Chapter 8
Toxic effects

Toxic and other adverse effects of sunscreens

In order for a sunscreen to have a toxic effect on living tissues, it must penetrate the skin. There is some evidence that this can occur (see p. 63 *et seq.*).

Human studies

No published studies of toxic effects in humans were available to the Working Group.

Contact sensitivity

There are numerous reports of cases of allergic reactions and photoreactivity to sunscreens, but the prevalence of this problem among sunscreen users is difficult to estimate. Since sunscreens are becoming more complex, with multiple active ingredients, fragances and other compounds, this problem could increase in the future. Reactions to sunscreens were found to be reasonably common among patients referred to a clinic because of suspected photosensitivity. Such patients are heavily exposed to sunscreen products and are thought to become more sensitive to chemicals than others (Green *et al.*, 1991; Bilsland & Ferguson, 1993; Stitt *et al.*, 1996; Berne & Ros, 1998).

The published reports of adverse effects range from case histories in one or several subjects (Schauder & Ippen, 1986; Knobler *et al.*, 1989; Motley & Reynolds, 1989; Murphy *et al.*, 1990; Torres & Correia, 1991; Buckley *et al.*, 1993; Collins & Ferguson, 1994; Kimura & Katoh, 1995; Parry *et al.*, 1995; Silva *et al.*, 1995; Marguery *et al.*, 1996; Ricci *et al.*, 1997; Zhang *et al.*, 1998) to studies of tens or hundreds of patients (Thune, 1984; English *et al.*, 1987; Lenique *et al.*, 1992; Szczurko *et al.*, 1994; Trevisi *et al.*, 1994; Gonçalo *et al.*, 1995; Ang *et al.*, 1998) and reviews (Dromgoole & Maibach, 1990; González & González, 1996; Schauder & Ippen, 1997). In the past, PABA and its esters were the most commonly reported contact and photoallergens in sunscreens (Funk *et al.*, 1997), and this finding contributed to a reduction in their use in sunscreens. The contact or photocontact allergen in sunscreens most frequently cited today is benzophenone-3, followed by dibenzoyl methanes. There have also been a few reports of contact allergy to excipients included in the formulations (Jeanmougin *et al.*, 1988; Nishioka *et al.*, 1995; Silvestre *et al.*, 1996). In a longitudinal, population-based study to reactions to sunscreens carried out in Australia, of the 603 people tested with a commonly used sunscreen formulation, 114 developed an adverse reaction (e.g. skin irritation). When they were patch tested, however, none was allergic to the active ingredients. A higher than expected proportion of the subjects who developed an adverse reaction had a personal history consistent with atopy (Foley *et al.*, 1993).

Overexposure to UVA

An obvious but not readily recognized adverse effect of sunscreens is interference with accommodation by the skin to UVR. Because most sunscreens absorb primarily UVB and, in some cases, short-wavelength UVAII (315–340 nm), the use of sunscreens changes the UVR spectrum to which the skin is exposed (Gasparro *et al.*, 1998). Since UVB is the primary stimulus for adaptation of the skin to sunlight, less adaptation might be expected to develop in individuals who use sunscreens regularly. The adaptive responses include thickening of the epidermis and transfer of melanin-containing granules to keratinocytes (tanning) (Fig. 44), which reduces the transparency of the skin to UVA and UVB (Fusaro *et al.*, 1966; Olson *et al.*, 1973). Several reports showed that UVR-induced injury, such as dermal connective tissue damage and sunburn cell formation, can occur in human epidermal cells in the absence of erythema and at doses that are far below the SPF of the sunscreen (Kaidbey, 1990; Kligman, 1997). Furthermore, prevention of sunburn by sunscreens may create a false sense of security, while allowing prolonged exposure to sunlight. An increasing number of studies indicate that, although UVB is the most damaging component of sunlight, UVA is responsible for numerous morphological, molecular and biochemical events which may contribute to photodamage of the skin (Kligman & Gebre, 1991; Scharffetter *et al.*, 1991; Wlaschek *et al.*, 1993; Lavker *et al.*, 1995b; Lavker & Kaidbey, 1997).

