Effect of a New Narrow-band UVB Lamp on Photocarcinogenesis in Mice

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Thirty lightly pigmented hairless (Hr/Hr) mice were irradiated 5 days per week for 30 weeks to assess the photocarcinogenicity of a new Philips TL 01 narrow-band (311 nm ± 2) UVB lamp. All mice were found to be tumour-bearing after 16 weeks and histologically, 83% of these had definite squamous cell carcinomas. Compared with our previous study where conventional broad-band Philips TL 12 UVB irradiation was used, tumours appeared earlier with the TL 01 lamp. The total irradiation dose was, however, several times greater in the TL 01 assay while the total MED dose was considerably less. ***Key word: Ultraviolet irradiation.***

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Photocarcinogenesis of non-melanoma skin cancer in Man is presumably dependent on the cumulative erythemally effective dose of ultraviolet radiation (UVR) (1,2). Animal experiments have clearly shown a dose-effect relationship for UV-carcinogenesis as described by $t_m \sim D^r$ where D is the daily (or weekly) dose of UVR administered and t_m is the median tumour appearance time, i.e., the interval between the first exposure and the moment when 50% of the animals have developed one or more tumours. The exponent r is approximately -0.6 and $\sim$ stands for direct proportionality (2).

Both carcinogenic and erythemal effectiveness increase strongly towards the shorter wavelengths within the UVB region (290–320 nm) (3). The objective of psoriasis therapy ought therefore to be to filter out as much as possible of these shorter wavebands and at the same time retain the irradiation which is therapeutically most effective.

Attempts to maximize the therapeutic effectiveness of UVB phototherapy of psoriasis have been limited, until recently, by the quality and purity of the available light sources. Several investigators (4–6) have determined that the therapeutic action spectrum for psoriasis lies in the range 295–313 nm and of these, Fischer (5) has shown that the longer wavelengths, around 313 nm, were particularly efficient. Taken together with the fact that the shorter UVB wavelengths are more erythemogenic and carcinogenic (7,8), a new UVB lamp (Philips TL 01) with a narrow, almost monochromatic emission at 311 nm (± 2 nm) was developed (9) (Fig. 1). The therapeutic effectiveness of this lamp has been recently assessed in a multicenter trial of psoriasis patients (10–12).

We report here on the carcinogenic effect of this new lamp on hairless mice, comparing the results with our previous study using the conventional broad-band Philips TL 12 lamp (13).

MATERIALS AND METHODS

The irradiance of the TL 01 lamp was measured with an UVX radiometer (UVP Inc., Calif., USA), and the results were standardized by the TL 01 lamp manufacturers, Philips, Netherlands, using a monochromator. An irradiance estimate of the emission spectrum of the combined wavelengths of the TL 01 lamp was thus established. At mid-tube level the irradiance was found to be 1.52 mW/cm^2 and at each end 0.84 mW/cm^2, at a distance of 40 cm. The irradiance of a Philips TL 12 broad-band lamp was similarly measured at a distance of 70 cm for comparison with a previous photocarcinogenesis study made with this lamp (13).

In order to establish the minimum erythema dose (MED), a pilot irradiation study involving 8 female lightly pigmented hairless (Hr/Hr) mice (Bomholdsgaard, Denmark) was first performed. The mice were anaesthesized by intraperitoneal injection of Midazolam (Dormicum®), using a dose of 2 mg/kg, and their backs covered by sections

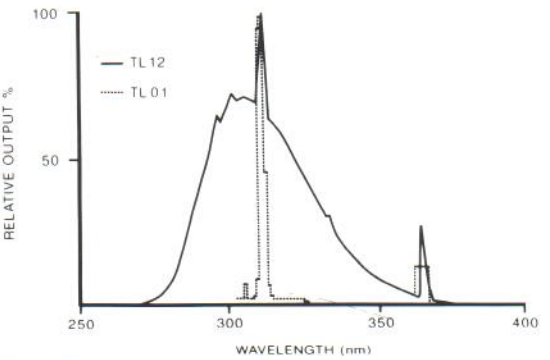

Fig. 1. The irradiation spectra used in the different experiments.

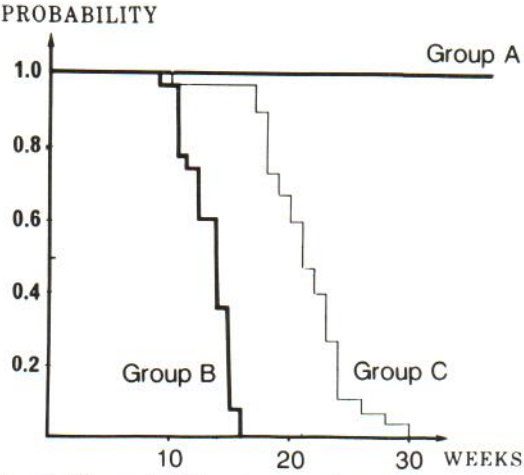

Fig. 2. The probability of tumour-free animals in three groups of hairless mice. *A*: Controls, *B*: UVB-irradiation from a TL 01 lamp, *C*: UVB-irradiation from a TL 12 lamp.

of Duoderm® dressing bandages, each with 6 punched-out round holes (diameter 6 mm). Each mouse was exposed to doses of UVR from the TL 01 lamp in increments of 50 mJ/cm^2 and examined for erythema after 24 h. The lowest dose causing a perceptible response in each animal was noted and the dose corresponding to the median rank, 1350 mJ/cm^2, was considered to be the MED, as described by Cole et al. (14). Although Midazolam is a benzodiazepine derivative, it is not phototoxic (15).

