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Nuclei Classification in Immunohistochemical Stainings for Tumor Microenvironment Analysis in Digital Pathology

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Tumor microenvironment (TME) is composed by the stromal cells surrounding cancers cells within a malignant tumor, including the immune system and the connective tissue. TME is being increasingly identified as an important factor in the dynamical behavior of a tumor. In histopathological imaging, the extraction of meaningful information describing the relationships between the tumor and its microenvironment relies on an accurate cell identification technique. In this work, we present an efficient approach for cell detection and classification from immunohistochemistry (IHC)-stained breast cancer tissue. The detected nuclei are classified in 3 types (cancer cells, fibroblasts and immune system cells) using Random Forest classifier based on morphologic, color and texture features.

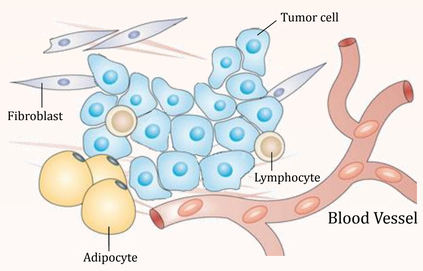
Keywords: Nuclei Classification, Breast Cancer, Tumor Microenvironment, Immunohistochemistry.

Introduction

Tumor Microenvironment (TME):

- Cellular environment in which a tumor develops.

Tumor cells ⟷ Complex interactions ⟷ Microenvironment cells



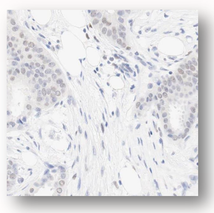
Characterization of heterotypic interactions from histopathology images?

➡ Cell detection and Classification

➡ Spatial Heterogeneity Analysis

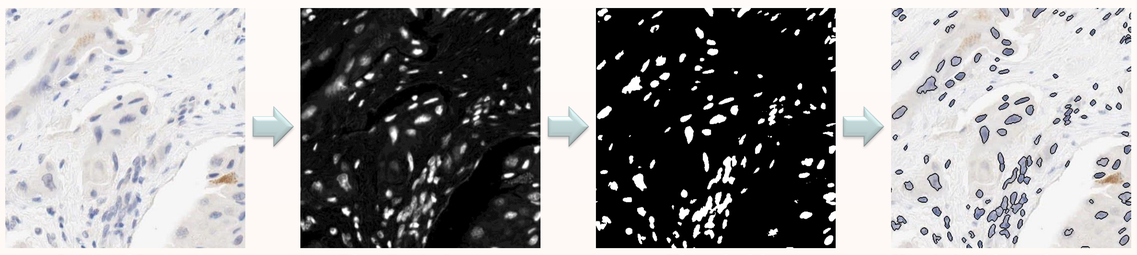
Data:

- Breast cancer slides stained with Phospho-Histone-H3 (PHH3) : Immunohistochemistry marker of mitotic cells and Haematoxylin counterstain
- 40 images (2000×2000 pixels) from 16 Whole Slide Images.
- 0.5µm/pixel resolution.



