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ASSESSING DIFFRACTIVE IMAGING FOR CYTOMETRY OF CELLS

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ABSTRACT

We consider the task of classifying cells as healthy or unhealthy from fluorescent microscopy images. We investigate the possibility to perform such a task in the conventional spatial domain or in a lens-less way via the Fraunhofer diffraction space. We demonstrate that despite the loss of phase in the spectral space, classification performances of images in spectral domain are of the same order of magnitude that the one obtained in the spatial domain.

Index Terms— Lens-less imaging, Fourier, cell classification.

1. INTRODUCTION

Lens-less imaging is a promising biomedical tool currently investigated for a large range of applications [1]. A specific use case of lens-less imaging consists in diffractive imaging where the modulus of the Fourier transform is directly captured as it corresponds to the Fraunhofer diffraction pattern. In this work, we propose to compare the sorting of 2D single cell images (healthy and unhealthy) in the spatial domain as recently illustrated in [2] to the modulus of the Fourier domain using textural feature spaces and a convolutional neural network (CNN).

2. METHODS

We applied the studied approaches on a set of real microscopy images of healthy and unhealthy (cancerous) cells. These images are 3D stacks of breast cell lines from multiple samples that were acquired using Aurox Clarity structured illumination/spinning disk laser-free confocal system in Imperial College of London and looking at mCherry labelled histone-H2B. 2D images were computed from 3D stacks by applying a sum z-projection transform. Finally, cells were segmented into single cell per image of 256×256 pixels. The modulus of the Fourier transform is computed then from the 2D images to simulate the diffractive imaging data set that would be produced with a lens-less imaging system. The total amount of single cell images is about 907 cancerous cell images and and 1007 healthy cell images for each studied imaging mode.

For cells classification in spatial and spectral spaces, we used the local binary pattern (LBP), the Haralick coefficient computed from gray-level co-occurence matrix (GLCM), the deep feature multi-scale method based on scattering transform convolution networks (scatnet) for textural feature extraction coupled with SVM classifier and also we included a CNN. Hyper-parameters of textural methods were selected as in [2] and a PCA was applied to reduce the number of features of LBP and scatnet to 14 equally to the GLCM. The architecture and hyper-parameters of CNN were optimized for both spatial and spectral images including regularisation by early stopping and random data augmentation with: rotation up to 180°, zoom in/out up to 20% and and brightness change up to 30% of original. Datasets were split into 80% for training and 20% for validation.

3. RESULTS

Table 1 shows the accuracy metric computed using 10-folds cross validation to evaluate classification performance. Globally, spectral space produces rather similar performance to spatial space when standard textural methods are used. The best results in spectral domain are obtained with the scattering transform. For CNN slightly higher classification performance for spatial and spectral spaces are observed with however a limited difference of few percent.

 Table 1. Classification results of used methods in both spatial and spectral domain.

| | Methods | | | |
|----------|---------|--------|---------|-------|
| Spaces | GLCM | LBP | Scatnet | CNN |
| Spatial | 73.9~% | 67.6~% | 73.3 % | 79.4% |
| Spectral | 73.6~% | 70.1 % | 74.1 % | 76.5% |

4. DISCUSSION AND CONCLUSION

The results demonstrated via simulation on real cells the possibility to use the Fraunhofer diffraction imaging mode to classify cells in healthy and unhealthy classes instead of using spatial space with rather limited loss of performance. This opens new directions for low-cost and fast cytometry performing cell sorting based on diffraction patterns without the need of close-up microscopic lenses.

5. REFERENCES

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