Assessment of cumulative exposure to UVA through the study of asymmetrical facial skin aging

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Background: Published studies assessing whether asymmetrical facial ultraviolet light exposure leads to underlying differences in skin physiology and morphology report only clinical observations. The aim of this study was to assess the visual impact on the skin of repeated ultraviolet-A (UVA) exposure through a window.

Methods: Eight women and two men presenting with asymmetrical signs of photoaging due to overexposure of one side of their face to the sun through a window over a long period of time were enrolled in the study. Split-face biometrologic assessments were performed (clinical scoring, hydration with Corneometer®, mechanical properties with Cutometer®, transepidermal water loss with AquaFlux®, skin relief with fringe projection, photography, stripping, and then lipid peroxidation analysis).

Results: Significant differences were observed in clinical scores for wrinkles, skin roughness assessed by fringe projection on the cheek, and skin heterogeneity assessed with spectrocolorimetry on the cheekbone. Other differences were observed for skin hydration, as well as skin laxity, which tended towards significance.

Discussion: This study suggests the potential benefit of daily UVA protection during nondeliberate exposure indoors as well as outside.

Keywords: UVA, asymmetry, photodamage, skin aging

Introduction
Terrestrial solar ultraviolet (UV) radiation comprises UVB (280–315 nm) and UVA (315–400 nm) because UV radiation within the UVC waveband (100–280 nm) is absorbed entirely within the atmosphere. During a summer day, approximately 3.5% of the UV radiation reaching the earth’s surface is UVB, whereas approximately 96.5% is UVA.

Whilst UVB radiation is absorbed almost entirely by ordinary glass, generally at least 50% of incident UVA is transmitted.¹⁻³ Photodaging is believed to account for most of the age-related changes in facial skin appearance, with underlying histopathology that reveals dystrophic elastic fibers, reduced collagen, and increased activity of matrix metalloproteinase-1.⁴⁻⁵ Many studies have explored the relative contributions of UVA and UVB to the aging phenotype and whilst UVA photons are on average 1000 times less energetic than UVB photons, they are capable of inducing aging changes even in the dermis, partly due to their greater average depth of skin penetration than UVB photons. Transmission of UV through car windows is more variable. Car windshields are made from laminated glass, which can filter most UVA (gray-tinted laminated glass is able to block about 99% of UVA), whereas nonlaminated side and rear windows
can transmit around one-half of incident UVA. This penetration is highly wavelength-dependent. For example, measurements obtained from the excised, nonirradiated epidermis obtained from the lower back of 16 Caucasian subjects showed average transmissions of 10%, 34%, 46%, 54%, and 67% at wavelengths of 290, 300, 320, 350, and 400 nm, respectively. Various studies are available that describe the ability of UVA to cause changes in cells in culture and in vivo. Human studies have demonstrated that even relatively low doses of UVA given repetitively can induce changes associated with photaging, including decreased elastic tissue content and stratum corneum thickening. These findings have also been replicated in whole animal studies, with hairless mice subjected to repeated UVA exposure developing wrinkles and other signs of photodamage, together with histologic changes in dermal tissues and increased levels of matrix metalloproteinase-1. These effects are primarily via reactive oxygen species, generated through endogenous chromophores such as trans-urocanic acid and porphyrins which act as photosensitizers. The resulting oxidative stress leads to damage to key structural proteins, lipids and DNA, and is further exacerbated by reduction in free radical scavenging enzymes, such as catalase. Oxidation in the epidermis could elicit signals that lead to damage in adjacent deeper cells.

Asymmetry has been observed with regard to photodamage, and premalignant skin lesions, with incidence higher on the side closest to a car side window. The photodamaging effects of UVA have also been demonstrated for a female office worker with extensive Favre-Roucouchot disease on her left cheek, with almost no photodamage observed on the other cheek. This patient’s left side had been near a window for 15 years. In the same way that twin studies are valuable for assessing the contribution of environment to appearance, those presenting with pronounced asymmetry and who have an unbalanced right-left side UV exposure history, provide an opportunity to assess the possible impact of chronic UV exposure without the interindividual variability brought by the different histories of total UV exposure. However, studies reporting only clinical observations are available in the literature or have been presented at scientific meetings, and they do not determine whether asymmetric facial UV exposure leads to any underlying differences in skin physiology and morphology.

