

ASSESSING BURN DEPTH IN TATTOOED BURN LESIONS WITH LASCA IMAGING

ÉVALUATION DE LA PROFONDEUR D'UNE BRÛLURE DE PEAU TATOUÉE PAR LASER

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SUMMARY. Tattoos are on the rise, and so are patients with tattooed burn lesions. A proper assessment with regard to burn depth is often impeded by the tattoo dye. Laser speckle contrast analysis (LASCA) is a technique that evaluates burn lesions via relative perfusion analysis. We assessed the effect of tattoo skin pigmentation on LASCA perfusion imaging in a multicolour tattooed patient. Depth of burn lesions in multi-coloured tattooed and untattooed skin was assessed using LASCA. Relative perfusion was measured in perfusion units (PU) and compared to various pigment colours, then correlated with the clinical evaluation of the lesion. Superficial partial thickness burn (SPTB) lesions showed significantly elevated perfusion units (PU) compared to normal skin; deep partial thickness burns showed decreased PU levels. PU of various tattoo pigments to normal skin showed either significantly lower values (blue, red, pink) or significantly increased values (black) whereas orange and yellow pigment showed values comparable to normal skin. In SPTB, black and blue pigment showed reduced perfusion; yellow pigment was similar to normal SPTB burn. Deep partial thickness burn (DPTB) lesions in tattoos did not show significant differences to normal DPTB lesions for black, green and red. Tattoo pigments alter the results of perfusion patterns assessed with LASCA both in normal and burned skin. Yellow pigments do not seem to interfere with LASCA assessment. However proper determination of burn depth both in SPTB and DPTB by LASCA is limited by the heterogenic alterations of the various pigment colours.

Keywords: laser speckle, laser Doppler, LPI, LDI

RÉSUMÉ. Les tatouages sont de plus en plus fréquents, et donc l'incidence des brûlures sur peau tatouée augmente. L'évaluation clinique correcte de la profondeur de la brûlure se heurte à la pigmentation du tatouage. Nous avons utilisé dans ce but l'imagerie laser, chez un patient porteur d'un tatouage multicolore, dans des zones le portant et ne le portant pas. La perfusion relative était mesurée en Unités de Perfusion (UP), comparée à des pigments colorés connus, et à l'aspect clinique. Les brûlures superficielles étaient plus perfusées que la peau normale, quand les brûlures profondes l'étaient moins. Les pigments bleus, rouges et rose entraînent une diminution des UP de la peau saine, les noirs une augmentation, l'effet artéfactuel des oranges et des jaunes étant non significatif. En cas de brûlure superficielle, les zones bleues et noires ont une perfusion diminuée, alors que le jaune ne change pas le résultat comparativement à une telle brûlure en zone non tatouée. L'aspect laser des brûlures profondes sur des tatouages noirs, rouges ou verts est semblable à celui observé avec les mêmes brûlures sur peau non tatouée. Les tatouages changent les résultats de l'évaluation laser de la perfusion cutanée, que la peau soit normale ou brûlée, sauf avec les pigments jaunes. En cas de tatouage multicolore, l'évaluation de la profondeur d'une brûlure par laser est prise en défaut.

Mots-clés: imagerie Doppler, tatouage multicolore, laser

Introduction

The number of people getting tattoos has been constantly on the rise in western countries over the last decade.^{1,2} Prevalence of tattoos in the 18 to 50-year-old population in Europe is estimated to be around 10%, and 25% in the United States.^{2,3} This is also reflected in an increased incidence of patients with tattoos admitted to burn units. Size and colouration of the tattoo may severely limit the possibility to properly assess the depth of the burn lesions just by visual examination. This in turn may negatively influence the subsequent treatment algorithm. Therefore, it is necessary to explore different modalities to

properly evaluate burned tattooed skin. Traditionally, assessment of the depth of a burn lesion is performed by subjective judgment of the skin and typical signs of corresponding burn lesions (*Table I*).⁴ For a trained and experienced physician, the distinction is typically obvious in 60-75% of cases.^{5,6} The most difficult distinction is between superficial partial thickness burn (SPTB) and deep partial thickness burn (DPTB). As the resulting treatment differs to a great extent – conservative treatment for the former, surgical treatment for the latter - numerous attempts have been made to objectify this distinction.⁷ One of the most promising and frequently applied methods in this regard is laser Doppler imaging (LDI).⁸ In the field of burn ther-

