[O R I G I N A L   R E S E A R C H]

In-office Painless Aminolevulinic Acid Photodynamic Therapy
A Proof of Concept Study and Clinical Experience in More Than 100 Patients

GEORGE M. MARTIN, MD
Dr. George Martin Dermatology Associates, Kihei, Maui, Hawaii

ABSTRACT

Objective: To evaluate the efficacy, safety, and pain of in-office "painless" aminolevulinic acid photodynamic therapy aimed at decreasing treatment-associated pain in patients undergoing removal of actinic keratoses. **Design:** Prospective split-face study comparing short aminolevulinic acid incubation times of 15 minutes followed by extended exposure (60 minutes) of continuous blue light versus conventional aminolevulinic acid photodynamic therapy. Prospective assessment of pain in patients undergoing in-office "painless" aminolevulinic acid photodynamic therapy. **Setting:** Clinical practice office. **Participants:** Three patients with actinic keratoses participated in the split-face study and 101 in the pain assessment study. **Measurements:** Evaluations in the split-face study included removal of actinic keratoses, skin temperature, and pain measured on a 10-point visual analog scale. Pain was assessed using the visual analog scale in the pain assessment study. **Results:** In the split-face study, in-office "painless" aminolevulinic acid photodynamic therapy resulted in a 52-percent reduction in lesions versus 44 percent for conventional aminolevulinic acid photodynamic therapy. Maximum pain scores of in-office "painless" aminolevulinic acid photodynamic therapy were all 0 at each time point, and the average score for conventional aminolevulinic acid photodynamic therapy was 7. Baseline skin temperatures increased from a baseline of 29 to 32ºC to 34 to 35ºC by minute 10 of blue light activation on both sides of the face. Results from the pain assessment study indicated no or minimal (scores 0–2) pain in nearly all patients who received in-office "painless" aminolevulinic acid photodynamic therapy as monotherapy or in combination with 5-fluoruacil or imiquimod used as pretreatments. **Conclusions:** In-office "painless" aminolevulinic acid photodynamic therapy appears to be effective for removing actinic keratoses and is associated with little or no pain in nearly all patients. This procedure should be evaluated in large-scale controlled trials. (*J Clin Aesthet Dermatol.* 2016;9(2):19–26.)

Actinic keratoses (AKs) are part of the spectrum between photodamaged skin and invasive squamous cell carcinoma (SCC).[1–5] They are a major health care concern because of their increasing prevalence worldwide,[6–10] economic impact,[9–11] and decreased quality of life of affected individuals.[10,12] Results from observational studies have indicated that AKs evolve into primary invasive SCC or *in situ* SCC at a rate ranging between 1/1,000 lesions per year[13] to 0.60 percent at one year and 2.57 percent at four years.[14] It is recommended that all AKs be treated because it is not currently possible to predict which will evolve into invasive SCC.[15–17]

A variety of therapeutic modalities are used to treat AKs.[1,18–20] Focally destructive therapies, such as cryotherapy,[21] electrodessication and curettage,[22] and shave excision[23] are most often used to treat individual AKs. Large areas of actinically damaged skin require "field therapies" such as 5-fluoruacil (5-FU),[24–26] imiquimod,[26–30] diclofenac gel,[31–33] ingenol mebutate,[34,36] aminolevulinic acid photodynamic therapy (ALA PDT)[35,37] and methyl-aminolevulinic acid PDT (MAL PDT),[38,39] chemical peels,[40] dermabrasion,[41–43] and laser resurfacing.[44,45]

PDT produces reactive oxygen species that result in tissue destruction[46] and it destroys AKs because of the

preferential accumulation of the photosensitizing molecule, protoporphyrin IX (PpIX) within AKs following topical application of pro-drugs ALA[47] and MAL.[48] PDT is safe and effective for treatment of large surface skin areas, provides good adherence because it is performed under supervision in a clinic setting, has minimal post-treatment downtime versus other AK field therapies, and produces good-to-excellent cosmetic outcomes with minimal potential for scarring.[49,50] PDT also has several drawbacks, most notably pain during the first few minutes of light activation phase.[51–53] Nearly two-thirds of patients undergoing ALA PDT report this pain as "moderate-severe" following 1, 2, or 3-hour ALA incubations.[54] Pain with PDT has been related to cellular destruction and inflammation and possibly a direct effect of PDT on nerve fibers[55–57]; it has now become clear that PDT-related pain is associated with PpIX tissue accumulation based on fluorescence and the fluence rate of the activating light source.[58] Topical anesthetics,[55] cooling devices,[59–61] nerve blockade,[61,62] and treatment interruption[63] have limited efficacy in managing PDT-related pain, which can lead to reluctance of patients to undergo future PDT treatments.

