Histological image processing and analysis

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Introduction

Biological image processing and analysis algorithms were created to evaluate diverse aspects of skin pathology in histological specimens of both human subjects and mouse models of human disease. These included:

* Measuring integrity in picrosirius-stained skin imaged with cross-polar microscopy to evaluate loss of collagen integrity. We measured degradation in collagen organisation (Figure 1, 2) and changes in bundle thickness in an ageing series (Figure 3), as well as in a model of diabetic skin.
* A technique to quantify collagen dynamics by assessing Herovici staining of old and young collagen fibres in skin from ageing and diabetic subjects (Figure 4).
* Quantification techniques to assess skin layer morphometrics (adipocyte hyperplasia and hypertrophy, and depth of each skin layer) to facilitate high-throughput analysis. (Figure 5, 6)
* Development of a technique to measure skin surface texture through the automated analysis of human skin impressions created from sun exposed and sun protected sites (Figure 7).
* Evaluation of changes in each of the three human skin layers in terms of morphology and structure using a variety of computational techniques.

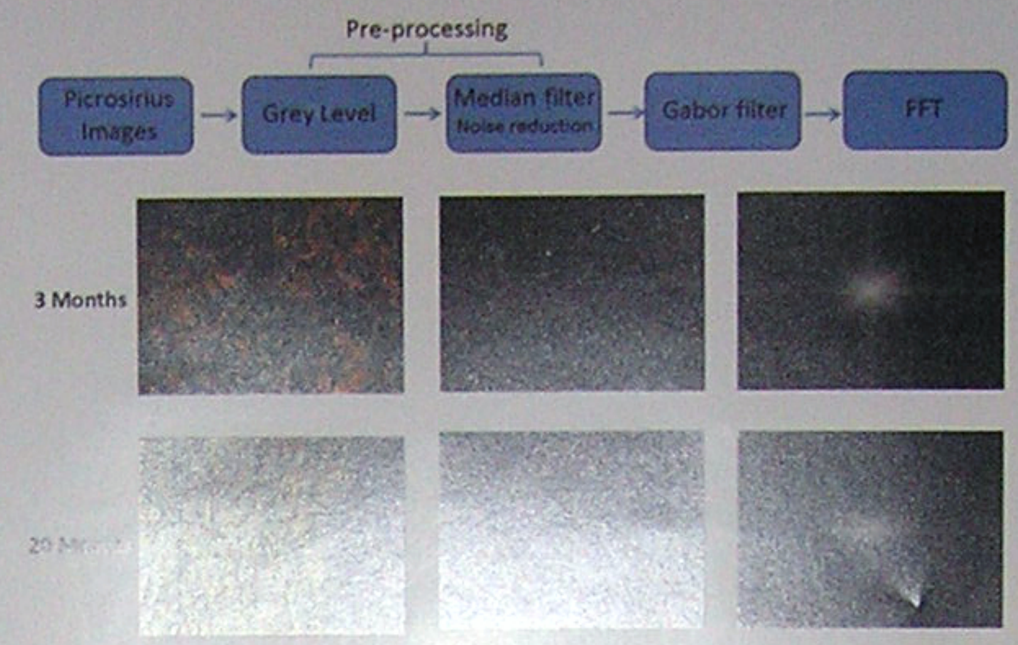


Figure 1: A novel method for measuring collagen orientation in skin. Picrosirius collagen images (left panels) are converted to Gabor filtered images in different directions (centre panels) and Fourier Transform (FFT) power spectra are created in the right panels, allowing comparison of young and old skin.

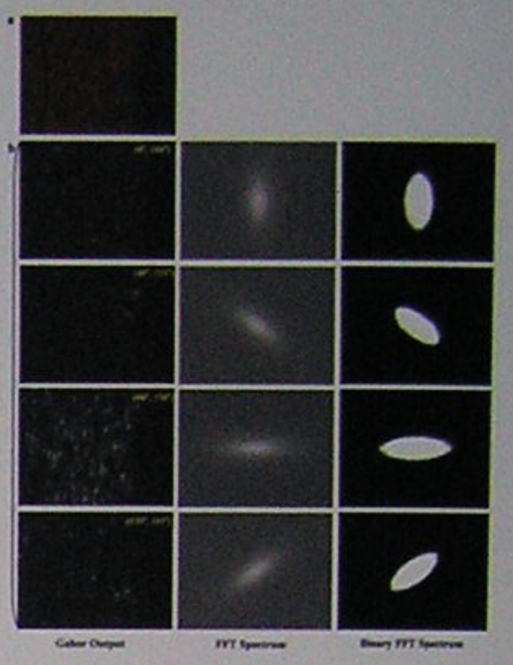


Figure 2: Measuring collagen orientation in a picrosirius stained skin image (a). (b) FFT elliptical analysis: Gabor filtered image in different directions (left panels), FFT power spectra (centre panels) and ellipses generated from each direction (right panels).

