Topical photodynamic therapy with endogenous porphyrins after application of 5-aminolevulinic acid

An alternative treatment modality for solar keratoses, superficial squamous cell carcinomas, and basal cell carcinomas?

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Background: Topical photodynamic therapy with endogenous porphyrins consists of irradiation of a tumor with visible light after the application of exogenous 5-aminolevulinic acid.
Objective: To assess the effectiveness of this modality, patients with precancerous conditions and various skin cancers were treated.
Methods: Thirteen patients with 70 skin lesions were enrolled. Standard treatment involved the topical application of 20% 5-aminolevulinic acid in an oil-in-water emulsion. The emulsion was applied under an occlusive dressing for 4 to 8 hours before exposure to photoactivating light.
Results: We observed a complete response after a single treatment for all 9 solar keratoses, 5 of 6 early invasive squamous cell carcinomas, and 36 of 37 superficial basal cell carcinomas. Only 1 of 10 noduloulcerative basal cell carcinomas completely resolved. Eight cutaneous metastases of malignant melanoma were therapeutic failures.
Conclusion: Topical photodynamic therapy with endogenous porphyrins is effective for superficial epithelial skin tumors.
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Photodynamic therapy (PDT), also known as photochemotherapy or photoradiation therapy, is effective for many forms of malignant tumors, including skin cancers.[1-6] Photoactivation of photosensitizing porphyrins, which selectively accumulate in malignant cells, leads to the release of cytotoxic substances and causes tumor destruction with minimal damage to surrounding normal tissue.[1-3] Unfortunately, when given systemically in PDT, the porphyrin compounds, hematoporphyrin derivative (HPD), or a mixture of its active components, commercially known as Photofrin II, lead to generalized skin photosensitivity for at least 4 weeks.[3,4,7] The risk of serious accidental phototoxic reactions is the main reason that PDT by intravenous injection of porphyrins is not used much more widely.

Recently, Kennedy et al.[8] described a novel method of topical PDT and showed favorable results in the treatment of selected superficial nonmelanoma skin cancers. 5-Aminolevulinic acid (5-ALA) was used, which is a precursor of endogenous porphyrins in the biosynthetic pathway for heme.[9] Topically applied to skin cancers, 5-ALA in aqueous solution passes readily through abnormal keratin and is metabolized by the tumor cells to photosensitizing concentrations of porphyrins.[8] The 5-ALA–induced photosensitivity is restricted to the abnormal epidermis. Subsequent exposure to photoactivating light selectively destroys skin cancers.

We describe our evaluation of the effectiveness of PDT after topical application of 5-ALA to epithelial precancerous lesions and various skin cancers.

PATIENTS

Thirteen patients (3 women and 10 men) between 39 and 89 years of age (average 66 years) participated in this study after giving signed informed consent. Patients with multiple skin lesions were preferentially enrolled. There were 70 lesions including 37 superficial basal cell carcinomas in 4 patients, 34 lesions on the trunk and 3 on the face; 10 noduloulcerative basal cell carcinomas in 5 patients, 8 lesions on the face and 2 on the trunk; 9 solar keratoses in 3 patients, all on the face or scalp; 6 early invasive squamous cell carcinomas (superficial squamous

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cell carcinomas with focal invasion of the papillary dermis) in 3 patients, 3 on the face of 1 patient, 2 on the trunk of a patient with Bowen's disease, 1 on the back of the hand of a patient with xeroderma pigmentosum (XP); 8 cutaneous metastases in 1 patient with advanced malignant melanoma, 5 melanotic and 3 amelanotic lesions, all on the left leg.

The greatest diameter of the lesions ranged from 0.5 to 3.0 cm for superficial basal cell carcinomas, from 0.5 to 5.0 cm for noduloulcerative basal cell carcinomas, from 1 to 2 cm for solar keratoses, from 1 to 6 cm for early invasive squamous cell carcinomas, and from 0.1 to 1 cm for cutaneous metastases of malignant melanoma.

Biopsies were always performed on single lesions before the treatment but, if the patient had more than two, biopsy was done on only a representative lesion.

MATERIAL AND METHODS

Therapeutic light source

A Leitz Pradovit slide projector (Leitz, Wetzlar, Germany) equipped with a Philips 250 W lamp (24 V, Philips, Germany) provided the source of irradiation. The total irradiance (i.e., full emitted spectrum before filtration) at 10 cm and 30 cm from the lens as measured by a calibrated photodiode BPW 34 was 100 and 50 mW/cm^2, respectively. Certain irradiations were performed with a Schott RG 570 long-wave-pass red color glass filter to eliminate wavelengths less than 570 nm.