A total of 60 female lightly pigmented hairless (Hr/Hr) mice of similar strain to that used in our broad-band study (13) were fed on standard laboratory feed (Ewos®, Sweden) and had free access to water. They were randomized into two groups of 30 mice each. Five animals were housed in each cage, and six cages constituted a group. Group A served as controls, while group B was irradiated with the narrow-band TL 01 lamp, equipped with three light tubes, at a distance of 40 cm. Since the UV emission at the ends of the lamps was lower than the mid-tube readings, the cages were rotated before each daily treatment. The starting dose measured at mid-tube level was 550 mJ/cm^2. It was increased by 25–30% every second week during the first ten weeks and thereafter remained constant at 1280 mJ/cm^2 (mid-tube level). This corresponded to an increase in exposure time from 6 min to 14 min. The animals were irradiated 5 days per week for 30 weeks.

The post-irradiation observation time was 10 weeks. Each animal was examined weekly by the same observer who had examined the mice in the previous study (13). A skin tumour was defined as a papule $\geq 1 \times 1 \times 1$ mm.

At the end of the observation period the animals were sacrificed and haematoxylin and eosin-stained sections of the tumours were examined by light microscopy. The histologic changes were classified as hyperplasia without atypia, atypical hyperplasia or squamous cell carcinomas with an indisputable stromal invasion.

Statistics

Time until event (tumour development) was analysed using the Kaplan & Meier method (16).

RESULTS

All mice survived the 30-week irradiation and subsequent observation period, and only irradiated mice developed tumours. The median tumour-induction time (t_m), when 50% of the mice were tumour-bearing, was 14 weeks, and all the irradiated mice had developed tumours by week 16 (Fig. 2). Histologically, 25/30 irradiated mice (83.3%) had invasive squamous cell carcinomas and the remaining 5 mice showed hyperplasia with nuclear atypia. Fig. 2 also depicts the results from a previous study (13) using conventional broad-band Philips TL 12 UVB radiation (Curve C). The t_m dose was 49 J/cm^2 (36.3 MED) in the present study compared with 17.2 J/cm^2 (137.6 MEDs) in the previous TL 12 lamp study (Table I).

DISCUSSION

The emission spectra of the two lamps have only a small number of wavelengths in common. That part of the TL 01 emission which is potentially carcinogenic is therefore of a dimension which requires a delivery of three times the energy dose (49 J/cm^2) of the TL 12 lamp (17 J/cm^2) in order to achieve a similar tumorigenic effect (Table I). However, the effect is achieved much earlier by the TL 01 lamp, in 2/3 of the time required by the TL 12 lamp. The erythemic potential (as measured by the MED) is the limiting factor in the case of the latter lamp, requiring an extended period of exposure (21 weeks) to t_m. The dose required to produce erythema (MED) is 11-fold greater in the case of the TL 01 lamp, allowing a greater no. of joules to be delivered, resulting in a shorter t_m.

Table I. *The time and radiation doses required to induce tumours in 50% of the mice*

Lamps	TL 01	TL 12
Median tumour-induction time (t_m)	14 wks	21 wks
MED (mJ/cm^2)	1350	125
Energy dose (× MEDs) to t_m	36.3	137.6
Energy dose (Joules) to t_m	49	17.2

Which one of these lamps then is the more carcinogenic? If the total dose delivered is a major factor in tumour induction, then the TL 01 lamp has, with its enhanced ability to deliver higher doses over shorter time periods, a greater tumorigenic potential, as shown here in terms of t_m (Table I).

This has a particular relevance in a clinical setting, where psoriasis patients, treated with the TL 01 lamps, will be exposed to higher daily and total irradiation doses. With this in mind, we chose to use higher daily irradiance doses in the present TL 01 study compared with those in the previous TL 12 study (13). In the latter, we started with a suberythemic dose of 80% MED and increased it with 20–25% increments every second week up to a maximum of 2 MED. In the case of the TL 01 lamp, the starting dose was much lower, as measured in MEDs, but a larger total exposure dose over the same 30-week period resulted in earlier tumour development in the TL 01 group (Fig. 2).

There have been few published reports of carcinogenic studies on TL 01 radiation of mice. Van Weelden & van der Leun (6) in a similar study, but using different doses and delivery times and a different mouse strain (Skh-hr 1), found the t_m for the TL 01 lamp to be 125 days with a t_m dose of 44.3 joules. The latter correlates well with our TL 01 result, while t_m was 2 weeks earlier in the present study. Van Weelden found a t_m dose (7 joules) for the TL 12 lamps, which is even less than in our TL 12 study (17.2 joules) and a t_m of 114 days compared with our 21 weeks.

Several factors are known to influence photocarcinogenesis in mice and must be taken into account in any assessment of a single study or comparison of separate studies. These include the mouse strain (17), the emission spectra of the lamps (8), the UVR dose delivered (13) and the mode of delivery in terms of dose fractionation and total delivery time (13, 18).

Despite differences in several of these parameters, there was close correlation between the present TL 01 study and the results of van Weelden's TL 01 study. The marked difference in the findings of these two studies using the TL 12 lamp may be due to differences in one or several of the above factors. Similar reasons do not permit a direct comparison of the relative tumorigenicity/carcinogenicity of the TL 01 and TL 12 lamps in the present study.

Nevertheless the results of this study and those of van Weelden et al. (10) established that the TL 01 low-erythemogenic lamp has a considerable carcinogenic potential in mice.

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