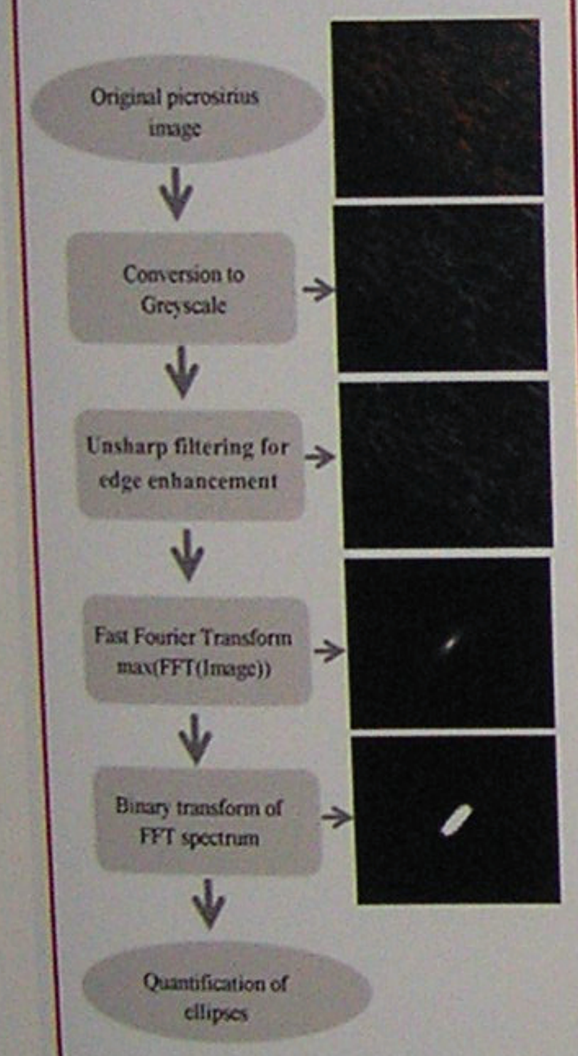


Figure 3: An overview of the measurement of collagen bundle thickness. Measurements were based on the maximum amplitude of the FFT spectrum.

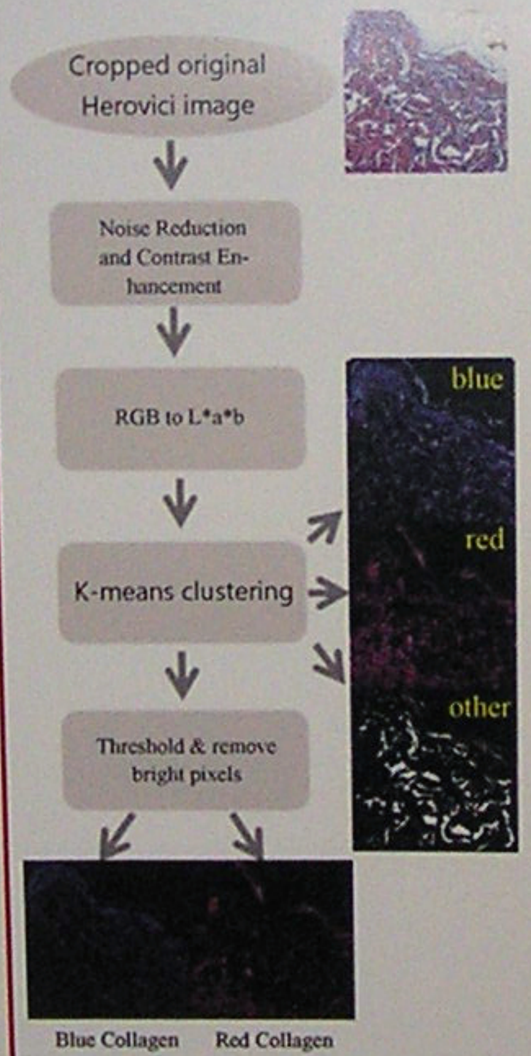


Figure 4: Measurement of collagen dynamics. Nascent (blue) and mature (red) pixels in Herovici images were segmented using *k*-means clustering-based methods.