Procedure

Standard treatment involved the topical application of 20% 5-ALA (Fluka Chemie AG, Buchs, Switzerland) dissolved in Doritin (Chemofux, Vienna, Austria), a proprietary oil-in-water emulsion. The emulsion was applied under occlusive dressing to the tumors and approximately 1 cm of adjacent skin to allow penetration of 5-ALA into the tissue and synthesis of endogenous porphyrins. The presence of endogenous porphyrins in the tumors was estimated by the visual evaluation of the characteristic red fluorescence of porphyrins under Wood's light in a darkened room. Fluorescence of the tumors could be observed as early as 1 hour after application of 5-ALA, but the intensity of the fluorescence was maximal at 4 to 6 hours, and then gradually decreased. Most tumors were exposed to photoactivating light 4 hours after application of 5-ALA. The remainder were irradiated at 6 or 8 hours after application. Exposure times for solar keratoses and superficial basal cell carcinomas were either 5 minutes at 100 mW/cm^2 or its equivalent, 10 minutes at 50 mW/cm^2, to unfiltered light (dosage 30 joules/cm^2) or 10 or 20 minutes, respectively, to red light (produced by filtration through RG 570). For two solar keratoses a dose as high as 100 joules/cm^2 of unfiltered light was given. Nodulolcerative basal cell carcinomas and squamous cell carcinomas and metastases of malignant melanoma were exposed either 15 minutes at 100 mW/cm^2 to unfiltered light (dosage 90 joules/cm^2) or 30 minutes to red light at 100 mW/cm^2 (before filtration).

Photographs were taken of all lesions before and after treatment. Tumor response was evaluated at 4 and/or 8 weeks after treatment. Complete tumor response was defined as absence of clinically evident tumor at the site of treatment. In cases in which the clinical assessment was uncertain, punch biopsies were performed. A partial tumor response was defined as marked reduction in tumor size as determined by clinical evaluation.

SELECTED CASE REPORTS

Case 1 (see also Wolf and Kerl[10]). A 47-year-old woman with XP had a hyperkeratotic plaque of 1.5 cm in diameter on the dorsum of her left hand (Fig. 1, *A*). A biopsy specimen indicated superficial squamous cell carcinoma. The lesion was exposed 30 minutes to red light at 100 mW/cm^2 4 hours after application of 5-ALA. Peak erythema and edema of the exposed area were abnormally delayed (72 hours, normal 8 to 24 hours). Erythema persisted for more than 2 weeks. The reaction on the adjacent skin was unusually strong with blistering. Four weeks later there was no sign of residual tumor. Erythema of the adjacent skin gradually resolved, leaving residual hyperpigmentation. There was no clinical evidence of tumor recurrence 6 months later (Fig. 1, *B*).

Case 2. A 59-year-old man, occupationally exposed to arsenic, had 32 superficial basal cell carcinomas on the trunk. In addition, he had a 1 cm ulcerative basal cell carcinoma on the right cheek. Biopsies were performed on two representative lesions on the lower back, two on the chest, and the lesion on the right cheek before therapy (Fig. 2, *A*). All tumors were exposed to 30 joules/cm^2 of unfiltered light 6 hours after application of 5-ALA. Immediately after exposure, the lesions became erythematous and edematous. Within 2 days the lesions developed a thin crust. Adjacent normal skin showed only moderate erythema. When the patient was reexamined 4 weeks later, there were only slight residual erythema or hypopigmented spots but no evidence of tumor in the treated areas. Three biopsies were performed on the back at the site of treated tumors. Histologic examination revealed fibrosis with an inflammatory cell infiltrate and no evidence of residual basal cell carcinoma (Fig. 2, *B*). After 8 weeks complete healing of all lesions with good cosmetic results had occurred.