A novel approach to minimizing discomfort during PDT, daylight-mediated PDT, uses a brief (30-minute) incubation period followed by 1.5 to 2.5 hours of daylight exposure.[64–66] The shortened incubation period is designed to minimize PpIX build-up in the targeted tissue prior to daylight PpIX activation, and photobleaching prevents further buildup of PpIX and minimizes patient discomfort.[67,68] Limitations to daylight-mediated PDT include dependence on favorable weather conditions and patient adherence to the treatment protocol outside the clinic.[65]

Based on the efficacy and improved tolerability of daylight-mediated PDT, an in-office painless (IOP) ALA PDT protocol was developed. It involves applying ALA topically to actinically damaged skin, incubating without occlusion for 15 minutes, and then 60 minutes of continuous blue light activation. This report summarizes results from a split-face comparison of IOP ALA PDT and a standard short (75 minute) ALA incubation protocol followed by the standard 1,000 seconds of blue light activation carried out in three patients, and assessment of pain associated with this treatment in 101 patients undergoing 121 treatments over a two-year time period. In the latter study, IOP ALA PDT was employed as either a full-face monotherapy or in combination with one week of prior treatment with 5% and 0.5% 5-FU and 3.75% and 5% imiquimod.

METHODS
Patients. All patients were selected from the private dermatology practice of the author (GMM). The study was conducted in accordance with the Declaration of Helsinki and patients provided informed consent prior to any study procedures.

All patients in the split-face study had moderate-to-severe actinic damage, were ≥18 years of age, not pregnant, not on immunosuppressant therapy within six months, had no history of photosensitizing skin disorders, not on any topical or systemic photosensitizing medications within 10 days of treatment, and had not received a field therapy for their actinic damage in the study treatment area in the preceding six months.

Patients with a history of herpes labialis were treated with oral valacyclovir 500mg daily for three days prior to their treatment date and continued on therapy for seven days post treatment. No cooling devices, topical or injectable anesthetics, or oral analgesics were used before, during, or after the procedure.

Procedures. *Pre-treatment.* On the day of the procedure, patients were instructed to shower prior to coming to the office and clean their faces with soap and water. Prior to their PDT, patients were instructed to wash their faces with Cetaphil Gentle Cleanser (Galderma Laboratories, Fort Worth, Texas), rinse, and dry. The patient's eyes were covered with disposable opaque eye shields (LASER-Aid Disposable Eye Shield; Honeywell Safety Products; Smithfield, Rhode Island) prior to and the entire time during blue light exposure.

Treatment. *Split-face study.* Patients in the intra-individual split-face study had 20% 5-ALA (Levulan Kerastick, DUSA Pharmaceuticals, Inc., Wilmington, Massachusetts) applied to the entire face. The 5-ALA was mixed and shaken for 30 seconds as recommended in the package insert,[69] applied, and left on without occlusion. The side of the face randomly selected for IOP APA PDT received a 15-minute ALA incubation followed by 60 minutes of blue light exposure ("BLU-U"; DUSA Pharmaceuticals, Inc., Wilmington, Massachusetts). The other side of the face remained covered with an opaque nonocclusive drape. At the end of the blue light exposure, the opaque dressing was removed from the untreated side and placed over the treated side. The untreated side, which had been incubating for 75 minutes following the ALA application, was then exposed to a standard dose of 1,000 seconds of blue light at 10J/cm². A registered nurse experienced in performing PDT performed the treatment protocol.

Pain assessment study. Patients in this study had 5-FU 0.5% (Carac; Valeant Pharmaceuticals North America) and 5% (Effudex; Valeant Pharmaceuticals North America), 3.75% imiquimod (3.75% Zyclara; Valeant Pharmaceuticals North America) and 5% imiquimod (Spear Dermatology Products Inc., Randolph, New Jersey) applied nightly for seven nights to the entire facial or non-facial treatment area prior to IOP ALA PDT delivered as described above on the eighth day.

