Abstract. Automatic classification of Human Epithelial Type-2 (HEp-2) cells staining patterns is an important and yet a challenging problem. Although both shallow and deep methods have been applied, the study of deep convolutional networks (CNNs) on this topic is shallow to date, thus failed to claim its top position for this problem. In this paper, we propose a novel study of using CNNs for HEp-2 cells classification focusing on cross-specimen analysis, a key evaluation for generalization. For the first time, our study reveals several key factors of using CNNs for HEp-2 cells classification. Our proposed system achieves state-of-the-art classification accuracy on public benchmark dataset. Comparative experiments on different training data reveals that adding different specimens, rather than increasing in numbers by affine transformations, helps to train a good deep model. This opens a new avenue for adopting deep CNNs to HEp-2 cells classification.

Keywords: Human Epithelial Type-2, Cell Classification, Deep Learning, Convolutional Architecture

1 Introduction

Indirect Immunofluorescence (IIF) is a commonly-used methodology to identify the presence of Anti-Nuclear Antibody (ANA) which facilitates the diagnosis of various autoimmune diseases. Until now, this diagnostic process is performed by specialists’ observing under a fluorescence microscope and it remains subjective and unreliable because it relies heavily on the experience and expertise of the physicians. Manual identification of these staining patterns suffers from intrinsic limitations related to visual evaluation performed by human\cite{3}. Thus, automatic classification with computer vision techniques has been increasingly demanded. During the past four years, various computer-aided systems have been designed with image analysis techniques in several classification challenges held by ICPR and ICIP\cite{4,2,10}. Current research in this topic is still in its early age and there is still room for improvement, especially for deep methods.

Due to the variations between different patients and the photo-bleaching effect caused by the light source\cite{14}, separated specimens in same patterns often have great variations between each other. As shown in Fig. 1, some different patterns can be easily misidentified (i.e., homogeneous vs speckled). Yet HEp-2
staining patterns recognition remains challenging. The existing shallow methods to address this problem mainly focus on three main separated aspects: handcrafting feature, (sparse) coding and classification. Each of those components has been well studied; for instance, the winner of ICPR 2014 Classification Contest utilized multiple types of local descriptors with multi-resolution combining with sparse coding and ensemble SVMs. For these systems, hand-crafted features in feature-extraction stage, as well as parameter selection in feature-coding stage, rely much on empirical selection.

Very recently, deep learning methods, such as CNNs, have been initially applied to HEp-2 cells classification. However, there are some key issues that have not been investigated: i) cross-specimen evaluation of CNNs model and ii) the key factors in adopting deep CNNs for cell image classification. It is important to note that the first issue is crucial to the evaluation of systems expected to be robust to different patients or specimens. Thus it is particularly valuable for HEp-2 cells computer-aided system.

In this paper, we proposed a deep convolutional architecture and evaluated our system on ICPR 2014 dataset Task-1. Our model is trained with extra images extracted from disjoint specimen dataset - ICPR 2014 Task-2 dataset. Experimental result shows the our framework is effective and outperforms several state-of-the-art methods. More importantly, we investigate further into the effects of different compositions of training set on deep model. We find that data augmentation by adding new specimens are much more powerful and efficient than by employing affine transformations alone, which was used in.

2 Classification Framework

2.1 Datasets and Experimental Protocols

Two public available dataset: Task-1 and Task-2 from I3A Contest were used in our experiments. Task-1 dataset contains 13,596 cell images extracted from 83 specimens, with cells categorised into six cell patterns namely Homogeneous,
Fig. 2: Extracting cells automatically from Task-2 dataset using ground truth bounding box (77 × 77). (a) A centromere specimen; (b) The segmentation mask for specimen in (a).

\[ \begin{array}{l|c|c|c} \text{Patterns} & \text{Task-1} & \text{Task-2} & \text{Task-2 augmented} \\ \hline \text{Homogenous} & 2494 & 11386 & 22772 \\ \text{Speckled} & 2831 & 11858 & 23716 \\ \text{Nucleolar} & 2598 & 9320 & 18640 \\ \text{Centromere} & 2741 & 10199 & 20398 \\ \text{NuMem} & 2208 & 4363 & 17452 \\ \text{Golgi} & 724 & 1501 & 12008 \\ \hline \text{Total} & 13596 & 48627 & 114986 \\ \end{array} \]

Table 1: Detailed compositions of Task-1, Task-2 and augmented Task-2 dataset.

"Speckled, Nucleolar, Centromere, Nuclear Membrane, and Golgi. Task-2 dataset contains 237 specimens images from the six patterns. As shown in Fig. 2, a simple automatic procedure was used to segment cells from each specimen given the segmentation mask in the dataset. We extracted 48,627 cell images from Task-2 dataset which were then used as an extra training set for deep model. For comparative study, we employed data augmentation on Task-2. Specifically, the number of images from first four patterns were doubled by rotating 90 degrees, while images from the last two patterns named NuMem and Golgi were rotated by 90, 180, 270 degrees and transposed respectively to generate eight times larger data, considering the last two patterns has relatively less images.

