1 General Notes

1.1 Image

1.1.1 Image Types

1. Pathology images
2. Fluorescence microscopy
3. Confocal images
4. H&E stained cancer images

1.1.2 Microscopy Models

1. Bright field
2. Fluorescence
3. Phase contrast

1.2 Random Summary

Seems a lot of the problems w/ different models are parameter selection. And usually, we can use some adaptive methods to optimally choose the parameter. Often, can be converted to optimization problems. And often can be exactly solved using DP like methods.

1.3 Software

1. ImageJ, detection and segmentation
2. CellProfiler, quantitative analysis on cell phenotypes.

1.4 Challenges

1.4.1 Touching/Overlapping

Many extant algorithms depend on datasets. Segmentation w/ shape preserving is very important.

1.4.2 Scalability to Large Number

Unnecessary to achieve cell segmentation on the whole slide images. But parameter choosing etc. are more accurate if whole image considered.

2 [1]

3 [2]

3.1 Detection

3.1.1 Distance Transform

Mechanism Local maxima = centroids of nuclei or cells. Often paired with "Watershed Segmentation".

Advantage

Disadvantage Only effective on regular shapes in a binary image. Susceptible to small changes. Complex image → variations → over-detection.

Improvement Gaussian filter, then trace gradient vector field. Accumulated pixels threshold to distinguish b/w local and non-local maxima.

Further Lin et al. gradient weighted-distance transform for 3D fluorescence image.

3.1.2 Morphology Operation

Mechanism Binary morphological filtering for images w/ certain structure element, circle, square, cross... Examining the geometrical and topological structures of objects w/ predefined shape. Four basic shift-invariant operators:

1. Erosion
2. Dilation
3. Opening
4. Closing

The four can be used to generate more basic morphological operations, boundary, hole, skeletonizing... Binary morphology can be extended to gray-scale morphology. Widely used operators: top-hat, bottom-hat transforms. For example, UE (Ultimate Erosion). Erosion until can't.

Advantage Can be used to basic image enhancement, preparing for further analysis. UE: can separate touching or overlapping cells.

Disadvantage UE: can produce multiple marker for each cell.

Improvement

1. Improved UE, Park et al. noise robust stopping criterion. Perform until convex. However, binarization.
2. Conditional Erosion: Yang et al. Coarse erosion preserves shapes, and fine erosion avoids under-segmentation

Further

Hodneland 3D fluorescence images. Adaptive threshold for ridge extraction, then link gaps.

Plissiti gray-scale, not converting.

3.1.3 H-minima/maxima Transform

Mechanism Based on morphology operation, used in local minima detection. Image A, depth value h, $H(A,h) = R^{\epsilon}(A+h)$, where R^{ϵ} is reconstruction by erosion. Some regional minima are suppressed. Initially connected parts can be split in terms of the detected minima, h leads to under/over-segmentation. Usually used to generate markers for watershed transform based segmentation.

Advantage Compared with DT (EDT), all minima $\rightarrow$ H-minima. Very popular in biomedical images.

Disadvantage Suppress minima, so needs enhancement beforehand. Properly defined h value is needed.

Improvement

1. Adaptive HIT., iteratively increase h until a region merging. Ignores nucleus size.
2. Jung and Kim, adaptively choose h to minimize segmentation distortion.
3. Variance in cell areas.

Further

3.1.4 LoG, Laplacian of Gaussian

Mechanism In medical image analysis, LoG is one of the most popular for small blobs.

Advantage

Disadvantage

1. Might fail in touching / overlapping objects.
2. Scale issue.

Improvement

1. Lindeberg introduces normalizing factor for multiscale LoG blob detector.
2. Kong generalized LoG, for elliptical structures (oblique elliptical Gaussian)
3. Hessian analysis to identify optimal scale
4. Unsupervised GMM can be used to refine blobs

Further

3.1.5 Maximally Stable Extremal Regions

Mechanism Maximally Stable Extremal Regions. Set of nested extremal regions based on level sets in the intensity landscape. Local intensity minimum-based criterion.

1. Generate sufficient number of extremal regions.
2. Recognize those regions corresponding to real nuclei or cells.
   (a) Eccentricity
   (b) Blob appearance + shape properties
   (c) Arteta formulates an optimization problem, candidates -¿ scores -¿ DP for maximal total score
   (d) Multilevel thresholding

Advantage

Disadvantage

Improvement

Further

3.1.6 Hough Transformation

Mechanism Circular/elliptical nuclei in pathological images. From xy-plane transform to circular a, b, r parameter space. Discrete voting strategy? Most votes corresponding to parameter? Locate the targets by seeking peaks in parameter space (e.g. gradient descent).

Advantage

Disadvantage False peaks due to noise, incorrect edge extraction, touching objects. Further analysis is needed.

Improvement Gaussian smoothing to denoise, morphology operations to reconstruct. SVM classifier. Optimization problem can be solved by some ILP.