Post treatment. A cool water-soaked facial cotton washcloth was applied to the treated skin, and this was followed by application of a topical clear aloe gel (Aftersun Aloe Vera Moisturizing Gel; CVS/Pharmacy, Inc. Woonsocket, Rhode Island). Treated areas were shielded from sunlight exposure using a scarf, broad-brimmed hat, and sunglasses. Patients were instructed to shower using Cetaphil Cleanser or simply rinsing with shower water applied to the treatment site upon returning home. Direct

and indirect sunlight were to be avoided for 48 hours post treatment.

Evaluations. All patients were evaluated in the clinic one week post treatment for occurrence of any adverse events following the procedure.

Split-face study. Photography. Non-hyperkeratotic AKs (minimally thick, easily seen, and felt clinically) were identified on examination by the dermatologist (GMM). The lesions were mapped and photographed using a Canfield VISIA system (Canfield Imaging Systems, Fairfield, New Jersey) at baseline and at the time of the final examination ≥8 weeks post therapy. The evaluating dermatologist was blinded during both the photographic review of the before and after photographs and during the final clinical examination.

Pain. Pain scores for were measured for both treatment sides using a 10-point visual analog scale (VAS) with patients instructed to score their maximum pain during the preceding treatment time as follows: 0=no sensation, 1–2=minimal (slight tingling or prickling sensation), 3–4=mild (mild stinging, prickling or burning sensation), 5–7=moderate (moderate stinging, prickling or burning sensation, 8–9=severe (severe stinging, prickling or burning sensation but tolerated without interruption), and 10=intolerable (severe stinging, prickling or burning sensation requiring interruption or premature termination of the procedure). Maximum pain scores were recorded at baseline and at 10-minute intervals for the 60-minutes of blue light exposure side and at baseline and at the completion of the standard 1,000-second blue light exposure.

Skin temperature. Skin temperatures were measured using an infrared temperature-measuring sensor on a Cry-Ac Tracker device (Cry-Ac Tracker Brymill Cryogenic Systems, Ellington, Connecticut). Patients underwent skin temperature measurements at four sites (midpoint of the midline of the forehead, left and right cheek 1cm below the infra-orbital rim in the mid pupillary line and in the midpoint of the chin) before and during the procedure. The blue light was turned off briefly during temperature measurements. For patients undergoing 15-minute incubations followed by 1-hour of blue light exposure, measurements were performed at baseline before ALA application, 15 minutes following ALA application, and at 10-minute intervals during 60 minutes of exposure to blue light. Temperature measurements for the side of the face receiving standard treatment were obtained at baseline, 15 minutes following ALA application, and just prior to and at the end of 1,000 seconds of blue light exposure.

Pain assessment study. Maximum pain scores were recorded at baseline and then at 10-minute intervals during treatment.

RESULTS

Patients. The split-face study included two men aged 54 and 57 years and one woman who was 50 years of age. The pain assessment study included 55 men (39 <70 years of age and 16 ≥70 years old) and 46 women (34 <70 years of age and 12 ≥70 years old).

Reduction of AK lesions. *Split-face study.* IOP ALA PDT resulted in individual lesion reduction from 27 AKs at baseline to 13 AKs on final assessment (52% reduction). The standard procedure resulted in individual AK lesion reduction from 32 to 18 (44% reduction).

Pain assessment study. No quantitative measurements of AK clearance were made in this study. Qualitative evaluations indicated that all patients had improvements in their overall actinically damaged skin. The greatest improvements were observed in those who underwent combination therapy.

Pain/temperature. *Split-face study.* Maximum pain scores for the side of the face receiving IOP ALA PDT were all 0 at each time point evaluated. The average maximum pain score for side of the face that received conventional ALA PDT was 7 (range = 6–8). Patient discomfort on this side diminished to minimal levels following discontinuation of the blue light activation. There was no increase in pain post treatment on either side of the face.

Baseline skin temperatures on both sides of the face, measured prior to topical ALA application, ranged between 29°C and 32°C. ALA had little effect on the skin temperature measured 15 minutes after application to both sides of the face in two of three patients. One patient had a temperature increase on the chin (29–35°C) and cheek (30–33°C). Temperatures measured at 10-minute intervals during 60 minutes of blue light activation showed an increase from pre-light activation temperatures to 34 to 35°C by minute 10 and the temperature remained constant in all patients throughout the course of light activation until the end. The contralateral side of the face, which underwent a 75-minute incubation under an opaque light-blocking nonocclusive barrier, demonstrated a rise in skin temperature to 34 to 35°C prior to the 1,000 seconds of light activation. The skin remained in this temperature range during and after this treatment.