RESULTS

Clinical response

The treatment was well tolerated by all patients without local anesthetics. Mild to strong stinging or burning was always said to occur at the site of pho-

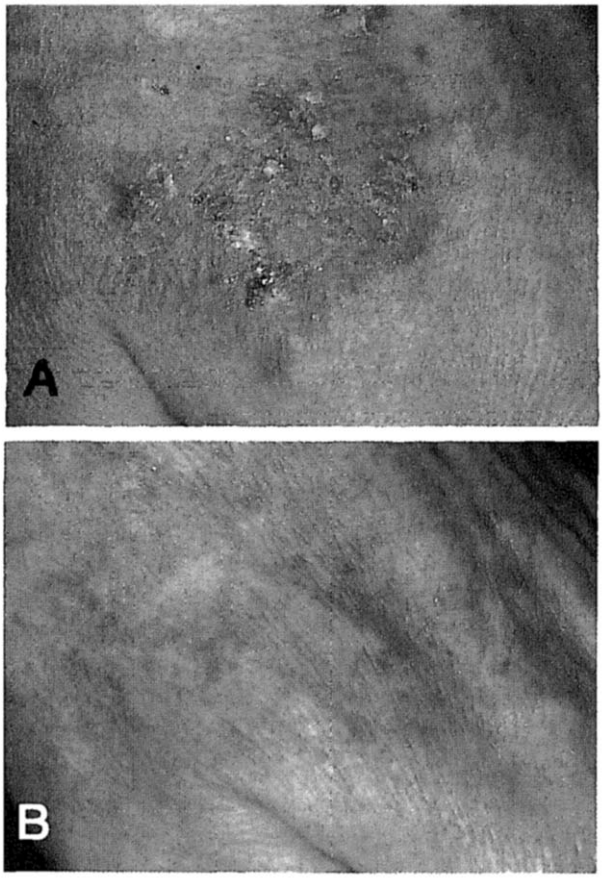

Fig. 1. Case 1. **A,** Superficial squamous cell carcinoma on back of left hand of patient with xeroderma pigmentosum. **B,** Six months after therapy.

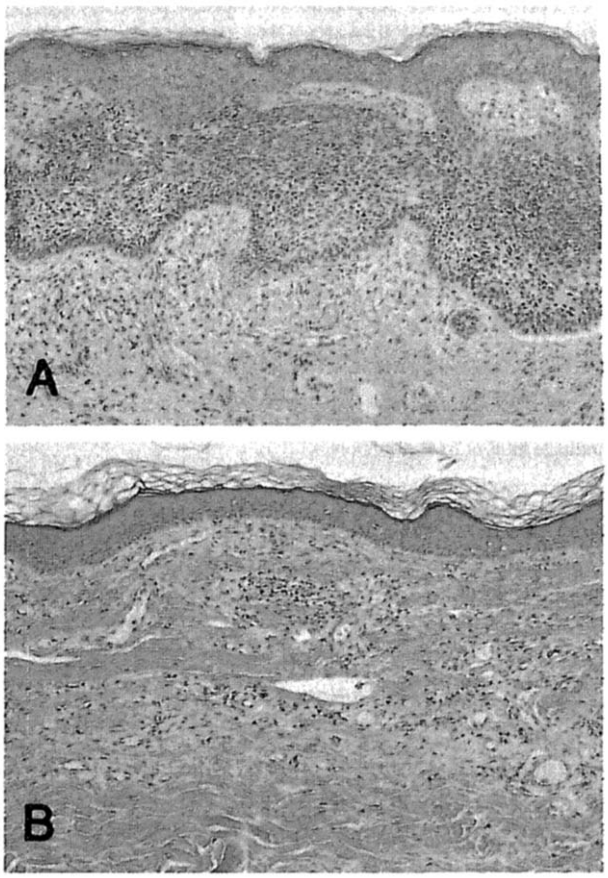

Fig. 2. Case 2. **A,** Photomicrograph of superficial basal cell carcinoma on back before therapy. **B,** Complete regression of tumor 4 weeks after therapy.

toirradiated areas during but not after exposure to photoactivating light. Pain was diminished when tumors were treated with red light. The treated lesions normally became erythematous, edematous or vesiculated during or immediately after therapy and later crusted with healing in 2 to 4 weeks. In general, the exposed areas of adjacent skin showed only a mild reaction to the treatment. However, patients with extensive sun-damaged skin on the face and the patient with XP had an unusually strong reaction in adjacent skin. Only limited areas were treated in these patients at one time, to minimize the skin reaction.