To assess the worth of a study on asymmetrical skin aging, a radio advertisement was broadcast appealing for men or women with significant differences in the clinical signs of facial aging that they attributed to sun exposure through a nearby window for a great part of their life, due to occupation or activities. The great variety of responders, with occupational histories suggestive of a causal link (drivers, teachers, shopkeepers, saleswomen) demonstrated the worth of performing a pilot study to assess the impact of cumulative UVA exposure through clinical skin measures.

Whilst slight asymmetry is commonplace, if those presenting with strong visible asymmetry have different left-right exposure histories, it does allow for an exploratory assessment of the impact that UV, and in particular UVA, might be having on skin.

**Materials and methods**

A pilot study was conducted on eight women aged 64.9 ± 5.4 years and two men aged 56–61 years living in the region of Besançon, France. The subjects were likely to have been exposed to a higher proportion of glass-filtered UV radiation on one side of their face.

Clinical evaluations were performed on bare skin in controlled environmental conditions (temperature 22.5°C ± 0.7°C, H 51.0% ± 4.5%). Eleven skin characteristics were assessed on the face by a trained dermatologist using photographic scales and ordinal scales (Figure 1). Furthermore, a questionnaire dedicated to the subjects’ history in terms of sun exposure and occupation, as well as sun protection was completed. For each side of the face, photographs were taken by VISIA® facial imaging (Canfield Scientific, Fairfield, NJ), skin relief by fringe projection (80 × 60 mm rectangular field; Eotech, France), skin color by spectrophotometry (D65 Illuminant, Minolta, France), skin elasticity by Cutometry (SEM 474, suction applied 3 sec/suction off 2 sec, 2 mm probe, 450 mbar pressure, five repeats, Courage and Khazaka, Germany), hydration by Corneometry (CM825, Courage and Khazaka, Cologne, Germany), and transepidermal water loss (TEWL) by AquaFlux® (AF200, Biox, UK).

Additionally, D-Squame® tape strippings (CuDerm Corporation, Dallas, TX) were collected (Figure 1) from both sides of the face. Tape strippings were stored until analyzed. Phospholipids were extracted and detected using LPO–CC (lipid peroxides) kits (Kamiya Biosciences, Seattle, WA). Detection of lipid peroxidation was carried out at 675 nm using a Multiskan RC colorimetric plate reader (Thermo Labsystems, Finland) with results normalized to protein content. Nonparametric tests were performed to compare data from exposed and nonexposed
areas (Wilcoxon test) with statistical significance achieved if $P < 0.05$ (5%).

Results

Ten volunteers with occupational and UV exposure histories suggestive of a link to the asymmetry were recruited (Table 1). Supporting the hypothesis of increased damage to and barrier impairment of photo exposed skin, statistically significant ($P < 0.05$) differences were observed when comparing window-exposed sides of the face to the nonwindow-exposed sides for:

- Clinical scores (Figure 2) for wrinkles on the cheek, wrinkles under the eyes, crow’s feet number and laxity; global clinical scores (Figure 3) which were higher on the window-exposed side than on the nonwindow-exposed side, indicating a worsening of each characteristic;
- Skin color, with eight subjects showing slightly less heterogeneity, ie, lower $\Delta E$ (Table 2), on the window-exposed side than the nonwindow-exposed side;
- Wrinkle volume and roughness on the cheek (Table 3 and Figure 4), with nine subjects showing deeper wrinkles and less roughness on the window-exposed side than on the nonwindow-exposed side.

Differences tending towards statistical significance ($P < 0.1$) were observed for:

- Clinical scores for crow’s feet depth and dullness of skin (Figure 2), which were higher on the window-exposed side than on the nonwindow-exposed side, indicating a worsening of each characteristic;
- Clinical scores for crow’s feet depth and dullness of skin (Figure 2), which were higher on the window-exposed side than on the nonwindow-exposed side, indicating a worsening of each characteristic;