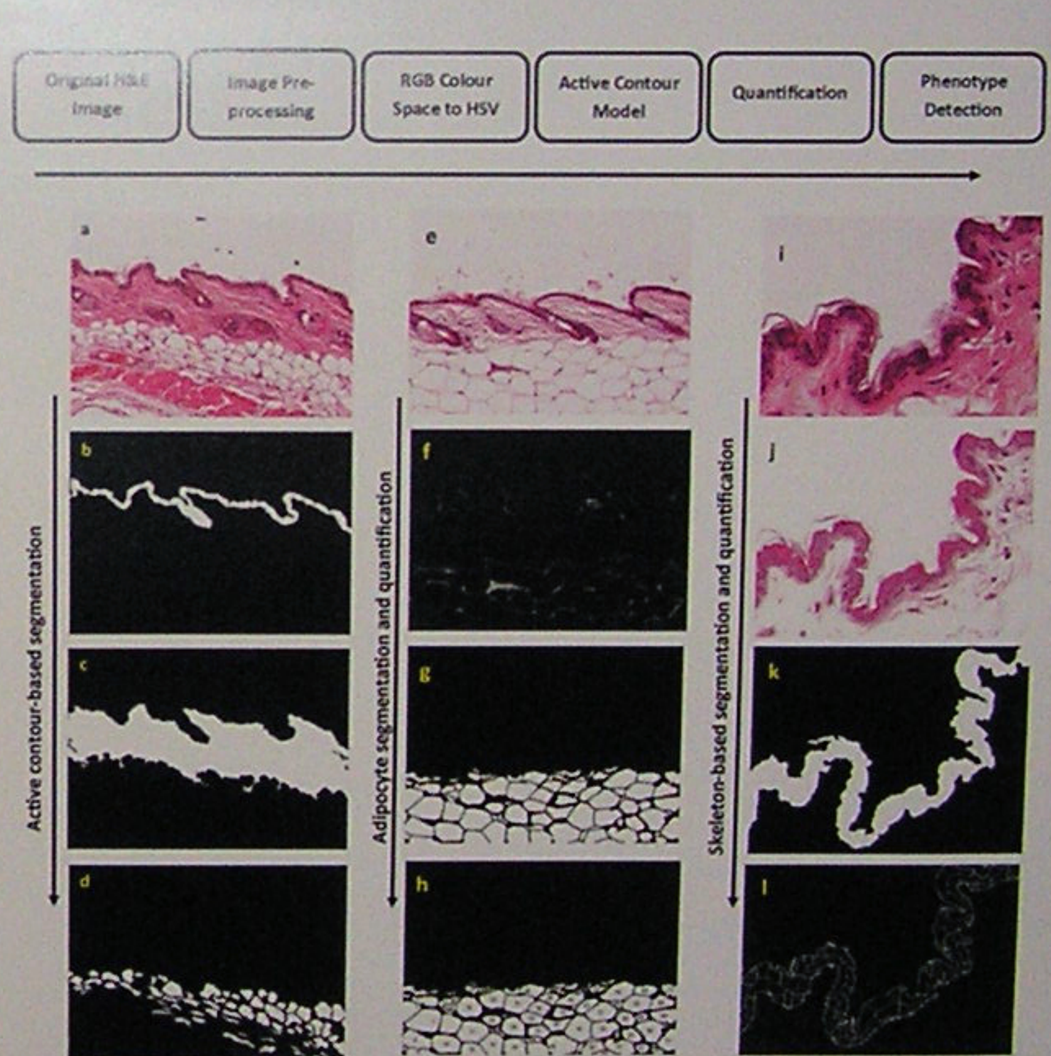


Figure 5: High-throughput cutaneous phenotype detection and quantification. Skin layer segmentation using an active contour method (a-d), quantification of adipocyte size and number (e-h), and epidermal and dermal layer thickness quantification (i-l).