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Table I - Criteria for visual clinical assessment of burn depth

	Skin	Colour	Recap	Histological depth	Scoring	Pain
First degree burn	intact	red	1-2 sec.	epithelial	bleeding	painful
SPTB	blistering	red, pink	1-2 sec.	epithelial x-y mm	bleeding	very painful
DPTB	blistering	white	no/slow recap	deep epithelial x-y mm	no bleeding	reduced pain
Third degree burn	hardened	white	no	dermal	no bleeding	no pain
Fourth degree burn	destroyed	grey/black	no	muscle, bone	no bleeding	no pain

apy, level of blood flow has been correlated to depth of the burn lesion,^{5,9} and therefore is used to assess the depth of burn wounds. LDI is based on the analysis of optical reflections of laser light on moving blood cells.¹⁰ However, skin colour alters perfusion values.¹⁰ Our hypothesis is that the kind of dye used for tattooing skin influences the result of laser-Doppler based imaging. On a patient with numerous multi-coloured tattoos and burn lesions of various depths, we studied the application of LASCA for burn depth assessment in tattooed individuals.

Material and methods

Primary burn lesions were assessed for clinical signs such as blistering, time of recapillarization and, in areas of uncertain depth, scoring with a needle. Our first step assessed all burn lesions in normal skin by grading them clinically, from first-degree burn to full thickness burn. We then evaluated burn lesions in tattoo areas in a similar fashion. Following clinical assessment, we used laser speckle contrast analysis (LASCA) imaging (PSI-Imager®, Perimed, Sweden) to analyse the skin on the first day after admission. The laser was always posi-

tioned at a distance of 20cm from the skin. In order to address the temperature sensibility of the device, room temperature was always kept at 20°C when measuring. To be able to compare measurable changes in perfusion with the device, we established a baseline of unburned skin areas without tattoos and unaffected tattooed skin for all given colours. The authors used the device to assess burned normal skin as well as burned tattooed skin in various colours. In each area of interest, normal skin sample analysis was performed in direct proximity to the burned/tattooed area to take local skin variations into account. A safety distance of 0.5cm from the burn lesion was respected to avoid distortion of the results due to hyperemia in the surrounding skin.

Laser Doppler imaging uses an invisible laser that creates a speckle pattern over a designated skin area and registers changes and variations of this pattern due to blood perfusion. Perfusion unit (PU) calculation as a measurement for LPI for the obtained images and data was measured using the PIMsoft (Perimed, Sweden) software analysis tool. For each analysed colour, three different areas within the same burn/skin zone were included (Fig. 1).

