Quantitative Analysis of Cheese Microstructure using SEM Imagery

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Structural properties of cheese highly influence its chemical, mechanical and nutritive properties. The analysis and quantification of relevant features in cheese imagery is the basis of modern food microscopy. Processing Scanning Electron Microscopy (SEM) images is a powerful tool to estimate microstructural cheese features.

In this paper, we present an ad-hoc method to analyse SEM cheese imagery to characterise quantitatively a number of features, using simple and efficient image processing techniques. The proposed method is currently under experimentation on a number of traditional Sicilian cheese varieties.

1 Introduction and Motivation

Cheese microstructure is the spatial arrangement of the casein particles that join together into clusters and chains to form a protein matrix throughout which are dispersed water, fat globules and minerals [1, 4]. Microstructure is one of the major controlling factors of texture and functional properties of cheese. Cheese microstructure analysis plays an important role in the quality control of the dairy products [3]. SEM imagery allows to study cheese

microstructure by qualitative visual evaluation as well as quantitative analysis. Image analysis and quantification of relevant features is the basis of modern food microscopy [1]. Unfortunately, due to the limitations of the acquisition process, the acquired images are noisy and present reflections that prevent the robust extraction of useful information by simple thresholding techniques. Even if some authors use commercial image-processing tools to manually threshold SEM images [6] (see Figures 1(c) 1(d)), the binarised images are not accurate enough for quantitative analyses. Therefore, SEM imagery must undergo an enhancing pre-processing step before binarisation.

We present an ad-hoc method to enhance and threshold SEM cheese imagery to quantitatively characterise a number of features, using simple and efficient image-processing techniques. Preliminary results on a number of traditional Sicilian cheese varieties will be shown at conference time.

2   Image Analysis

Given a SEM scan of a cheese sample, our objective is to compute a number of statistical descriptors about its microstructure. Since SEMs give a set of regularly spaced samples, scans can be regarded as digital greyscale images. In particular, we investigate the structure of micro-pores in the protein matrix in order to quantify useful measures, such as porosity and a number of shape descriptors.

In order to compute statistics about the microstructure of cheese samples, the SEM scans must be binarised to classify which pixels belong to pore or to protein matrix regions. Due to scanning limitations, simple manual or automatic thresholding techniques produce noisy images. Hence, a SEM scans must be enhanced before binarisation. This is the main topic of our paper. The proposed method is composed of four simple phases: 1) denoising and flattening, 2) pore shape regularisation, 3) binarisation, 4) quantification of relevant features.

SEM images often present speckle noise. Even if sophisticated despeckle methods exist [2] we found that a 3×3 median filter gives sufficiently accurate results for our application. We further apply a Gaussian smoothing filter with a small kernel to remove noise deriving from different sources 2(b).

After removing noise, some images still present reflections causing strong luminance gradients that rule out global thresholding, since the greyscale value of highly illuminated pores can be greater than that of lowly illuminated protein matrix areas 2(c). In order to remove this effect we flatten the image by removing low frequencies (i.e., the carrier signal) using a bandpass filter 2(e). Since automatically finding a global threshold in flattened images

Parameter name	Formula	Parameter name	Formula
Form Factor	$\frac{4\pi \cdot A}{p^2}$	*Convexity*	$\frac{p_{convex}}{p}$
Roundness	$\frac{4A}{\pi \cdot D_{max}^2}$	*Solidity*	$\frac{A}{A_{convex}}$
Aspect Ratio	$\frac{D_{max}}{D_{min}}$	*Hole Fraction*	$\frac{A_{fill}-A_{net}}{A_{fill}}$
Elongation	$\frac{l_F}{w_F}$	*Radius Ratio*	$\frac{d_{inscr}}{d_{circum}}$
Curl	$\frac{l}{l_F}$	*Directionality*	gradient direct.

Table 1: Shape quantitative descriptors. Meaning of the parameters. A: net area; A_{convex}: area of the convex hull; A_{fill} area of the filled polygon; p: perimeter; p_{convex}: perimeter of the convex hull; D_{max}, D_{min}: maximum and minimum dimension; d_{inscr} and d_{circum}: diameters of the inscribed and circumscribed circle; l: length of the feature along the farthest points in the convex hull; l_F, w_F: fiber length and fiber width (measured from the skeleton of the feature [8]).

may be tricky, care must be taken using this operation in order to avoid unsatisfactory results. For the time being, the process is assisted by the user who enables this operation only when actually needed.

