Lycopene-rich products and dietary photoprotection

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Plant constituents such as carotenoids and flavonoids are involved in the light-protecting system in plants and contribute to the prevention of UV damage in humans. As micronutrients they are ingested with the diet and are distributed into light-exposed tissues where they provide systemic photoprotection. β-Carotene is an endogenous photoprotector, and its efficacy to prevent UV-induced erythema formation has been demonstrated in intervention studies. Lycopene is the major carotenoid of the tomato and is a very efficient singlet oxygen quencher in the group of carotenoids. Following ingestion of lycopene or tomato-derived products rich in lycopene, photoprotective effects have been demonstrated. After 10–12 weeks of intervention a decrease in the sensitivity towards UV-induced erythema was observed in volunteers. Dietary carotenoids may contribute to life-long protection against harmful UV radiation.

Introduction

UV exposure of the skin leads to chemical and biological reactions either damaging or adaptive to light-induced stress.[1,2] Primarily, light of an appropriate wavelength interacts with a suitable chromophore which may be directly damaged or act as a photosensitizer. Short-lived electronically excited species initiate subsequent reactions. In the presence of oxygen, secondary reactive oxygen species are generated extending the range of photodamage. Photooxidative damage affects cellular lipids, proteins and DNA and is involved in the pathobiochemistry of erythema formation, premature aging of the skin, development of photodermatoses, and skin cancer.

Sunburn is a visible dermal reaction following excessive exposure to sunlight, called UV-induced or solar erythema and is characterized by tenderness, sometimes painful blistering and second degree burns.[3] Direct and indirect damage resulting from photochemical reactions leads to vasodilation of dermal vessels and edema and causes increased blood flow in the affected area. Damage to proteins and DNA accumulates within skin cells, and morphological changes occur in keratinocytes and other skin cells. When a cell becomes irreversibly damaged by UV exposure, cell death follows *via* apoptotic mechanisms, leading to the appearance of so-called sunburn cells in the epidermis.[4,5]

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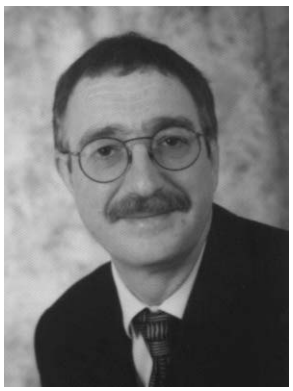

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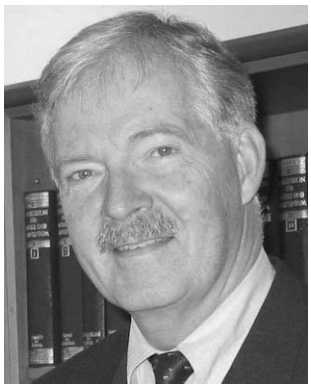

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The UV-B part of the electromagnetic spectrum of the sun is highly erythematogenic and considered to be the major cause of typical sunburn, which starts to develop a few hours after irradiation, culminating about 18–24 h post-irradiation.

The sensitivity of an individual towards erythematogenic UV exposure is determined by the minimal erythemal dose (MED), defined as the threshold dose required to cause a perceptible reddening of the skin 24 h after exposure.[6] MED values differ between individuals and depend on the actual endogenous protection by melanin and on skin type. Melanin levels determine the skin color and are related to some extent to the skin type which is often categorized following the Fitzpatrick scale ranging from type I to VI. Skin type I is assigned to people with white or freckled skin, green or light-blue eyes, red hair and high sensitivity to sun light; skin type VI shows black skin, dark brown eyes and black hair, almost never experiencing sunburn.[7]

Photoprotection

Several strategies are applicable for protection against hazardous light and subsequent impairment of molecular and cellular functions.[8] Avoidance of sun exposure and protective covering and topical application of sunscreens with a high sun protection factor is recommended during times of intense exposure. A major contribution to endogenous protection of the human skin is provided by melanins, endogenous pigments which scatter and absorb UV light.[9] In epidermal melanocytes the production of melanin is increased by exposure to sunlight (tanning).

