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RESEARCH REPORT

A pilot study on the percutaneous absorption of microfine titanium dioxide from sunscreens

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SUMMARY

Many Australians are being advised to apply microfine titanium dioxide sunscreen daily from the cradle to the grave. However, there is a surprising lack of data on the percutaneous absorption of microfine titanium dioxide. A prospective pilot study was conducted to analyse the percutaneous absorption of microfine titanium dioxide from sunscreens. Selected patients scheduled to have skin surgery, applied titanium dioxide sunscreen to the skin daily for 2–6 weeks prior to their operation. After excision, the stratum corneum of the sample was stripped and the titanium concentration of the remaining epidermis and dermis was measured by inductively coupled plasma-mass spectrometry. The results from this pilot study showed that levels of titanium in the epidermis and dermis of subjects who applied microfine titanium dioxide to their skin were higher than the levels of titanium found in controls. Studies with larger cohorts are necessary to establish if this absorption is statistically significant.

Key words: skin, toxicity, ultrafine.

INTRODUCTION

Many modern sunscreens contain microfine titanium dioxide. Titanium dioxide is suitable for use in sunscreen products as it scatters and reflects both ultra violet (UV) A light (wavelength 320–400 nm) and UVB light (wavelength 290–320 nm).[1] Micronizing and then microfining titanium dioxide produces an effective and cosmetically acceptable reflectant. Microfine particles may have a greater potential to be percutaneously absorbed compared with commercial grade titanium dioxide.

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Titanium dioxide is believed to be a compound of low potential toxicity although its metabolism and biological effects on human tissues are not clearly defined.[2,3] It is used in face powder, beauty creams, lipsticks, toothpaste, emulsion bases, ointments, a wide variety of ingested products, and sunscreens. Cutaneous exposure from skin products has been associated with metallic skin papules,[4] granulomatous inflammation and fibrosis.[5] Reports of granulomatous lung disease with alveolar metaplasia,[6] and black pigment deposition and cyst formation in association with artificial joints[7,8] and cardiac pacemakers[9] exist.

There is little recent clinical or toxicological information on the percutaneous behaviour of titanium dioxide, particularly in microfine form. Young Australians are being recommended to use these sunscreens lifelong on a daily basis. This might result in the annual application of kilograms of titanium dioxide sunscreens.

We have conducted a pilot study to determine whether titanium dioxide is percutaneously absorbed from microfine sunscreen.

MATERIALS AND METHODS

Patients

The subjects were 13 consecutive patients who presented between November 1993 and March 1994, scheduled to have surgery for skin lesions. Subjects were required to have an adequate amount of skin surplus removed during the operation in order to be able to provide a 4 mm punch biopsy. All subjects gave informed consent and the study was approved by the Westmead Hospital Human Ethics Review Committee.

Dosage and duration of therapy

Subjects applied sunscreen containing 8% microfine titanium dioxide complex in the morning and at midday to the skin surrounding the lesion to be excised, for a period of 2–6 weeks. This was ceased 2 days before their operation and the area was washed with soap and water twice per day. No other sunscreens, makeup, lotions, creams or cleansers were applied. The tubes of sunscreen were weighed before and after application to evaluate compliance.

Collection of samples

The skin for excision was vigorously scrubbed and cleaned with 70% isopropanol. After surgery, excess skin away from

the lesion was removed from the excised specimen to become samples for our analysis.

Controls

Ten cadaver skin specimens were obtained from hip skin at postmortem. These were used to evaluate the background levels of titanium normally present in skin from a body location not likely to have been exposed to titanium dioxide cream.

Removal of stratum corneum

Precautions were taken in collection and storage of samples to prevent titanium contamination. The stratum corneum was stripped using cyanoacrylate ester and elastic adhesive plaster in a modification of the skin surface biopsy technique described by Marks and Dawber.[10] A thin layer of cyanoacrylate ester was placed on the epidermis and elastic adhesive plaster was pressed onto this for 1–5 min and then rapidly pulled away, removing layers of stratum corneum cells. Previous histological studies (data not shown) demonstrated that we could completely remove the stratum corneum using two applications of cyanoacrylate ester for 5 min each. The skin sample was then placed upside down on a clean surface and a 4 mm punch taken. This allowed for a minimum of 10 mg to be analysed.

Tissue preparation

Approximately 10 mg wet weight of tissue was digested using 100 μL of H_2O_2 (AnalR, 30%; Crown Scientific, Moorebank, NSW, Australia) and 2 mL of concentrated HNO_3 (Suprapur, Merck Chemicals, Kilsyth, Vic., Australia) in a Milestone microwave system.

