Pushing the Boundaries of Deep Learning in Cell Imaging: From Pixels to Understanding and Beyond

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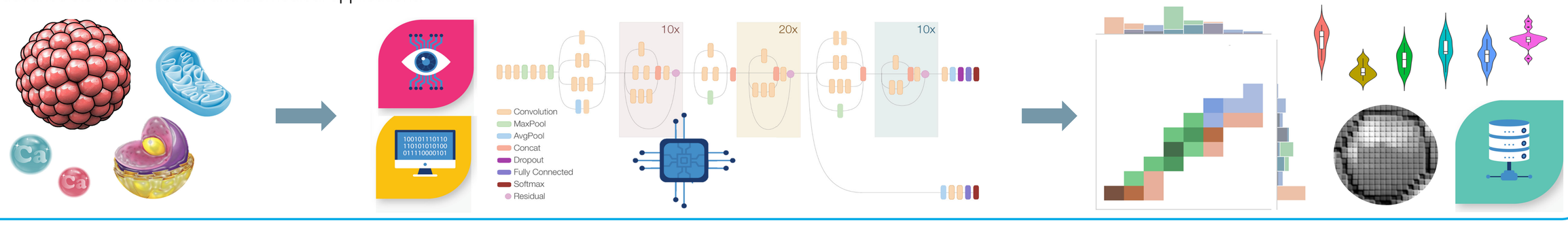
Summary: The integration of artificial intelligence (AI) into computational imaging analysis in cell biology has emerged as a transformative force, unveiling unprecedented insights and revolutionizing the way to understand the cellular structures and functions. Here, a series of computer vision and image processing tools have been adeptly employed to develop the advanced algorithms, with the aim to analyze the dynamic process of cell and substrate interaction across multi-, single- and sub-cellular levels. This work demonstrated the multitude of benefits of AI-based approach for cell imaging analysis, thereby accentuating its capacity to advance stem cell research and biomedical applications.



Computer vision and biological image processing tools

- An image moment is a specific weighted average moment of the pixel intensities within an image, or a function derived from these moments. These are often chosen for their desirable properties or interpretations [1]. Leveraging image moments, along with Gaussian blur, the Canny algorithm, and dilation and erosion functions within the OpenCV module in Python, we identified human induced pluripotent stem cell (hiPSC) colonies using various ways, including bounding boxes, convex hulls, and contours, as depicted in Figure 1.

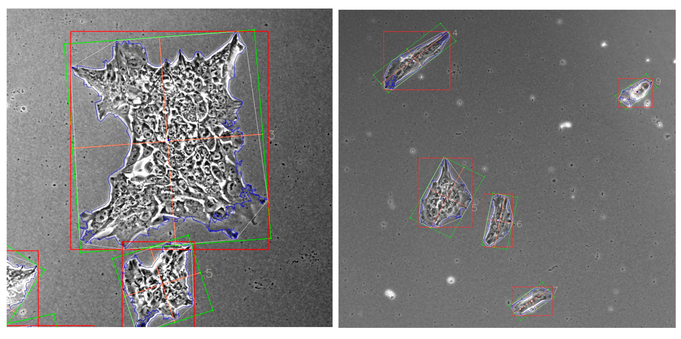


Figure 1. Recognition of human induced pluripotent stem cell (hiPSC) colonies using bounding boxes (in red and green), convex hulls (in white), and contours (in blue) imaging moment tools within the OpenCV module in Python.

- Changes in nuclear shape can be induced by various factors in extracellular environments, often associated with alterations in transcriptional regulation [2]. We introduced modifications to the extracellular environment by Polydopamine (PDA) coating and PDA film. We aimed to investigate whether these different PDA modifications could lead to changes in nuclear shape. Our results, as depicted in Figure 2, indicate that PDA film had the most pronounced effect in elongating the nuclei, suggesting a significant alteration in nuclear organization. This can result in changes in nuclear function.

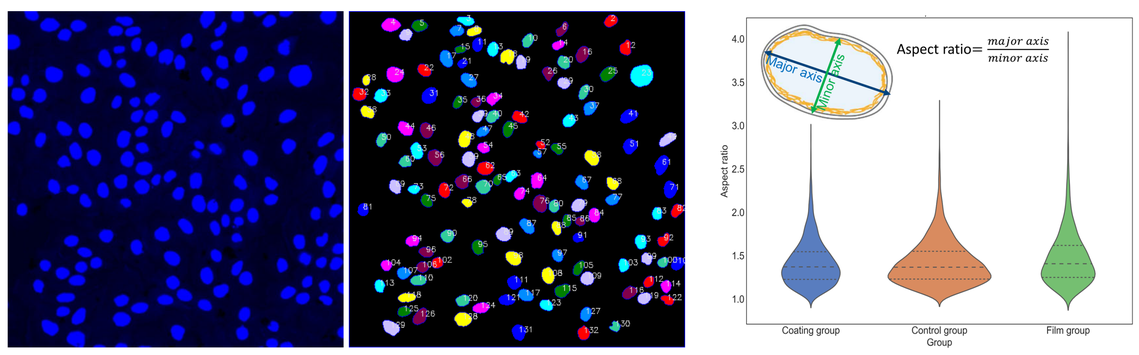


Figure 2. Precise detection of nuclei in human epidermal keratinocytes (HaCaT) from the Control, Coating, and Film groups for evaluating essential nucleus parameters, including aspect ratio.