Endogenous or systemic photoprotection may be enhanced supplying dietary or non-dietary compounds with photoprotective properties. Although the concept of additional endogenous protection was proposed about 30 years ago[10] the development of the concept of systemic photoprotection gained momentum only recently.[11] In addition to some drugs like psoralens or antimalarial agents, mainly dietary constituents have been investigated. Depending on the desired mechanism of action, specific structures and photochemical properties are required for a suitable photoprotectant.[12] Furtheron, toxicological aspects and pharmacokinetic characteristics must be taken into consideration. In order to increase the barrier for UV light, the compound should absorb UV light over a broad range of wavelength with high efficacy. Sufficient photostability is a prerequisite in this context. Antioxidants protect relevant molecular targets by scavenging reactive oxygen species including excited singlet oxygen and triplet state molecules. For the induction of repair systems dealing with UV-induced damage, compounds which interfere with stress-dependent signaling are demanded. Suppression of cellular and tissue responses like inflammation also require the application of compounds which affect intra- and intercellular signaling pathways.

A number of efficient antioxidants among the major micronutrients are capable of directly scavenging lipophilic and hydrophilic prooxidants or serving as constituents of antioxidant enzymes. Carotenoids, tocopherols, flavonoids and other polyphenols as well as vitamin C contribute to antioxidant defense and thus contribute to endogenous photoprotection.

Carotenoids in photoprotection

Carotenoids are accessory light harvesting pigments and play an essential role in the protection of plants against excess light and photooxidative stress.[13] Carotenoids exhibit a long central chain of conjugated double bonds carrying acyclic or cyclic substituents. Xanthophylls, also called oxo-carotenoids, additionally contain functional oxygen groups.[14]

The extended system of conjugated double bonds is crucial to the antioxidant properties of carotenoids.[15–17] Carotenoids are among the most efficient natural scavengers of singlet molecular oxygen.[16,18] It has been suggested that UV-A dependent skin aging is mainly associated signaling pathways associated with singlet oxygen formation and that the effects of β-carotene on signaling are at least in part related to singlet oxygen quenching properties.[19] At low oxygen tension carotenoids also scavenge peroxyl radicals[20] inhibiting the process of lipid peroxidation.

The most abundant carotenoids in the human organism are β-carotene, α-carotene, lycopene, as well as the xanthophylls lutein, zeaxanthin, α- and β-cryptoxanthin.[21,22] Carotenoid levels in human skin are in the range of 0.2 to 0.6 nmol (g wet tissue)$^{-1}$,[23] however, there are significant differences regarding the level of single carotenoids and distribution of carotenoids within different skin areas.[24]

β-Carotene supplements are applied as oral sun protectants and the protective effects are thought to be related to their antioxidant properties. Data from human studies on the photoprotective effects of orally applied β-carotene are contradictory. In some of the studies moderate photoprotection was determined while no effects were found in others. It has been noted that the efficacy of β-carotene in systemic photoprotection is dependent on the duration of treatment before exposure and on the dose. In studies where protection was found, treatment with carotenoids was for at least 10 weeks, and the dose was higher than 20 mg of carotenoids per day.[10,25–27] In studies reporting no protective effects carotenoids were applied for only 3–8 weeks.[28,29] Based on these findings it has been concluded that the application of moderate doses of β-carotene alone is not sufficient to obtain sustained photoprotection.[29]

The use of high doses of β-carotene in supplements for photoprotection has been discussed controversially due to safety concerns.[30] In two intervention trials with individuals at a high risk for lung cancer, a higher cumulative index for lung cancer was observed in the groups that received β-carotene.[31–33] Therefore, other carotenoids or dietary sources providing considerable amounts of other carotenoids may be suitable for endogenous photoprotection. Supplementation with a daily dose of 24 mg of carotenoid mix comprising the three main dietary carotenoids, β-carotene, lutein and lycopene (8 mg day^{-1} each) provides protection against UV-induced erythema; the effect was comparable to that of β-carotene alone applied at 24 mg day^{-1}.[34] Because of its chemical and biological properties especially lycopene is an interesting dietary constituent with possible photoprotecting activity.

