MORPHOLOGICAL SCHEME FOR MORPHOMETRIC ANALYSIS OF EPIDERMAL BIOPSY IMAGES

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Abstract.
This paper addresses the problem of morphometric analysis of microscope images from cutaneous biopsy samples. A morphological scheme is applied for the automatic measurement of histologic parameters of the epidermis. It consists in an unsupervised segmentation approach that is strongly based on an 'a priori' model of the images. The watershed algorithm has proven to be a very powerful tool for the introduction of such 'a priori' information, because the segmentation process can be conveniently guided by some strategic markers in order to perform the detection of the desired structures. This permits an automatic measurement of some objective parameters which are highly correlated with the evolution of some skin diseases.

Key words: Morphometric analysis, cutaneous biopsy, skin layers, segmentation

1. Introduction

Morphometric analysis of epidermal biopsies permits an objective study of several skin diseases. Skin morphology in patients under treatment can be accurately analyzed by means of image processing techniques, avoiding less reliable methods such as visual inspection. In this paper, we present a morphological scheme for the analysis of epidermal biopsy samples. The target is the detection of the main transitions that are visible on the sample. Such transitions separate the different skin layers whose associated morphometric parameters have to be measured.

The proposed technique relies on three different steps: prefiltering, detection of the transitions and the measurement itself. In the first step, alternate sequential filters are used for image simplification in order to enhance significant transitions and to remove small noisy components. The second step is performed by the watershed algorithm. The set of markers imposed to constrain the watershed algorithm allows the introduction of helpful 'a priori' information about the images being processed. Once the main transitions in the image have been detected, the morphometric mea-

sures are carried out.

The paper is structured as follows: section 2 describes the application problem and the work images. In section 3, an outline of the proposed morphological scheme for morphometric analysis is discussed. Finally, section 4 presents some results and the conclusions.

2. Morphometric Analysis of Epidermal Biopsy Samples

Figure 1 shows two samples of skin biopsies. Apart from the brighter band at the top of the images, which is simply air, from top to bottom three different layers can be observed: the corneal stratum, the epidermis and the dermis. Between these last two layers, there is an extremely thin cellular tissue where epithelial cells germinate: the dermo-epidermal junction (DE-junction). From this place, epithelial cells grow and migrate outwards, through the epidermis, until they reach the corneal stratum. Once here, the cells die and come off the skin in the form of scales. Different skin diseases are associated with changes in this process. Most of them can be observed as an alteration of the morphology of the skin layers. As an example, the right image in figure 1 is a sample of 'psoriatic skin' at the same magnification. 'Psoriasis' is characterized by epidermal hyperplasia with elongation of the interpapillary folds. In the right image of figure 1 the dermis is so ripply that in some points it even touches the corneal stratum, producing bleeding wounds. The bidimensional section also produces isolated dermal lobes located inside the epidermis and apparently disconnected from the dermis.

Morphometric analysis of skin biopsies permits an objective measurement of these alterations. The usual parameters to be computed are the average thickness of the epidermis and the degree of ripple of the DE-junction. Such parameters are highly correlated with the evolution of the disease, and so, of importance for several medical applications.

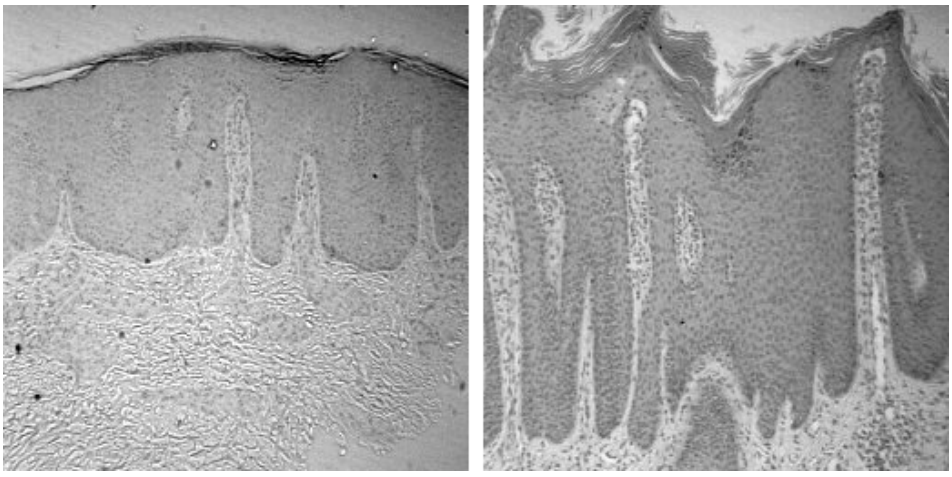

Fig. 1. Skin biopsy samples (H&Ex200). a) left: normal skin; b) right: psoriasis.