Chemical analysis

The diluted tissue digests were analysed for titanium levels on an Elan 5000 Inductively Coupled Plasma Mass Spectrometer (Perkin Elmer-Sciex, North Ryde, NSW, Australia) using Scandium 45 as an internal standard against a standard curve covering the range 5–25 μg/L titanium (Merck Chemicals Atomic Spectroscopy Standard, Kilsyth, Victoria, Australia).

Statistical analysis

Student's *t*-test was used to assess the statistical significance of differences in mean titanium concentration between patients and controls, and between sites of sunscreen application. Pearson's correlation coefficient was used to assess the relationship between duration of sunscreen application and titanium concentration in the skin.

RESULTS

The subjects consisted of four females and nine males, mean age 71 years (range 59–82), and all were Caucasians. One subject had three samples taken from three different lesions. Another subject had two samples taken from two different lesions. A total of 16 samples was obtained.

Subjects applied the titanium dioxide-containing sunscreen for a period ranging from 9–31 days. Subjects did not sustain any trauma or rash to the area to which the sunscreen was applied, and no adverse reactions to the sunscreen were recorded. We found that the levels of titanium in the dermis of subjects who had applied sunscreen containing microfine titanium dioxide to their skin were higher than the levels of titanium found in the cadaver controls (Fig. 1). The difference in these levels did not reach statistical significance ($P = 0.14$); analysis was confounded by the presence of one outlier cadaveric control sample with a titanium concentration twice as high as any other control sample (Fig. 1). If this one extreme value is excluded, there was a statistically significant difference in titanium concentrations between patients and controls ($P = 0.006$).

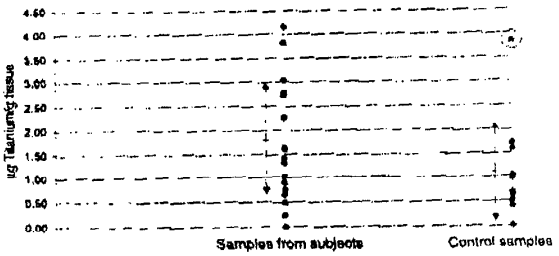

Figure 1 The concentration of titanium (μg titanium per gram wet weight tissue) is shown for each of the 16 samples and 9 cadaver controls. The double headed arrows display the mean (centre of arrows) and standard deviation (ends of arrows) of each distribution. The circled control point (top right) is the outlier control sample refered to in the text.

There was no correlation between the duration of sunscreen application and the concentration of titanium in the samples, and no relationship between the site of sunscreen application and concentration of titanium in the samples.

DISCUSSION

Percutaneous absorption is a process involving slow passive diffusion through the stratum corneum, followed by rapid diffusion through the viable epidermis and papillary dermis into the microcirculation of the skin.[11] Penetration through the stratum corneum is limited by molecular size. The average width of the intercellular space is approximately 100 nm.[12] The aqueous gap between lamellar bilayers that occupy the intercellular space measures only 0.5–1.0 nm.[13] However, these domains can enlarge into bigger clefts when filled with topically applied materials,[14,15] making possible the penetration of 10–50 nm particles of microfine titanium dioxide.

This pilot study has provided the first tentative evidence that the use of sunscreens containing microfine titanium dioxide can result in the percutaneous absorption of titanium. We found the concentration of titanium in the dermis of subjects exposed to microfine titanium dioxide was higher than that of control cadaver specimens unlikely to have been exposed. However, the pilot nature of this study has limitations: the sample size was small, the control group consists of unmatched cadaveric samples, and the magnitude of the differences between subjects and controls failed to reach statistical significance at the $P = 0.05$ level when all control samples were included. Although there was an outlier control specimen we are reluctant to exclude this data point without explanation.

The concentrations of titanium encountered in the tissue samples are close to, or at, the calculated analytical limits of detection of titanium even using the most sensitive available analytical techniques such as inductively coupled plasma-mass spectrometry. There are also potential analytical interferences to the measurement of titanium from minor isotopes of calcium, such as ^{44}Ca, ^{46}Ca and ^{48}Ca.

Other limitations include our use of elderly Australians, application of sunscreen to less than healthy skin, samples taken from different sites varying in surface topography, thickness of stratum corneum and number of skin appendages. All these factors may have influenced percutaneous absorption and, hence, the concentration of titanium detected in skin. The lack of dose-response relationship and the overlap between subjects and control data may also indicate that percutaneous absorption of titanium is a non-linear or idiosyncratic process.

Australians are being increasingly encouraged to adopt life-long use of sunscreen products, many of which contain microfine titanium dioxide. The difficulties of the experimental design notwithstanding, there is a need for others to follow up this pilot study and to demonstrate the safety and lack of toxicity from microfine titanium dioxide-containing sunscreen products.

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