Lycopene

More than 80% of lycopene consumed in the United States is derived from tomato products, although apricots, papaya, pink grapefruit, guava, and watermelon also contribute to dietary intake. Lycopene content of tomatoes can vary significantly, depending on type of tomato and ripening. In the reddest strains of tomatoes, lycopene levels are close to 50 mg kg^{-1} compared with only 5 mg kg^{-1} in the yellow strains. In most cases, bioavailability of

lycopene from dietary sources is increased by thermal processing and coingestion of dietary lipids.[35,36] Processing of food helps to release lycopene from the food matrix, thus improving accessibility of the lipophilic compound for the formation of lipid micelles together with dietary lipids and bile acids. Cooking and food processing enhance the bioavailability of carotenoids; *e.g.* lycopene uptake is higher after ingestion of processed tomatoes (tomato paste) as compared to fresh tomatoes.[37]

Many of the reported health benefits of lycopene are attributed to its ability to protect cells against oxidative damage. Although there has been less research focused on lycopene compared to other carotenoids, in vitro studies show that lycopene is a very efficient quencher of singlet oxygen and a potent scavenger of oxygen radicals.[18,38,39]

In the following paragraphs we describe a series of studies performed in our laboratory investigating the photoprotective effects of lycopene and lycopene-rich products in human intervention trials.

Study design

Some of the data from human intervention studies which are summarized here have been published earlier, and methodological details have been described.[40,41] All volunteers that participated in the studies were of skin type II, which was evaluated based on the coloration of skin, hair and eyes and the history of sensitivity to sun exposure.[42] Further criteria for inclusion were healthy condition, body mass index (BMI) of 18–25 kg m^{-2}, no pregnancy or lactation, no supplementation with vitamins, and no medication during the study.

For intervention, the volunteers consumed lycopene from differents sources for a period of 10 to 12 weeks. Lycopene sources were:

Tomato paste: 40 g of tomato paste were consumed once a day together with 10 g of olive oil, providing 16 mg lycopene; duration of the study was 10 weeks.

Carrot juice: carrot juice (Fruchtsaft Bayer & Co, Ditzingen, Germany) was prepared from the variety "Nutrired" which contained 2.5 mg lycopene and 1.3 mg β-carotene (100 ml)$^{-1}$. Volunteers ingested 2 × 200 ml of juice providing a total dose of 10 mg of lycopene and 5.1 mg of β-carotene per day over a period of 12 weeks.

Lycopene supplement: soft gel capsules containing a tomato extract (Lyc-o-Mato®, LycoRed, Natural Prod. Industr. Ltd., Beer-Sheva, Israel) with 4.9 mg lycopene and 0.2 mg β-carotene. Over a period of 12 weeks two capsules wer ingested per day (daily dose: 9.8 mg lycopene and 0.4 mg β-carotene).

Lycopene drink: the lycopene drink was prepared from tomato extract (Lyc-o-Guard-Drink, LycoRed, Israel). The content of carotenoids in 250 ml of the beverage was: 4.1 mg lycopene, and 0.2 mg β-carotene. Volunteers consumed 2 × 250 ml of the lycopene drink providing a total dose of 8.2 mg of lycopene and 0.4 mg of β-carotene per day over a period of 12 wk.

Synthetic lycopene: volunteers ingested two hard shell capsules per day which contained synthetic lycopene encapsuled as beadlets (5.1 mg capsule^{-1}) amounting to a total dose of 10.2 mg day^{-1}.

