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Non-invasive skin sample collection: Comparing D-squame and the Smart Sticker.

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Abstract

Collection of stratum corneum *via* tape stripping is a minimally invasive procedure applied in a number of fields, including identification of epidermal biomarkers in skin cancers and inflammatory disorders. Tape stripping is an established technique; however, the number and types of tapes used in different studies vary widely. As an example, the most commonly used tape, D-squame skin sampling disc (CuDerm), is applied between 10-35 consecutive times to the sampling site. Repeated tape stripping is likely to result in increased local inflammation, potentially complicating comparison and interpretation of changes in biomarkers. To optimize data translatability, there is a need to define a robust operating protocol for epidermal tissue collection, including the definition of the least invasive sampling tape. In this study, we have compared the Smart Sticker™ (DermTech) to the D-squame skin sampling disc (CuDerm) by collecting non-overlapping adjacent volar forearm skin of twenty healthy volunteers. The performance was tested by measuring the epidermal tissue content on tapes, levels of induced erythema, and the quantity of extracted nucleic acids. We report significantly lower levels of erythema when stripping four consecutive times with the Smart Sticker™ compared to the four D-squame skin sampling discs. While having a smaller adhesive surface, four Smart Stickers™ lifted significantly higher amounts of tissue per tape, and yielded more nucleic acids per cm2, compared to four D-squame sampling discs. Our data highlights the applicability of the Smart Sticker™ for fast, highly efficient, and truly non-invasive epidermal tissue collection.

Disclosures & Acknowledgements

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Methods

Skin Sampling

The DermTech adhesive skin collection kit (DermTech, La Jolla, CA) and D-Squame skin sampling discs (CuDerm, Dallas, TX) were used to collect skin samples from 20 healthy volunteers on adjacent non-overlapping sites on volar forearms. The target skin was prepped with an alcohol pad to remove oils and then dry wicked with a gauze pad to remove any remaining moisture. Placement of different sets of sampling tapes is shown in Figure 1. Superior/inferior position of each tape type was alternated between the subjects. The target skin location was marked with a skin-safe pen, to guide placement of the tapes and ensure stripping from the same spot. To help the adhesion to target skin, five seconds of constant pressure were applied to each tape with the D-squame pressure instrument (CuDerm, Dallas, TX) before removing the tapes. Once removed, tapes were individually placed onto the tri-fold sample collector (DermTech, La Jolla, CA) and stored at -70ºC or colder until further processing.

RNA Extraction and Gene Expression Analysis

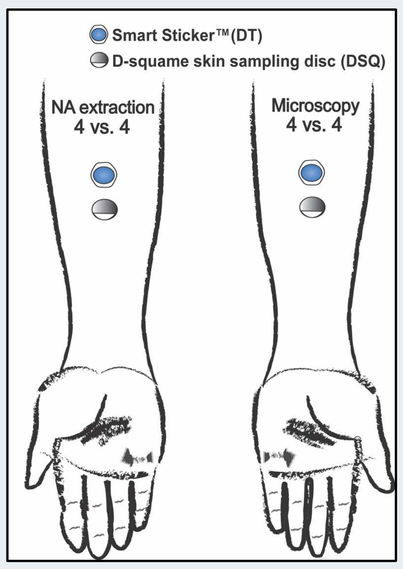
RNA was extracted from Smart Stickers™ using a closed-tube, bead-based method. Briefly, Smart Stickers™ were enzymatically digested to extract the genomic material from the adhesive and nucleic acid isolated using magnetic beads on a KingFisher Duo Prime (ThermoFisher Scientific, Waltham, MA). The quantity of total human RNA was determined by RT-qPCR with a gene expression analysis assay (Hs01060665_g1) that uses human β-actin (ACTB) mRNA as a quantified marker. The quantity of human genomic DNA (gDNA) was determined by qPCR with a human gene copy number analysis assay (Hs03023880_g1) that uses human ACTB gene in gDNA as a marker.