Further

1. Can deal with arbitrary shapes.
2. 3D transformation can be done.
3. Randomized version

3.1.7 Radial Symmetry Based Voting

Mechanism Locate the centroids of nuclei or cells. High radial symmetry points highlighted.

Advantage

Disadvantage High computational complexity. False peaks due to clustered nuclei. Radius range. What if not circular?

Improvement FRST. Candidates, thresholding. Affine transform to deal with non-circular.

Further

3.1.8 SVM

3.1.9 Random Forest

3.1.10 DNN, esp. CNN

Mechanism

Ciregan Mitotic cell detection in breast cancer histology images.

1. Probability map of being centroid of a mitotic cell.
2. Smoothed w/ disk kernel
3. Non-maxima suppression

Alternatively, can be formulated into an optimization problem.

1. Candidates by LoG, MSER, iterative voting, etc
2. Score by CNN
3. Best subset of candidates

Advantage

Disadvantage Computationally expensive for large-scale images.

Improvement Fast scanning.

Further

3.2 Segmentation

Methodologies:

1. Separate fore and back grounds, and then splits
2. Markers, then expand
3. Generate candidates, then select

Algorithms:

1. Thresholding
2. Morphology Operation
3. Watershed transform
4. Deformable models
5. Clustering
6. Graph-based models
7. Supervised learning

3.2.1 Intensity Thresholding

First and simplest method.

Mechanism Assumption: intensity distributions for fore- and back- grounds are sufficiently and consistently distinct. Convert to binary with global threshold, or locally adaptive threshold. Usually empirical. Can also be some optimization problem. Inter-variance for example.

Advantage

Disadvantage How to choose threshold

Improvement Dividing into sub-images. However, introduce other need-to-defined parameters.

Further Convert RGB to gray-scale, Callau

3.2.2 Morphology Operation

Mechanism Top down erosion and bottom up dilation. Erosion until markers are obtained. Grows the markers w/ dilation to reconstruct, while preventing merging.

Advantage

Disadvantage Under-segmentation in dense cell clumps.

Improvement Modeling w/ shapes

Further Always used to facilitate subsequent segmentation.

3.2.3 Watershed Transformation

Mechanism most popular region accumulation method. Seed points, then add pixels. Flood water in the regional minima, while preventing merging building dams. Highest point. Boundaries -¿ watershed lines, slitting the landscape into regions.

Advantage Gradient magnitude images, also gray intensity images, distance transform maps, and other gray scale images.

Disadvantage Over segmentation.

Improvement Merge false segmentations based on real nuclei or cells.

Further

3.2.4 Deformable Models

Mechanism One of the most popular. Pre-specified region, active contour evolves to boundary by minimizing energy functional, achieves segmentation when reaches boundary. $EG(v) = E_{int}(v) + E_{image}(v) + E_{con}(v)$. Internal smoothness, image energy encouraging towards feature, constraint for interaction with user.

Advantage Great tradeoff b/w efficiency and flexibility.

Geodesic Models

Parametric Models

3.2.5 Clustering

Mechanism Different levels of similarity, internal, and outside. Often followed by edge extraction.

1. K-means: iterative descent $argmin \sum_{i=1}^{N} \sum_{k=1}^{K} r_{ik} \|x_i - \mu_k\|^2$
2. Fuzzy C-means: Not hard as K-means, one object to plural clusters, membership degree.
3. Expectation-Maximization: also soft. Mixture of Gaussians: $N(x_i|\mu_k, \Sigma_k)$ Maximize log likelihood over the Gaussian parameters and weights.

Disadvantage What metrics to choose, Euclidean distance, correlation, 0-1 error, etc.

Improvement

3.2.6 Graph-Based Methods

Model one image as a weighted graph, each node is a pixel, or subpixel, edge weights = similarity b/w pixels. By certain criterion, partitioned into multiple sets, representing segmentation.

1. Max-flow/Min-cut: Graph-cut algorithm minimizes an energy function. $EG(L) = \sum_{v \in V} D_v(L_v) + \sum_{(v,u) \in N} S_{v,u}(L_v, L_u)$, N penalty plus interaction potential that controls the spatial smoothness.
   Favoring partitioning out small sets of nodes, which are undesired.

2. Normalized cut: Avoid unnatural bias. Somehow normalizes the cut.

3. Conditional Random Field: Variant of Markov random field, set of random variables represented by a graph.

4. Random Walk: graph edge weight represents the likelihood that a random walker will cross the edge.

3.2.7 Supervised Learning

Disadvantage Pixel-wise usually unable to handle touching objects. Superpixel-wise partitions into set of candidate regions, then discriminates. Highly depends on the superpixels.

1.

References

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[2] F. Xing and L. Yang, "Robust nucleus/cell detection and segmentation in digital pathology and microscopy images: A comprehensive review," *IEEE Reviews in Biomedical Engineering*, vol. 9, pp. 234–263, 2016.