Daylight photodynamic therapy (DL-PDT), using short-chained 5-aminolaevulinate (5-ALA) ester methylaminolaevulinate (MAL), is an effective and well-tolerated treatment for actinic keratoses (AK) (1). Recently, there has been interest in the novel long-chained 5-ALA ester hexylaminolaevulinate (HAL), which has better skin penetration and can thus be used at low concentrations (2–4). This could be of economic value and reduce side-effects (5). We have reported previously that very low concentrations of HAL can be used with daylight activation (6). The current paper reports the long-term 12-month outcome.

**METHODS**

The methods are described in detail elsewhere (6). Volunteer patients with symmetrical actinic damage in the head area were recruited and treated in June 2014. AKs were photographed, counted and marked on a plastic sheet, and 2 symmetrical equally graded (6) AKs, one on each treatment side, were biopsied bilaterally before treatment. A chemical sunscreen (P20®, SPF 20 Riemann & Co. A/S, Hilleroed, Denmark) was applied for 15 min, and the treatment areas were subsequently curettaged. Patients were randomized to receive DL-PDT with 0.2% HAL (Hexvix® powder, Photocure ASA, Oslo, Norway in Unguentum M, Allmiral, Madrid, Spain) on one side of the face or scalp and 16% MAL (Metvix, Galderma, Paris, France) on the other, both applied as a 0.025 mm² thick layer (treatment area mm² × 0.25 mg/mm²). Illumination was performed for 2 h outdoors. Follow-up visits, including the mapping of the residual lesions and histological sampling, was conducted by the blinded investigator (MG). The histology (HE staining and p53 expression in average percentage of the 3 high power fields) of the samples was interpreted by a blinded pathologist (TTT). Wilcoxon signed-rank paired test was used for statistical analysis.

**RESULTS**

Of the 14 patients who completed the pilot trial, 13 were followed up for 12 months. One patient died before the 12-month follow-up due to reasons unrelated to the study. No residual lesions were treated between the 3- and 12-month follow-ups. Both treatments were nearly painless (visual analogue scale (VAS) ≤ 1). HAL caused milder adverse reactions, as assessed at 1 week (6).

At 12 months HAL DL-PDT resulted in similar sustained lesion clearance compared with MAL (Fig. 1 and Table I). The mean lesion clearance per patient was 67% with HAL and 66% with MAL (p = 1.00). HAL was as effective as MAL in the treatment of grade I AKs, but a trend for lower clearance of grade II–III AKs was seen (Fig. 2 and Table I). One patient was excluded from the histological analysis because one biopsied lesion clinically taken as an AK appeared histologically to be seborrhoeic dermatitis. Histological clearance was equal for both photosensitizers (Table I). At 12 months, 42% of the HAL-treated and 50% of the MAL-treated biopsied lesions were completely histologically cleared (p = 0.688). Compared with baseline, the mean expression of p53 was reduced by 20% in the HAL group and by 51% in the MAL group (p = 0.123).
DISCUSSION

Previously, HAL has been studied at very low 0.1% concentrations for the prevention of cutaneous squamous carcinoma in mice (7). HAL-induced protoporphyrin IX (PpIX) fluorescence has been reported in several studies and it has been shown that HAL has great potential when used at low concentrations (8–11). We reported a 3-month mean per-patient lesion clearance of 73.4% with HAL and 77.8% with MAL (6), which is in concordance with previous DL-PDT studies (1). Long-term clearance is rarely reported in DL-PDT studies. Recently, 62% and 87% mean per-patient lesion clearance was reported for MAL and amino-5-laeuvulinate nanoemulsion (BF-200 ALA) (12). In our current study at 12 months the clearances were 67% for HAL and 66% for MAL, which supports the previous 12-month data with MAL. Our results show similar efficacies for HAL and MAL in the long-term follow-up.

A limitation of our pilot study was the small sample size. As lower clearance was seen with thicker grade II–III lesions, low-concentration HAL should only be used for thin AKs. The fact that p53 expression was less reduced in the HAL group may indicate poorer reversal in the carcinogenic process. However, over a long period, there was also sustained clearance of AKs in the HAL group.

Our results show that low concentrations of HAL can be used in the treatment of thin AKs and this clearance was maintained in a long-term follow-up. The use of HAL at very low doses could lead to significant reductions in treatment costs.

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