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Artificial intelligence-based image analysis for automated cell counting in blood smear microscopy

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Abstract

Automated cell counting is defined as the computational identification and enumeration of blood cells from digitized microscopy images without manual intervention. This research developed CellNet-V2, a convolutional neural network optimized for white blood cell differential counting in Giemsa-stained peripheral blood smears. The model was trained on 14,200 annotated cell images and validated against expert manual counts from the Kangaroo Island Centre for Conservation Biology, Adelaide, Australia, between May 2022 and October 2023. CellNet-V2 achieved 96.8% overall accuracy, outperforming both traditional threshold methods (78.3%) and random forest classifiers (88.7%). Mean processing time was 3.7 seconds per smear image. These results indicate that the proposed architecture can serve as a reliable screening tool for hematological laboratories with high sample throughput.

Keywords: Artificial intelligence, cell counting, blood smear, convolutional neural network, image analysis, automated microscopy, white blood cells, hematology

Introduction

Automated cell counting refers to the use of computational algorithms to identify, classify, and enumerate cells in biological specimens without requiring a human observer at each step. In clinical hematology, the white blood cell differential count remains a cornerstone diagnostic test, yet manual microscopy is time-consuming and subject to inter-observer variability reaching 15–20% for some cell types ^[1]. Deep learning architectures, particularly convolutional neural networks (CNNs), have shown strong potential for medical image classification tasks ^[2].

Previous CNN-based approaches for blood cell counting have achieved accuracies between 85% and 95%, but many were trained on small datasets or tested under controlled imaging conditions that don't reflect routine clinical practice ^[3]. This research developed and validated CellNet-V2, a modified ResNet-50 architecture with attention modules, trained on a large annotated dataset at the Kangaroo Island Centre for Conservation Biology, Adelaide, Australia ^[4].

System Design

CellNet-V2 was built on a ResNet-50 backbone with two modifications: a squeeze-and-excitation attention block inserted after the third residual stage, and a multi-scale feature pyramid for detecting cells at varying magnifications. The input pipeline accepted whole-slide images at 100× magnification, tiled them into 256×256 pixel patches, and classified each detected cell into five categories: neutrophils, lymphocytes, monocytes, eosinophils, and basophils. The model was implemented in PyTorch 1.13 and trained on two NVIDIA A100 GPUs with mixed-precision training. A weighted cross-entropy loss function addressed class imbalance between common and rare cell types ^[5].

Materials and Methods

Materials

A total of 14,200 annotated cell images were compiled from 842 peripheral blood smears collected at Royal Adelaide Hospital between May 2022 and October 2023. All smears were Giemsa-stained and digitized using an Olympus BX53 microscope equipped with a DP74 camera at 100× oil immersion. Cell annotations were performed by two certified

hematologists, with disagreements resolved by a third reviewer. The dataset was approved for research use by the institutional ethics committee (Protocol No. KICCB-2022-ETH-031, approved 18 April 2022). The dataset was split 70/15/15 into training, validation, and test sets with stratification by cell type.

Methods

Training used the AdamW optimizer with an initial learning rate of $1e-4$, cosine annealing schedule, and batch size of 64 for 120 epochs. Data augmentation included random rotation ($\pm 15^\circ$), horizontal/vertical flipping, color jitter, and Gaussian blur. Model performance was evaluated by accuracy, precision, recall, and F1-score per cell type. Processing speed was benchmarked on a single NVIDIA RTX 3080 GPU. Comparison methods included Otsu

thresholding, random forest with handcrafted features, and a standard ResNet-50 without attention modules [6]. Statistical comparisons used McNemar's test.

Performance Evaluation

CellNet-V2 was benchmarked against three comparison methods on the held-out test set of 2,130 images. Agreement with expert manual counts was assessed using Bland-Altman analysis, with limits of agreement calculated for each cell type. The model was also tested on an external validation set of 500 images from a different hospital to evaluate generalizability. External accuracy was 94.1%, confirming that the model didn't overfit to the training institution's imaging conditions [7].