Leave-one-specimen-out (LOSO) is an important setting to test the generalization performance of a computer-aided system [13,12]. In this paper, we adopt this setting in our evaluation. In the LOSO strategy, each time all cell images from one of the 83 specimens are used for testing, the rest combined with the data from Task 2 or augmented Task 2 are used for training."
(MCA) is used as evaluation metric based on the 83 splits, which is defined as

\[
MCA = \frac{1}{K} \sum_{k=1}^{K} CCR_k
\]

where \( CCR_k \) is the correct classification rate for class \( k \) and \( K \) is the total number of classes. This metric was required by the I3A HEp-2 classification contest.

### 2.2 Deep Convolutional Architecture

Our deep CNNs share the basic architecture of the recent works\[15,9\]. Specifically, it contains ten layers as illustrated in Figure 3. Convolutional layers with 1 × 1 kernel size are heavily used in our model to increase the depth. The network is initialized with small random numbers (around 0.001). Rectified linear units (ReLU) are employed as activation functions after convolutional layers. The model is trained with batch size of 200 and 50 epoches, and the learning rate is set to 0.002. These settings are deliberately kept fixed for all experiments to observe effects of changes in the training data. In this work, we use MatConvNet\[16\] library to build up deep convolutional architecture. Usually, normalization step is not an essential part of CNNs as the networks have capacity to handle variations. Therefore, we did not implement normalization on the data. Each image was resized to 60 × 60 to guarantee a uniform scale of all the images used for training. We augment the training set in two ways: 1) by affine transformations, e.g. rotations; 2) by importing extra data from Task-2 dataset.

To classify a test image, firstly the image is resized to 60 × 60. Then the image is forward-propagated through the network, and the score of this cell for each class is obtained. To further improve the robustness of our system, we select the last three epoches, that is, the 48th, 49th, and 50th epoch to jointly classify a cell image. The predicted label is the one with the maximum output score averaged over the three votes.

### 3 Comparison with State of the Arts

The general performance of our system was evaluated across different specimens. Experiments were conducted under leave-one-specimen-out setting shown in Section 2.1.
A pooled set of 48,627 images from Task-2 and training set of Task-1 were used for training. Then a mean-class-accuracy of 82.90% was obtained. We further employed data augmentation on Task-2 by rotations illustrated in Section 2.1. By training with a pooled set of 114,986 images and training set of Task-1, a slightly better classification accuracy of 83.55% was obtained. Table 2 reported the confusion matrix. The Golgi class had poor results (70.30%). We further compare our approach to several state-of-the-art systems: 1) Shape index histograms with donut-shaped spatial pooling[8]; 2) multi-resolution patterns combined with ensemble SVMs (the winner of ICPR14 contest)[12]; and 3) Gaussian scale theory with improved Fisher Vector[13]. These methods were top performers on Task-1 dataset.

| Patterns    | Homogenous | Speckled | Nucleolar | Centromere | NuMem | Golgi |
|-------------|------------|----------|-----------|------------|-------|-------|
| Homogenous  | 85.93      | 8.02     | 0.20      | 0          | 3.65  | 2.21  |
| Speckled    | 9.71       | 79.97    | 3.50      | 5.58       | 1.24  | 0     |
| Nucleolar   | 2.19       | 3.66     | 85.84     | 6.66       | 0.92  | 0.73  |
| Centromere  | 0.04       | 10.58    | 4.16      | 84.93      | 0.11  | 0.18  |
| NuMem       | 1.45       | 1.18     | 0.68      | 0.09       | 94.34 | 2.26  |
| Golgi       | 5.11       | 5.52     | 13.54     | 0.97       | 4.56  | 70.30 |

Table 2: Confusion matrix under Task-1 under leave-one-specimen-out protocol

| Methods                                             | Mean Class Accuracy |
|-----------------------------------------------------|---------------------|
| Shape index histograms with donut-shaped spatial pooling[8] | 78.70%              |
| Multi-resolution patterns with Ensemble SVMs[12]     | 81.10%              |
| Gaussian scale theory with improved Fisher Vector[13] | 82.03%              |
| Our proposed method (Task-1 + Task-2)               | 82.90%              |
| Our proposed method (Task-1 + Task-2 augmented)     | 83.55%              |

Table 3: Comparisons with state-of-the-art

As shown in Table 3, our proposed approach slightly outperforms three state-of-art systems. Different from our framework, the above methods use Task-1 dataset only. This indicates deep CNNs requires large dataset with sufficient specimens for training. And deep convolutional neural network is competitive compared to systems based on hand-crafted features on HEp-2 classification problem.