Vitamin D depletion

Vitamin D is produced when UVB absorbed by the epidermis causes 7-dehydroxycholesterol to form previtamin D_3, which isomerizes spontaneously to vitamin D_3 before entering the circulation, where it is metabolized by the liver into 25-hydroxyvitamin D_3 and by the

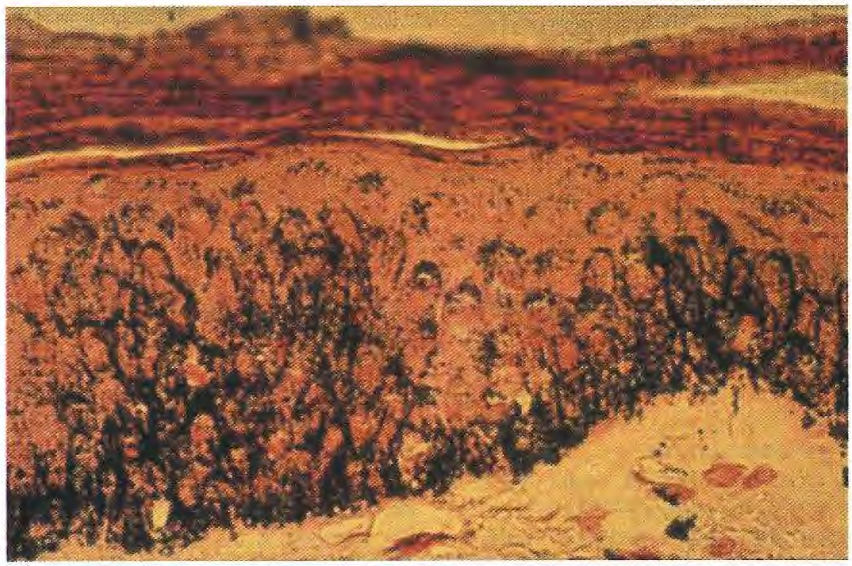

Figure 44 Increased melanin deposition induced by repeated exposure to the sun can be visualized throughout the epidermis by Fontana Masson staining

kidneys into 1,25-dihydroxyvitamin D_3. The latter is the most biologically active form. Vitamin D can also be supplied by the diet. With parathyroid hormone, it regulates calcium homeostasis. There has been concern that reduction of UVB absorption by the epidermis by sunscreen use could suppress vitamin D production, thus affecting calcium metabolism.

In one study, six women and two men received whole-body exposure to 1 MED UVR (Westinghouse sunlamps FS72T12, 260–360 nm) with or without protection from 5% PABA (SPF 8). The serum concentration of vitamin D in unprotected subjects increased from 1.5 ± 1.0 to 26 ± 6.7 ng/ml 24 h after exposure to UVR. In the sunscreen-protected volunteers, serum vitamin D was unaltered by exposure, with values of 5.6 ± 3.0 and 4.4 ± 2.4 ng/ml before and 24 h after exposure, respectively. PABA also completely inhibited previtamin D_3 production from 7-dehydroxycholesterol in triplicate samples of human skin exposed to UVR *in vitro* (Matsuoka *et al.*, 1987).

In a subsequent study, serum vitamin D was measured in groups of four healthy subjects 1 h before and 24 h after exposure to 0.8 MED from the same UVR source (Matsuoka *et al.*, 1990). The volunteers received either no sunscreen or sunscreen applied to increasing areas of the body. Whole-body protection completely prevented the UVR-induced increase in serum vitamin D, and selective protection of increasing skin areas correlated with the serum vitamin D concentration.

Twenty persons with a history of skin cancer (mean age, 64.6) who had been using PABA-based sunscreens for more than 1 year had a significantly lower serum vitamin D concentration (40 ± 3.2 nmol/L) than 20 healthy controls matched for age and exposure to sunlight (91 ± 6.2) (Matsuoka *et al.*, 1988).

In a study of eight patients with xeroderma pigmentosum who took extreme measures to protect themselves from light, including minimizing the time spent in sunlight, protective clothing and constant sunscreen use, the serum vitamin D concentration monitored over 6 years was found to be at the lower end of the normal range. Nevertheless, the serum calcium concentration was within the normal range, as was that of parathyroid hormone, which might have been expected to be increased if the vitamin D level was low (Sollitto *et al.*, 1997).

