Changes of the skin barrier and bacterial colonization after hair removal by clipper and by razor

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Abstract. Background: Inappropriate hair removal increases the risk of surgical site infections which are associated with a higher morbidity and mortality of surgical patients. Here, the effects of a clipping device and a disposable razor on the skin barrier, microbial burden and surface structure were compared. Methods: Changes in bacterial colonization, transepidermal water loss, antioxidant status and the skin surface structure were investigated on the calves of 12 healthy volunteers. Measurement time points were at baseline (t_{base}) and 24 hours after hair removal (t₂₄). *Results:* Both, the disposable razor and the clipper showed a decrease in log colony-forming units count from t_{base} (mean(t_{base}) ± standard deviation = 2.6 ± 1.27, median ± standard error = 2.6 \pm 0.37) to t₂₄ at p_{razo} = 0.05 and p_{clipper} = 0.06 respectively. At t₂₄ clipping resulted in a higher reduction of log colony-forming units (mean(t_{24}) = 1.76 ± 0.8, median = 1.69 \pm 0.23) compared to the use of the disposable razor (mean(t₂₄) = 1.84 \pm 0.85, median = 1.91 ± 0.24). Furthermore, the razor-treated group showed an increase in colony-forming units from t₀ to t₂₄, whereas clipping lead to a continuous decrease in colony-forming units from t_0 to t_{24} . An enhanced appearance of microlesions and a significant increase of transepidermal water loss after shaving using the disposable razor (p = 0.005) were found indicating skin barrier disruptions. Clipping showed no significant effect on transepidermal water loss. Conclusion: Hair removal using the clipping device results in less disruption of the skin barrier compared to the razor, avoiding the development of microlesions. This could be favorable for the prevention of surgical side infections and postoperative wound management. © 2016 Journal of **Biomedical Photonics & Engineering.**

Keywords: preoperative hair removal, skin barrier disruption, post-operative, postsurgical infection, clipping.

Paper #2997 received 2016.03.31 revised manuscript received 2016.05.27; accepted for publication 2016.05.30; published online 2016.06.14. <u>doi: 10.18287/JBPE16.02.020303</u>

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1 Introduction

Preoperative hair removal within the scope of surgical disinfection has been controversially discussed and is a critical factor for the prevention of postsurgical wound infections. Preoperative hair removal is practiced to ensure a clear surgical site to facilitate the peri- and postoperative wound care and the application of wound dressings as well as to prevent the contamination of the surgical site with hair sticking pathogens [1-3].

However, several clinical studies have shown that hair removal by conventional razors can cause incisions and other microscopic injuries, to the epidermis, which serve as a reservoir for bacterial colonisation. This may favour bacterial contamination of the surgical field and lead to an increased risk of surgical site infections (SSI) [4-12].

The prevention of SSI is of great importance, since surgical wounds show a delayed wound healing process after infection prolonging the duration of the hospital stay [13]. Furthermore this can cause unnecessary pain, it can have a negative impact on the operation result and is associated with substantial morbidity and mortality [14-16]. It has been shown in several trials that the infection risk after shaving is increased in comparison to hair clipping [4, 6, 7, 17-19]. Apart from the hairremoving device, the point in time may also play a crucial role for the prevention of postsurgical infections. If hair removal is desired, previous investigations indicate that the preferable time is immediately before surgery [13, 20]. However, only few studies have been conducted investigating the most convenient point in time for clipping and these studies did not include the effect on the outcome in surgical patients [1].

In the present study, the effect of hair removal on the skin barrier and microbial burden was studied at up to two time points, directly after (t0) and 24h after (t24) hair removal and compared to baseline measurements (tbase), which were performed immediately before hair removal. A surgical clipper with a disposable shaving head and a conventional single-use shaver have been investigated. We compared the skin surface of treated subjects using laser scanning microscopy (LSM) (t24), measuring the transepidermal water loss (TEWL) (t24), the antioxidant status of the skin in form of carotenoid concentration (t24) and the bacterial growth (t0 and t24) after hair removal. TEWL is a well-suited indicator of changes in the skin barrier function showing increased values in case of a disrupted skin barrier. In vivo LSM is a non-invasive spectroscopic method to visualize the skin surface structure and evaluate the skin barrier at the cellular level [21]. Furthermore, using resonance Raman spectroscopy the antioxidant status of the skin was determined non-invasively indicating changes in the production of free radicals due to cellular impairment or inflammatory processes. Moreover, taking into consideration that the highest concentration of carotenoid antioxidants can be detected in the uppermost layers of the stratum corneum [22], the razor can potentially influence the epidermal carotenoids.