Table 1

| Subject (Number) | Age | Gender | Phototype | Profession/activity accounting for asymmetry | Years of asymmetrical ultraviolet exposure/years of life (%) |
|------------------|-----|--------|-----------|---------------------------------------------|-------------------------------------------------------------|
| 1                | 62  | F      | III       | Medical saleswoman                          | 53.2                                                        |
| 2                | 66  | F      | II        | Childminder                                 | 49.2                                                        |
| 3                | 56  | M      | IV        | Lorry driver                                | 32.1                                                        |
| 4                | 59  | F      | III       | Medical saleswoman                          | 59.3                                                        |
| 5                | 75  | F      | II        | Teacher                                     | 49.3                                                        |
| 6                | 64  | F      | III       | Bus driver                                  | 39.1                                                        |
| 7                | 70  | F      | II        | Shopkeeper/(shop window)                    | 48.6                                                        |
| 8                | 63  | F      | II        | (Sales)/two hours driving every day          | 47.6                                                        |
| 9                | 61  | M      | II        | Salesperson + lorry driver                  | 24.6                                                        |
| 10               | 60  | F      | II        | Medical saleswoman                          | 50.0                                                        |
Figure 2 Mean clinical scores assessed on each side of the face. Most of the clinical items studied were higher on the window-exposed side (worsening of skin characteristics).

Notes: **Significant differences (P < 0.05) *Tendency (0.05 < P < 0.1).

- Hydration of cheek skin, with eight subjects showing more dryness (Table 4) on the window-exposed side than on the nonwindow-exposed side;
- Laxity of skin on the cheekbone, with seven subjects showing more lax skin, ie, an increase of Uf (Table 2), on the window-exposed side than on the nonwindow-exposed side.

Other results (P > 0.1) worthy of note were found for:
- EWL on the cheek (Table 4), with six subjects showing a higher TEWL on the window-exposed side than on the nonwindow-exposed side;
- Elasticity, ie, Ur/Ue (Table 2), on the cheekbone, with seven subjects showing lower skin elasticity on the window-exposed side than on the nonwindow-exposed side;
- Wrinkle volume measured under the eyes (Table 3 and Figure 5), with seven subjects showing deeper wrinkles on the window-exposed side than on the nonwindow-exposed side (difference of 26% on average between both sides [7%–50%]);
- The amount of lipid peroxidation (Table 4) was lower in seven subjects on the window-exposed side than on the nonwindow-exposed side (decrease of 42% [7%–67%]).

Figure 3 Global score calculated from the sum of the 11 clinical scores for each subject (significant difference between window-exposed and nonwindow-exposed sides, P = 0.0039).
**Table 2** Results from biometrologic assessments performed on the cheekbone. ($\Delta E_{\text{mean}} =$ the mean colorimetric distance computed from the three measures (CIELAB 1976), $\Delta E_{\text{max}} =$ the maximal distance between the three measures)

|       | Ue          | Ur          | Uf          | Ur/Ue       | $\Delta E_{\text{mean}}$ | $\Delta E_{\text{max}}$ |
|-------|-------------|-------------|-------------|-------------|--------------------------|--------------------------|
| WE    | 0.263 ± 0.114$^a$ | 0.100 ± 0.022$^a$ | 0.339 ± 0.126$^a$ | 0.449 ± 0.204 | 2.7 ± 1.4$^a$ | 3.9 ± 2.2$^a$ |
| NWE   | 0.178 ± 0.068 | 0.091 ± 0.022 | 0.249 ± 0.076 | 0.567 ± 0.212 | 3.9 ± 1.4 | 5.6 ± 2.0 |

**Notes:** Data are expressed as mean ± standard deviation. $^a$Differences tend to be significant (0.05 < $P$ < 0.1). $^b$Significant differences ($P < 0.05$).

**Abbreviations:** WE, window-exposed side; NWE, nonwindow-exposed side.

**Discussion**

This study indicates the impact of cumulative UVA exposure on skin. All subjects had histories suggestive of unilateral UVA exposure and, although the primary clinical sign was not the same for all subjects (some had more erythrosis [data not shown], others had more wrinkles on the cheeks, some had more wrinkles on the eye contour), significant differences were systematically observed for most of the assessed parameters.

Lowe et al have shown that even suberythemal doses of repetitive UVA are capable of producing photodamage.$^{10}$ Cumulative UVA induces thickening of the viable epidermis and deposition of lysozyme in elastic fibers.$^{17}$ Our results demonstrate possible consequences of these previous histologic findings by showing a significant decrease of skin laxity and elasticity on window-exposed skin, as well as a decrease in skin color heterogeneity. Chromophores may be less visible due to increased epidermal thickness. No significant differences were observed for TEWL, lipid peroxidation levels, or wrinkles under the eyes by fringe projection, although this may be a function of low sample size. It would be worth undertaking further work with a larger panel, because differences appear in more than 60% of the enrolled volunteers.

