FR-Be-GE

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FR-Be-GE: SUMMARY

Our approach [2] for cell tracking focuses on the segmentation of cells in phase contrast microscopy images. The key feature of our algorithm is that it strongly favors dark-to-bright transitions at the boundaries of the (arbitrarily shaped) segmentation mask, which is effectively found by a fast min-cut approach. The small but essential difference to standard min-cut based approaches is that our graph contains directed edges with asymmetric edge weights. The tracking algorithm applies segmentation propagation to promote temporal consistency. Label propagation is performed by a greedy association of segments between two consecutive frames. We provide an open-source implementation for Matlab on our homepage [1].

FR-Be-GE: PREPROCESSING

Image intensities are normalized to the interval [0, 1] first. Then, images are background corrected by subtracting the smoothed image (large Gaussian kernel with standard deviation σ_{bgr} of 20 pixels) from the original image.

FR-Be-GE: SEGMENTATION

Our segmentation approach exploits the fact that the true cell borders in positive phase contrast microscopy always appear as a dark-to-bright transition in outwards direction. It means that all borders with an inverse transition (bright-to-dark) are definitely not the sought cell borders. We set up a segmentation energy functional for a mask $M:\Omega \rightarrow \{0,1\}$ with $\Omega \subset \mathbb{R}^2$ and the given image $I:\Omega \rightarrow \mathbb{R}$. The functional contains a data cost $C_{obj}:\mathbb{R} \rightarrow \mathbb{R}$ that depends on the intensity, and an edge cost $C_{edge}:\mathbb{R} \rightarrow \mathbb{R}$ that depends on the intensity gradient at the mask border in outwards direction

$$E(M) = \lambda \int_{\Omega} M(x) \cdot C_{\text{obj}}(I(x)) dx + \int_{\Omega} C_{\text{edge}}(\langle \nabla M(x), -\nabla I(x) \rangle) dx$$

where we define ∇M to be a unit normal vector on the mask boundary and **0** elsewhere. The data cost for a gray value v is derived from the foreground intensity histogram P(v|O) and background intensity histogram P(v|B) from training regions. We define it as $C_{obj}(v) = (P(v|B) - P(v|O))/(P(v|O) + P(v|B))$. The edge cost for the intensity derivative d is computed as

$$C_{\text{edge}}(d) = \begin{cases} \exp\left(-\frac{d^2}{2\sigma^2}\right) & \text{if } d > 0, \\ 1 & \text{otherwise.} \end{cases}$$

This asymmetric edge term favors dark-to-bright transitions at the mask borders. To optimize the energy in (1), we discretize the edge term into eight directions (8 pixel neighborhood) and solve it by a min-cut [3] using the maxflow algorithm MATLAB-interface [4]. The min-cut parameters λ and σ are estimated by the best performing parameters on the training data found by grid-search. Region histograms (with N_{bins} = 120 bins) for computing the data costs in (1) are obtained from manual foreground and background scribbles drawn into a single frame of a training sequence (that is not contained in the segmentation ground truth). We use both training sequences (2 frames in total) for dataset **PhC-C2DH-U373** and only one training sequence (1 frame in total) for dataset **PhC-C2DL-PSC**. Finally, we optionally apply a hole-filling algorithm (*holeFilling*) and discard small segments below a pixel area threshold a_{\min} .

FR-Be-GE: TRACKING

Our tracking algorithm consists of two parts. Segmentation propagation promotes temporally consistent segmentation by propagating segmentation information to subsequent frames. Label propagation transfers the label of each segment to subsequent frames using a greedy association.

Segmentation propagation. Min-cut segmentation yields a binary segmentation mask. Segmentation information is propagated from frame t to frame t + 1 in two fashions:

- <u>Foreground propagation (FP)</u>: The eroded mask is set as hard foreground constraint for the mincut segmentation in the next frame. This adds robustness to the region term in case of insufficient foreground evidence. The size of erosion must be chosen at least as large as the expected motion of the object boundary pixels between frames. The erosion is computed using a disk-shaped structuring element (radius s_{erosion}).
- <u>Non-merging constraint (NM)</u>: If it can be assumed that cells do not merge, it is reasonable to
 prevent separate objects from merging in the next frame. We achieve this by computing a
 distance transform on the segmentation mask and applying watershed transform seeded at the
 object locations. The boundaries of the herewith computed "support regions" of each object are
 set as hard background constraint.

Label propagation. For propagating labels we use a greedy algorithm. Each segment in frame t propagates its label to the segment in frame t + 1 with the highest overlap (measured as intersection over union). If a segment in frame t + 1 receives multiple labels, it prefers the segment in frame t with the highest overlap and discards the other labels. If a segment receives no label, a new label is assigned. Additionally, the provided field of interest (FOI) specification is used to discard segments that lie completely outside the FOI (specified by the value *E*). However, we still use the tracking information from the full view to add parent links in case segments, which are tracked on the full view, reenter the FOI.

FR-Be-GE: POST-PROCESSING

No post-processing is carried out after tracking.

REFERENCES

- 1. http://lmb.informatik.uni-freiburg.de/resources/opensource/CellTracking/
- 2. Bensch R, Ronneberger O. Cell segmentation and tracking in phase contrast images using graph cut with asymmetric boundary costs. In *Proceedings of the 12th IEEE International Symposium on Biomedical Imaging*, 1220-1223 (2015).
- 3. Boykov Y, Funka-Lea G. Graph cuts and efficient n-d image segmentation. *International Journal of Computer Vision* **70**, 109-131 (2006).
- Boykov Y, Kolmogorov V. An experimental comparison of min-cut/max- flow algorithms for energy minimization in vision. *IEEE Transactions on Pattern Analysis and Machine Intelligence* 26, 1124-1137 (2004).