2 Methods

2.1 Study design

12 healthy male subjects aged between 20 and 40 years have been enrolled in the study. The investigations have been performed on hairy skin areas of the calves. The hair removal devices have been randomly assigned to either the right or left calf of the subject.

On both calves the observation areas for the measurements were marked with a template and assigned to the different measuring devices and time points. All measurements took place prior to the hair removal and 24 hours afterwards. Bacterial growth was additionally investigated directly after hair removal to further investigate the temporal course of bacterial colonization. After baseline assessments before hair removal both calves were shaved and clipped with the assigned method according to the manufacturer's instructions of use. The subjects were asked to indicate the painfulness of the hair removing procedure on a scale from 1 to 10. All volunteers participating in the study had given their written informed consent. The study had been approved by the Ethics Committee of the Charité - Universitätsmedizin Berlin and was performed according to the declaration of Helsinki.

2.2 Hair removal devices

The Clipper Professional 9681 from 3M Medica is a commercially available medical device, which is intended for pre- and perioperative hair removal. It was applied with the disposable shaving head 9680 from the

same manufacturer and compared with a conventional single-use razor (Wilkinson Sword GmbH, Solingen, Germany).

2.3 Transepidermal water loss (TEWL)

TEWL measurements were conducted using a standard protocol before and 24h after hair removal. Prior to TEWL measurements the volunteers needed to acclimatize in standardized climatic conditions. For acclimatization the calves of the measured subjects were uncovered and relaxed at a temperature of 20 ± 2 °C and $50 \pm 10\%$ relative humidity (rH) for at least 30 minutes before the actual measurements began. Measurements were conducted using a Tewameter® TM 300 (Courate-Khazaka, Cologne, Germany) with appropriate software.



Fig. 1 Clipper Professional 9681 (3M Medica).



Fig. 2 Disposable razor (Wilkinson Sword GmbH).

2.4 Resonance Raman spectroscopy

Resonance Raman spectroscopy was used in order to determine changes in the carotenoid concentration of the skin before and 24h after hair removal [23]. It has been shown in previous investigations that carotenoids can be measured as marker substance for the whole antioxidant status of the skin, since the antioxidant substances in the skin form protective chains that work against the destructive effects of free radicals [24, 25]. Based on carotenoid absorption maxima, which lies in the blue-green range of the spectra, the excitation laser radiation was set at 488 and 514 nm on the skin [26]. As a result, the resonance amplification of the Raman signal of cutaneous carotenoids occurs and is easily detected on the high fluorescence background. The twowavelength excitation scheme is used to determine most prevalent cutaneous carotenoids beta-carotene and lycopene separately [27]. The technical description of the utilized device, its advantages and limitations in comparison to other measuring techniques, were previously described in detail by our group [27, 28].

2.5 Bacterial colonisation

The bacterial colonisation was determined prior to (t_{base}) , directly after (t_0) and 24 hours after (t_{24}) application of the clipper and the razor respectively.

The number of colony-forming units (CFU) was determined according to a standardized protocol [29]. A sterile stainless steel ring with a diameter of 2 cm was applied on the respective skin sites on the calves of each subject. Then 1 ml of a solution, consisting of 50% phosphate buffer solution (Dulbecoo's PBS, Laboratory GmbH, Graz, Austria) and 50% egg yolk, were filled inside the ring. Afterwards the solution was homogeneously distributed inside the ring with an aseptic applicator for 30 seconds followed by the removal of 0.5 ml of the supernatant. The 0.5 ml supernatant were afterwards diluted in 4.5 ml basic solution, resulting in a ratio of 1:10 and vortexed homogeneously. 0.5 ml of the 1:10 solution were then applied on an agar plate (TSA with 5% sheep blood) and incubated for 24 hours at 37°C. Afterwards the number of CFU of each plate was counted.

2.6 Laser scanning microscopy (LSM)

In vivo LSM was applied in order to evaluate the skin surface and to visualize potential microlesions in the epidermal layers. The Vivascope® 1500 (MAVIG GmbH, Munich, Germany) with an optical lateral resolution of less than 1.25 μ m and a vertical resolution of 3 to 5 μ m was used in this study. Due to a pinhole attenuating the light from out-of-focus planes this high resolution of reflectance and fluorescence confocal laser scanning microscopy in vivo with three different excitation wavelengths (488 nm, 658 nm and 785 nm) and three filter sets detecting reflectance, fluorescence and a combination of both signals.