In order to help thresholding, pore borders can be strengthened using simple morphological operations. We employ a closure operator with small support to regularise the shape of pores and to enhance pore borders. We found that better results can be obtained if the dilation and erosion operations are decoupled and thresholding is done before erosion. The well-known Otsu thresholding algorithm [5] is used for automatic binarisation 2(f).

The most relevant measure for the analysis of the microstructure of porous materials (e.g., cheese) is *porosity*, defined as the percentage of pore area with respect to the total sampled area. Anyway, a number of other important measures are used in food technology [8]. Some examples of feature shape descriptors are shown in Table 1. The descriptors *Form Factor*, *Roundness*, and *Aspect Ratio* quantify the departure of a feature from roundness. The former assigns a higher value to features exhibiting uneven edges, while the last two quantify elongations toward more elliptical shapes. *Elongation* accounts for the elongation of fiber features along the main dimension. *Curl* gives a measure of how much a fiber feature appears "curly", i.e., is bent along its main dimension. *Convexity*, defined as the ratio between the perimeter of the convex hull and the perimeter of the structural unit, shrinks as the two perimeters depart and is maximum for a perfectly convex feature. *Solidity* quantifies to which extent the feature area covers the convex hull area. *Hole Fraction* measures the fraction of feature area that is occupied by holes. *Ra-*

dius Ratio is an attempt to measure the compactness of the feature shape. Finally, *Directionality* is computed as the distribution of gradient directions.

We discard fiber descriptors (i.e., *Elongation* and *Curl*) since pores in the cheese matrix show a typical round shape. The most relevant for our application are the shape descriptors *Form Factor*, *Roundness*, *Aspect Ratio*, and *Convexity*, as well as the area descriptors *Solidity* and *Hole Fraction*.

Obviously, these descriptors quantify the aspect of a single structural unit. Statistical parameters such as mean, standard deviation, skewness, and kurtosis can be effectively used to extract useful information on the distribution of the collected data. Due to the lack of space, details about these measures and some pieces of data will be given at conference time.

For simplicity of use, the proposed method has been implemented as a plugin for ImageJ [7], a widely-used Open-Source tool for image analysis.

3 Conclusion

We have presented a simple and efficient binarisation method for SEM cheese imagery. We have also briefly discussed a number of shape descriptors to characterise quantitatively cheese features. The proposed method is currently under experimentation on a number of traditional Sicilian cheese varieties.

References

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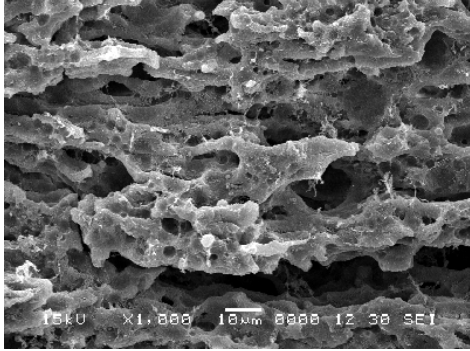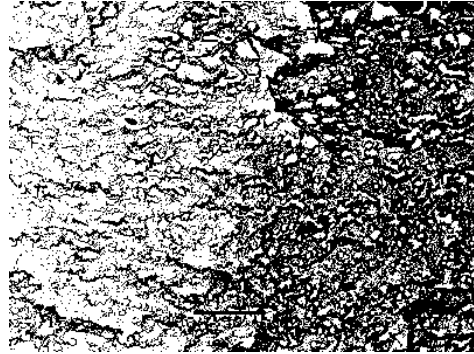
(a) SEM scan of a sample of Palermitano cheese.
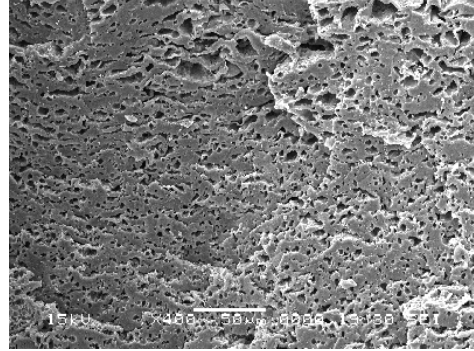
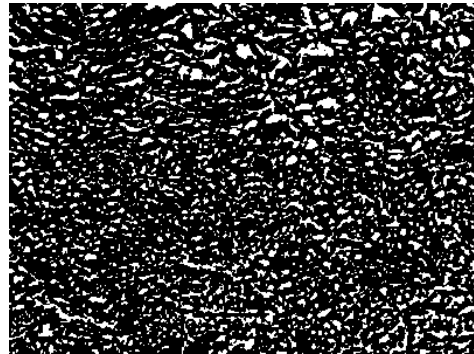(b) SEM scan of a sample of Ragusano cheese.