The response of the tumors to the topical PDT after application of 5-ALA is summarized in Table I. When examined 4 to 8 weeks after a single treatment a complete response was observed for all solar keratoses, 5 of 6 early invasive squamous cell carcinomas, 36 of 37 superficial basal cell carcinomas, but only 1 of 10 noduloulcerative basal cell carcinomas. Partial tumor response was observed for all other nonmelanoma skin cancers. There was no observable difference in tumor response with 100 mW/cm^2 irradiance for 5 or 10 minutes or 50 mW/cm^2 for 10 or 20 minutes. In some cases of partial tumor response after a single PDT weekly repeated treatments were performed up to 3 times. However, only in one case (superficial basal cell carcinoma on the abdomen) was a partial tumor response followed by complete tumor eradication.

In amelanotic cutaneous metastases of malignant melanoma only superficial tumor necrosis was observed in posttreatment biopsy specimens; in melanotic metastases the treatment was ineffective (both types were exposed to the same dose of red light).

The nonresponding tumors were excised or treated with radiotherapy.

Follow-up

The longest available follow-up is with the first two patients who entered the study. They have reached 12 months of follow-up; the median fol-

Table I. Tumor response to topical photodynamic therapy with unfiltered visible light (full spectrum) and red light

Tumor type	Unfiltered light	Red light
Superficial BCC	32 (32)*	4 (5)
Noduloulcerative BCC	1 (5)	0 (5)
Solar keratosis	9 (9)	—
Superficial SCC	4 (5)	1 (1)
Metastases of MM	—	0 (8)

BCC, Basal cell carcinoma; *MM*, malignant melanoma; *SCC*, squamous cell carcinoma.
*Numbers indicate complete tumor responses; numbers in parentheses indicate total of treated tumors.

low-up in all patients is 7 months (range 3 to 12). There was one recurrent superficial basal cell carcinoma; no recurrence was observed in the other tumors.

DISCUSSION

This study suggests that superficial epithelial carcinomas of the skin are highly sensitive to PDT after topical application of 5-ALA. A complete response was observed for all treated solar keratoses, 5 of 6 early invasive squamous cell carcinomas, and 36 of 37 superficial basal cell carcinomas. Neither generalized photosensitivity nor other severe adverse reactions were noted and the patients' acceptance of the treatment modality was good. There was only one recurrent tumor (superficial basal cell carcinoma).

Noduloulcerative basal cell carcinomas did not respond well to the therapy. Only 1 of 10 tumors completely resolved. The results of our clinical trial are in agreement with the study of Kennedy et al.[8] They also observed complete response for nearly all treated superficial lesions, including solar keratoses, selected superficial basal cell carcinomas, and either in situ or early invasive squamous cell carcinomas, but only partial response for most noduloulcerative basal cell and thick squamous cell carcinomas. The ineffectiveness of the treatment in noduloulcerative basal cell carcinomas and cutaneous metastases of malignant melanoma may be because these lesions were partly or completely covered by normal epidermis that did not allow penetration of 5-ALA and subsequent production of endogenous porphyrins. In melanotic metastases, pigment within the lesions may also have prevented adequate light penetration.[11-13]

In the biosynthetic pathway for heme, 5-ALA is a precursor of photoactivable porphyrins, including uroporphyrins, coproporphyrins, and protoporphyrins.[9, 14] However, protoporphyrin IX (Pp IX) has been shown to be the main active photosensitizer after treatment with exogenous 5-ALA.[8, 15-17] Red fluorescence and phototoxic damage is characteristic for tissues accumulating 5-ALA–induced Pp IX when exposed to appropriate wavelengths of photoactivating light.[8, 14, 15, 17]

PDT normally includes the intravenous administration of a photosensitizer and the subsequent exposure of tumors to appropriate wavelengths of light.[1-6] Unfortunately, the photosensitizers in common use, HPD or Photofrin II, persist in the skin for several weeks to several months after injection, which results in generalized photosensitivity.[1, 3, 4, 7] In contrast, the skin photosensitivity caused by topical application of 5-ALA is restricted to the tumor and vanishes within 24 hours after treatment.[15]

Most clinical applications of PDT have used red light (630 nm) to treat tumors because of the increased ability of longer wavelengths to penetrate tissues compared with shorter wavelengths of visible light, that is, the optical penetration depth is about 8 to 10 mm at 630 nm compared with 1 to 2 mm at 400 to 500 nm.[1-6] However, 5-ALA–induced photosensitivity has a porphyrin-like spectrum with maximum excitation at 410 nm (Soret band) and three smaller peaks in the visible range (at 510, 545, and 580 nm).[16] Because for thin skin lesions the depth of penetration of shorter wavelengths of visible light was effective,[3] most superficial skin tumors in our study were treated by full-spectrum visible light (400 to 760 nm) rather than by red light with wavelengths longer than 570 nm (produced through the Schott RG 570 filter). In the treatment of superficial basal cell carcinomas, full-spectrum visible light was effective in all 32 lesions, and red light in 4 of 5 treated lesions (see Table I); however, shorter exposure times could be given with full-spectrum visible light. Both were ineffective in the treatment of noduloulcerative basal cell carcinomas.