At the beginning of the study (week 0) and after 4 and 12 weeks of supplementation blood samples were collected; sampling times in the study with tomato paste were day 0, week 4, and week 10. Serum levels of lycopene and β-carotene were determined by means of high performance liquid chromatography as described previously.[21] Levels of total carotenoids in the skin were measured at the same time points by means of reflection spectroscopy.[24]

Erythema prevention and measurement of skin color

Prior to the start of the study the minimal erythemal dose (MED) was determined for each subject. At the time points of blood sampling, dorsal skin was irradiated with UV light (1.25 times the MED) to induce erythema; for irradiation a blue-light solar simulator (Hönle, Munich, Germany) was applied. The test for photoprotection is based on the recommendations of the COLIPA work group "Sun-Protection-Measurement".[43] At each time point skin color was evaluated before and 24 h after irradiation. Skin color was determined by chromametry (Chromameter Minolta CR 200, Ahrensburg, Germany) using the three-dimensional color system (*L*,*a*,*b*-values). *L*-values are a parameter for lightness of skin and *b*-values (blue–yellow axis) are indicative of pigmentation. *a*-Values (red–green axis) are a measure of skin reddening. Δ*a* values were calculated subtracting the *a*-value measured before irradiation from the *a*-value determined after irradiation. They are directly related to UV-induced erythema formation and were used to quantify skin responses to UV irradiation. Upon successful intervention, Δ*a* values at the end of the study should be lower than at the beginning. The *a*-values differ between individuals due to skin sensitivity and basal skin colour. For better comparison, the Δ*a* values at the beginning of the study were set at 100% and the others calculated as the percentage of basal numbers.

Lycopene serum and skin levels

The daily intake of lycopene was different between groups. The highest lycopene dose was 16 mg day^{-1} in the tomato paste group; about 10 mg day^{-1} were ingested in the synthetic lycopene (10.2 mg day^{-1}), the lycopene supplement (9.8 mg day^{-1}), and the carrot juice (10 mg day^{-1}) group. With the lycopene drink 8.1 mg lycopene day^{-1} were ingested. In the different groups basal levels of lycopene in serum ranged from 0.28 to 0.36 nmol ml^{-1} (Fig. 1A) which is within the range described in the literature.[21,22] After 4 weeks of supplementation lycopene was increased and reached values between 0.55 and 0.84 nmol ml^{-1} which is in accordance with blood levels reported in the literature after prolonged intake of tomato products or lycopene supplements.[44] Between weeks 4 and 12 (or 10), lycopene levels hardly increased any further. At the end of the study basal levels were almost doubled except for the carrot juice group where an increase of about 1.5 times was determined. The differences observed between the groups may be due to differences in bioavailability of lycopene from the various sources but may also be related to interindividual variations with respect to lycopene uptake and/or distribution and metabolism.

In contrast to the serum, carotenoid skin levels were less affected (Fig. 1B). However, increases were determined in all study groups. At the end of the study, skin levels were elevated 1.2 to 1.4 times compared to baseline. By means of reflection spectroscopy in the mode applied here, only total carotenoids in skin could be quantified. Thus, increases in carotenoid skin levels could not be assigned to a single carotenoid, *e.g.* lycopene.