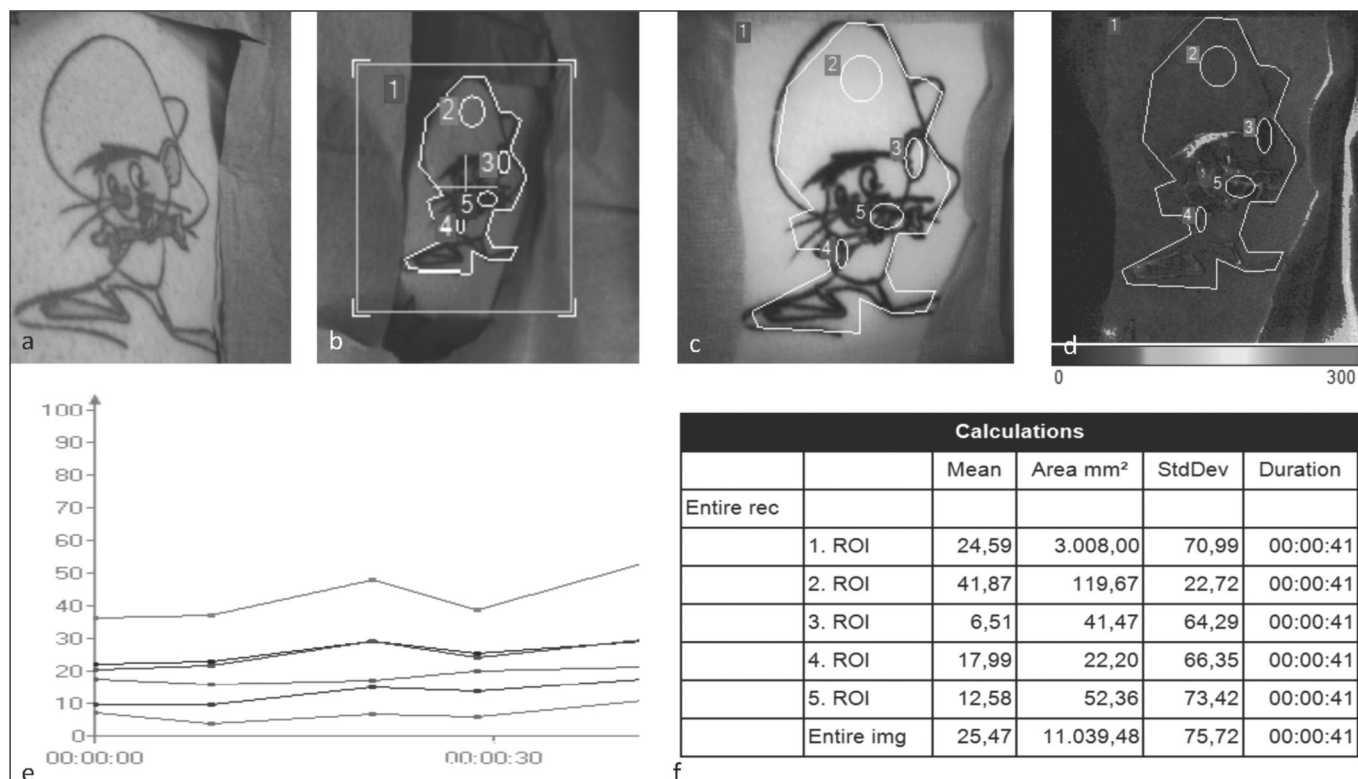


Fig. 1 - a) Multi-colour tattoo on healthy skin, normal photograph; b) the same multi-colour tattoo as a photograph through a laser-doppler device, circles indicate exemplary regions of interest within the tattoo (1 = healthy skin without tattoo, 2 = yellow pigment, 3-5 = pink pigment); c) grayscale image of the area; d) laser speckle contrast image of the tattoo; e-f) table and graph with corresponding perfusion units per region of interest (ROI) as indicated above.

The treatment was solely based on clinical assessment: second, first and SPTB lesions were treated with polyhexanid hydrogel and fatty gauze dressings with daily changes, and DPTB lesions received surgical necrosectomy with split-skin grafting. This was done in order to be able to retrospectively correlate LASCA results with clinical assessment.

We first compared PU of different burn depths to normal skin, and secondly different tattoo pigmentations of healthy skin to normal skin. We then compared PU of burned tattoos

to healthy tattoos and untattooed burn lesion PUs. Lastly, we compared PU results to the clinically determined burn depths.

The authors further calculated a LASCA score for the pigments with sufficient values, where PU of SPTB and DPTB were divided by the PU for the respective unburned normal skin.

Statistical analysis using Student's t-test and ANOVA was performed with Prism 6 (Graphpad Software Inc., USA).

Results

A 51-year-old Caucasian patient was admitted with burn lesions due to flames from flying sparks from an angle grinder that he had used for construction work in his basement. The affected burn TBSA on admission was 25%. Our patient had multiple multi-coloured tattoos all over his body, depicting numerous Looney Tunes characters among others (Figs. 2a-b). Some of these tattoos were part of the burned skin in varying burn depths.