Skin biopsy images are generally noisy and low contrasted. Sometimes, even for an expert observer, the transition lines between the layers are difficult to draw. Morphometric analyses are usually performed manually, either by means of a ruler superimposed on photographic enlargements, or with the aid of a digitized tablet. This approach is extremely time-demanding and does not permit to obtain many repeated measures to ensure the reliability of the results. The automatic analysis proposed in this paper greatly simplifies this task.

3. Morphological Filtering and Segmentation

The prefiltering step is aimed at the simplification of the image. Alternate sequential filters [1] can remove both bright and dark noisy components of small size. In this application, open (close) filters by reconstruction of erosion (dilation) of increasing sizes [2] are used in the filtering sequence, in order to change as least as possible the structure of the remaining components. The results of applying these filters to the images of figure 1 are presented in figure 2.

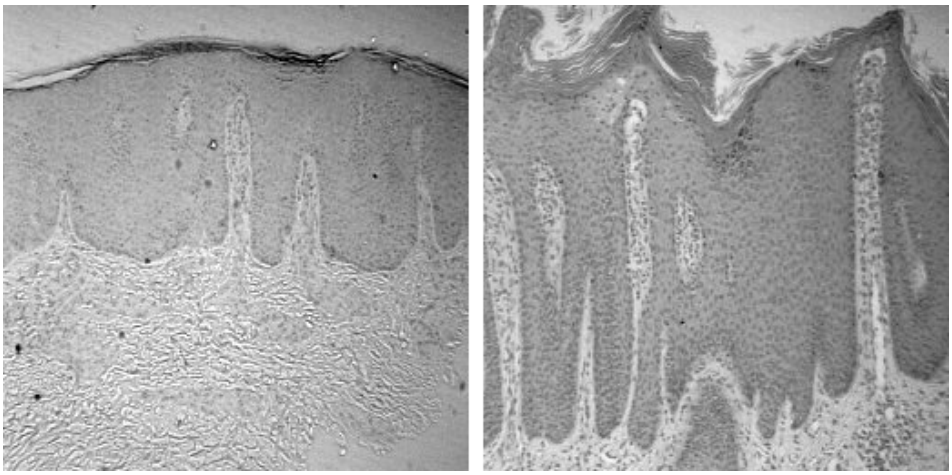

Fig. 2. Application of an alternate sequential filter by reconstruction of size 2.

The watershed algorithm [3] is then applied in three steps to detect one by one the different transitions between the above mentioned layers. This process is strongly guided by the 'a priori' model of the image, which is assumed to be crossed by three horizontal transitions among the four layers. Such a supposition imposes that biopsy samples should be upright and correctly centered for acquisition, what does not restrict very much the image acquisition process while allowing the introduction of specific markers in the segmentation step.

The first layer to be detected is the corneal stratum because it is the most outstanding feature in the image. Being darker and narrower than the others, the black top-hat transform [4] performs as an appropriate enhancement operator. The watershed of the result, with two markers placed at the bottom and top lines of the enhanced image, gives the result plotted in the left image of figure 3 with an axis-

line locating the corneal stratum. The inner and outer contours of this layer are found by running again the watershed, this time on the morphological gradient of the enhanced image (fig.3, right) using the previous watershed line as a new marker.

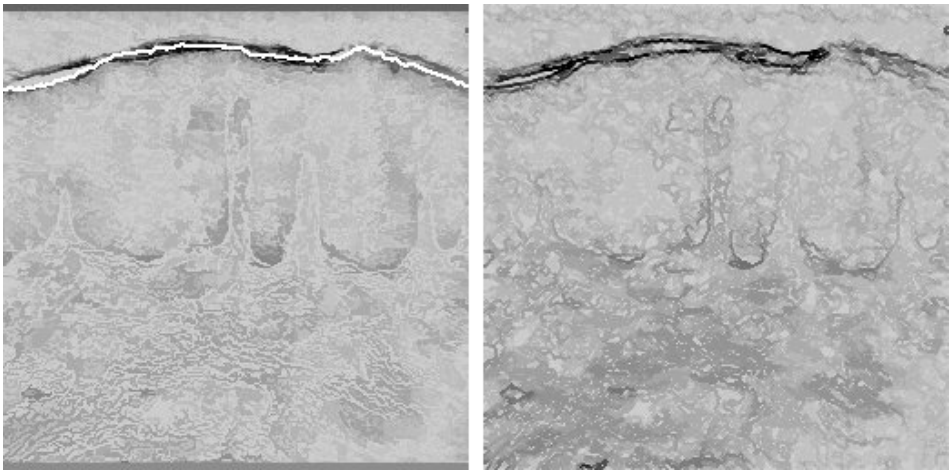

Fig. 3. Detection and segmentation of the corneal stratum for the sane skin sample. a) left: watershed on the black top-hat with 2 markers; b) morphological gradient of the top-hat.

