

# Unsupervised Active Contour Model for Biological Image Segmentation and Analysis

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*Abstract*— The active contour models or snakes are widely used for image segmentation, especially in case of biological images. This is because the importance of biological image analysis demands quality segmentation which the active contour models can provide. However, active contours suffer from the serious problem of initialization and tend to wander off toward other image features if initialized far off from the actual object. Since *a priori* information about the regions of interest is generally not available in biological images, in this paper, we propose to use the conventional hyperstack based multi-resolution image segmentation technique to extract information about the regions of interest. This helps in initializing the contours close to the actual object boundaries. The active contour model in the subsequent stage refines the initial contours. Experimental results demonstrate the effectiveness of the proposed scheme in terms of segmentation quality.

## 1. INTRODUCTION

Image segmentation is a fundamental step in many computer vision and pattern recognition applications. During the last three decades computerized segmentation techniques have been studied in numerous scientific fields including medical imaging, Landsat image analysis, machine vision, etc. Most of these image analysis processes perform segmentation as a first step towards producing a description of the image. This step in image analysis determines the eventual success or failure of the algorithm. Thus, for successful image analysis applications there is a need for robust image segmentation algorithms.

Image segmentation finds widespread use in the analysis of Landsat images, image registration, target location and detection in radar/sonar images and so on. Segmented image data are also useful in three-dimensional visualization and multimodal visualization. With the rapid development of multimedia applications in modern times, there is a renewed interest in image segmentation with potential applications to content based search and object based coding.

Segmentation also appears to be a key issue in modern biomedical image analysis enabling numerous clinical applications. With the introduction of digital imaging devices in medicine, computerized tissue recognition and classification have become important in clinical diagnosis systems. One such application is where one may be interested in locating and segmenting anatomical structures in a medical image. Image segmentation is also necessary in volumetric analysis of particular tissues and structures, reconstruction of anatomical models, 3D visualization of separate neural structures in magnetic resonance (MR) images, etc. Another very important and related application of image segmentation is automated biological image understanding. For example, it plays a vital role in evaluating the shape and size of microorganisms in a microscopic image. Thus, image segmentation plays a vital role in biological/biomedical image understanding and analysis.

Image segmentation may be defined as the partitioning of the spatial domain on which the image is defined into regions of interest (ROI). The segmented region may be a complete object or a part of it. All segmentation algorithms make systematic use of image features to segment out the regions of interest. Conventional segmentation algorithms are either pixel based or region based. Accordingly, most image segmentation approaches can be placed in one of the following three classes.

- Characteristic feature clustering
- Region growing
- Boundary detection

Characteristic feature clustering is often ineffective because it does not exploit spatial information. The popular *k*-means and ISODATA algorithms are examples of clustering algorithms.

Region growing approach is based on the concept of thresholding and extracting the regions over which a homogeneity criterion is satisfied. Some commonly used region based segmentation techniques are histogram analysis, region growing by pixel aggregation, the split and merge algorithm, watershed algorithm, multiresolution approach, etc. However, with straightforward region growing, spectrally similar but spatially disjoint regions

are never associated together, thus complicating their identification. Also, it is often not clear at what point the region growing process should be terminated, resulting in under- and over-segmentation. In addition, region growing approach generally tends to be a computationally intensive process.

Boundary detection approach is mainly based on the detection of discontinuities in an image commonly known as contour or edge detection. It exploits spatial information through examining local edges found throughout the image. However, conventional techniques for edge detection, e.g., Sobel, Prewitt and Laplacian operators, generally yield a lot of false edges (whose removal requires further processing) and are computationally expensive. Also, while for simple noise-free images, detection of edges results in straightforward boundary delineation, edge detection on noisy and complex images often produces missing edges. As a result, the detected boundaries do not necessarily form a set of closed connected curves that surround connected regions.

One efficient and promising approach for contour-based image segmentation is by using deformable models [1], [2] that has emerged as a vigorously researched model-based approach for computer-aided image analysis. Deformable models are capable of accommodating the often significant variability of structures over time and across different images. The name *deformable models* stems primarily from the use of elasticity theory at the physical level, generally within a Lagrangian dynamics setting. The physical interpretation views deformable models as elastic bodies which respond naturally to applied forces and constraints.