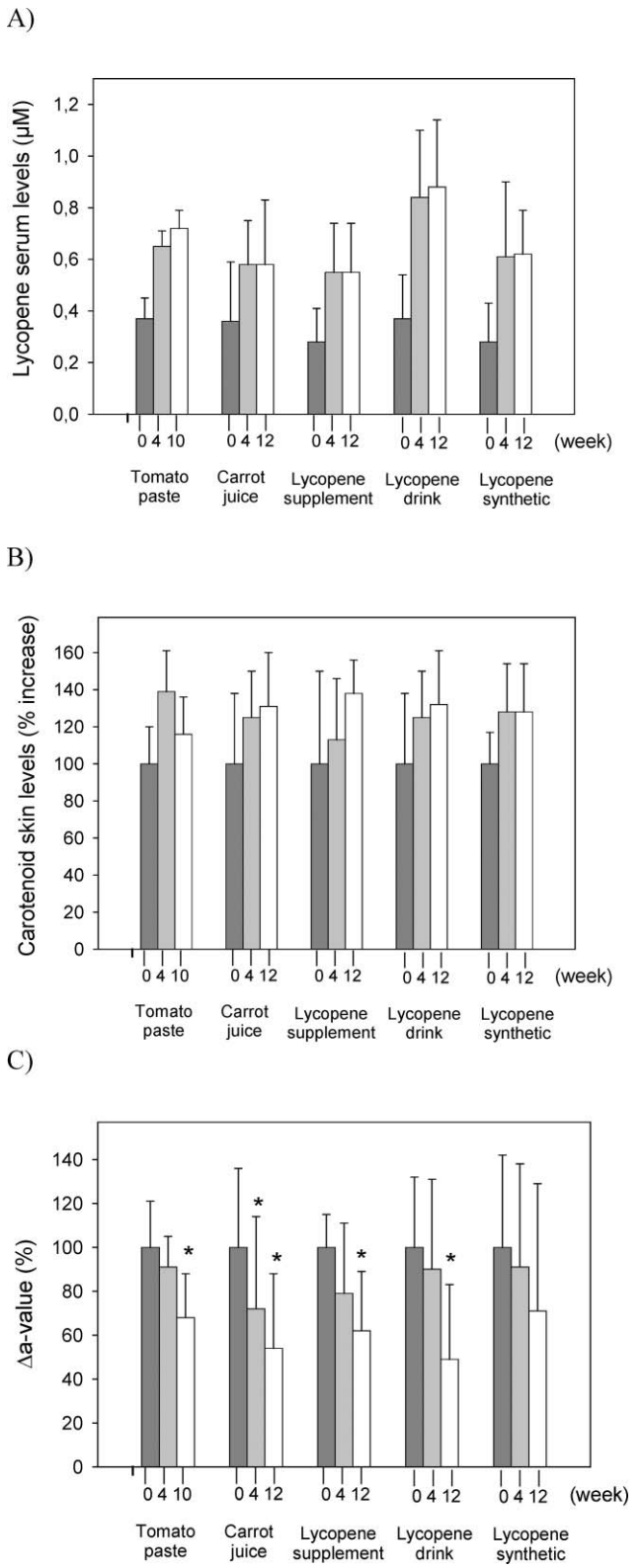

Fig. 1 (A) Lycopene levels in serum (in µM) on day 0, week 4 and week 10 (or week 12) of the studies in the different supplementation groups. (B) Total carotenoids in skin (% increase from baseline) on day 0, week 4 and week 10, respectively wk 12 of the studies in the different supplementation groups. (C) Δ*a*-values of skin of volunteers (% baseline) on day 0, week 4 and week 10, respectively week 12 of the studies in the different supplementation groups. * Significantly different from week 0 ($p < 0.05$).

Photoprotective effects following the consumption of lycopene-rich products

The difference between chromametry *a*-values after and before irradiation (Δ*a*) were taken as a measure for UV-response of the skin, namely erythema formation (Fig. 1C). Decreasing Δ*a*-values in comparison to week 0 (set to 100%) reflect a protection against UV-induced erythema.

Following the ingestion of tomato paste, a decrease in the Δ*a*-values from week 0 to week 4 and week 10 was determined. The difference was most pronounced and statistically significant on week 10. In comparison to baseline the Δ*a*-value was lowered by about 40%.

The photoprotective effect was even more pronounced in the group which ingested the lycopene-rich carrot juice. The Δ*a*-values were significantly lower on week 4 and week 12 compared to baseline. At the end of the study the decrease was about 45%.

In the group consuming the lycopene supplement, Δ*a*-values were lower on wk 4 and 12 than on wk 0. The differences to week 0 were statistically significant only on week 12.

Upon consumption of the lycopene drink also a decrease of the Δ*a*-values from week 0 to week 4 and week 12 was observed. The difference in comparison to week 0 was statistically significant on week 12 of supplementation and the Δ*a*-value was lowered by about 50%.

Treatment with synthetic lycopene for a period of 12 weeks led to a decrease in the Δ*a*-value after 12 weeks. However, the difference was statistically not significant.