A randomized double-blind controlled trial of 113 healthy adults over 40 years of age who used a sunscreen or a placebo cream included analyses of serum vitamin D concentrations over 7 months, including summer (Marks *et al.*, 1995). The broad-spectrum sunscreen had an SPF of 17 and contained 8% ethylhexyl methoxycinnamate and 2% butyl methoxydibenzoylmethane and was applied to the head and neck, forearms and dorsum of each hand at least once a day. The concentrations of 25-hydroxyvitamin D_3 rose to a similar extent in the groups given sunscreen and placebo over the summer period, whereas those of 1,25-dihydroxyvitamin D_3 increased in the group given the placebo but not in those given the sunscreen, although they did not fall below the normal range. This suggests that although vitamin D synthesis was reduced by the sunscreen it was not reduced sufficiently to cause deficiency.

This finding is in agreement with that of another study (Farrerons *et al.*, 1998) in which serum vitamin D, parathyroid hormone and bone biological markers were assessed in 24 users of a sunscreen (SPF 15) and compared with those in 19 controls over 2 years. Whereas significantly lower levels of vitamin D were observed in the sunscreen users, there were no changes in parathyroid hormone or bone biological markers.

Experimental studies

Whole animals and cells

All UVR filters used in over-the-counter sunscreen products are subjected to extensive testing for toxicity and safety, and the results are evaluated by regulatory bodies, including the Scientific Committee on Cosmetics and Non-food Products for the Commission of the

European Union (Loprieno, 1992) and the Food and Drug Administration in the USA (Food & Drug Administration, 1999). Only compounds proven to be safe and without significant toxicological effects receive approval for use in sunscreens. This information is supplied to the regulatory bodies by manufacturers but is not publicly available and could therefore not be reviewed by the Working Group.

A study of the safety of benzophenone-3 found that it was practically non-toxic when administered orally to rats and was not toxic when applied to the skin of rabbits at doses up to 16 g/kg bw, with no significant lesions at autopsy (Cosmetic Ingredient Review, 1983). It did not irritate the skin or eyes of rabbits and was not phototoxic in guinea-pigs and rabbits when applied five times per week for 2 weeks. When dissolved in petroleum jelly base and applied topically to the skin of male Sprague-Dawley rats twice daily for 4 weeks at 100 mg/kg bw per day, benzophenone-3 did not cause any observable toxicity. There was no effect on body weight, organ:body weight ratios or haematological, clinical chemical or histological parameters (Okereke *et al.*, 1995). When fed to rats, it had an LD_{50} > 13 g/kg bw, and the no-effect level over 90 days of feeding was found to be 0.1%, corresponding to 0.33 g/kg bw per day (Lewerenz *et al.*, 1972).

Groups of Hr/Hr pigmented female hairless mice treated with 0.1 ml of a commercial sunscreen containing ethylhexyl methoxycinnamate and benzophenone-3 on 4 days/week for 12 months developed some toxic side-effects, including amyloidosis, eczema-like oedema and ulceration and pigment deposition (Wulf *et al.*, 1982). The specific component could not be identified. Application of the same sunscreen to the eyes of mice caused significant hyperplasia of the eyelid skin and acute inflammation of the cornea (Vangsted, 1985). A benzophenone-3-containing sunscreen was also reported to exacerbate dermal damage caused by chronic exposure to long-wavelength UVA (> 340 nm) (Kligman & Zheng, 1994).

Isoamyl-*para*-methoxycinnamate administered to pregnant Wistar rats on days 6–15 of gestation caused the death of 10% of the animals due to gastrointestinal erosion and haemorrhage when given at a dose of 2.25 but not 0.75 or 0.25 g/kg bw per day by intragastric instillation. The remainder of the animals at this dose lost weight and had reduced food and increased water consumption and hair loss. Mild hair loss and reduced food intake were observed in the group receiving 0.75 g/kg bw per day (Jekat *et al.*, 1992).

Moderate skin irritation was caused by octocrylene applied topically at a dose of 264 mg/kg bw per day to New Zealand white rabbits, but not at lower doses (Odio *et al.*, 1994). Reduced weight gain was also observed, but there were no macroscopic or histopathological abnormalities in blood cells, kidney or liver. Octocrylene did not induce mutation in lymphoma cells *in vitro*.

TiO_2-coated mica, 10–35 µm, added to the diet of Fischer 344 rats at up to 5% for 130 weeks did not cause consistent changes in any end-point studied, including survival, body-weight gain, haematological or clinical chemical parameters or histological appearance (Bernard *et al.*, 1990). Intratracheal exposure of rats to 2 mg of ultrafine TiO_2 particles (< 30 nm), however, caused inflammation and cytotoxicity to pulmonary alveolar macrophages (Afaq *et al.*, 1998).