One important class of deformable models are the active contours or *snakes* proposed by Kass et al [3], [4]. Snakes are planar deformable contours that are useful in several image analysis tasks. They are often used to approximate the locations and shapes of object boundaries in images based on the reasonable assumption that boundaries are piecewise continuous or smooth. The potency of active contour models comes from their ability to segment, match and track images of structures by exploiting constraints derived from the image data together with *a priori* knowledge about the location, size and shape of these structures. This means, the active contour model approach is a supervised technique in the sense that *a priori* knowledge about the location, size and shape of the ROIs are necessary and the contours require to be initialized accordingly.

Image segmentation using deformable models have been used successfully in medical image analysis [5]. Among all the different deformable models proposed, the active contour models have attracted the most attention to date especially in case of biological images. This is

because the importance of biological image analysis demands quality segmentation which the active contour models can provide. However, the performances of the active contour models highly depend on how close the initial contours are to the actual boundaries of the ROIs. Unfortunately, *a priori* information about the ROIs is generally not available in biological images. This makes it difficult to employ active contour models in biological image segmentation. Consider a medical imaging based automated clinical diagnosis system where the task is to detect tumor or any other anatomical deformity. Since it is not possible to have prior information about the location and form of any such abnormality that may be present, active contour models can not be used reliably in this case. The problem exists in other typical biological images as well: The position of micro-organisms in a microscopic image is random, anatomical structures of two different persons are in general similar but not exact, and so on. Hence, an unsupervised active contour model is desired so as to guarantee quality segmentation even in the absence of any *a priori* information about the ROIs.

In this paper, we propose a hybrid scheme that combines multi-resolution based image segmentation algorithm with the active contour models. The multi-resolution approach helps in estimating the initial contour points while subsequent application of the active contours refines the segmentation result. So, we may say that the proposed hybrid scheme is basically an unsupervised active contour model approach to image segmentation that suits our requirement in case of biological image analysis, i.e., high quality of segmentation while no *a priori* information about the ROIs is available.

## 2. PROPOSED UNSUPERVISED ACTIVE CONTOUR MODEL

In this paper, we develop a novel approach for unsupervised image segmentation using the active contour models. In the first stage of the algorithm, initial contour points may be derived through any unsupervised image segmentation approach. However, since coarse initial contours are enough to satisfy our requirements, we would like to reduce the number of pixels participating in the segmentation process thereby speeding up the algorithm considerably. This may be accomplished through down-sampling of the original image. Hence, multi-resolution image segmentation is proposed for the purpose. Many algorithms have been formulated for multi-resolution image segmentation, viz., multiscale approaches like the wavelet based multiresolution approach [6], the stack [7] and the hyperstack [8] approaches, the pyramidal approach [9], etc. Among these, the hyperstack algorithm is chosen for its computational simplicity.

The hyperstack based multi-resolution segmentation helps in initializing the contours close to the actual object boundary. This is followed by boundary detection so as to provide the initial control points for the active contours in the following stage. Boundary detection may be accomplished by using any edge-linking procedure available in the literature. The object boundaries so obtained is subsequently refined using active contours. Thus, we develop an unsupervised active contour model that may be used reliably in biological image segmentation and analysis.

### The Hyperstack Algorithm

Hyperstack allows for outer scale reduction that provides reduction in the number of pixels as the scale increases thereby speeding up the process of hyperstack building. There are four steps in the hyperstack algorithm as follows.

*Blurring:* A hyperstack is a pixel-based multi-scale data structure whose levels are constructed by convolving the original image with a Gaussian kernel of increasing width. Suppose, we denote the original image by  $L_0(\mathbf{v}, \sigma_0)$ , where  $\sigma_0$  is the inner scale of the image. Then the  $\sigma$ -blurred replica of the original image is given as

$$L(\mathbf{v}, \sigma_0 \oplus \sigma) = L_0(\mathbf{v}, \sigma_0) * G(\mathbf{v}, \sigma) \quad (1)$$

where  $G(\mathbf{v}, \sigma)$  is the Gaussian kernel of width  $\sigma$  and, following the semigroup property,  $\sigma_0 \oplus \sigma = \sqrt{\sigma_0^2 + \sigma^2}$ .

*Linking:* Child-parent linkages are established between pixels at adjacent scale levels according to a model-directed linkage scheme. Thus, a scale tree of linkages is created that has active and passive pixels. Active pixels are those with at least one child or parent reference. All other pixels that do not participate in the linking process are passive. For each active pixel at level  $n$ , a parent is sought in a search volume at level  $n+1$ . The parents are selected on the basis of their affection or linkage strength to a given child; the candidate parent with the highest affection value is selected to become the child's parent. The three affection components of linking scheme are: (1) Intensity proximity, (2) the ground volume measured as the number of pixels to which a parent is connected at the ground level (original image), and (3) the ground volume mean intensity.