- Numerous intracellular processes are known to be associated with calcium signaling. In our investigation, we aimed to determine whether PDA coating or PDA film could induce changes in intracellular calcium levels compared to cells not exposed to PDA. We assessed intracellular calcium using a fluorescence dye, as fluorescence intensity is positively correlated with calcium concentration [3]. Upon comparing the fluorescence intensity, we could discern relative changes in intracellular calcium concentration. For the film group, a pattern emerged in the mean fluorescence intensities of intracellular calcium in certain cells over time, showing an increase as time passed (Figure 3).

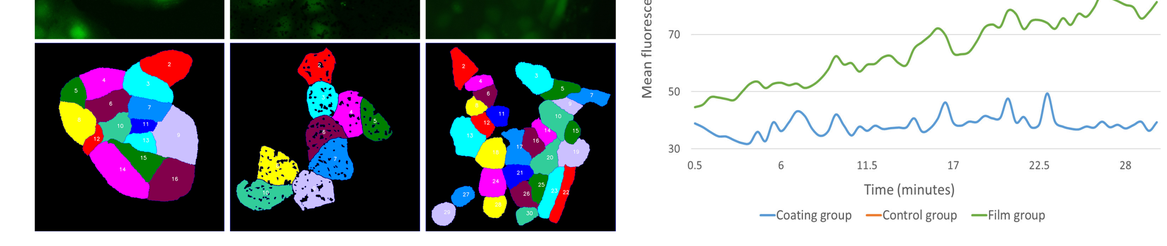


Figure 3. Calcium influx assay in skin keratinocytes cultured on different materials: A. Images depicting calcium influx in keratinocytes and the segmentation of cells within the colony. B. The curves illustrate the time course of calcium influx in keratinocytes cultured on different materials.

Deep convolutional neural network (DCNN) architectures

- This end-to-end data-driven approach employs a DCNN-based system for the detection of hiPSC in photomicrographs using transfer learning. The process started with the pre-processing of hiPSC images and the generation of ground truth data in JavaScript Object Notation format (JSON). The hiPSC dataset were then divided into training, validation, and testing sets. Subsequently, data augmentation techniques are applied to the training set images. To enhance performance, pre-trained weights from ImageNet are integrated into the Mask R-CNN architecture [4] through transfer learning, as illustrated in Figure 4. Finally, the model's performance is assessed using the testing sets.

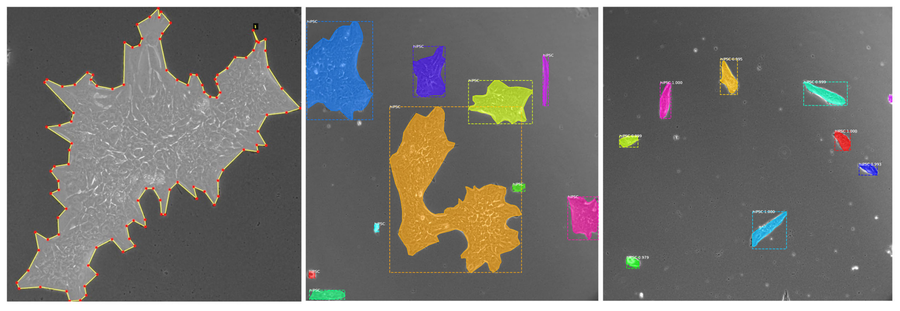


Figure 4. An image along with its corresponding Ground Truth used for fine-tuning pre-trained ImageNet weights in the Mask R-CNN architecture and performing instance segmentation of hiPSC on the testing set.

- Deep learning, which leverages data patterns for predictive purposes, holds the potential to revolutionize nearly every industry. One of the significant challenges in advancing deep learning methodologies is the availability of substantial, meticulously curated, and well-labeled datasets. The goal of this work is to design an efficient DCNN architecture for the identification of hiPSC-derived endothelial cells. To evaluate its effectiveness, the proposed model is compared with existing models (Figure 5), such as DenseNet [6].

