ACTA STEREOL 1993; 12/2: 277-282
PROC 6ECS PRAGUE, 1993
ORIGINAL SCIENTIFIC PAPER

GREY LEVEL IMAGE ANALYSIS USING A TOPOLOGICAL TREE OF CONNECTED FIGURES SELECTED IN A SET OF SERIAL BINARY SECTIONS

Elena G. MARKOVA[1], Igor A. MARKOV[1], Dick van VELZEN[2] and Vyvyan C. HOWARD[2]

[1] Brain Research Institute Russian Academy of Medical Sciences, Per.Obukha 5, Moscow, 103064, Russia
[2] Department of Fetal and Infant Pathology, University of Liverpool, P.O. Box 147, Liverpool, L69 3BX, England

ABSTRACT

A method of grey level image analysis is proposed. It consists in the extraction and labeling of connected components (figures) in a set of serial binary sections, establishing connectivity between figures in adjacent sections, forming of image description, building of a topological tree of connected figures. The topological tree is analyzed for extraction of objects of interest. The parameters which characterize the location and size are calculated for each figure. Topological tree analysis can be made taking into account the figure dimensional parameters and the tree furcations, which characterize grey level image topological features. Parameters filtering allows to erase the information about those figures which do not satisfy the appropriate constraints, including the figures which extraction is the result of noise. For extracted objects of interest the extended set of parameters (area, perimeter, coordinates of gravity center, shape and orientation parameters and so on) is calculated, which may be used for image recognition.
The method was realized in the image processing system consisting of a light microscope, a TV camera, a Frame Grabber card built in IBM PC 386/ 20 Mhz and image processing software package. The system enables to fulfil segmentation of a grey level image by 512 x 512 x 8 format using 64 binary sections for the time which does not exceed 35 seconds. This method is effective for the extraction of low contrast objects located at uneven background. The method was used for image analysis of glial cells in the substantia nigra of infant brains.

Keywords: binary section, grey level image analysis, segmentation, thresholding, topological tree.

INTRODUCTION

Grey level image analysis consists of the image segmentation and image recognition or description. Terms used in this paper have the same meanings as the ones used by Rosenfeld (1987).
Segmentation is the stumbling-block of image analysis process.

In addition pure algorithmic difficulties arise while extracting of the objects of interest, which may considerably differ by contrast and background levels. In some tasks it is necessary to get information about internal structure for reliable recognition. Furthermore, image may contain redundant information, resulting from noise.
The first step of segmentation is grey level image transformation by image comparison with the constant threshold level or with such threshold level, which is adapted to local or integrated image properties. As a rule, it is impossible to detect all the objects of interest simultaneously, when a constant threshold level is used and there is uneven background in an image.
In the proposed method an attempt was made to eliminate shortcomings mentioned above. Besides, the method contains additional possibilities for the extraction of objects, which are characterized by complex internal structure.

METHODS

The method is based on 3-D representation of 2-D image. The third coordinate Z characterizes the brightness of image pixels. Image analysis consists of three steps. At the first step the description of grey level image is formed. The analyzed image considered as pseudo volume structure is cut by a set of serial sections XY, which locations are defined by a set of grey levels Z. The number of sections and their locations depend on image character and necessary precision of object selection.
In every section the connected components (figures) are selected and labeled. Besides, the connectivity between figures in adjacent sections is determined. In other words the connectivity between pseudovolume image fragments in the third dimension is determined. The information of connected figures in adjacent sections is stored in such a way that the label of each figure is fixed for all the figures which are connected with this one and located at the upper section. Parameters which characterize the location and sizes of the circumscribed rectangle (Feret diameters) and an area are calculated for each figure.
At the second step the topological tree of connected figures is built using information of connectivity between figures in adjacent sections. As an example in Fig. 1 you can see a picture of the analyzed image of GFAP-immunoreactive glial cells. In Fig. 2 the graphical representation of a connected figures tree fragment is displayed. Each rectangle symbol represents a selected figure and linking lines show the figures connectivity. The symbols corresponding to the figures of the one and the same level are located on the same diagram line. The picture shows only a tree fragment. The whole tree consists of about 5000 figures extracted in 26 sections. For further analysis this information is often redundant, as the main part of figures appears as a result of noise. It is possible to apply the parameters filtering of the topological tree to reduce this redundancy. The filtering consists in erasing from the topological tree the information about those figures which don't meet the defined requirements. For example, a minimum area of figures or Feret diameters may serve as a threshold to make a decision for erasing. Another way of filtering consists in erasing from the topological tree those figures which are the branch ends. In most cases such figures are extracted as a result of noise influence. The problem of topological tree analysis has a common character so it was studied in a lot of papers. The approach used by van Pelt and

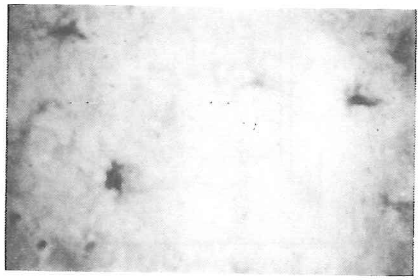


Fig. 1. An analyzed image of GFAP-immunoreactive glial cells.