The technique is based on the reflected and fluorescent light from examined skin structures within an area of 500 μ m x 500 μ m with different refractive indices and reflection patterns that are translated and displayed as greyscale images of 1000 x 1000 pixels. In this study, sodium fluorescein was used as fluorescence-active dye using the excitation wavelength at 488nm to visualize structural changes of the skin surface. Image processing and analysis was conducted using customised VivaScope software.

2.7 Statistical analysis

The statistical analysis of the measured results by TEWL, bacterial colonization and resonance Raman spectroscopy was conducted using IBM SPSS vs. 19. Standard deviation was calculated as a measure of spread for mean values; standard error was calculated for median values. Wilcoxon signed-rank test was used for analysis of related samples. Shapiro-Wilk test was performed to test normality assumptions. Statistical analysis of CFU data was performed based on log reduction. Tbase values were calculated from individual average log10 CFU counts of both calves.

3 Results

3.1 Antioxidant status of the skin

Raman signal intensities of carotenoids and thus relative concentrations of carotenoids in the skin showed no significant differences before and after hair removal. Neither the conventional razor nor the clipper lead to statistically significant changes in beta-carotene or lycopene signal intensities.

3.2 Bacterial colonisation

The average log10 CFU of both unshaved calves at tbase showed a high individual variance and showed a mean count of 2.6 ± 1.27 (median = 2.6 ± 0.37). Both the conventional razor and the clipper showed a reduction of bacterial colonisation 24 hours after hair removal. Shaving by using the disposable razor lead to a significant reduction in log reduced CFU values from t_{base} to t₂₄ (mean = 1.84 ± 0.85 , median = 1.91 ± 0.24) significant at p = 0.05, while the clipper showed an even higher mean reduction in CFU from t_{base} to t₂₄ (mean = 1.76 ± 0.8 , median = 1.69 ± 0.23) at p = 0.06 (Figure 3). The change from before clipping and shaving to 24 hours afterwards was found at 0.4 to 0.5 logs.

The investigations of CFU immediately after hair removal at time point t0 showed a notable reduction in both treatment groups that was not significant. After 24 hours though, shaving with the clipper lead to a further and continuous decrease in CFU at time point t24 compared to t0, while the razor-treated group showed an increase in CFU from t0 to t24 (Figure 3).

3.3 TEWL

The TEWL values increased significantly (p = 0.005) from t_{base} (mean = 5.72 ± 1.5, median = 6.1 ± 0.43) to t_{24} (mean = 10.93 ± 4.45, median = 10.53 ± 1.28), after shaving with the disposable razor, indicating a disruption of the skin barrier (Figure 4). Shaving by clipper showed no increase in TEWL results, but stable values at t_{base} (mean = 5.02 ± 1.21, median = 5.42 ± 0.35) and t_{24} (mean = 5.25 ± 1.14, median = 5.33 ± 0.33) indicating an intact skin barrier function after shaving with the clipper.

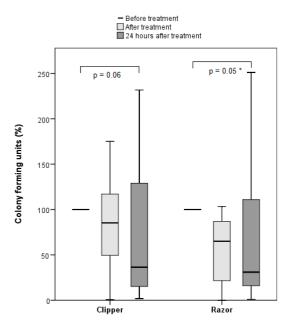


Fig. 3 Colony-forming units (CFU) before treatment (tbase), immediately after treatment (t_0) and 24 hours after treatment (t_{24}) on a log10 scale.

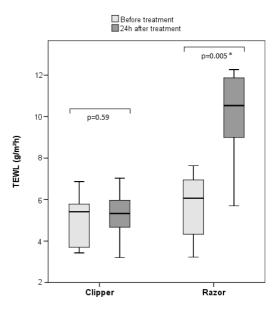


Fig. 4 Transepidermal water loss (TEWL) before (t_{base}) and 24 hours after (t_{24}) shaving using the clipper and the razor. The difference in the razor treatment was

significant (p = 0.005), whereas clipper showed similar TEWL values before and after shaving.

3.4 LSM

In the LSM analysis it was shown that the disposable razor causes lesions of the stratum corneum (Figure 5 (b), (d), (f)), ranging even into the deeper epidermal layers of the stratum granulosum and stratum spinosum. After the application of the clipper the stratum corneum was largely intact (Figure 5 (a), (c), (e)).

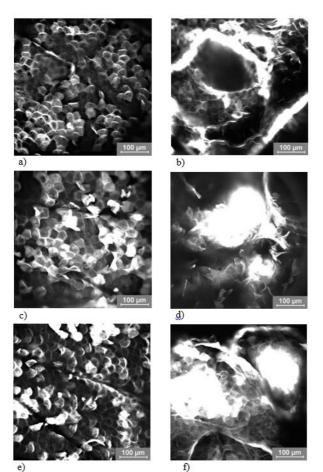


Fig. 5 LSM of the skin surface 24 hours after shaving with the clipper (a, c and e) and the razor (b, d and f).