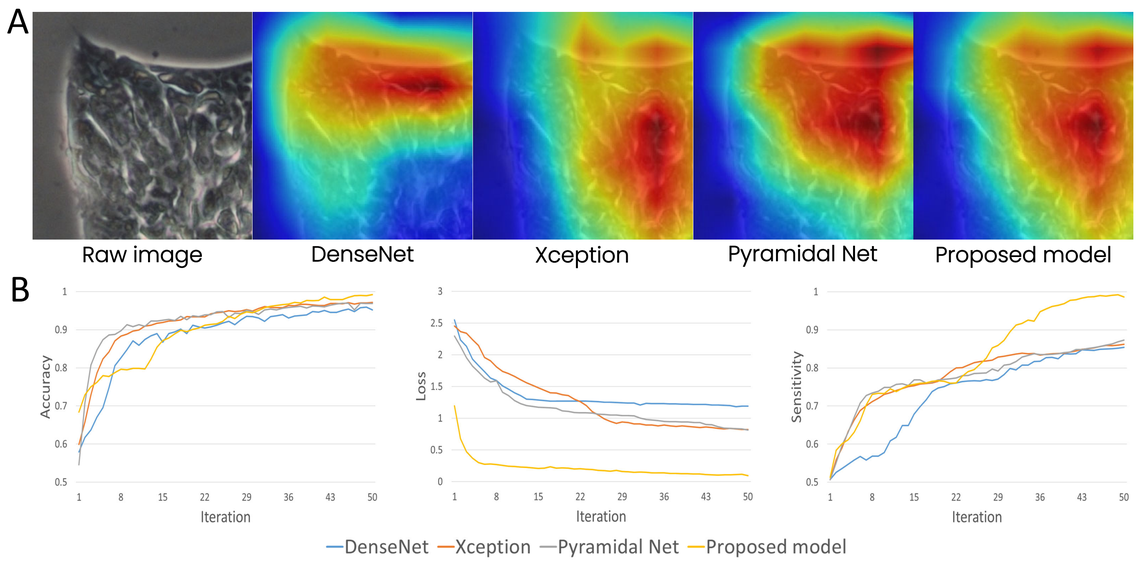


Figure 5. A. Visualization of significant regions (in red) and trivial regions (in blue) using global average pooling layer of several models. B. Comparison of accuracy, loss and sensitivity of different models.

- Mitochondria play pivotal roles in cellular processes, and their morphology and distribution offer valuable insights into cell function, health, and dynamics. Precise segmentation facilitates quantitative analysis of mitochondrial properties, encompassing size, shape, spatial organization, and cellular distribution. The dataset represents a 5×5×5 µm section extracted from the CA1 hippocampus region of the brain, resulting in a volume of 165×1024×768. It was acquired by Graham Knott at École polytechnique fédérale de Lausanne. The core motivation behind this work is the imperative need for accurate mitochondria segmentation (Figure 6), achieved through the U-Net.

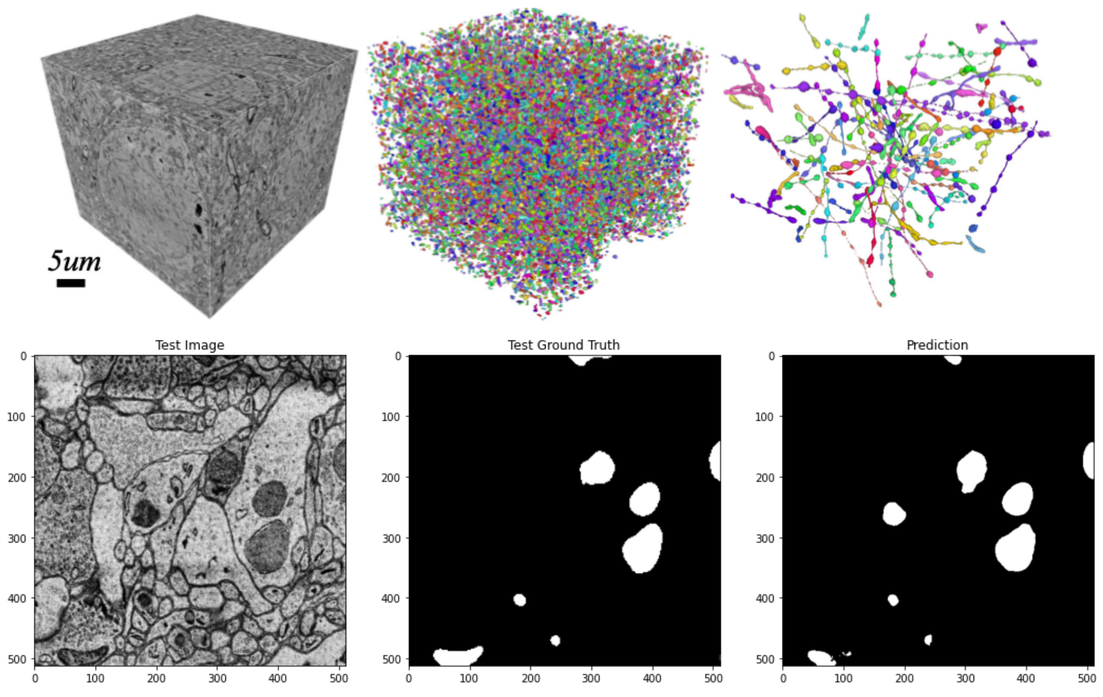


Figure 6. Typical images from the testing set of the Electron Microscopy datasets [5], along with the Ground Truth and the predictions generated by the U-Net DCNN architecture for mitochondria segmentation on the test set images.

References

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