This is an electronic reprint of the original article.
This reprint may differ from the original in pagination and typographic detail.

Author(s): Neittaanmäki-Perttu, Noora; Neittaanmäki, Eerika; Pölönen, Ilkka; Snellman, Erna; Grönroos, Mari

Title: Safety of Novel Amino-5-laevulinate Photosensitizer Precursors in Photodynamic Therapy on Healthy Human Skin

Year: 2016

Version:

Please cite the original version:

All material supplied via JYX is protected by copyright and other intellectual property rights, and duplication or sale of all or part of any of the repository collections is not permitted, except that material may be duplicated by you for your research use or educational purposes in electronic or print form. You must obtain permission for any other use. Electronic or print copies may not be offered, whether for sale or otherwise to anyone who is not an authorised user.
SHORT COMMUNICATION

Safety of Novel Amino-5-Laevulinate Photosensitizer Precursors in Photodynamic Therapy on Healthy Human Skin

Noora Neittaanmäki-Perttu1, Eerika Neittaanmäki2, Ilkka Pöllönen1, Erna Snellman3 and Mari Grönroos4
1Department of Dermatology and Allergology, Helsinki University and Helsinki University Hospital, FIN-00029 Helsinki, 2Department of Pharmacy, Helsinki University, Helsinki, 3Department of Mathematical Information Technology, University of Jyväskylä, 4Department of Dermatology, Tampere University and Tampere University Hospital, Tampere, and 5Department of Dermatology and Allergology, Päijät-Häme Social and Health Care Group, Lahti, Finland. E-mail: noora.neittaanmaki@fimnet.fi

Accepted Apr 27, 2015; Epub ahead of print May 5, 2015

Photodynamic therapy (PDT) is a highly effective treatment for superficial skin cancers and skin cancer precursors (1, 2). In PDT, the administration of a photosensitizing drug, followed by its activation by a specific wavelength of light matching the absorbance of the sensitizer, leads to a phototoxic reaction destroying the tumour cells (3). Adverse effects include pain during the illumination, and erythema and crustng after treatment. Recently, several attempts have been made to improve the tolerability of PDT (4–6). Several topical porphyrin-based photosensitizer precursors are available for PDT. Currently, amino-5-laevulinate (5-ALA) and its methyl ester (MAL) are widely used. Novel photosensitizer formulations include 5-ALA nanoemulsion (BF-200 ALA) and a long-chain lipophilic 5-aminolaevulinate hexyl-ester (HAL). These novel formulations can be used at low concentrations, which may increase the tolerability and reduce the costs of the treatment (7, 8). This non-sponsored double-blinded pilot study tested the safety of PDT with BF-200 ALA and HAL at 2 different concentrations compared with MAL on healthy human skin.

MATERIALS AND METHODS

The study was approved by the local ethics committee. We included 7 healthy volunteers, aged between 26 and 34 years, with skin phototypes I–II, to receive PDT on the sun-protected area and dividing it by the fluorescence of untreated skin of the same patient. The severity of the reactions at 1 and 2 days after illumination was evaluated from photographs by a blinded observer (NNP). The fluorescence images were taken before and immediately after the illumination using a visual analogue scale (VAS). Fluorescence images were taken before and immediately after the illumination using Wood’s light (Philips Burton®, Somerset, USA) and a digital camera (Canon Ixus 10 megapixels). Fluorescence intensity and photobleaching were calculated from the images using the MatLab® (Mathworks, Natick, MA, USA). The fluorescence index in arbitrary units (AU) was calculated by measuring the mean fluorescence of the treated area and dividing it by the fluorescence of untreated skin of the same patient. Friedman’s test (9). Friedman’s test and Pearson’s correlation were used for statistical analyses.

RESULTS

Mean maximal pain during the illumination was significantly lower for HAL 0.2% and HAL 2% compared with BF-200 ALA and MAL (p = 0.043). No significant difference was found in pain due to BF-200 ALA and MAL.