### 4 Comparative Study on Data Augmentation

Data augmentation is a key step for training CNN when data is limited. This is also the case of recent study of using CNN for HEp-2 cell classification. However, only affine transformations for argumentations was ever considered in this problem. In this section we investigate the effect of argumentations by adding specimens for training deep convolutional model on HEp-2 cells classification problem. Then we show that CNNs is superior when sufficient specimens are available.
We use the datasets containing 1) the data extracted from Task-2 dataset and 2) training set in Task-1 for testing. In total, three group of experiments were designed to explore the effects of additional specimens under leave-one-specimen-out evaluation: 1) set-1: only Task-1 dataset was used; 2) set-2: data augmentation based on Task-1 only; 3) Set-3: data augmentation with additional 237 specimens from Task-2. In building set-2, each image in Task-1 was rotated by angle 0°, 90°, 180° and 270° and transposed respectively to generate new images. The size of set-2 is eight times larger than Task-1. In building set-3, we used Task-1 dataset and additional 237 specimens from Task-2. For each specimens, 41 cells images were randomly selected within each specimen. Thus we obtained a dataset with size similar to Task-1 but more specimens contained.

| Patterns      | set-1 | set-2 | set-3 |
|---------------|-------|-------|-------|
| Homogenous    | 2494  | 19952 | 2629  |
| Speckled      | 2831  | 22648 | 2788  |
| Nucleolar     | 2598  | 20784 | 2706  |
| Centromere    | 2741  | 21928 | 2747  |
| NuMem         | 2208  | 17664 | 1476  |
| Golgi         | 724   | 5792  | 574   |
| Total         | 13596 | 108768| 13120 |

Table 4: Detailed compositions of data set for three experiments

By comparing the results on set-1 and set-2, we see that model trained with data augmentation outperforms the former one by 4.15%, demonstrating that data augmentation is necessary and effective for small training sets in CNN frameworks. Notably, the model trained on set-3 with less images but more specimens than set-1, outperforms one trained on set-2, which used eight times larger dataset. It indicates that data augmentation by adding new specimens are more powerful and more efficient than by employing affine transformations alone. And employing affine transformations alone has limited impact on the overall performance.

In previous CNN-based framework [5], data augmentation was based on affine transformations. It was shown effective under k-fold cross-validation protocol. However, under k-fold cross-validation protocol, the identities of specimen from which cells were extracted were totally disregarded. As one can observe from each specimen, the cells inside have little variation but positions and orientations, which causes that the training set and test set are highly similar. The task was artificially made easier in this setting. The mean-class-accuracy of our CNNs under this setting is 91.16% without data augmentation. Therefore, one system achieving high recognition accuracy under k-fold cross-validation may achieve poor performance under cross-specimen test.

5 Conclusions

In this work, we presented a detailed study of a deep convolutional architecture to address HEp-2 cell classification problem. We evaluated our systems in cross-specimen experiments. Experimental results show that the proposed system outperforms several state-of-the-art frameworks. In particular, we investigated the effect of different data
Table 5: Confusion matrix of results on set-1. The mean class accuracy obtained is 70.52%.

| Patterns     | Homogenous | Specckled | Nucleolar | Centromere | NuMem | Golgi |
|--------------|------------|-----------|-----------|------------|-------|-------|
| Homogenous   | 70.89      | 20.49     | 1.04      | 0.12       | 4.13  | 3.32  |
| Specckled    | 16.43      | 63.55     | 5.55      | 12.61      | 1.77  | 0.11  |
| Nucleolar    | 3.00       | 8.89      | 75.25     | 4.77       | 6.85  | 1.23  |
| Centromere   | 0.04       | 15.18     | 6.31      | 78.33      | 0.11  | 0.04  |
| NuMem        | 8.92       | 3.53      | 1.04      | 0.09       | 82.20 | 4.21  |
| Golgi        | 7.04       | 0.97      | 22.24     | 1.38       | 15.46 | 52.90 |

Table 6: Confusion matrix of results on set-2. The mean class accuracy obtained is 74.67%.

| Patterns     | Homogenous | Specckled | Nucleolar | Centromere | NuMem | Golgi |
|--------------|------------|-----------|-----------|------------|-------|-------|
| Homogenous   | 66.00      | 20.89     | 3.49      | 0.48       | 7.69  | 1.44  |
| Specckled    | 10.73      | 67.64     | 10.56     | 4.31       | 7.12  | 2.38  |
| Nucleolar    | 1.23       | 7.31      | 77.64     | 4.31       | 7.12  | 2.38  |
| Centromere   | 0.18       | 11.60     | 2.37      | 84.86      | 0.04  | 0.95  |
| NuMem        | 5.84       | 4.39      | 2.26      | 0.18       | 85.33 | 1.99  |
| Golgi        | 2.07       | 0.97      | 23.20     | 1.65       | 5.52  | 66.57 |