In contrast, HeLa cells and T-24 human bladder cancer cells grown *in vitro* were killed by a suspension of TiO_2 particles and exposure to 300–400-nm UVR (Cai *et al.*, 1992; Kubota *et al.*, 1994). Furthermore, when TiO_2 was injected into the tumour and the animals were irradiated with the same UVR source, the growth of both tumour cell lines transplanted subcutaneously into athymic mice was inhibited.

PABA, ethylhexyl methoxycinnamate and benzophenone-3 inhibited cell growth and DNA synthesis, retarding cell cycle progression from G_1 when added to cultured cell lines at doses of 50–100 µg/ml. As these doses could be achieved *in vivo* in sunscreen-treated skin, these effects may be of biological relevance (Xu & Parsons, 1999). PABA at a dose of 328 µmol/L was reported to inhibit platelet aggregation *in vitro* (Barbieri *et al.*, 1999).

A mouse lymphoma cell line had decreased survival in a suspension of 0.1% PABA after exposure to 313 nm UVR (Osgood *et al.*, 1982).

Production of reactive oxygen species by sunscreens

PABA has been reported to scavenge singlet molecular oxygen species (Fig. 45) (Allen *et al.*, 1995). It also protected calf thymus DNA from damage by free radicals induced by exposure to UVR at 254 nm for 1 h at 1.9 mW/cm^2, due to either its sunscreening or its reactive oxygen quenching properties (Hu *et al.*, 1995).

In contrast, irradiation of aqueous solutions of PABA, ethylhexyl PABA, octocrylene and ethylhexyl methoxycinnamate but not benzophenone-3 or benzophenone-8 with solar-simulated UVR (provided by a filtered 1000-W xenon arc lamp) generated singlet molecular oxygen (Allen *et al.*, 1996a,b). In another study, benzophenone-3 interfered with antioxidant defence in the skin. Benzophenone-3 in a sunscreen (SPF 25) applied to human skin was photooxidized to benzophenone-3 semiquinone after 20 min of exposure to sunlight. The latter reacted with thiol groups on proteins, such as thioredoxin reductase and reduced glutathione, involved in antioxidant defence, causing their inactivation (Schallreuter *et al.*, 1996).

Uncoated TiO_2 particles exposed to UVR can form reactive oxygen species (Sclafani *et al.*, 1990), including hydroxyl radicals (Brezova & Stasko, 1994). TiO_2 particles extracted from commercial sunscreens and irradiated with UVR