Quantification of Skin Erythema and Epidermal Tissue Content on Tapes

Photographs of subjects' skin were taken before and after stripping with 4 consecutive tapes by using the VHX-7000 microscope handheld camera (Keyence). Erythema levels were determined by quantifying the change in skin redness intensity between the baseline image and after stripping with 4 tapes. Tapes were evaluated for epidermal tissue content by acquiring 2D and 3D images of whole tapes using the VHX-7000 microscope main head camera (Keyence). Particle analysis was used to determine the percent of tape area covered by tissue in 2D (Keyence VHX-7000 analysis software). Height of tissue on tapes was determined by 3D image analysis (Keyence VHX-7000 analysis software) and the final quantification of tissue content was performed in Fiji with the Color Profiler 3D plugin.

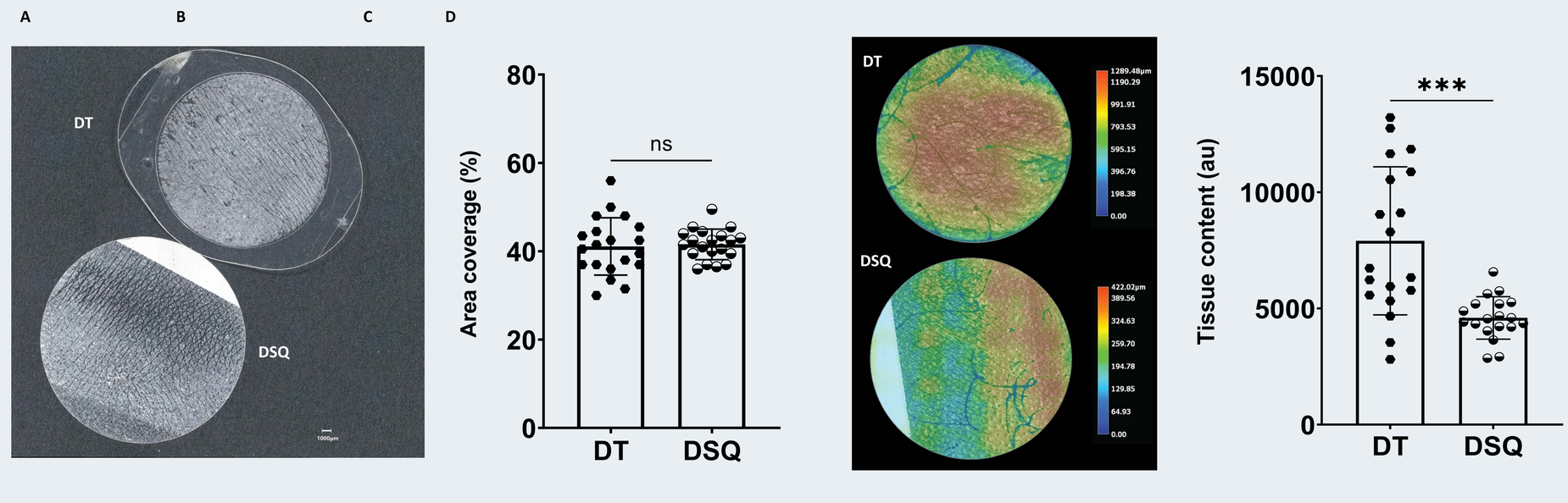
Results

Figure 1. Tape placement on target skin



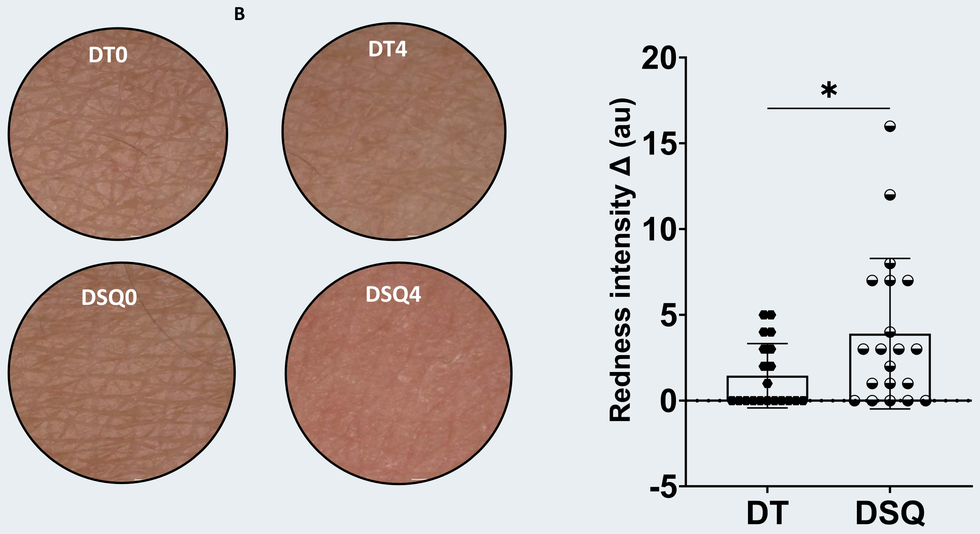
Samples collected with the Smart Sticker™ or D-squame skin sampling discs were analyzed by microscopy and nucleic acid (NA) extraction and quantification.