Table 7: Confusion matrix of set-3. The mean class accuracy obtained is 79.13%.

| Patterns     | Homogenous | Specckled | Nucleolar | Centromere | NuMem | Golgi |
|--------------|------------|-----------|-----------|------------|-------|-------|
| Homogenous   | 86.16      | 9.06      | 0.56      | 0          | 4.21  | 0     |
| Specckled    | 12.86      | 70.96     | 8.72      | 5.30       | 2.01  | 0.14  |
| Nucleolar    | 2.61       | 3.27      | 86.87     | 3.23       | 2.54  | 1.46  |
| Centromere   | 0          | 10.94     | 4.85      | 85.62      | 0.36  | 0.22  |
| NuMem        | 5.48       | 3.13      | 0.68      | 0.14       | 85.42 | 5.16  |
| Golgi        | 11.88      | 4.14      | 7.73      | 1.10       | 13.39 | 61.74 |
augmentation strategies. Our results show that the key factor for training a good deep model is by adding sufficient specimens rather by affine transformation, which could calibrate further development of using CNN in classifying HEp-2 cell images.

References

1. Bayramoglu, N., Kannala, J., Heikkila, J.: Human epithelial type 2 cell classification with convolutional neural networks. In: Bioinformatics and Bioengineering (BIBE), 2015 IEEE 15th International Conference on. pp. 1–6. IEEE (2015)
2. Foggia, P., Percannella, G., Saggese, A., Vento, M.: Pattern recognition in stained hep-2 cells: Where are we now? Pattern Recognition 47(7), 2305–2314 (2014)
3. Foggia, P., Percannella, G., Soda, P., Vento, M.: Early experiences in mitotic cells recognition on hep-2 slides. In: Computer-based medical systems (CBMS), 2010 IEEE 23rd international symposium on. pp. 38–43. IEEE (2010)
4. Foggia, P., Percannella, G., Soda, P., Vento, M.: Benchmarking hep-2 cells classification methods. Medical Imaging, IEEE Transactions on 32(10), 1878–1889 (2013)
5. Gao, Z., Wang, L., Zhou, L., Zhang, J.: Hep-2 cell image classification with deep convolutional neural networks. arXiv preprint arXiv:1504.02531 (2015)
6. Hobson, P., Percannella, G., Vento, M., Wiliem, A.: Competition on cells classification by fluorescent image analysis. In: Proc. 20th IEEE Int. Conf. Image Process.(ICIP). pp. 2–9 (2013)
7. Krizhevsky, A., Sutskever, I., Hinton, G.E.: Imagenet classification with deep convolutional neural networks. In: Advances in neural information processing systems. pp. 1097–1105 (2012)
8. Larsen, A.B.L., Vestergaard, J.S., Larsen, R.: Hep-2 cell classification using shape index histograms with donut-shaped spatial pooling. Medical Imaging, IEEE Transactions on 33(7), 1573–1580 (2014)
9. Lin, M., Chen, Q., Yan, S.: Network in network. arXiv preprint arXiv:1312.4400 (2013)
10. Lovell, B.C., Percannella, G., Vento, M., Wiliem, A.: Performance evaluation of indirect immunofluorescence image analysis systems, ICPR 2014 (2014)
11. Manivannan, S., Li, W., Akbar, S., Wang, R., Zhang, J., McKenna, S.J.: Hep-2 cell classification using multi-resolution local patterns and ensemble svms. In: Pattern Recognition Techniques for Indirect Immunofluorescence Images (I3A), 2014 1st Workshop on. pp. 37–40. IEEE (2014)
12. Manivannan, S., Li, W., Akbar, S., Wang, R., Zhang, J., McKenna, S.J.: An automated pattern recognition system for classifying indirect immunofluorescence images of hep-2 cells and specimens. Pattern Recognition 51, 12–26 (2016)
13. Qi, X., Zhao, G., Chen, J., Pietikäinen, M.: Hep-2 cell classification: The role of gaussian scale space theory as a pre-processing approach. Pattern Recognition Letters (2015)
14. Song, L., Hemminki, E., Young, I.T., Tanke, H.J.: Photobleaching kinetics of fluorescein in quantitative fluorescence microscopy. Biophysical journal 68(6), 2588 (1995)
15. Szegedy, C., Liu, W., Jia, Y., Sermanet, P., Reed, S., Anguelov, D., Erhan, D., Vanhoucke, V., Rabinovich, A.: Going deeper with convolutions. In: CVPR 2015 (2015) [http://arxiv.org/abs/1409.4842]
16. Vedaldi, A., Lenc, K.: Matconvnet: Convolutional neural networks for matlab. In: Proceedings of the 23rd Annual ACM Conference on Multimedia Conference. pp. 689–692. ACM (2015)