The last transition to be detected is tinier than the former ones. The watershed algorithm is then applied on the morphological gradient of the prefiltered image, but some folds of the DE-junction may be lost, as shown in the left image of figure 4. There is a physiological reason that explains such a behaviour of the watershed algorithm: the inflammatory infiltrate generates noisy points (reaction points) inside the dermis, which are responsible for this wrong detection. An alternate sequential filtering of larger size is performed in order to remove this noise. The iteration of the watershed algorithm, now on the gradient of this strongly filtered image constrained by the two markers (the cornea layer and the previous approximation just obtained for the dermis) yields the final result. The right image of figure 4 shows this new gradient image for the second run of the watershed with the constraints imposed by the new markers.

The isolated interior lobes of the epidermis should also be detected. They are part of the dermis and their contours are also considered part of the DE-junction. A morphological contrast transform between an opening and a closing by reconstruction [5] is used for the enhancement of these components. Note that these lobes are of the same grey level than the dermis layer and darker than the surrounding epidermis. Therefore, the set of markers for the segmentation is obtained by two operations: thresholding and size selection on the enhanced image. The threshold level is estimated from the average values of the regions obtained in the previous steps, whereas the size parameter is selected according to the size of the smaller lobes that are going to be found. Finally, the application of the watershed algorithm on the gradient image with the obtained markers allows the precise location of the contours of the interior lobes.

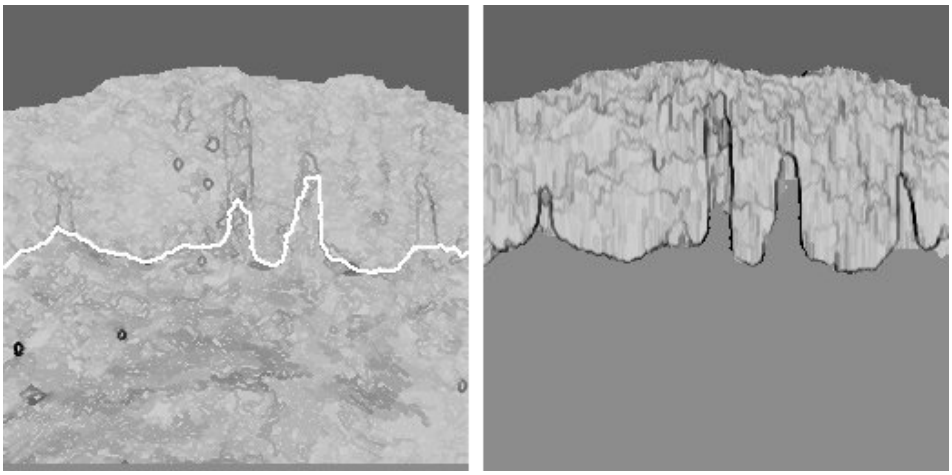

Fig. 4. Location of the DE-junction. a) left: watershed on the morphological gradient; d) right: gradient image for the second run of the watershed (with constraints).

Figure 5 presents a flow chart summarizing the methodology for the segmentation of the epidermal biopsy samples. Please note that the information about the 'a priori' model of the image is introduced by the markers in the decision step (the watershed).

4. Results and Conclusions

The results for the two images of figure 1 are shown in figure 6, with the transition lines superimposed on the original images. Notice than the transition between the corneal stratum and the air has not been very accurately located, but this is the least important transition for the morphometric analysis due to the presence of scales coming off the skin.