Pain assessment study. Results from this study indicated no or minimal (scores 0–2) pain in nearly all patients who received IOP ALA-PDT as monotherapy or in combination with 5-FU or imiquimod (Table 1). Pain scores of 3 were recorded for three applications, scores of 5 were recorded for two applications (one after Levulan administration, prior to light activation), and a score of 6 was recorded after one application.

Safety. *Split-face study.* No significant adverse events were reported following treatment. All three patients reported erythema and mild-to-moderate edema on both sides of the face during the 24 to 48 hours after treatment. This was followed by desquamation over the subsequent seven days and resolution of the erythema and edema.

Pain assessment study. No safety assessments were carried out in this study.

DISCUSSION

The results of the proof-of-concept intra-patient split-face study demonstrated that IOP ALA PDT was associated with less pain than conventional ALA PDT and that its

TABLE 1. In-office painless aminolevulinic acid photodynamic therapy

	PDT FULL-FACE MONOTHERAPY	PDT WITH 0.5% 5-FU	PDT WITH 5% 5-FU	PDT WITH 3.75% IMIQUIMOD	PDT WITH 5% IMIQUIMOD
Number of treatments	49	24	24	22	2
Number of patients	41	22	17	20	1
MAXIMUM PAIN SCORES					
0–2	47	19	22	21	2
3	1	4	2		
4					
5		1		1*	
6	1				
7					
8					
9					
10					
* Post-Levulan					

efficacy for removal of AKs may be equivalent to standard ALA PDT treatment. No new safety signals were detected with IOP ALA PDT. Results from pain assessment study also indicated that IOP ALA PDT was associated with little or no pain when used as monotherapy or in combination with either 5-FU or imiquimod in a total of 121 treatments carried out over a two-year period. In the latter study, little or no pain was recorded when IOP ALA PDT was used to treat non-facial areas, including the scalp, trunk, and extremities, either separately or along with the face in the same treatment session.

The present results appear to be substantially different from those obtained in conventional MAL or ALA PDT in which pain during illumination is the most important and common side effect.[36,52–56] In the pivotal Phase 3 ALA PDT trial involving 14- to 18-hour ALA incubation prior to blue light activation, 90 percent of patients experienced moderate-to-severe discomfort.[36] Results from studies with shorter ALA incubation periods (1–3 hours) indicated that approximately 60 percent of patients experienced moderate-to-severe pain as moderate to severe.[54]

Multiple mechanisms, including inflammation resulting from cell death in targeted tissue and activation of Aδ and/or C fibers resulting from free radicals produced by light-activated PpIX, have been proposed to explain PDT-related pain.[55,58] The most important factor in PDT-associated pain appears to be the amount of PpIX buildup in targeted tissues prior to and during light activation.[55,58] Protocols that limit this

buildup by early and continuous light activation following application of MAL result in significant reductions in patient discomfort.[64-67] Reduced PpIX buildup in target tissue may prevent diffusion into surrounding tissues containing intact sensory nerve fibers. An additional theoretical benefit of continuous activation of PpIX produced within the target tissue is that prolonged light activation might result in efficient destruction of the targeted cells while minimizing adjacent tissue damage. The benefit of limiting PpIX accumulation prior to and during PDT for pain reduction has been supported by multiple studies that have employed daylight-mediated PDT.[64-67] The pivotal study using this approach randomized 120 patients with 1,572 AKs of the face and scalp to either 1.5 or 2.5 hours of Nordic daylight exposure during the months of June to October within 30 minutes following an in-office application of MAL to the skin followed by application of a SPF 20 sunscreen. This treatment was uniformly well-tolerated, and the authors reported a 77-percent AK clearance rate for grade I lesions three months post treatment.[66] This approach was effective for minimizing pain with 92 percent of patients reporting no or mild pain. Ambient weather influenced treatment outcomes in this study—sunnier weather and a resultant higher light dose was associated with more intense pain.[66] In contrast, results of a second study of daylight-mediated PDT indicated a linear correlation between increasing light doses and response rate, but no correlation between pain and treatment efficacy.[65]

