$_2$ measured in these sunscreens ranged from 0.05 to 3.21 %. This is far lower than the COLIPA-approved level of a maximum of 25 % in suncare products.[53] Judging from our results, we conclude that the levels of TiO$_2$ in sunscreen products available on the South African market are well within the internationally stipulated limits.

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