(c) Simple manually-adjusted thresholding.

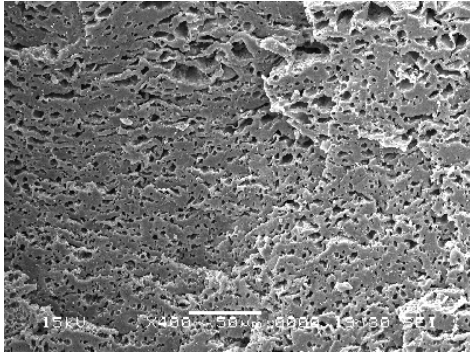(d) Simple manually-adjusted thresholding.

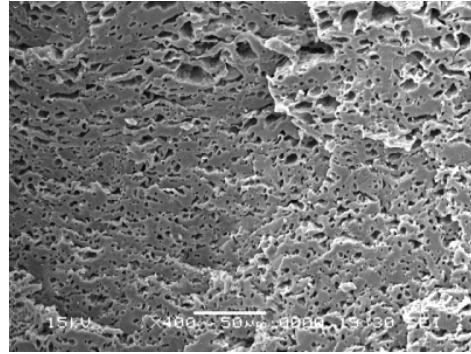(e) Automatic binarisation using our method.

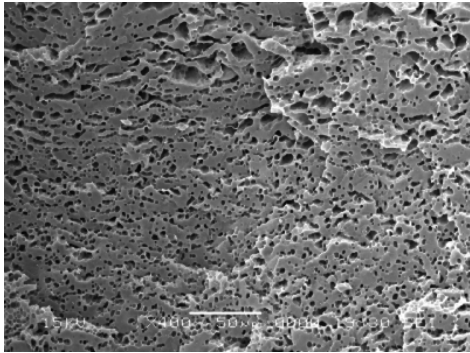
(f) Automatic binarisation using our method.

Figure 1: Comparison between a simple manual thresholding technique and our ad-hoc automatic binarisation.
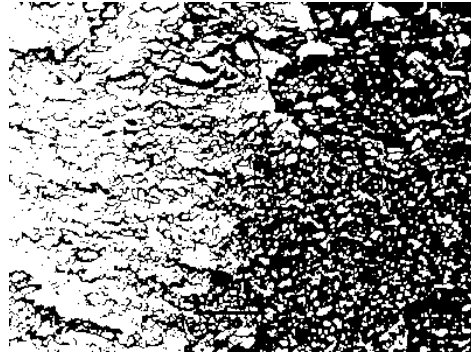
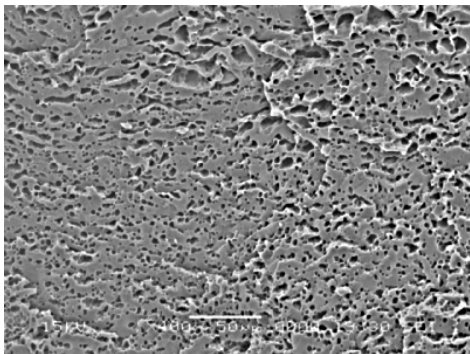(a) SEM scan of a sample of Ragusano cheese.

(b) Removal of noise.

(c) Morphological dilate.

(d) Automatic thresholding. Wrong result due to electron reflections.

(e) Bandpass filtering.

(f) Automatic thresholding. Correct result after bandpass filtering.

Figure 2: Electron reflections and bandpass filtering.