While daylight-mediated PDT is an attractive approach for treatment of AKs, it does have significant limitations. Wide variations in the regional, seasonal, and daily weather patterns create logistics issues in implementing daylight-mediated PDT. Treatment cancellations due to inclement weather can impact office scheduling. Sudden changes in weather, such as rain or fluctuations in sunlight intensity, could potentially create delays between ALA and MAL applications and sunlight exposure, interrupt treatments, and affect the level of discomfort. Compliance issues could potentially arise, as patients are required to follow precise instructions during and after active sunlight exposure.[65] Issues involved in daylight-mediated PDT requiring further exploration include specific light exposure times during the day and season needed to photoactivate PpIX, the effect of specific weather conditions on treatment outcomes, information on the type of sunscreen use, the effect of ambient temperatures on PpIX production, protocols for photosensitizers other than MAL, and occlusion versus non-occlusion.[66]

While IOP ALA PDT eliminates issues pertaining to weather and patient adherence associated with daylight-mediated PDT, it may result in increased clinician and staff time and higher in-office space requirements. From a reimbursement standpoint, United States healthcare providers are required to follow the current PDT code, which requires the use of the BLU-U® (Dusa Pharmaceuticals, Inc., Wilmington, Massachusetts) and time spent by the provider performing the procedure. Daylight-mediated PDT does not include either of the prerequisites for proper coding and billing for reimbursement.

The efficacy for the IOP ALA PDT protocol for clearing AKs appeared similar to that for conventional ALA PDT based on results from the three patients included in that split-face study. The IOP ALA PDT protocol resulted in a clearance rate of 52 percent versus 44 percent for conventional ALA PDT. Results in the very small-scale study reported here are consistent with AK clearance rates following a single initial treatment in a Phase 2 study of ALA PDT (35.7% reduction for 47 patients treated with a 1-hour ALA incubation and 50.0% in 48 patients who received a 2-hour ALA incubation, each followed by standardized blue light exposure period of 1,000 seconds).[54] Longer ALA incubation times of 14 to 18 hours have been shown to result in higher clearance rates of 83 percent at eight weeks and 91 percent at 12 weeks post 1 or 2 treatments.[54] While effective, this long incubation period resulted in significant patient discomfort and logistic problems, and resulted in shortened incubation times of 1 to 3 hours becoming the standard of care.[71]

The AK clearance rates with IOP ALA PDT appeared less than those reported using either daylight-mediated MAL PDT (75–77%)[66] or standard red light MAL PDT (69–83%).[38,39] Differences in incubation and light activation times, both of which allow for the intracellular formation of PpIX in the targeted tissue, may in part be responsible for the increased clearance rates seen in daylight-mediated PDT versus IOP ALA PDT.

The biologic conversion of ALA and MAL to active metabolites via the heme synthesis pathway to PpIX is temperature dependent.[72-75] Higher skin temperatures are believed to be associated with increased skin penetration of PpIX, higher cellular uptake, and elevated intracellular enzyme activity. Increasing the skin temperature from 29.4ºC to 38.8ºC by using heating blankets improved the efficacy of ALA PDT in clearing AKs located on the extremities.[76]

Both IOP ALA PDT and conventional ALA PDT resulted in elevations in skin temperature from 29 to 32ºC and 34 to 35ºC. It is reasonable to suggest that these elevations in skin temperature contributed to an overall increase in PpIX tissue levels and enhanced AK clearance for both protocols.

In summary, the present results support the efficacy of IOP ALA PDT for safely removing AKs while decreasing patient discomfort. Little or no pain was observed in patients receiving IOP ALA PDT as monotherapy or in conjunction with 5-FU or imiquimod. This procedure has efficacy that appears similar to that of conventional short incubation ALA PDT as demonstrated by results from the three patients included in the split-face study. Large-scale controlled trials evaluating the IOP ALA PDT protocol are needed to further validate the observed reduction in patient discomfort and efficacy in eliminating AKs, which might include a standardized washout period for topical or systemic photosensitizing agents prior to PDT. Areas for future research might include finding the number of treatments needed to optimize the complete, partial

(≥75%), and individual lesion AK clearance rates; establishing the optimal wavelength for performing IOP ALA PDT; identifying the optimal intensity and duration of the activating light source; gaining experience using photosensitizers other than ALA, such as MAL; evaluating the effect of skin temperature on treatment outcomes; establishing the value of occlusion versus non-occlusion of the treatment site; identifying the optimal protocols for combining other AK field therapies (5-FU, imiquimod, and ingenol mebutate) with IOP ALA PDT; increasing the number of study subjects in order to determine whether there are significant differences in efficacy of IOP ALA PDT versus conventional ALAS PDT; and standardizing pre- and post-therapy skin care regimens for optimal safety, efficacy, and compliance.

ACKNOWLEDGMENTS

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