Phototoxic damage in PDT is believed to be mediated via cytotoxic substances, mainly excited singlet oxygen.[2, 3] The main targets of PDT are cell membranes.[2, 3] However, the mechanism of tissue destruction in topical PDT with endogenous porphyrins may be different from the mechanism in PDT after the intravenous injection of HPD or Photofrin II. In addition to direct tumor cell cyto-

toxicity and inflammatory response, vascular effects including endothelial cell damage, blood cell aggregation, reduction and/or cessation of blood flow, and hemorrhage are believed to contribute to the effectiveness of the latter treatment.[2] In 5-ALA–induced PDT, however, we observed in posttreatment biopsy specimens (24 and 48 hours after treatment) epidermal necrosis and acute dermal inflammation and edema, but vascular damage was not noted.

REFERENCES

1. Dougherty TJ, Kaufmann JE, Goldfarb A. Photoradiation therapy for the treatment of malignant tumors. Cancer Res 1978;38:2628-35.
2. Dougherty TJ. Photosensitizers: therapy and detection of malignant tumors. Photochem Photobiol 1987;45:879-89.
3. Henderson B. Photodynamic therapy—coming of age. Photodermatology 1989;6:200-11.
4. Manyak MJ, Russo A, Smith PD, et al. Photodynamic therapy. J Clin Oncol 1988;6:380-91.
5. Waldow SM, Lobraico RV, Kohler IK, et al. Photodynamic therapy for treatment of malignant cutaneous lesions. Lasers Surg Med 1987;7:451-6.
6. Robinson PJ, Carruth JAS, Fairris GM. Photodynamic therapy: a better treatment for widespread Bowen's disease. Br J Dermatol 1988;119:59-61.
7. Zalar GL, Poh-Fitzpatrick M, Krohn DL, et al. Induction of drug photosensitization in man after parenteral exposure to hematoporphyrin. Arch Dermatol 1977;113:1392-7.
8. Kennedy JC, Pottier RH, Pross DC. Photodynamic therapy with endogenous protoporphyrin IX: basic principles and present clinical experience. J Photochem Photobiol B Biol 1990;6:143-8.
9. Berlin NI, Neuberger A, Scott JJ. The metabolism of delta-aminolaevulinic acid. 1. Normal pathways, studied with the aid of ^{15}N. Biochemistry 1956;64:80-90.
10. Wolf P, Kerl H. Photodynamic therapy in a patient with xeroderma pigmentosum [Letter]. Lancet 1991;337:1613-4.
11. Carruth JAS. Photodynamic therapy in the treatment of diseases of the skin. Lasers Med Sci 1990;5:199-201.
12. Bernstein EF, Thomas GF, Smith PD, et al. Response of black and white guinea pig skin to photodynamic treatment using 514-nm light and dihematoporphyrin ether. Arch Dermatol 1990;126:1303-7.
13. Nelson JS, McCullough JL, Berns MW. Photodynamic therapy of human malignant melanoma xenografts in athymic nude mice. J Natl Cancer Inst 1988;80:56-60.
14. Malik Z, Lugaci H. Destruction of erythroleukaemic cells by photoactivation of endogenous porphyrins. Br J Cancer 1987;56:589-95.
15. Divaris DXG, Kennedy JC, Pottier RH. Phototoxic damage to sebaceous glands and hair follicles of mice after systemic administration of 5-aminolevulinic acid correlates with localized Protoporphyrin IX fluorescence. Am J Pathol 1990;136:891-7.
16. Pottier RH, Chow YFA, LaPlante J-P, et al. Non-invasive technique for obtaining fluorescence excitation and emission spectra in vivo. Photochem Photobiol 1986;44:679-87.
17. Malik Z, Ehrenberg B, Faraggi A. Inactivation of erythrocytic, lymphocytic and myelocytic leukemic cells by photoexcitation of endogenous porphyrins. J Photochem Photobiol B Biol 1989;4:195-205.