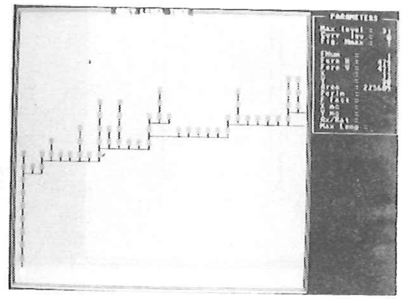
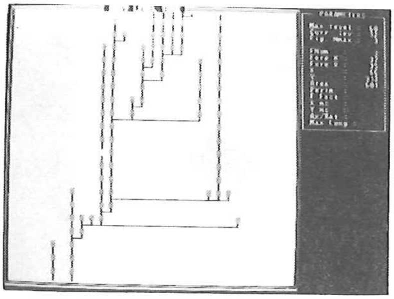


Fig. 2. Fragment of the grey level tree.

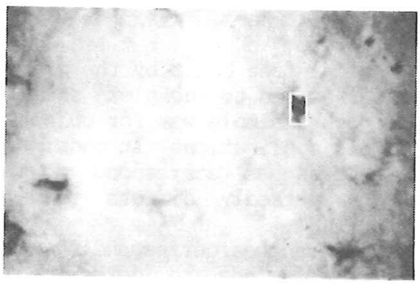
Verwer (1987) may be applied to a topological tree built by the proposed method for branches extraction, which correspond to complex structure objects of interest. But we can propose a simple way for objects of interest selection, which may have multiple applications. It consists of the longest tree branches search. These branches correspond to those grey level image fragments which intensity markedly differs from the surrounding background.

After the extraction of topological tree branches corresponding to the objects under analysis the problem arises as how to determine the informative figure (or the set of figures) in each extracted branch. A number of approaches can be proposed. The first one consists of the analysis of figure area meanings, which belong to one and the same branch. Figures of any branch are enclosed in each other. Their area meanings are arranged in a descending order. The slowest changes of figures dimensions correspond to the sharpest change of intensity on the object border. The analysis of figure areas changes allows to define the optimum thresholding level and consequently a necessary figure. Another way of optimum thresholding level definition consists of the analysis of figures form factor, which belong to extracted objects. The sharpest change of intensity in object image will correspond to the local maximum of form factor. But this way is connected with the necessity of perimeter calculation in the process of image description forming. A brightness meaning histogram may be used for the optimum thresholding level definition. It is built for the image region which is restricted by the lowest tree branch. Thresholding level is defined by the most prominent histogram local minimum, which corresponds to the sharpest brightness change in object image.

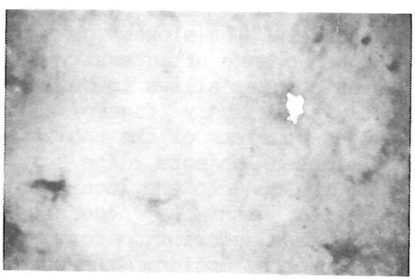
At the third step of the grey level image analysis the extended set of parameters is calculated for each extracted object: perimeter, convex perimeter, form factor, gravity center coordinates, min/max diameters, moments, orientation etc. Besides, parameters characterizing pseudo volume can be calculated. These are object volume (which is equivalent to absorbing substance amount), coordinates of gravity center, vertical cross-section parameters etc. The information contained in the topological tree also allows to define more complex object parameters which describe their topology. It may appear useful while describing objects internal structure.



a)

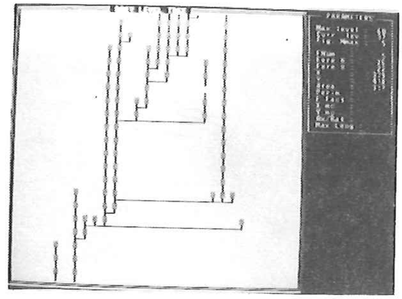


b)

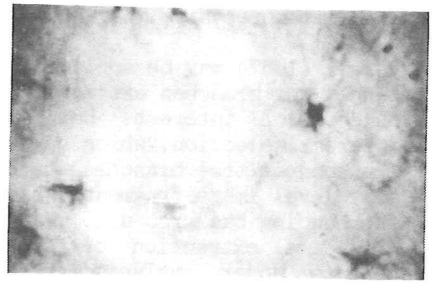


c)