Mean perfusion units (PU) for normal skin were 41.1 (SD 1.6). PU in first and superficial partial thickness burns (SPTB) on normal skin were 200.5 (SD 37.9) and PU values in deep partial thickness burns (DPTB) were 99.3 (SD 51.3). When compared to normal skin, SPTB values were significantly higher ($p=.002$), whereas DPTB values were elevated but not significantly so ($p=.162$).

Unburned tattooed skin could be analysed for black, blue, red, yellow, pink and orange pigmentation. Mean PU results for black were 84.5 (SD ± 3.8), blue 27.8 (SD ± 1.6), red 12.2 (SD ± 0.9), yellow 41.9 (SD ± 1.8), pink 11.9 (SD ± 3.6) and orange 50.4 (SD ± 1.4). Comparison to normal skin showed significant alterations in ANOVA ($F(6/2) = 176.9, p=.001$). Black pigments showed significantly higher PU. Blue, pink and red pigments' PU were significantly reduced, whereas yellow and orange pigments did not show significant differences (Fig. 3).

Tattoo pigments accessible for analysis in clinically determined SPTB were black, blue and yellow. Their respective PU values were 86.4 (SD ± 18.6), 35 (SD ± 2.4) and 159.5 (SD ± 83.5). One-way ANOVA did not show overall significantly altered PU when compared to normal SPTB burned skin (PU 200.5; $F(3/2) = 5.805, p=.134$); PU of black and blue pigments were however significantly reduced when analysed individually (Figs. 4a-b). When compared to normal skin, PU levels of



Fig. 2 - a) 51y patient with multiple burn lesions of various depths on normal skin and multi-coloured tattoos; b) close up of burned tattooed lesions, clinically suspected to be full thickness second degree burn.

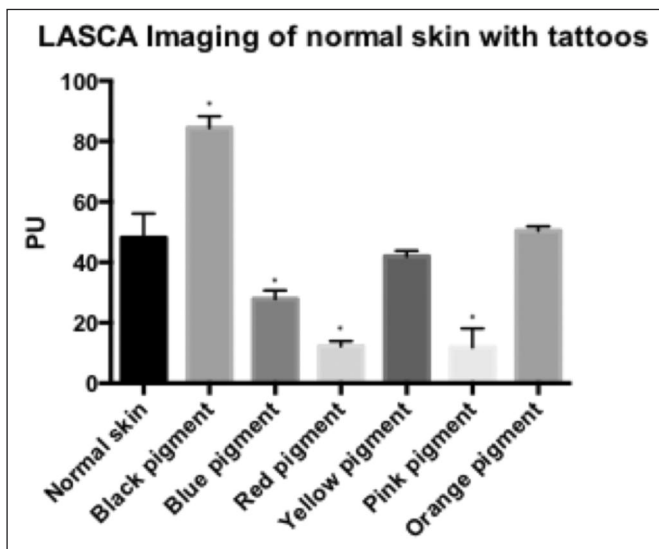


Fig. 3 - Laser Speckled Contrast Analysis (LASCA) of normal skin with tattoos; * = significantly different to normal skin.