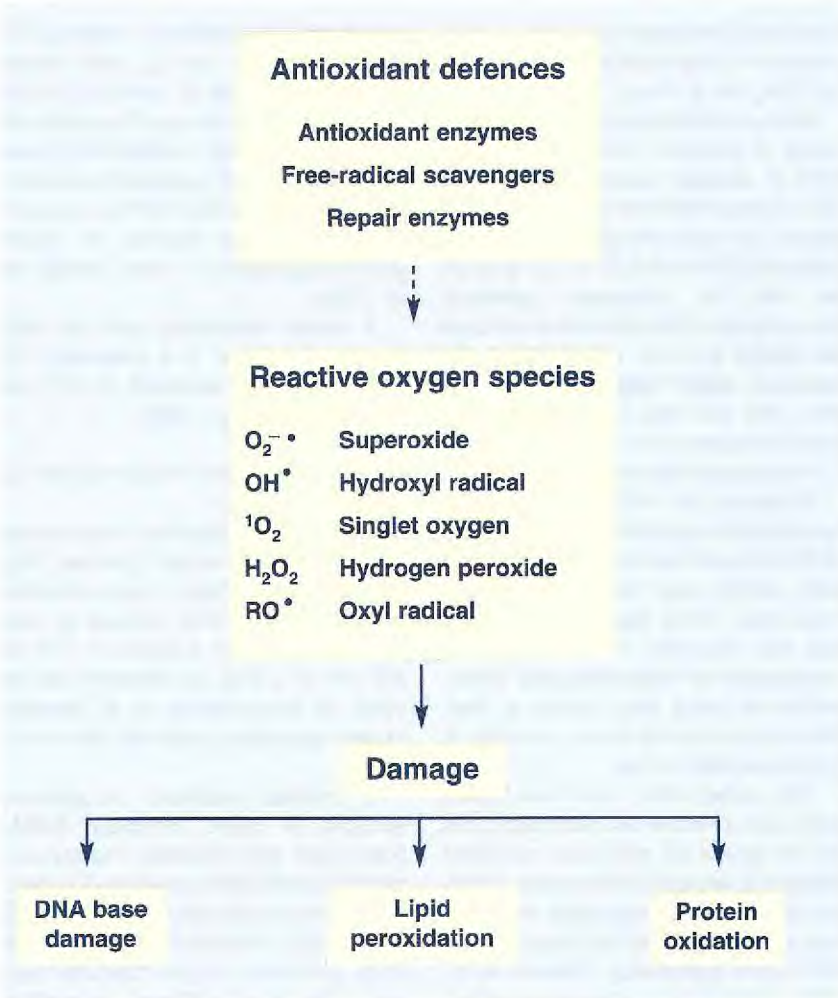

Figure 45 Reactive oxygen species: endogenous defences and damaging effects

(310–400 nm) oxidized organic substrates, indicating that sunlight-irradiated TiO_2 induces biological damage mediated by reactive oxygen species (Dunford *et al.*, 1997). When hydroxylation of guanine was used as a biomarker for reactive oxygen-mediated damage to nucleic acids, 0.45 µm of anatase TiO_2 particles irradiated with UVA (320–400 nm) induced oxidative damage to the RNA but not the DNA of cultured skin fibroblasts. The finding that these cells accumulated the TiO_2 particles in the cytoplasm but not the nucleus suggests that reactive oxygen species generated by irradiated TiO_2 particles induce damage only at the site of their production (Wamer *et al.*, 1997).

Huang *et al.* (1997) found that uncoated, 10-nm TiO_2 particles exposed to 300–400 nm radiation induced oxidative damage to DNA, leading to cell death. This could be prevented by the addition of reactive oxygen scavengers.

Intratracheal exposure of rats to 2 mg of ultrafine, uncoated TiO_2 particles (< 30 nm) caused lipid peroxidation and hydrogen peroxide production associated with enhancement of antioxidant enzyme activity and cytotoxicity to pulmonary alveolar macrophages (Afaq *et al.*, 1998). This suggests that the TiO_2 particles kill the macrophages by increasing oxidative stress which cannot be overcome by the increase in antioxidant enzymes. Scavengers of reactive oxygen have also been shown to inhibit the killing of tumour cells by UV-irradiated uncoated TiO_2 (Cai *et al.*, 1992; Kubota *et al.*, 1994).

Immune system

Application of 8% ethylhexyl dimethyl PABA, 8% ethylhexyl methoxycinnamate or 7.2% microfine TiO_2 in an oil-in-water emulsion 5 days/week for 4 weeks suppressed the induction of contact sensitivity to trinitrochlorobenzene in female C3H/HeJ but not BALB/c mice, in the absence of UVR. This immune suppressive effect of the sunscreens could be overcome by supplementation with oxygen radical scavengers (Bestak *et al.*, 1995). A similar result was observed in a study in which three commercial sunscreens, applied topically to mice for 3 consecutive days suppressed the induction of contact sensitivity by about 50% (Reeve, 1997). It is not clear from either study whether the immune suppression was due to the UVR filter or another component of the sunscreen product. In contrast to the above findings, 0.2% 4-isopropyldibenzoylmethane, but not 5% PABA or 1% homosalate, induced contact sensitization and mild irritation in unirradiated Hartley outbred guinea-pigs after occlusion for 2 h. PABA, but not the other two agents, induced photoallergy when the guinea-pigs were irradiated with 100 kJ/m^2 UVA (320–400 nm) (Gerberick & Ryan, 1989). Thus, it appears that sunscreens can be immune suppressive, sensitizing or photoallergenic under some conditions.

Reproductive and developmental effects

Human studies

No epidemiological study has been conducted showing that sunscreen use has any reproductive or developmental effects.

Experimental studies

PABA injected intraperitoneally into pregnant rats on days 1–6, 6–16 or 1–16 of gestation at 5 mg/kg bw per day did not damage the fetuses (Stroeva & Popov, 1998). Benzophenone-3 dissolved in acetone and applied topically at doses ≤ 400 mg/kg bw per day had no toxic effects on the reproductive organs of male B6C3F_1 mice. Sperm concentration and motility, reproductive organ weight and histological appearance were normal (Daston *et al.*, 1993). When benzophenone-3 was fed to Swiss CD-1 mice at 1.8, 4 or 9 g/kg bw per day, the two higher doses caused reduced body weight, a reduced number of live pups per litter, reduced pup weight and increased mortality among lactating dams (Chapin *et al.*, 1997).

