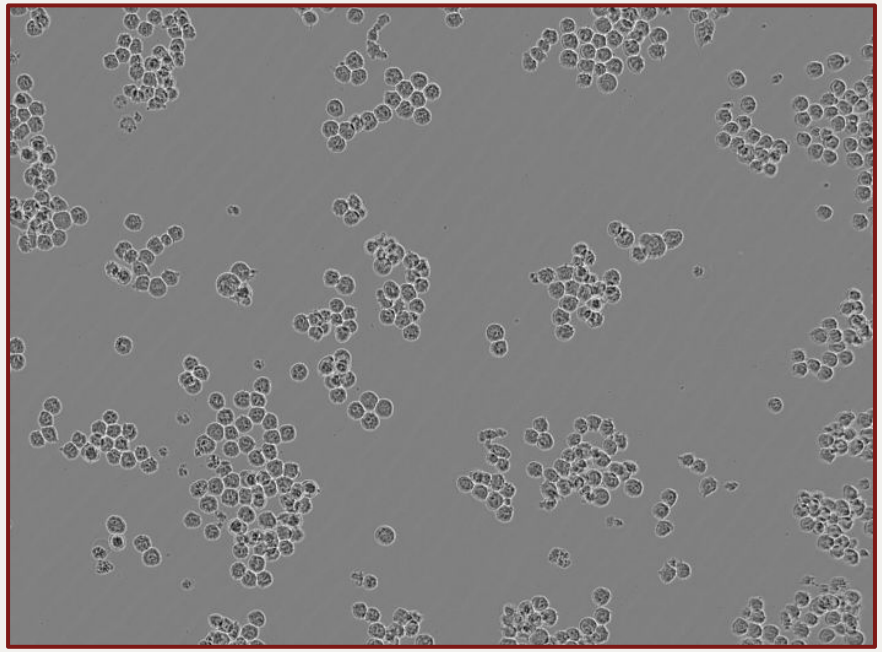
Ahmedabad University

Label-Free Cell Detection Using Traditional Image Processing on the LIVECell Dataset

Group 5
Nirjara Jain, Aashi Shah, Diya Patel, Bansi Mahkana

Introduction

- Automated cell detection and counting
- Traditional image processing
- Based on LIVECell label-free microscopy dataset
- Challenges:
  - Low contrast
  - Uneven lighting
  - Overlapping cells
- Aim: count individual cells accurately without using fluorescent labels and deep learning



Literature Review

- Object counting is key in biomedical imaging and automation
- LIVECell dataset: large-scale, label-free images
- Traditional image processing: fast & explainable
- Deep learning: accurate but needs large labeled datasets
- Need for label-free, classical methods that work without rely on labeled data

RESOURCE
https://doi.org/10.1038/s41592-021-01249-6
nature methods
Check for updates
OPEN

LIVECell—A large-scale dataset for label-free live cell segmentation

Christoffer Edlund[1,5], Timothy R. Jackson[2,5], Nabeel Khalid[3,5], Nicola Bevan[2], Timothy Dale[2], Andreas Dengel[3], Sheraz Ahmed[3], Johan Trygg[1,4] and Rickard Sjögren[1,4] ✉

Light microscopy combined with well-established protocols of two-dimensional cell culture facilitates high-throughput quantitative imaging to study biological phenomena. Accurate segmentation of individual cells in images enables exploration of complex biological questions, but can require sophisticated imaging processing pipelines in cases of low contrast and high object density. Deep learning-based methods are considered state-of-the-art for image segmentation but typically require vast amounts of annotated data, for which there is no suitable resource available in the field of label-free cellular imaging. Here, we present LIVECell, a large, high-quality, manually annotated and expert-validated dataset of phase-contrast images, consisting of over 1.6 million cells from a diverse set of cell morphologies and culture densities. To further demonstrate its use, we train convolutional neural network-based models using LIVECell and evaluate model segmentation accuracy with a proposed a suite of benchmarks.