3.5 LSM

After treatment with the clipper and disposable razors the subjects were asked to provide their subjective sensation of pain on a scale from 1 to 10 where the value 1 indicated "no pain" and the value 10 "strongest pain". Three (25%) of the twelve subjects indicated a slight to moderate pain (scale values 2-4) for the razor. Five (41.7%) described a slight discomfort during the shaving with the disposable razor and nine subjects (75%) described even a burning sensation after the shaving. Regarding the clipper, all of the twelve subjects rated no pain (scale value 1) for the treatment. Only one subject (8%) described a "light scratching" after treatment with the clipper.

4 Discussion

Preoperative hair removal should only be applied in case it is required by the surgical procedure. There are several clipping and shaving devices on the market, of which we compared only two within this study. The results confirmed that clipping is the preferable option of hair removal. Both the TEWL changes and the skin lesions analysed by LSM favour the use of the clipping device to the use of a razor.

It could be shown that the skin barrier is significantly compromised by the use of single-use razors, but hardly no change in TEWL values was seen after shaving using the clipper. The disruption of the skin barrier may be one of the main causes for an increased risk of postsurgical infections [30]. The TEWL measurements show that shaving using the razor causes considerable damage of the skin barrier, while after clipping the TEWL remains stable.

Regarding the measurements of the antioxidant status of the skin, there were no significant changes in the skin observed, meaning that the mechanical stress on the skin caused by both clipper and razor is relatively low. Another explanation is that resonance Raman spectroscopy is a technique to measure carotenoids in a defined skin volume in a depth of up to 150µm. Both the clipper and the razor are in contact with the skin surface only, where the upper epidermal layers can be damaged as seen by LSM. Therefore, changes of the carotenoid concentration that occur only in the uppermost epidermal layers might not be detectable in relation to the high measuring volume. Confocal Raman microscopy might be a more suitable method to identify these changes in carotenoid changes in the uppermost layers in future studies. It should also be taken into consideration that carotenoid-rich egg yolk [31] utilized for collection of bacteria from the skin surface could potentially influence the Raman measurements of cutaneous carotenoids. This influence was avoided by the different measurement areas on the calves' skin.

Concerning the number of superficial CFU it was shown that the razor initially lead to a more significant reduction in CFU, which can be due to the fact that the hair and hair follicles are less affected by clipping. After 24 hours though the shaving using the disposable razor lead to an increase in CFU from t0 to t24, which could not be found in the clipper-treated group, where a continuous and further decrease in CFU could be found. Previous studies indicate that a disruption of the skin barrier can favour bacterial skin colonization, which is associated with cutaneous inflammation [32]. Therefore, the increase in CFU after shaving with the razor at t24 could presumably be due to the disruption of the skin barrier function found also in the TEWL measurements. The barrier damage caused by the razor was also confirmed by LSM, where superficial lesions in the form of cavities and gaps were detected after shaving. These cavities and gaps may allow both bacterial and fungal spores to accumulate and to reach the viable epidermis by passing stratum corneum. Such skin lesions were not detected in the case of the clipper.

Furthermore the discomfort and burning sensation after shaving with the razor stated by the majority of the subjects also confirm the mechanical irritation caused by the single-use razor.

From previous studies it is known that considerable amounts of bacteria and fungi can be found within the hair follicles and that they reach the skin surface with the sebum flow [33]. It is assumed that the higher rate of local infections after shaving using a razor compared to clipping is due to microlesions of the skin during the process of shaving. By using the razor that is applied in closer direct contact with the skin surface, microlesions are formed, which can be used as portal of entry by pathogens of the skin flora [2, 7]. These pathogens can also reach the systemic bloodflow leading to SSI even if the lesions are remote from the operation site. The formation of microlesions can be avoided by the use of a clipping device. Until the regeneration of these microlesions pathogens from the deeper skin layers and especially from the hair follicles can be continuously released through these lesions.

5 Conclusions

In summary it can be stated that the application of the clipper shows a number of advantages over the use of a razor. The clipper, in contrast to the razor, does not cause skin lesions and a damage to the skin barrier, which allows a hair removal on the previous day of the surgery without an enhanced risk of SSI. This means a higher flexibility and simplicity in the preparation of surgical interventions.

Acknowledgments

The clipping device was provided by 3M Medica.