![Fig. 1.](image-url) (a) Mean maximal pain values during the illumination. Hexyl-ester (HAL) 0.2% and HAL 2% caused significantly less pain compared with amino-5-laevulinate (5-ALA) nanoemulsion (BF-200 ALA) and the methyl ester (MAL) of 5-ALA (p = 0.043), while no difference was found in the pain between BF-200 ALA and MAL. (b) Erythema% compared with the baseline. Directly after illumination the erythema values differed between all 4 photosensitizers, p = 0.0041. During 2 post-treatment days HAL 0.2% caused significantly less erythema compared with BF-200 ALA (p = 0.003 at the first and p = 0.001 at the second post-treatment day) and MAL (p = 0.043 at the first and p = 0.023 on the second post-treatment day). No significant difference was found between MAL and BF-200 ALA or HAL 2%.
The mean ± SD maximal pain scores (VAS 1–10) were 4.2 ± 2.4 for BF-200 ALA, 3.8 ± 2.8 for MAL, 2.3 ± 2.2 for 2% HAL and 1.6 ± 2.0 for 0.2% HAL (Fig. 1a).

The erythema values are shown in Fig. 1b. A significant difference was found in the erythema between HAL 0.2% and BF-200 ALA (p = 0.003 on the first and p = 0.001 on the second post-treatment day), and between HAL 0.2% and MAL (p = 0.043 on the first and p = 0.023 on the second post-treatment day). No significant difference was found between MAL and BF-200 ALA or HAL 2%.

The treatment reactions of the investigated compounds are summarized in Table I. The reactions were similar on the second post-treatment day (Fig. S1).

Reduction in the initial protoporphyrin fluorescence induced by the photosensitizers, i.e. photo-bleaching, can predict the treatment efficacy (10). After 3 h occlusion the fluorescence was equal in the BF-200 ALA, MAL and HAL2% groups (ns), but lower in the HAL 0.2% group (p = 0.043). Photobleaching was equal with BF-200 ALA, MAL and HAL2%, while significantly lower photobleaching was seen with HAL 0.2% (p = 0.003 and p = 0.023 compared with BF-200 ALA and MAL).

There was a strong positive correlation between photo-bleaching and the clinically assessed reaction severity, and a weaker positive correlation between photobleaching and erythema measured with the spectrometer. There was no correlation between mean maximal pain and photo-bleaching, erythema or total protoporphyrin IX (PpIX) fluorescence.

DISCUSSION

These results show that low-concentration HAL was better tolerated than BF-200 ALA or MAL with regards to pain and erythema. Interestingly, while better tolerated, HAL 2% produced similar fluorescence and photobleaching to that of MAL and BF-200 ALA. The fact that all photosensitizers induced pain and erythema on non-photo-damaged healthy skin indicates that the effect of PDT is not completely specific to cancerous tissues.

A limitation of our trial was the small sample size and the lack of an accurate fluorescence imaging system. As our study was limited to healthy human skin, further research is needed into the safety and efficacy of these novel photosensitizers in cancerous skin.

Previously PDT with MAL 20% was less painful and caused less erythema compared with ALA 20% on healthy sun-exposed human skin (11). We showed that BF-200 ALA was as well tolerated as MAL. This may be due to the lower concentration of 5-ALA in this nano-formulation. In UV-exposed mice, HAL 2% induced similar epidermal fluorescence to that of MAL 20% (12). Our finding of strong fluorescence and photobleaching with only 2% HAL supports these findings and further indicates the better tolerability of HAL.

These pilot results show better tolerability of low-concentration HAL compared with BF-200 ALA and MAL on healthy human skin. However, further research is required to determine whether the treatment might be sufficiently effective without erythema and pain.

ACKNOWLEDGEMENTS

The authors would like to thank our healthy volunteers for participating in the study and MSc Reeta Neitaaamäki for statistical assistance. The study was supported by the Foundation for Clinical Chemistry Research.

Conflicts of interest. NNP has received travel grants from Biofrontera and Galderma and speaker honoraria from Biofrontera and Desitin Pharma. EN, IP, MG, and ES have no conflicts of interest.

REFERENCES

7. Peng Q, Berg K, Moan J, Kongshaug M, Nesland JM.