Figure 2. Epidermal tissue content on Smart Sticker™ (DT) and D-squame (DSQ) tapes



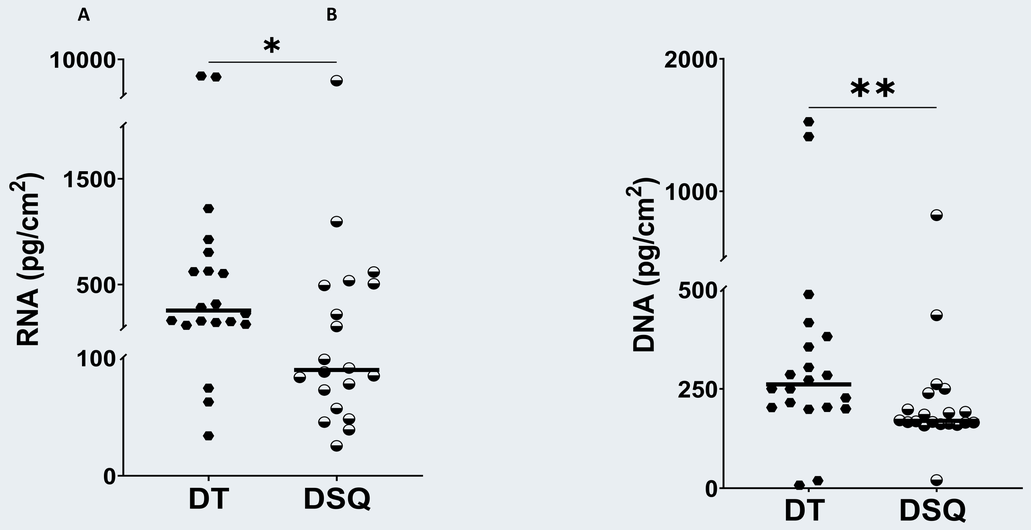
A. Adhesive area coverage with skin tissue after tape-stripping with the Smart Sticker (DermTech-DT) and D-squame skin sampling disc (CuDerm-DSQ). Adhesive area measures 2.8cm^2 for the SmartSticker™ and 3.5cm^2 for the D-squame disc. **B.** Quantification of the percent tape area covered by tissue after sampling. Graph shows combined data for the 1st and 4th tape. **C**. Representative image of the tissue amount on each type of adhesive tape. The indicated RGB color scale reflects tissue height in µm. **D.** Quantification of the whole tissue amount per tape by color profiling. Graph shows combined data for the 1st and 4th tape.

Figure 3. Skin erythema after sampling with four consecutive tapes



A. Images of skin erythema before (DT0, DSQ0) and after sampling with 4 consecutive tapes (DT4, DSQ4). Bar is 1mm.
B. Quantification of skin erythema expressed as a difference in skin redness intensity between the baseline and the 4th tape.

Figure 4. Quantity of nucleic acids extracted from four DT or DSQ tapes



A, B. Nucleic acids were extracted from four DT or DSQ tapes and quantified using RT-qPCR.

Conclusions

- **Skin sampling with the Smart Sticker™ was associated with more tissue collection, higher yields of nucleic acids, and lower levels of skin irritation.**
- **Smart Sticker™ is proposed as a tape of choice for applications requiring fast, highly efficient and truly non-invasive collection of epidermal tissue.**