However, larger panel size may demonstrate a significant impairment of skin barrier properties on the window-exposed side. Lower lipid peroxidation levels could indicate depleted lipid and/or sebum production, leading to reduced TEWL and poorer hydration. This would be consistent with reduced lipid content of the stratum corneum in photoaged skin.$^{28}$

**Table 3** Results from fringe projection assessments

|        | Volume (mm$^3$) | Sa (μm)  |
|--------|----------------|---------|
| Cheek  | WE 24.8 ± 9.7$^a$ | 82.9 ± 32.6$^a$ |
|        | NWE 15.8 ± 5.0 | 52.9 ± 16.8 |
| Wrinkles under eyes | WE 10.0 ± 3.8 | 88.2 ± 33.8 |
|        | NWE 8.9 ± 2.5 | 78.6 ± 22.2 |

**Notes:** Data are expressed as mean ± standard deviation. Differences tend to be significant (0.05 < $P$ < 0.1). $^a$Significant differences ($P < 0.05$).

**Abbreviations:** WE, window-exposed side; NWE, nonwindow-exposed side.

Whilst in this study we focused on the role of cumulative UVA exposure on skin aging, in recent years there has been increasing interest in its ability to promote carcinogenesis.
following published research using fish models of sunlight-induced malignant melanomas. It is now recognized that UVA is able to induce oxidative DNA damage, generating 8-oxoguanine and cyclobutane pyrimidine dimer photo-products which, in turn, can lead to mutagenesis. Whilst the relative contribution of UVA exposure to carcinogenesis in human skin is unclear, there is growing evidence of a strong association, particularly amongst people subjected to higher UVA levels. For example, a higher incidence of both nonmalignant and malignant skin lesions has been found on the window side of the face of drivers. Additionally, frequent use of sun beds (which emit predominantly UVA radiation) is known to increase the risk of melanoma. Sunscreens, whilst offering good levels of protection against predominately UVB-induced skin burning, still transmit significant levels of UVA. This may lead to accumulation

Table 4 Results from biometrologic assessments performed on lower cheek

|        | HI        | TEWL      | LPO       |
|--------|-----------|-----------|-----------|
| WE     | 34.8 ± 11.1* | 21.9 ± 6.6 | 0.33 ± 0.14 |
| NWE    | 39.0 ± 9.8  | 19.6 ± 4.6 | 0.44 ± 0.18 |

Notes: Data are expressed as mean ± standard deviation. *Differences tend to be significant (0.05 < P < 0.1). *Significant differences (P < 0.05).

Abbreviations: WE, window-exposed side; NWE, nonwindow-exposed side; LPO, lipid peroxidation.

Figure 5 Photographs for subject number 3 (male) who had driven a lorry for a third of his life for 9–10 hours per day. A) Nonwindow-exposed side (his right side). B) Window-exposed side (his left side) where the wrinkles of the crow’s foot are more numerous and more marked.

Figure 6 Photographs of the subject number 1 (female) who had spent more than half of her life working as a medical saleswoman and driving a car 2–5 hours per day. A) Nonwindow-exposed side (her right side). B) Window-exposed side (her left side): wrinkles of cheek, crow’s feet and wrinkles under the eyes are more numerous and deeper.
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of high doses of deeper skin-penetrating UVA and contribute to melanoma risk.35

The great variability of occupations affected by asymmetrical facial aging (Table 1) suggests that more attention should be given to this type of indirect irradiation. The effect of UV radiation is now well recognized, and whilst people are aware of the usefulness of sun protection during peak periods, people are not yet familiar with indirect sun exposure. Moreover, whereas tinted windows in vehicles are now widely provided by car designers, contemporary architectural design incorporates ever larger window areas. This pilot study provides some evidence for the contribution of UVA to photoaging. We cannot exclude the influence of UVB exposure when car windows are opened, for example, nor each subject's own intrinsic asymmetry. However, this study does suggest that daily protection against nondeliberate UVA exposure indoors, as well as outside, may be an important function of any daily sunscreen.36

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