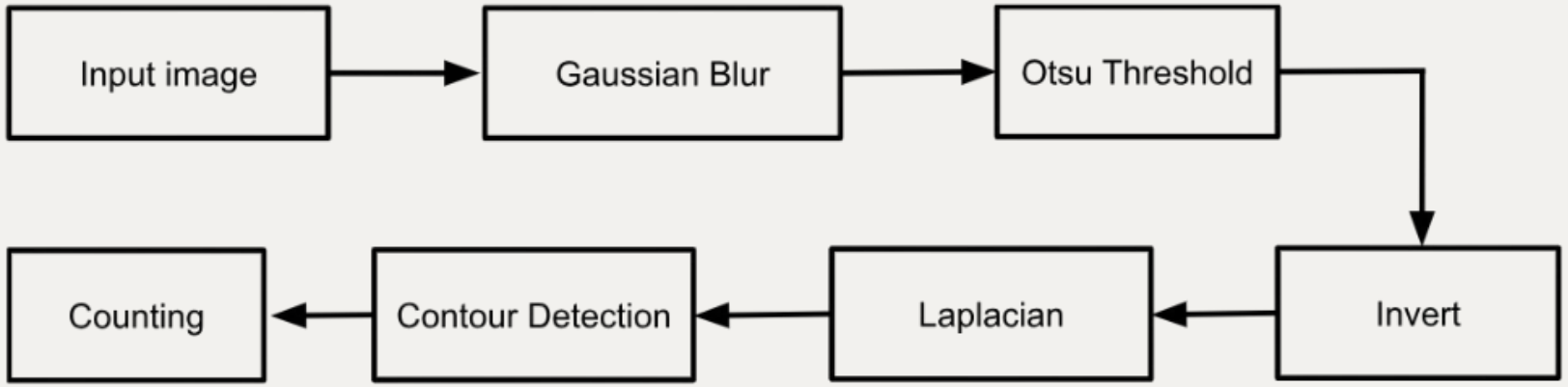
Quantitative imaging offers unequaled spatial and temporal resolution when measuring biological phenomena, which has led to its wide use in cell biology and biomedical research. Two-dimensional (2D) cell monolayer models of mam- contrast for resolving cells grown in a monolayer. Furthermore, the morphology of a cells in culture can vary dramatically, not only across cell types, but also due to genetics and epigenetics, microenvironmental factors, stages in the cell cycle or differentiation

Dataset Discussion

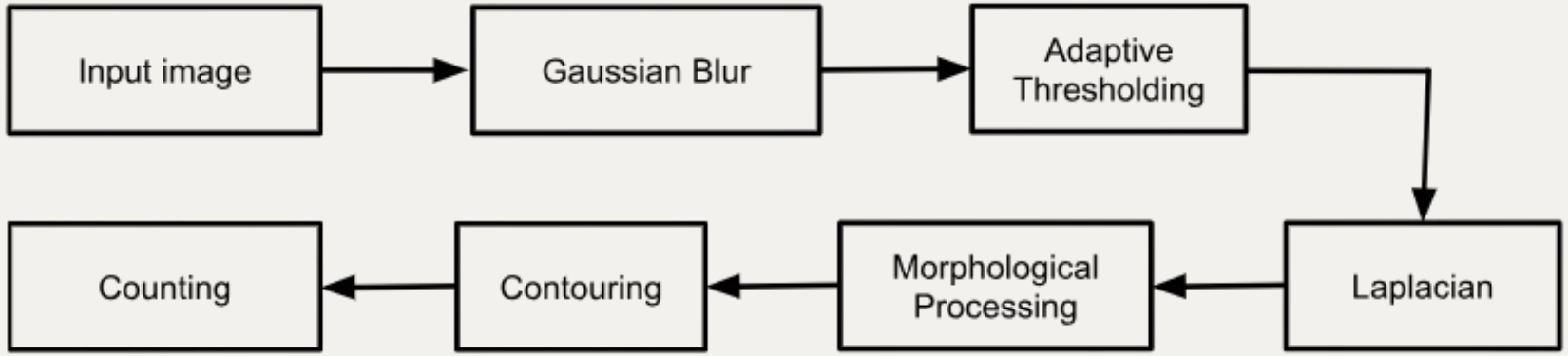
Name	LIVECell Dataset
Image Type	Label-free phase-contrast microscopy images
Size	5,239 images, ~1.6M annotated cells
Cell Types	8 mammalian cell lines A172, BV-2, BT-474, Huh7, MCF7, SH-SY5Y, SKBr3, SK-OV-3
Challenges	Low contrast, overlapping & irregular cell shapes, halo artifacts
Annotations	Pixel-level expert annotations for each cell is provided
Goal	Achieve accurate cell detection per image without any deep learning model