Isoamyl-*para*-methoxycinnamate given to pregnant Wistar rats on days 6–15 of gestation by intragastric instillation had a significant effect on reproduction or embryo development only at the highest dose tested, 2.25 g/kg bw per day (Jekat et al., 1992). The two lower doses, 0.75 and 0.25 g/kg bw per day, had no observable effect. The highest dose increased the rate of intrauterine deaths, decreased fetal weights and caused some signs of retarded development but no signs of teratogenicity.

Octocrylene at doses up to 267 mg/kg bw per day had no discernible reproductive or developmental effects when applied topically to New Zealand white does on days 6–18 of gestation, and no signs of toxicity were observed on the male reproductive system. In female CD-1 mice given octocrylene orally by gavage on days 8–12 of gestation, doses ≤ 1000 mg/kg bw per day had no effect on pup survival or litter weight (Odio *et al.*, 1994).

Genetic and related effects

Human studies

No data were available to the Working Group.

Experimental systems

Since sunscreens are exposed to UVR, it is important to consider the potential damaging effects of the compounds alone and in combination with UVR and visible light, including photosensitized damage and mutation. Consideration must be given to the wavelength distribution and energy of the source(s) employed. In all studies, it is crucial to define not only the exact chemical nature and concentration of the sunscreen ingredient(s) but also the precise nature of the test system.

Most sunscreens have now been tested for their ability to modify genetic material in a variety of test systems in the absence of UVR (Table 26). Those that have shown evidence of direct DNA damaging properties are ethylhexyl methoxycinnamate (or more likely an unidentified contaminant of the sunscreen preparation, Bonin *et al.*, 1982) and a nitrosamine contaminant (2-ethylhexyl 4-*N*-methyl-*N*-nitrosamino-benzoate) of ethylhexyl dimethyl PABA, originally reported by Loeppky *et al.* (1991) but not confirmed by Dunkel *et al.* (1992). Benzophenone-3 was also shown to have some mutagenic properties (French, 1992), but the result was not confirmed in a further study (Robison *et al.*, 1994). A review of data on the mutagenicity of ethylhexyl methoxycinnamate (Trueman & Schüpbach, 1982) shows that the results differ according to batch, even within a single laboratory. This finding lends credence to the idea that the clear positive results observed in certain studies are the result of a contaminant.

PABA has been shown to cause differential killing of repair-deficient bacteria (Hodges *et al.*, 1977), and similar toxicity was later reported in mouse lymphoma (L5178Y) cells (Osgood *et al.*, 1982). PABA also caused photosensitized formation of pyrimidine dimers in DNA (Sutherland & Griffin, 1984). Although PABA and UVR or visible light were not mutagenic in bacteria, a preliminary report showed that both conditions could lead to chromosomal aberrations in mammalian cells (Dean *et al.*, 1991; Table 27). Ethylhexyl dimethyl PABA (Knowland *et al.*, 1993) can cause genetic damage in combination with UVR or visible light. A classic example of genotoxicity in the presence of UVR is that of methoxypsoralens, which were previously used in sunscreens (Ashwood-Smith *et al.*, 1980; Dean *et al.*, 1991; Chételat *et al.*, 1993a,b).

Phenylbenzimidazole sulfonic acid generated guanine-specific damage in DNA when a mixture of the compound and a synthetic oligodeoxyribonucleotide were irradiated with UVB (Stevenson & Davies, 1999), but studies have not yet been conducted in cells or *in vivo*.

TiO_2 was considered to be non-mutagenic (IARC, 1989), but an increased frequency of sister chromatid exchange in CHO-K1 cells and a slight increase in the frequency of micronuclei have since been shown after treatment with non-lethal doses of TiO_2 (Lu *et al.*, 1998). Nakagawa *et al.* (1997) demonstrated that TiO_2 particles have no or weak genotoxicity in the absence of UVR or visible light, but significant DNA damage was found in the Comet assay and in the chromosomal aberration test after irradiation with a solar simulator.