*Root labeling:* In the resulting tree-like data structure, roots are formed to indicate the most plausible locations in the scale space. A root is a pixel in the scale tree that represents a single segment in the ground level. They are formed by selecting the segmentation level; all active pixels in the segmentation level are roots.

*Downward projection:* The final segmentation is obtained by tracing back the linkages for all the roots. All

the ground pixels that are connected to a common root are classified as a single segment.

### Active Contour Models

An active contour model or *snake* is defined as energy minimizing spline, with its energy dependent upon its shape and location within the image. Typically, local energy minima correspond to desired image properties. The energy function to be minimized is a weighted combination of internal and external energy forces. The standard active contour energy function is usually given with relation to the entire contour.

An active contour or snake can be open (like a length of string) or closed (more like balloon) and can be represented by a parameterized curve  $\mathbf{v}(s) = (x(s), y(s))$  that represents the contour coordinates  $(x, y)$  as a function of arc length  $s$ . The best fit between a snake and an object's shape is obtained by minimizing the total energy  $E$ . Energy in the active contour model is represented by three energy terms, viz.,  $E_{int}$ ,  $E_{img}$  and  $E_{con}$ , and is given as

$$E = \int_{\mathbf{v}} E_{snake} ds = \int_{\mathbf{v}} (E_{int} + E_{img} + E_{con}) ds \quad (2)$$

Each of the terms that make up the energy model can be intuitively realized in a physical sense.  $E_{int}$  is the internal spline energy caused by stretching and bending. Minimization of the internal energy avoids excessive curvature and elongation of the snake thus providing continuity and smoothness.  $E_{int}$  is given as

$$E_{int} = \alpha(s) \left| \frac{d\mathbf{v}}{ds} \right|^2 + \beta(s) \left| \frac{d^2\mathbf{v}}{ds^2} \right|^2 \quad (3)$$

where  $\alpha(s)$  and  $\beta(s)$  are the measures of elasticity and stiffness of the snake, respectively. The first term in the expression accounts for continuity that likes to maintain equal distances between points in the snake. From implementation point of view, this may be formulated by taking the difference between the average segment length and the current snake segment length, i.e.,

$$\frac{d\mathbf{v}}{ds} = D_{avg} - |\mathbf{v}_i - \mathbf{v}_{i-1}| \quad (4)$$

where  $D_{avg}$  is the average distance between the points  $\mathbf{v}_i$  on the snake. This has the effect of maintaining an even spacing of the points. The second term is the curvature which is a measure of the change in external angle with respect to arc length. This may be estimated by

$$\frac{d^2\mathbf{v}}{ds^2} = \mathbf{v}_{i-1} - 2\mathbf{v}_i + \mathbf{v}_{i+1} \quad (5)$$

$E_{img}$  denotes the image energy which determines the effect of the image on the form of the spline. The image energy is derived from the image data and is given as

$$E_{img} = w_{line}E_{line} + w_{edge}E_{edge} + w_{term}E_{term} \quad (6)$$

where  $w_i$  are appropriate weighting functions. Commonly, the line functional  $E_{line}$  is defined simply by the image function  $f(x, y)$  so that if  $w_{line}$  is large positive the spline is attracted to light lines (or areas) and if large negative then it is attracted to dark lines (or areas). The edge functional  $E_{edge}$  is defined by  $|\nabla f(x, y)|^2$ . Hence, the spline is attracted to large image gradients, i.e., parts of the image with strong edges. The third term  $E_{term}$  is the termination functional that allows terminations (i.e., free ends of lines) or corners to attract the snake. Thus, the snake may be attracted to lines, edges or terminations. Finally, the constraint energy  $E_{con}$  is determined by external constraints that may come from higher knowledge about the image in question.

Once the snake parameters are chosen, the snakes are placed on the image. They require starting shapes and locations, preferably near to that of the desired objects. The resulting forces are calculated iteratively, thereby moving the snakes across the image until some stable position is reached. Thus, snakes lend themselves to object delineation, particularly where guided by higher level understanding processes (whether interaction with human or guidance from an automatic system). With the above described method it is, in principle, possible to track the boundary of any arbitrarily shaped object in an image. The advantage of using snakes is that the final form of the contour can be influenced by feedback from a higher level process.