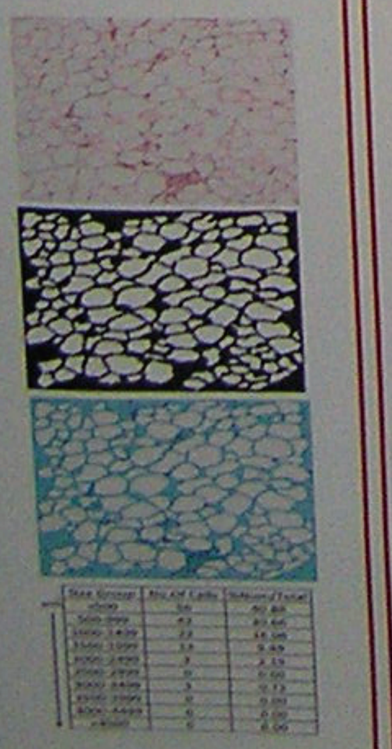


Figure 6: Automated adipocyte size and number quantification. Adipocyte segmentation was achieved with piecewise linear transformation before counting and area measurements.

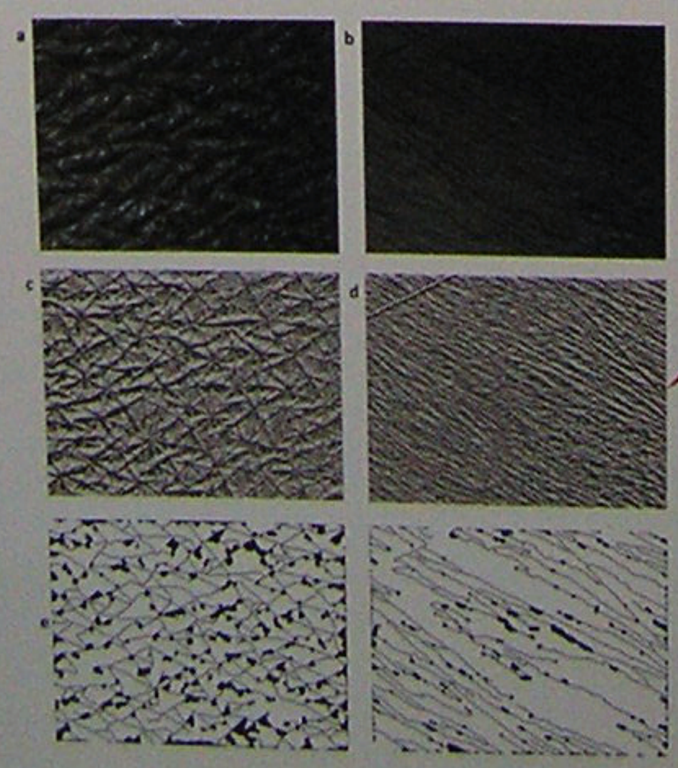
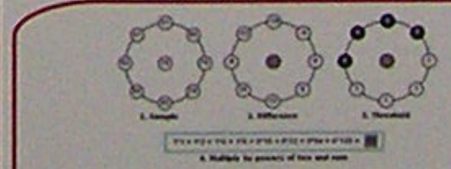


Figure 7: Non-invasive assessment of skin surface structure. Original skin mould, LBP image and inverse skeletonised images of sun-protected arm (a,c and e) and sun exposed face (b, d and f).



Calculation of the Local Binary Pattern LBP value for each pixel

Conclusions

* We have developed automated, high-throughput image analysis tools to enhance, segment and quantify various features of histological images of skin.
* These methods were able to analyse images from various imaging platforms (including whole-slide scanners) in an automatic, adaptive manner to handle variations in colour distribution and intensity.
* Automated image analysis and quantification techniques have proved reliable and accurate without the need for user interaction. Moreover, the techniques are easy to use with little or no special knowledge of image processing required by the user.
* Our methodologies are adaptable to a wide range of high-throughput morphometric projects.

Publications

* Osman OS, Selway JL, Harikumar PE, Stocker CJ, Wargent CJ, Cawthorne MA, Jassim S, Langlands K. A Novel Method to Assess Collagen Architecture in Skin. BMC Bioinformatics 2013
* Osman OS, Selway JL, Kępczyńska MA, Stocker CJ, O'Dowd JF, Cawthorne MA, Arch JR, Jassim S, Langlands K. A novel automated image analysis method for accurate adipocyte quantification. Adipocyte 2013; 2:160 - 164; http://dx.doi.org/10.4161/adip.24652
* Osman OS, Selway JL, Harikumar PE, Jassim S and Langlands K. Automated analysis of collagen histology in ageing skin. *In Proceedings of the International Conference on Bioimaging*, France, 3rd – 6th March 2014, 265-270.

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