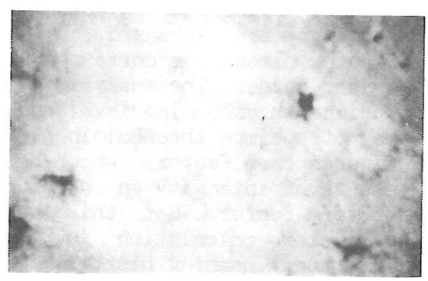
Fig. 3. Extraction of a high contrast object.



a)



b)



c)

Fig. 4. Extraction of a lower contrast object.

a) Cursor (light rectangle) shows the figure corresponding to the optimum threshold level of defined object of interest.
b) Light rectangle frame shows the extracted object of interest on the screen of halftone monitor.
c) The extracted object according to optimum threshold level defined for this object of interest.

RESULTS

The proposed method was realized in an image processing system, consisting of a light microscope, a TV camera, a Frame Grabber card (Visionetics Inc.), built in IBM PC 386/20 Mhz and an original image processing program package. Image processing program package is written in Turbo C (Borland). Functions from the enclosed library were used for communication with Frame Grabber video memory. The system provides segmentation, compressed description forming for image in the format of 512 x 512 x 8 bit and using of a number of binary sections from 1 to 255. Processing time depends on the number of extracted figures of an image which in its turn depends on image complexity: noise, number of objects, objects shape complexity and grey level range. Nevertheless, we shall give some results to show the method productivity. Two images contained 1 and 150 objects, respectively, were analyzed. Segmentation time was 5" and 19" for 32 threshold levels when 32 and 4800 figures were extracted, respectively. When the number of threshold levels was increased to 64 this time was 6" and 33" (64 and 9600 figures were extracted).
For image segmentation a special algorithm was developed which allowed to extract the connected components of an arbitrary shape and to define connectivity between the figures of adjacent sections during one pass scanning. A researcher has the opportunity, working only with the built and displayed tree, to extract objects of interest and then to calculate the expanded set of their parameters. Figure selection is carried out by cursor moving along the tree displayed on PC screen (Fig. 3a). On the greyscale monitor a selected image region is restricted by a rectangular frame which corresponds to the location and size of the rectangular, circumscribed around the selected figure (Fig. 3b). For a more detailed control a researcher can extract the figure in its borders correctly (Fig. 3c). Then the expanded set of image parameters can be calculated. Fig. 4 illustrates extraction of the object, which has a significantly lower contrast as compared to the previous one.
The method was used to study the glial fibrillary reaction in the substantia nigra (SN) of intrauterine growth retarded (IUGR) infant brain in comparison with age-matched normally developed one. Glial cell somata were analyzed in systematically random sections of the SN stained with immunocytochemical method for glial fibrillary acidic protein (GFAP). The obtained data have revealed a significant increase in the cross-sectional cell area and shape-factor but a decrease in the anisotropic factor (the semi-axes ratio of inertia ellipse) in the SN of IUGR case as compared to the control one. It may suggest that there is a hypertrophy of glial cells of the SN in the IURG infant brain as a result of development under chronic hypoxic condition.

DISCUSSION

Experiments have shown that the proposed method can be effectively used for the extraction of low contrast objects located at uneven background. Objects extraction criteria can be sufficiently formalized which allows to perform image processing in an automatic mode.
The analysis of noisy grey level images may be fulfilled without image preprocessing (noise smoothing, border sharping, contrast enlarging and so on), applied for the image enhancing. All these operations enhancing the image quality may bring some distortions which bias the results of parameters calculations. The filtering of parameters used in the method

effectively improve the image description, excluding noise influence. The method has no any restrictions on the number of extracted figures. The volume of operative PC memory may appear insufficient for the storage of obtained information when a lot of thresholding levels are used and there is a significant noise in an analyzed image. In this case hard disk may be used, which leads to some image processing time increase. As practical studies of biological samples showed, the operative memory may overflow when large format images (512 x 512 pixels) and large number of thresholding levels (more than 64) are used. The method may be used not only for 2-D grey level images. To our opinion, it may be applied for the analysis of 3-D images. It will be the subject of future study.
The proposed method of grey level image analysis is intended to be applied in mass cheap image processing PC based systems, providing good processing time.

REFERENCES

Rosenfeld A. An overview of image processing and analysis. Acta Stereol 1987; 6/III: 939-44.

Van Pelt J, Verwer RWH. Mean order as a measure for branching pattern topology. Acta Stereol 1987; 6/III: 393-7.

Van Pelt J, Verwer RWH. Multifurcations in topological trees: growth models and comparative analysis. Acta Stereol 1987; 6/III: 399-404.