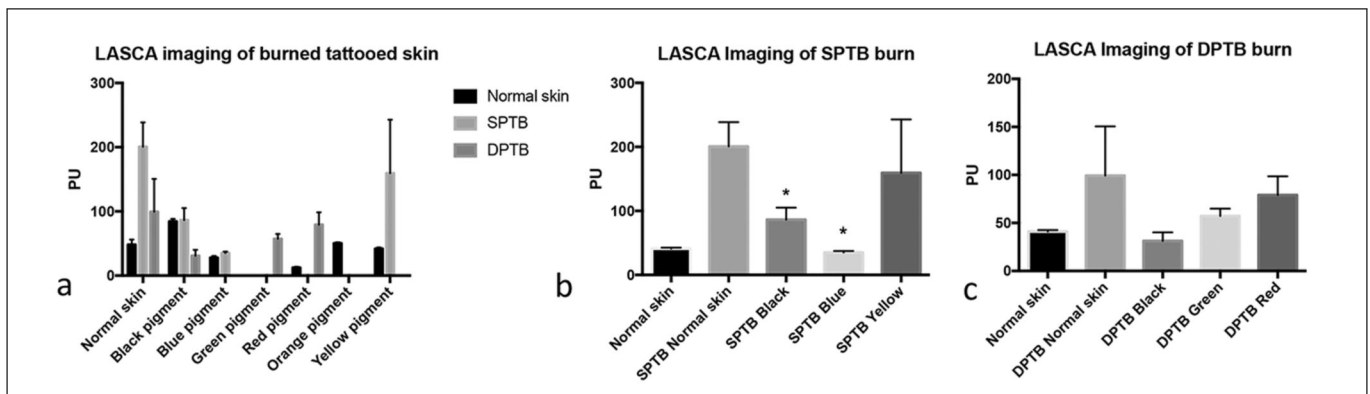


Fig. 4 - a) LASCA of tattooed normal skin and tattooed burned skin – superficial partial thickness burn (SPTB) and deep partial thickness burn (DPTB); b) LASCA of SPTB burn lesions with tattoo pigmentation; c) PU values of LASCA of DPTB; * = significantly decreased perfusion units (PU) compared to normal skin SPTB.

Table II - LASCA score for normal and pigmented skin with ratio of perfusion units of superficial/deep partial thickness burn (SPTB/DPTB) to normal skin for selected pigments

	Normal skin ratio	Black pigment ratio	Blue pigment ratio	Red pigment ratio	Yellow pigment ratio
Normal skin / SPTB	4.86	1.02	1.26		3.8
Normal skin / DPTB	2.41	0.37		6.45	

the pigmented SPTB were not significantly different (ANOVA $F(3/2) = 4.870, p = .155$).

Comparing normal skin with DPTB lesions (PU 99.3, SD ± 51.3) to pigmented DPTB lesions showed no statistical differences (ANOVA $F(3/2) = 3.361, p = .181$) in the PU levels of black pigment (31, SD ± 9.1), green pigment (57.2, SD ± 7.6) and red pigment (79.1, SD ± 19.5). This remained when compared to unpigmented normal skin (PU 41.1) also (ANOVA $F(3/2) = 10.04, p = .0748$) (Fig. 4c).

The LASCA score showed a 4.9-fold increase of SPTB lesions to unburned skin for untattooed skin, and a 2.4-fold increase for DPTB. Ratios for black pigments were 1x (SPTB) and 0.4x (DPTB), 1.3x for SPTB blue pigments, 6.5x for DPTB for red pigments and 3.8x for SPTB for yellow pigments (Table II).

Discussion

Our study demonstrates alterations in the perfusion patterns of the skin in varying burn depths and for multiple colour pigments. Perfusion units measured by the LASCA device seem to be strongly influenced by different colour pigments, as shown by the heterogeneity of PU of different pigments in healthy unburned skin. Interestingly, black pigments led to increased PU in our patient, whereas blue pigments showed reduced PU. Pigments of a spectrum close to that of Caucasian skin, such as yellow and orange, seem to have similar perfusion characteristics to normal skin – showing only a little interference with the regular speckle pattern. Looking at untattooed burned skin of various burn depths, SPTB lesions showed an almost 5-fold increase in perfusion units, and DPTB a 2.4-fold increase, indicating relative hyperemia in these lesions. When compared to the PU of tattooed burns, only yellow pigments showed a comparable increase of PU in SPTB, whereas black and blue pigments remained at a comparable level to their normal skin values. PU in DPTB did not correlate with the values for unpigmented skin either. While red pigments showed a 6-fold increase of PU, black pigments showed a 0.4-fold decrease as shown by the LASCA score. These alterations are very heterogenic and seem to be different for every colour, irrespective of burn depth.

Unlike the previous study by McGill et al.,¹¹ we were able to detect LASCA signals also in black pigments, with contradictory results, as we had already seen increased PU in healthy skin. This may be due to application of different chemical agents used as tattoo pigments (Table III): there is a wide variety, even for similar colours, potentially due to mixed pigments. Tattoo colours are derived from a large spectrum of pigments that are injected into the dermis or dermal-epidermal border. They can be classified as organic or inorganic substances (Table III) and comprise a number of substances specifically designed for that purpose, as well as unadvised substances at times. These pigments are not standardized, neither in production nor in application, and can differ widely among countries. The visible colour of a tattoo does not only depend on the pigment but also on the amount of pigment used, depth of injection as well as skin type.