Of the popular models proposed for snakes, the models proposed by Kass et al [3], [4], Amini et al [10] and Williams and Shah [11] are particularly important. The model proposed by Kass used a variational approach to solve the problem of energy minimization of the active contour. However, Amini et al pointed out problems with the minimization procedure used, such as instability and a tendency for points to bunch up on a strong portion of an edge. They proposed a dynamic algorithm for minimization of the energy functional that allows addition of hard constraints to obtain more desirable behavior of the snakes. But, this algorithm is expensive in terms of computational cost as well as storage. Subsequently, Williams and Shah proposed an algorithm that is stable, flexible, allows hard constraints and runs much faster than the dynamic programming method.

### 3. EXPERIMENTAL RESULTS

In order to demonstrate the performance of the algorithm described above, the algorithm was tested on some different types of biological images, viz., a microscopic

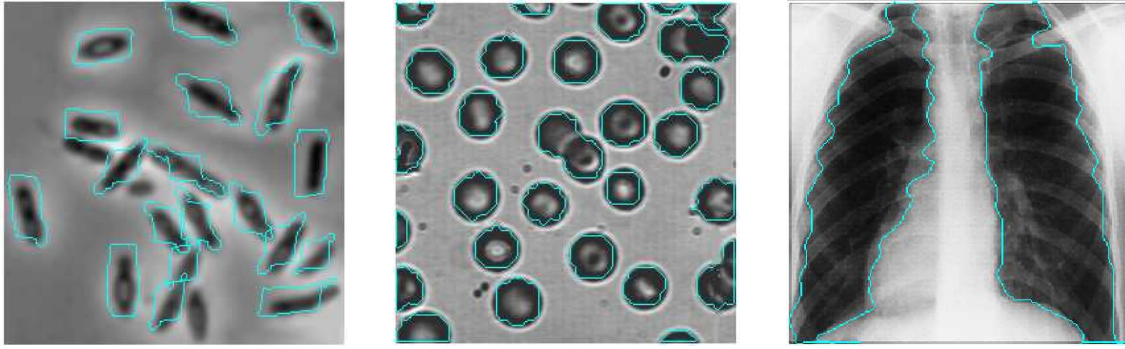
view of bacteria culture, a pathological image of blood cells and an X-ray image of the lungs. Figure 1 shows the initial contours on the test images, obtained after applying down-sampling, multi-resolution image segmentation and boundary tracing. Figure 2 gives the final refined contours obtained after applying active contour model. As we observe, the hyperstack based multi-resolution approach gives rough contours around the objects in the images while the final contours are smooth and capable of capturing all the regions of interest.

### 4. CONCLUSION

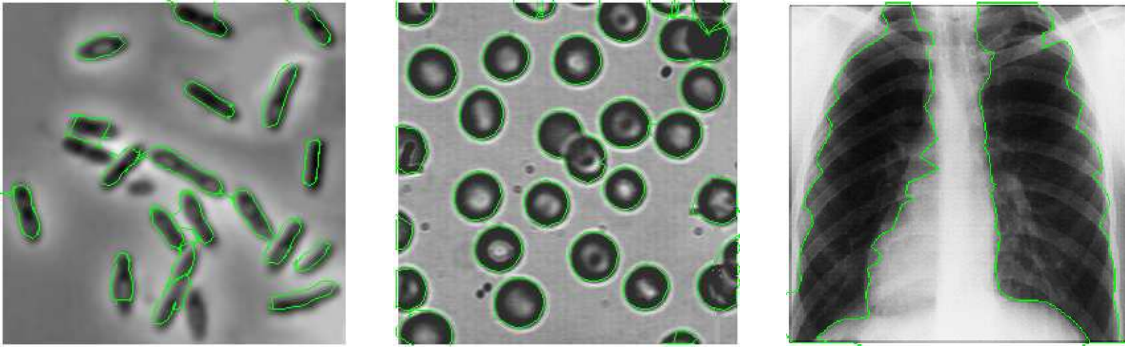
A method for segmenting biological images is presented. The problem of requiring initial contours in the active contour models has been taken care of by the hyperstack approach which yields initial coarse contours. The computational complexity in this stage is reduced by using a down-sampled version of the original image while Gaussian blurring helps to remove strong portions of edges where the contour points might bunch up. Application of active contour model in the final stage yields accurate results. Therefore, the proposed scheme proves to be an ideal choice in biological image segmentation. This is corroborated by our experimental results. However, the algorithm works well only on images with good contrast, i.e., images which have the regions of interest well distinguished in gray-scale from the background. Therefore, image enhancement is necessary as a preprocessing step for images with poor contrast, e.g., X-ray images.

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**Figure 1.** Initial contour obtained after segmentation using multi-resolution approach



**Figure 2.** Final contour obtained after segmentation using active contour model

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