

**Presented at: *Joint Microscopy Meeting, Lille, France, June 24-28, 2002***

P. Van Osta\*, K. Ver Donck\*, L. Bols\*, J. Geysen\* J.M. Geusebroek\*\*, B.M. ter Haar Romeny\*\*\*

\*Union Biometrica N.V., European Scientific Operations, Ciplastraat 3, B-2440 Geel, Belgium

\*\*Intelligent Sensory Information Systems, Faculty of Science, UvA, Amsterdam, The Netherlands

\*\*\*BioMedische Technologie, Technische Universiteit Eindhoven, Eindhoven, The Netherlands

***Title: Application of linear scale space and the spatial color model in microscopy***

**Abstract:**

### **Introduction**

Structural features and color are used in human vision to distinguish features in light microscopy. Taking these structural features and color into consideration in machine vision often enables a more robust segmentation than based on intensity thresholding. Linear scale space theory and the spatial color model provide a powerful framework for feature extraction in microscopy images. Differential geometry is applied in image analysis by convolving the image with Gaussian derivatives of the appropriate scale ( $\sigma$ ) for the objects of interest.

### **Linear scale space**

Grayscale microscopy images acquired with a B/W camera contain structural features for which grayscale scale space provides a robust detection framework. The work of J. Koenderink provides the theoretical basis for this approach<sup>1</sup>. From this work we know that the linear scale space framework offers robust extraction of features for most computer vision tasks, despite the presence of noise. Feature detectors can be constructed based on differential invariants, which are relatively insensitive to changes in illumination condition and signal to noise ratio, which is an important feature in light microscopy. Several differential invariants can be used in light microscopy. Figure 1 illustrates the use of a detector for dark ridges, the scale of the Gaussian kernel ( $\sigma$ ) is 1.0 (Equation 1)<sup>2</sup>.

$$L_{xx} + L_{yy} - \frac{1}{2} \sqrt{(L_{xx} - L_{yy})^2 + 4L_{xy}^2} > 0 \quad (1)$$

L denotes the image itself and  $L_x$  and  $L_{xx}$  the first and second Gaussian derivative in the x direction of the image.

### **Spatial color model**

For color light microscopy we use the spatial color model as proposed by Koenderink and Geusebroek to select different colored regions and objects<sup>3</sup>. Differential invariants can be constructed which are insensitive to changes in illumination color temperature and illumination intensity. Figure 2 shows the detection of colored microspheres in fluorescence microscopy, the scale ( $\sigma$ ) is 1.0. For detection of the different colors in the image, the following sets of equations are applied to the image (Equation 2)<sup>4</sup>.

