UP-PT

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UP-PT: SUMMARY

We track cells in 2D and 3D microscopy image sequences based on Laplacian of Gaussians (LoG) local detection and detection-association tracking. The detection-association is based on a Euclidean distance nearest neighbor search for detections between consecutive frames. The cells in all data modalities are detected using LoG filtering which enhances the image's blob like structure which corresponds to cell locations [2]. To improve the cell's shape estimation after Log detection we resort to automatic image threshold or to the Sliding Band Filter (SBF) as better suited to the data modality under analysis [1]. The SBF is a local convergence filter, capturing edge convergence on a band around a certain location, which can also detect blob like structure, but which can adapt to a wider range of shapes than the LoG filter.

UP-PT: PREPROCESSING

We rescale each image (*imScale*) for speed purposes. We reduce image noise by applying a Gaussian filter with a specific standard deviation (*LOGsigma*).

UP-PT: SEGMENTATION

We perform detection using scale non-maxima suppression, where we vary the scale of the LoG filter between the expected range of the cell radius (*Rmin*, *Rmax*) [1, 2]. In the first frame, we specify the cells to track by considering local maxima from the filter response above a specific threshold (*initDetectTH*). In the remaining frames, we consider all the detections with a filter response above a lower threshold (*detectTH*) to ensure the detection of all the cells being tracked. Given all detections, we then perform cell shape estimation based on a local convergence filter (SBF option) [1, 2] or based on image thresholding (Otsu option) specifying the method to use in the parameter *getCellShape*. For SBF parametrization, we set the range of filter scales to be the same as in the LoG (*Rmin* and *Rmax*), setting the bandwidth of the filter (q) and the number of orientations (N) to values verified to be adequate to the data [1, 2]. The SBF is applied in the image only to the locations of LoG detection for a better cell shape estimation for each detection. When a good shape estimation can be obtained using automatic

thresholding, we apply Otsu's method, in which we decide if we want to smooth the image again or not (*filterShape*) previous to image thresholding. For each LoG detection, we select a region (ROI) around the specific cell location where the size of this region is given by multiplying the estimated cell size by a specific factor (*windowSize*). Finally, within the selected ROI we focus on the bigger segmented region (in terms of area) and we get the boundary information, which corresponds to the boundary estimate for the detected cell. In case of 3D data, the detection and shape estimation is performed in all the slices of each data volume that has image entropy higher than a predefined value (*entropyE*). Detections that appear near the same location (x,y) within z-axis are considered imaging slices of the same cell. Cells that are detected in less than a certain number of slices (*minSlices*) are discarded. During the process of saving the results, we also have the possibility of increasing the estimated cell size by dilating *sizeComp* times the segmentation result.

UP-PT: TRACKING

Our tracking is based on a detection-association approach [3]. We perform the association of the closest detections in consecutive frames based on the Euclidean distance. If the detection does not have a neighbor within a minimum distance (4*detection_radius), we stop tracking it.

UP-PT: POST-PROCESSING

Based on the detection-association result, we get the final cell tracking information by performing the next steps: merging of incomplete tracks that are in close spatio-temporal vicinity; removal of tracks that have less than four frames; merging each track that does not start in the first frame with the closest track that starts in a previous time point (mitosis event). For the **Fluo-N3DL-DRO** dataset, we only consider tracks that start from the first frame and we remove all the others.

REFERENCES

- Esteves T, Quelhas P, Mendonça AM, Campilho A. Gradient convergence filters and a phase congruency approach for in vivo cell nuclei detection. *Machine Vision and Applications* 23, 623-638 (2012).
- 2. Esteves T, Oliveira MJ, Quelhas P. Cancer cell detection and morphology analysis based on local interest point detectors. In *Proceedings of the 6th Iberian Conference on Pattern Recognition and Image Analysis*, 624-631 (2013).

3. Esteves T, Oliveira MJ, Quelhas P. Cancer cell detection and tracking based on local interest point detectors. In *Proceedings of the 10th International Conference on Image Analysis and Recognition*, 434-441 (2013).