Methodology

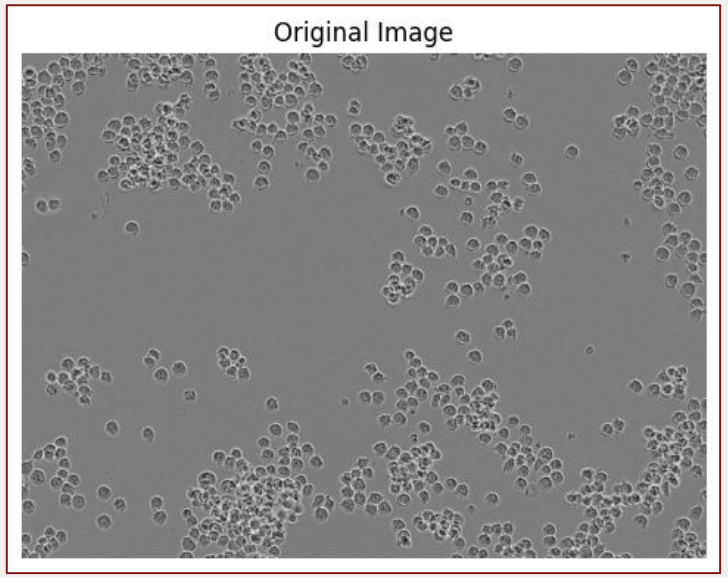
Otsu's Thresholding



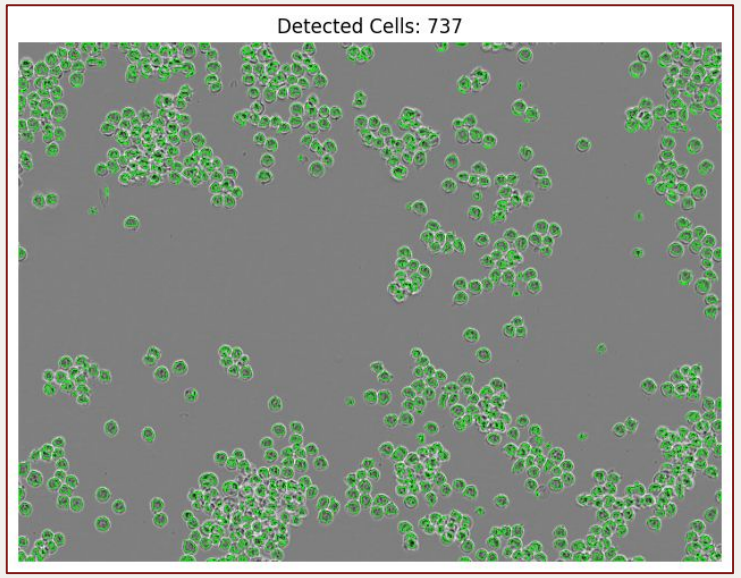
Adaptive Thresholding



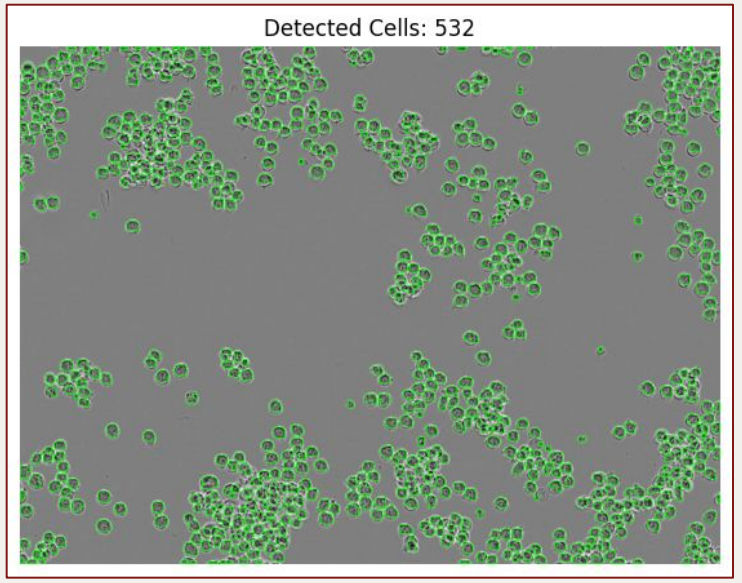
Results



Original

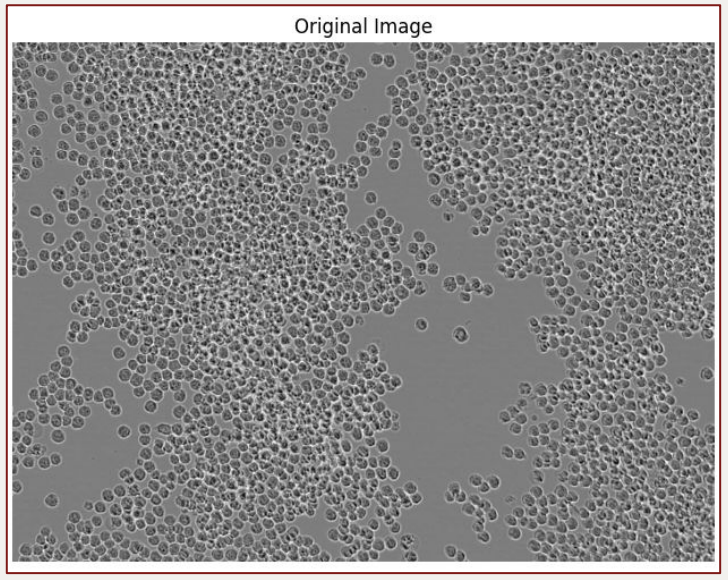


From Approach 1

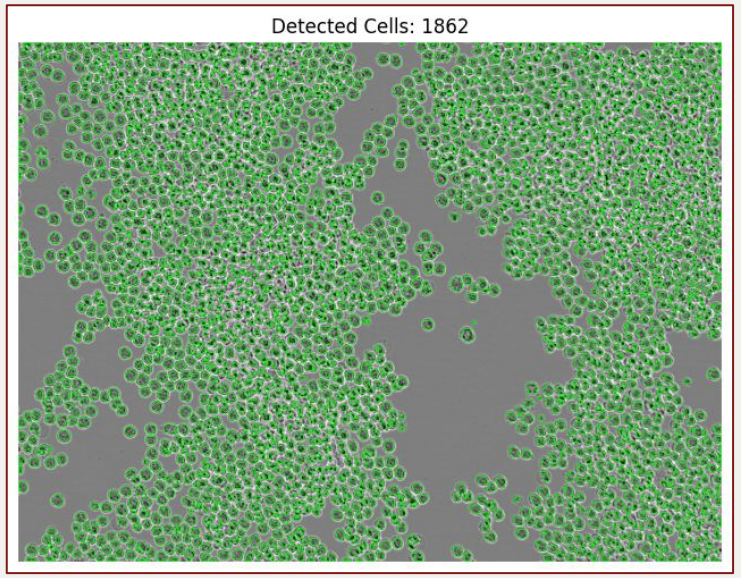


From Approach 2

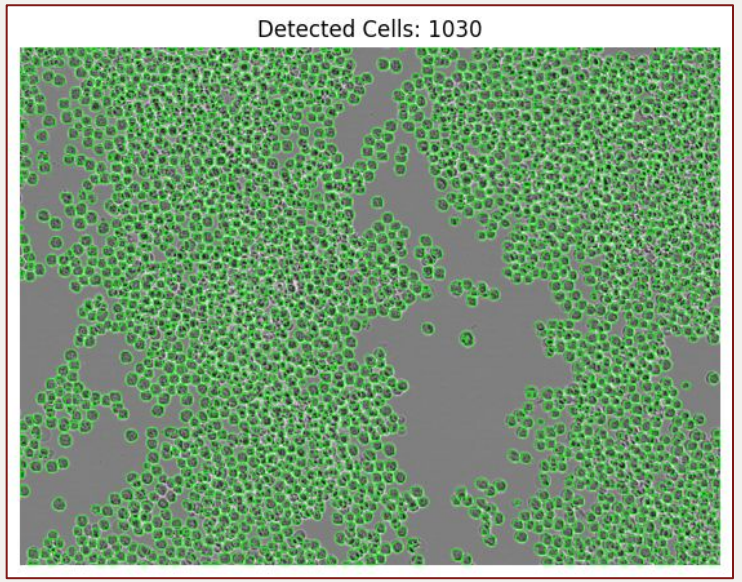
Results



Original



From Approach 1



From Approach 2

Conclusion

- Automated cell detection using traditional image processing.
- Good results despite low contrast and overlapping cells.
- Adaptive Thresholding performed better than Otsu’s.
- Gaussian blur and morphological operations improved segmentation.
- Classical methods were efficient, simple, and interpretable.

Next set of work

- To extend our analysis to all remaining cell types and prepare a model that can be trained and evaluated on the entire LIVECell dataset.
- Develop a conversion pipeline to transform the JSON annotation files (ground truth) into mask or label images for training and evaluation.
- Compare the segmented outputs with the provided ground truth annotations to calculate accuracy metrics such as IoU and precision.

References

- Edlund, C., Jackson, T.R., Khalid, N. et al. LIVECell—A large-scale dataset for label-free live cell segmentation. Nat Methods 18, 1038–1045 (2021). https://doi.org/10.1038/s41592-021-01249-6

- Senthilkumaran, N., & Vaithegi, S. (2016). Image segmentation by using thresholding techniques for medical images. *Computer Science & Engineering: An International Journal*, *6*(1), 1-13. https://d1wqtxts1xzle7.cloudfront.net/43439997/Image_segmentation_by_using_thresholding_techniques_for_medical_images