Method

❑ Nuclei Segmentation



Original Image

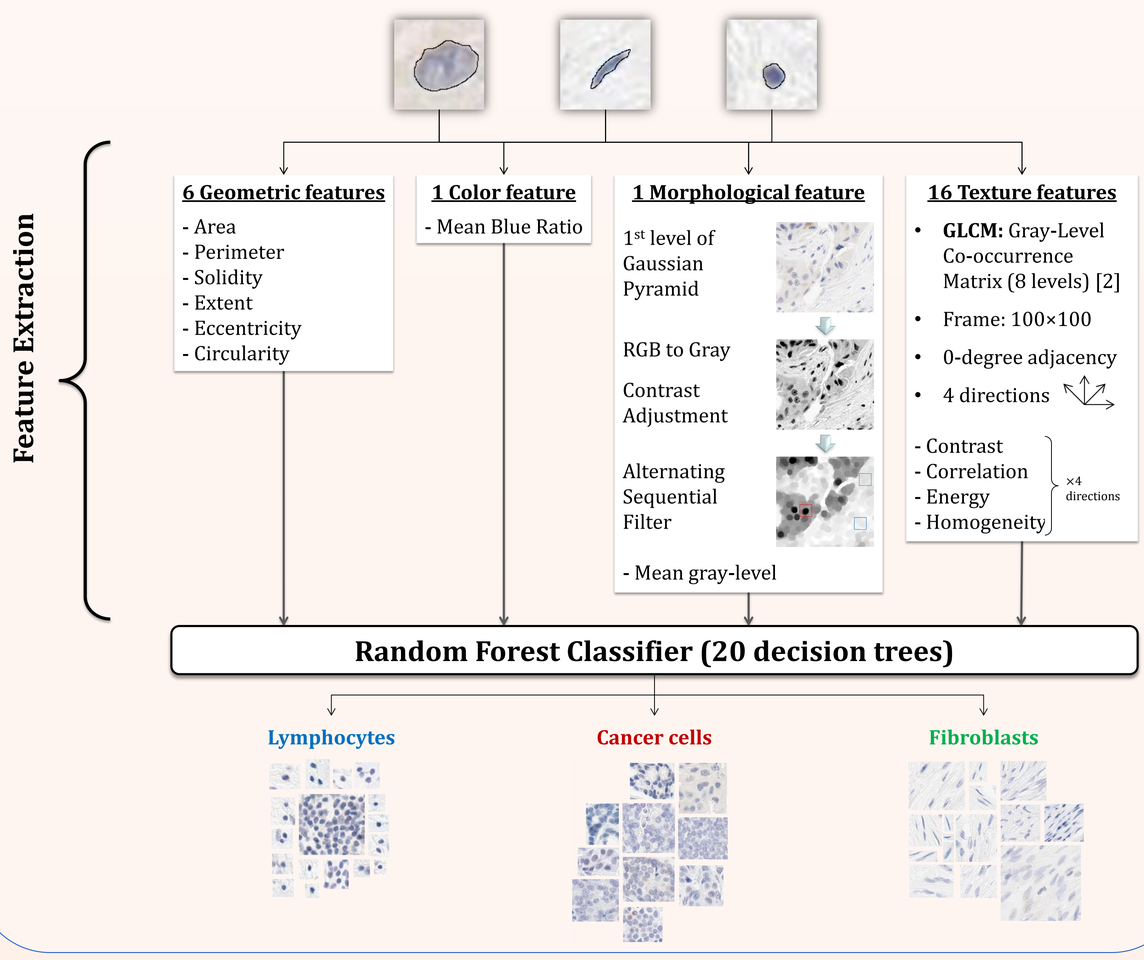
Blue Ratio Image

$$\frac{100 \times B}{(1+R+G)} \times \frac{256}{(1+R+G+B)}$$

Threshold (Otsu's method)

H-minima based marker-controlled Watershed [1] ($H = 0.85$)

❑ Nuclei Classification

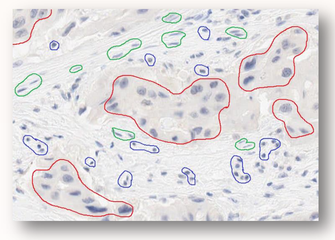


Results

❑ Ground Truth Generation

10154 nuclei manually labelled:

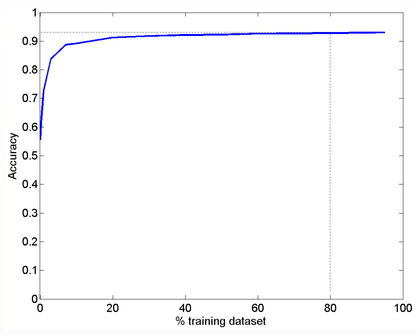
- 3332 Cancer Cell Nuclei (—)
- 3516 Fibroblasts (—)
- 3306 Lymphocytes (—)



- Nuclei that are detected inside the labeled regions are used for ground-truthing.
- The class of a nucleus = the class of its labeled region.

❑ Quantitative & Qualitative Results

Model performance vs training dataset

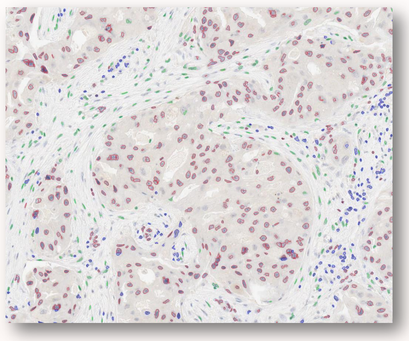


Confusion Matrix

Actual Class \ Predicted Class	Cancer Cell Nuclei	Fibroblasts	Lymphocytes
Cancer Cell Nuclei	647	26	14
Fibroblasts	29	625	36
Lymphocytes	21	46	587

Quantitative measures for nuclei classification [3]

Average Accuracy	Precision	Recall	F-score
$\frac{1}{3}\sum_{c=1}^{3}\frac{TP_c + TN_c}{TP_c + TN_c + FP_c + FN_c}$	$\frac{\sum_{c=1}^{3} TP_c}{\sum_{c=1}^{3} TP_c + FP_c}$	$\frac{\sum_{c=1}^{3} TP_c}{\sum_{c=1}^{3} TP_c + FN_c}$	$\frac{Precision \times Recall}{Precision + Recall}$
0.9418	**0.9132**	**0.9125**	**0.9129**



Example of nuclei segmentation and classification result

Conclusion

- This work presents an efficient approach for nuclei classification in IHC-stained histopathology images.
- Texture, color, morphology and geometry of nuclei were studied to extract meaningful features.
- The proposed algorithm has been tested on a large dataset of nuclei that were manually labeled.
- **Future works:** This result represents a fundamental part of a broader study dedicated to tumor heterogeneity, focusing in particular on spatial distribution quantification of the tumor microenvironment using graph theory and sparse sets' mathematical morphology.

References

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