Four different sources were used to supply lycopene to volunteers and investigate UV-protective effects over a period of 10–12 weeks. The daily dose of lycopene was comparable between groups, ranging from 8.2 to 16 mg day^{-1}. In one of the groups (lycopene synthetic) only lycopene and no other carotenoid was present. The supplements derived from tomato-based products contain a number of other constituents including further carotenoids such as phytofluene and phytoene which are precursors of lycopene in the biosynthetic pathway. These compounds may well contribute to the photoprotective effects since they absorb in the UV-range.

Conclusion

Carotenoids are suitable compounds for photoprotection in the human. In addition to β-carotene, other carotenoids like lycopene or lutein can be used as photoprotectants. Photoprotective effects can be achieved with a diet rich in carotenoids. However, it should be noted that other components of the diet such as further carotenoids (phytoene, phytofluene) or non-carotenoid constituents contribute to photoprotection.

Endogenous protection associated with the ingestion of dietary carotenoids is not comparable to the topical use of a sunscreen with a high sun protection factor. However, increasing the basal protection systemically contributes to life-long defense against UV-dependent skin damage.

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References

1 S. R. Pinnell, Cutaneous photodamage, oxidative stress, and topical antioxidant protection, *J. Am. Acad. Dermatol.*, 2003, **48**, 1–19.
2 J. Wenk, P. Brenneisen, C. Meewes, M. Wlaschek, T. Peters, R. Blaudschun, W. Ma, L. Kuhr, L. Schneider and K. Scharffetter-Kochanek, UV-induced oxidative stress and photoaging, *Curr. Problems Dermatol.*, 2001, **29**, 83–94.
3 G. J. Clydesdale, G. W. Dandie and H. K. Muller, Ultraviolet light induced injury: immunological and inflammatory effects, *Immunol. Cell. Biol.*, 2001, **79**, 547–568.
4 J. P. Cesarini, Sunburn and apoptosis in *Skin Cancer and UV Radiation*, ed. P. Altmeyer, K. Hoffmann and M. Stücker, Springer-Verlag, Berlin, 1997, pp. 94–101.
5 D. Kulms and T. Schwarz, Molecular mechanisms of UV-induced apoptosis, *Photodermatol. Photoimmunol. Photomed.*, 2000, **16**, 195–201.
6 D. Orentreich, A.-S. Leone, G. Arpino and H. Burack, Sunscreens: practical applications, in *Sun Protection in Man*, ed. P. U. Giacomoni, Elsevier, Amsterdam, 2001, pp. 535–559.
7 L. Andreassi, M. L. Flori and P. Rubegni, Sun and skin. Role of phototype and skin colour, *Adv. Exp. Med. Biol.*, 1999, **455**, 469–475.
8 P. Wolf and A. Young, Photoprotection, in *Dermatological Phototherapy and Photodiagnostic Methods*. (ed. J. Krutmann, H. Hönigsmann, C. A. Elmets and P. R. Bergstresser), Springer Verlag, Heidelberg, 2001, pp. 303–326.
9 J. P. Ortonne, Photoprotective properties of skin melanin, *Br. J. Dermatol.*, 2002, **61**, 7–10.
10 M. M. Mathews-Roth, M. A. Pathak, J. A. Parrish, T. B. Fitzpatrick, E. H. Kass, K. Toda and W. Clemens, A clinical trial of the effects of oral beta-carotene on the responses of human skin to solar radiation, *J. Invest. Dermatol.*, 1972, **59**, 349–353.
11 H. Sies and W. Stahl, Nutritional protection against skin damage from sunlight, *Annu. Rev. Nutr.*, 2004, **24**, 173–200.