Table III - Most common organic and inorganic pigments used for tattoos

Organic pigments	Inorganic pigments
azo pigments	titanium dioxide
polycyclic pigments	barium sulfate
pigment black	iron oxide

In our case, the heterogeneous results of the different pigments - even in normal unburned skin - are difficult to interpret. We can only speculate that the unknown pigment substances lead to unique reflection patterns, which are responsible for these results.

The significant increase in perfusion in superficial partial thickness burns is distinctive for this lesion and has been previously described.¹²⁻¹⁴ This correlates well with the clinical presentation of SPTB with reddish hyperemia in the lesion. With regard to the relative increase of perfusion in DPTB, we speculate that the reactive hyperemia as physiological reaction is still taking place, but its visibility is limited due to the deeper burn of epidermal tissue. Clinical wounds of DPTB present with a whitish lesion, which reflects reduced perfusion compared to SPTB.

In the pursuit of optimal tools that will help burn specialists to better assess difficult burn injuries such as tattooed lesions, our results contribute to a better understanding of promising techniques such as laser Doppler imaging and their limitations in these settings. To our knowledge, the results of LASCA in multi-coloured tattoos, both in normal and in superficial burn lesions, have not yet been studied by any other group. In the light of our data, LASCA may not be able to reliably help to determine burn depth in tattooed burns.

One technical limitation is that laser Doppler imaging by itself is not able to determine burn depth - but it has been shown that the measured perfusion of burned areas correlates well with burn depths and is superior to the clinical judgment of an experienced surgeon.^{5,15-17}

We tried to circumvent potential confounders by taking control skin LASCA values from intact skin in direct proximity to the pigmented and/or burned lesion. The distance was far enough away to avoid false elevated values due to hyperemia in skin areas directly surrounding burn lesions, as has been described by Jeng et al.¹⁸ We also used three different areas within the same colour or lesion to create mean values to counteract the partially huge standard deviations, which may be the result of the above-mentioned challenges with tattoo pigment substances.

As the utility of speckle-based perfusion imaging was unclear at the beginning of the measurements, clinical evaluation and determination of burn depth was performed. This took into account conventional signs such as blistering, time of recirculation and, in areas of uncertain depth, scoring with a needle. Due to the nature of tattooed burn lesions, this clinical assessment is challenging, subjective and one of the limitations of this study. Furthermore, not all colours were consistently represented in all skin areas of interest (normal skin, SPTB,

DPTB), which limits the conclusions we can draw from the perfusion patterns. Even though we only had one patient to assess for this study, we consider this to be more of an advantage due to consistent skin type and most likely also inter-individually comparable tattoo pigments. Current understanding of speckle-based perfusion imaging to assess burn lesions is still limited, partially due to reported experiences mainly in Caucasian patients. Current studies lack data regarding the impact of race-related skin pigmentation or correlation with Fitzpatrick skin types. If and how tattoo pigmentation in other skin types may alter LASCA imaging results can only be speculated.

With regard to the rising prevalence of tattoos in the general population and hence in the population of burn victims, we consider this to be an important piece of the puzzle in determining ideal, objectifiable assessment tools for burn lesions in tattooed patients and subsequent treatment.

In summary, we see major limitations in the application of LASCA for standard evaluation of burn depths in tattooed lesions. In areas where pigments are in a yellow-orange colour spectrum, it may yield proper assessment comparable to normal unburned skin.

Even though future research is warranted, it will be challenged by the diversity of tattoo pigments available and their individual representation.

Conclusion

LASCA analysis of tattooed burn lesions may not help in correctly assessing burn depth. Variability of the pigments used for tattoos and their individual presentation are likely to be the main reason. Tattoos in yellow to orange colours can be assessed with LASCA as an additional tool to differentiate lesions with unclear clinical evaluation.

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