Two sample parameters obtained for these images are given in table I: the *equivalent thickness* of the epidermis and the *index of rugosity* of the DE-junction. Both parameters are representative of the morphology of the epidermal layer and have been defined as follows:

The equivalent thickness of the epidermis is defined as the apparent surface of this layer divided by the width of the image, expressed as a percentual ratio over the image height, i.e.,

$$thickness(\%) = \frac{dermis\_surface}{image\_width} \cdot \frac{100}{image\_height}$$

The index of rugosity of the DE-junction measures the degree of ripple of such transition. Its value should be 0% for a completely flat transition and 100% if the transition was so ripply that filled up the whole image with folds of height equal to the image height. A possible expression for this parameter is the next one:

$$ripple(\%) = \frac{transition\_length - image\_width}{dermis\_surface - image\_width} \cdot 100$$

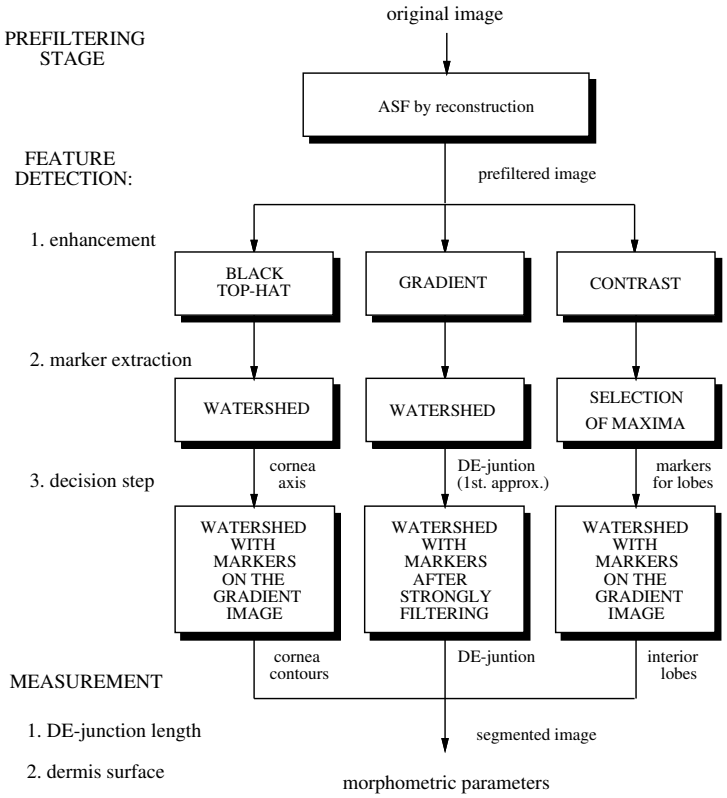

Fig. 5. Methodology for the segmentation of skin biopsy samples.

TABLE I
Morphometric parameters obtained for the images of figure 1

IMAGE	thickness(%)	ripple(%)
sane skin image (fig.1a)	35.1%	1.6%
'psoriasis' image (fig.1b)	52.6%	5.1%

The values shown in table I are objective measures of the morphology of the skin layers. The same parameters have been computed for a wide range of biopsy samples and have proven to be useful in order to follow the evolution of alterations in the epidermal layer.

As a conclusion, it has been shown that the measurement of histologic parameters from epidermal biopsy samples can be efficiently performed by unsupervised morphological segmentation techniques. The watershed algorithm allows the straightforward introduction of the model of the image to be segmented, so that the available 'a priori' information can be used in order to guide the detection of the structures of interest. The degree of correlation of the values of these parameters with the usual

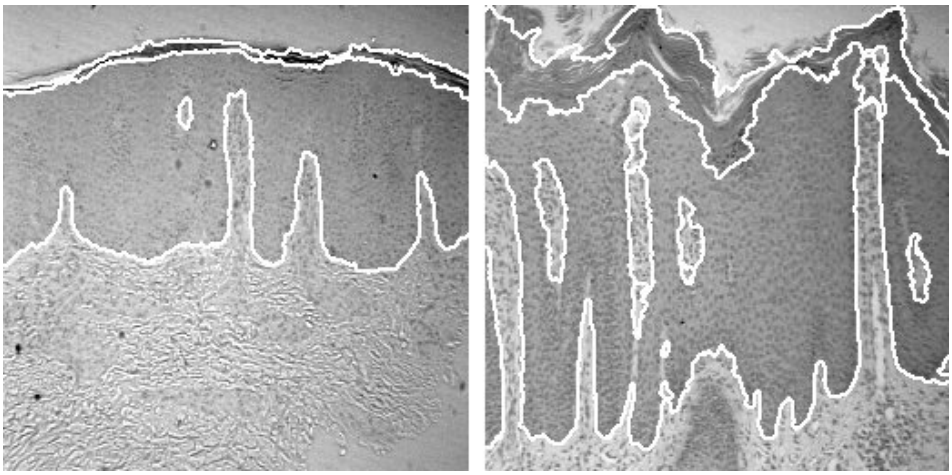

Fig. 6. Final results for the original images of figure 1.

histopathologic findings has been found to be sufficient in order to guarantee the reliability of the proposed automatic method.

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