12 H. S. Black and L. E. Rhodes, Systemic photoprotection; dietary intervention and therapy, in *Sun Protection in Man*, ed. P. U. Giacomoni, Elsevier, Amsterdam, 2001, pp. 573–591.
13 B. Demmig-Adams and W. W. Adams, III, Antioxidants in photosynthesis and human nutrition, *Science*, 2002, **298**, 2149–2153.
14 J. A. Olson and N. I. Krinsky, Introduction: the colorful fascinating world of the carotenoids: important physiologic modulators, *FASEB J.*, 1995, **9**, 1547–1550.
15 A. J. Young and G. M. Lowe, Antioxidant and prooxidant properties of carotenoids, *Arch. Biochem. Biophys.*, 2001, **385**, 20–27.
16 W. Stahl and H. Sies, Antioxidant defense: vitamins E and C and carotenoids, *Diabetes*, 1997, **46**, S14–S18.
17 N. I. Krinsky, The antioxidant and biological properties of the carotenoids, *Ann. NY Acad. Sci.*, 1998, **854**, 443–447.
18 A. Cantrell, D. J. McGarvey, T. G. Truscott, F. Rancan and F. Bohm, Singlet oxygen quenching by dietary carotenoids in a model membrane environment, *Arch. Biochem. Biophys.*, 2003, **412**, 47–54.
19 K. Wertz, P. B. Hunziker, N. Seifert, G. Riss, M. Neeb, G. Steiner, W. Hunziker and R. Goralczyk, beta-Carotene interferes with ultraviolet light A-induced gene expression by multiple pathways, *J. Invest. Dermatol.*, 2005, **124**, 428–434.
20 G. W. Burton and K. U. Ingold, β-Carotene: an unusual type of lipid antioxidant, *Science*, 1984, **224**, 569–573.
21 W. Stahl, A. R. Sundquist, M. Hanusch, W. Schwarz and H. Sies, Separation of β-carotene and lycopene geometrical isomers in biological samples, *Clin. Chem.*, 1993, **39**, 810–814.
22 F. Khachik, C. J. Spangler, J. C. Smith, L. M. Canfield, A. Steck and H. Pfander, Identification, quantification, and relative concentrations of carotenoids and their metabolites in human milk and serum, *Anal. Chem.*, 1997, **69**, 1873–1881.
23 Y.-M. Peng, Y.-S. Peng and Y. Lin, A nonsaponfication method for the determination of carotenoids, retinoids, and tocopherols in solid human tissues, *Cancer. Epidemiol. Biomark. Prev.*, 1993, **2**, 139–144.
24 W. Stahl, U. Heinrich, H. Jungmann, J. von Laar, M. Schietzel, H. Sies and H. Tronnier, Increased dermal carotenoid levels assessed by noninvasive reflection spectrophotometry correlate with serum levels in women ingesting Betatene, *J. Nutr.*, 1998, **128**, 903–907.
25 H. P. M. Gollnick, W. Hopfenmüller, C. Hemmes, S. C. Chun, C. Schmid, K. Sundermeier and H. K. Biesalski, Systemic beta carotene plus topical UV-sunscreen are an optimal protection against harmful effects of natural UV-sunlight: results of the Berlin-Eilath study, *Eur. J. Dermatol.*, 1996, **6**, 200–205.
26 J. Lee, S. Jiang, N. Levine and R. R. Watson, Carotenoid supplementation reduces erythema in human skin after simulated solar radiation exposure, *Proc. Soc. Exp. Biol. Med.*, 2000, **223**, 170–174.
27 W. Stahl, U. Heinrich, H. Jungmann, H. Sies and H. Tronnier, Carotenoids and carotenoids plus vitamin E protect against ultraviolet light-induced erythema in humans, *Am. J. Clin. Nutr.*, 2000, **71**, 795–798.
28 M. Garmyn, J. D. Ribaya-Mercado, R. M. Russell, J. Bhawan and B. A. Gilchrest, Effect of beta-carotene supplementation on the human sunburn reaction, *Exp. Dermatol.*, 1995, **4**, 104–111.