Samples of a TiO_2 sunscreen catalysed the photooxidation of phenol, and sunlight-irradiated TiO_2 induced DNA damage *in vitro* and in human fibroblasts, as measured in the Comet assay (Dunford *et al.*, 1997). Further information on the genotoxicity of TiO_2 can be found in *The US Pharmacopeia* of 1999.

Table 26. Genetic effects of sunscreen ingredients in the absence of UVR or visible light

Test substance	Test system[a]	Result	Metabolic activation	Concentration	Reference
Ethylhexyl methoxycinnamate	G SA8	+	–	?	Bonin *et al.* (1982)
	G DMX	+	–	?	
	S SIC[b]	+	–	0.18 mol/L	
Ethylhexyl dimethyl PABA contaminant: 2-ethylhexyl 4-*N*-methyl-*N*-nitrosaminobenzoate	G SA0	–	–	0–50 μmol/plate	Loeppky *et al.* (1991)
		+	+	0–50 μmol/plate	
	G SA5	–	–	0–50 μmol/plate	
		+	+	0–50 μmol/plate	
	G SA0	–	+/–	2.3–34.2 μmol/plate	Dunkel *et al.* (1992)
	G SA5	–	+/–	2.3–34.2 μmol/plate	
	G SA8	–	+/–	2.3–34.2 μmol/plate	
	G SA9	–	+/–	2.3–34.2 μmol/plate	
	G G5T	–	–	3.42×10^{-6} mol/L	
		–	+	1.06×10^{-4} mol/L	
Benzophenone-3	M MVR	–	–	3–50 $\times 10^3$ ppm in feed	French (1992)
	G SA0	–	+/–	0–1 mg/plate	
	G SA5	–	+/–	0–1 mg/plate	
	G SA7	–	+/–	0–1 mg/plate	
	G SA9	–	+/–	0–1 mg/plate	
	S SIC	–	–	1.7–17 μg/ml	
		+	+	5–50 μg/ml	
	C CIC	–	–	9.4–93 μg/ml	
		+	+	9.4–75 μg/ml	
	G DMM	–	–	$3.0–3.5 \times 10^3$ g/L	Robison *et al.* (1994)
	C CBA	–	–	0.5–5.0 g/kg bw	
PABA	C CIC	+	–	1900 μg/ml	Dean *et al.* (1991)

[a] See Appendix 2 for explanation of codes.

Table 27. Genetic effects of sunscreen ingredients in the presence of UVR or visible light

Test substance	Test system[a]	Result	Metabolic activation	UVR or visible light source	Concentration	Reference
PABA	C CIC	(+)	–	UVA/UVB	1500–1700 μg/ml (1900 μg/ml toxic)	Dean *et al.* (1991)
Ethylhexyl dimethyl PABA	D SSD	+	–	Solar simulator	50 μmol/L	Knowland *et al.* (1993)
	G SCR	+	–	Solar simulator	50 μmol/L	
	D DIH (human keratinocytes)	+	–	Solar simulator	50 μmol/L	Gulston & Knowland (1999
5-Methoxypsoralen	G ECW	+	–	UVA (320–380 nm) black light bulbs	40 μg/ml	Ashwood-Smith *et al.* (1980)
	S SIC	+	–	UVA (320–380 nm) black light bulbs	40 μg/ml	
8-Methoxypsoralen	G SA0	+	–	UVA and UVA/UVB	6.25–5- μg/plate	Dean *et al.* (1991)
				UVA/UVB	50–1000 μg/plate	
	C CIC	+	–	UVA/UVB	50 μg/ml	
	G SA0	+	–	Solar simulator	0.3–3 μg/plate	Chételat *et al.* (1993a)
	G SA2	+	–	Solar simulator	0.3–3 μg/plate	
	R SCG	+	–	Solar simulator	0.8–5 μg/ml	
	C CIC	+	–	Solar simulator	2 μg/ml (5 μg/ml phototoxic)	Chételat *et al.* (1993b)
TiO_2	R SCG	+	–	Solar simulator	0–3200 μg/ml	Nakagawa *et al.* (1997)
	G SA0	–	–	Solar simulator	0–40 mg/ml	
	G SA2	–	–	Solar simulator	0–40 mg/ml	
	G SA9	–	–	Solar simulator	0–40 mg/ml	
	G G51	–	–	Solar simulator	0–2000 μg/ml	
	C CIC	+	–	Solar simulator	0–50 μg/ml	

[a] See Appendix 2 for explanation of codes.