29 F. McArdle, L. E. Rhodes, R. A. Parslew, G. L. Close, C. I. Jack, P. S. Friedmann and M. J. Jackson, Effects of oral vitamin E and beta-carotene supplementation on ultraviolet radiation-induced oxidative stress in human skin, *Am. J. Clin. Nutr.*, 2004, **80**, 1270–1275.
30 H. K. Biesalski and U. C. Obermueller-Jevic, UV light, beta-carotene and human skin-beneficial and potentially harmful effects, *Arch. Biochem. Biophys.*, 2001, **389**, 1–6.
31 The ATBC-Study Group, The effect of vitamin E and beta carotene on the incidence of lung cancer and other cancers in male smokers, *N. Engl. J. Med.*, 1994, **330**, 1029–1035.
32 G. S. Omenn, G. E. Goodman, M. D. Thornquist, J. Balmes, M. R. Cullen, A. Glass, J. P. Keogh, F. L. Meyskens, B. Valanis, J. H. Williams, S. Barnhart, M. G. Cherniack, C. A. Brodkin and S. Hammar, Risk factors for lung cancer and for intervention effects in CARET, the beta-carotene and retinol efficacy trial, *J. Natl. Cancer Inst.*, 1996, **88**, 1550–1559.
33 G. E. Goodman, M. D. Thornquist, J. Balmes, M. R. Cullen, F. L. Meyskens, Jr., G. S. Omenn, B. Valanis and J. H. Williams, Jr., The Beta-Carotene and Retinol Efficacy Trial: incidence of lung cancer and cardiovascular disease mortality during 6-year follow-up after stopping beta-carotene and retinol supplements, *J. Natl. Cancer. Inst.*, 2004, **96**, 1743–1750.
34 U. Heinrich, C. Gartner, M. Wiebusch, O. Eichler, H. Sies, H. Tronnier and W. Stahl, Supplementation with beta-Carotene or a Similar Amount of Mixed Carotenoids Protects Humans from UV-Induced Erythema, *J. Nutr.*, 2003, **133**, 98–101.
35 W. Stahl and H. Sies, Lycopene: a biologically important carotenoid for humans?, *Arch. Biochem. Biophys.*, 1996, **336**, 1–9.
36 A. W. Williams, T. W. M. Boileau and J. W. Erdman, Factors influencing the uptake and absorption of carotenoids, *Proc. Soc. Exp. Biol. Med.*, 1998, **218**, 106–108.
37 C. Gärtner, W. Stahl and H. Sies, Lycopene is more bioavailable from tomato paste than from fresh tomatoes, *Am. J. Clin. Nutr.*, 1997, **66**, 116–122.
38 P. Di Mascio, S. Kaiser and H. Sies, Lycopene as the most efficient biological carotenoid singlet oxygen quencher, *Arch. Biochem. Biophys.*, 1989, **274**, 532–538.
39 W. Stahl and H. Sies, Antioxidant activity of carotenoids, *Mol. Aspects Med.*, 2003, **24**, 345–351.
40 W. Stahl, U. Heinrich, S. Wiseman, O. Eichler, H. Sies and H. Tronnier, Dietary tomato paste protects against ultraviolet light-induced erythema in humans, *J. Nutr.*, 2001, **131**, 1449–1451.
41 O. Aust, W. Stahl, H. Sies, H. Tronnier and U. Heinrich, Supplementation with tomato-based products increases lycopene, phytofluene, and phytoene levels in human serum and protects against UV-light-induced erythema, *Int. J. Vitam. Nutr. Res.*, 2005, **75**, 54–60.
42 M. A. Pathak, Sunscreens: topical and systemic approaches for protection of human skin against harmful effects of solar radiation, *J. Am. Acad. Dermatol.*, 1982, **7**, 285–312.
43 *COLIPA sun protection factor test method*, The European Cosmetic Toiletry and Perfumery Association–COLIPA, Brussels, 1994, ref. 94/289.
44 I. Paetau, D. Rao, E. R. Wiley, E. D. Brown and B. A. Clevidence, Carotenoids in human buccal mucosa cells after 4 weeks of supplementation with tomato juice or lycopene supplements, *Am. J. Clin. Nutr.*, 1999, **70**, 490–494.