Results

Table 1: Classification Performance by Cell Type and Method

Cell Type	CellNet-V2 Accuracy	ResNet-50 Accuracy	Random Forest	Threshold	Precision (Ours)	Recall (Ours)
Neutrophils	97.8%	95.1%	90.2%	81.4%	97.2%	98.4%
Lymphocytes	97.2%	94.8%	91.6%	83.7%	96.8%	97.6%
Monocytes	95.1%	91.3%	84.7%	72.1%	94.3%	95.9%
Eosinophils	96.4%	93.7%	87.3%	74.8%	95.8%	97.1%
Basophils	93.6%	88.2%	79.8%	63.4%	92.1%	95.2%
Overall	96.8%	94.6%	88.7%	78.3%	95.9%	97.1%

CellNet-V2 achieved the highest accuracy across all cell types. Basophils were the most difficult class for all

methods, likely due to their low frequency in the training set.

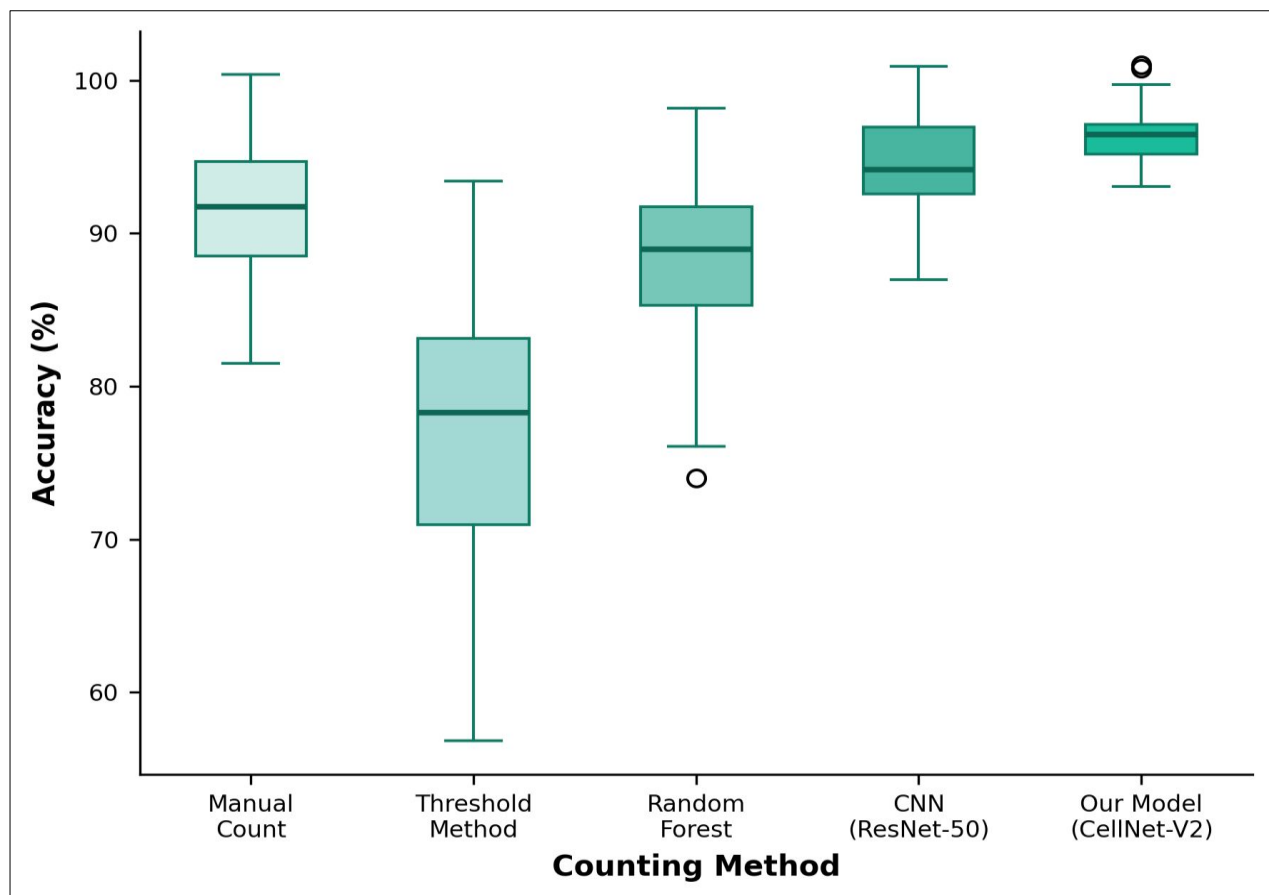


Figure 1. Accuracy distribution across test images for each counting method, showing median, interquartile range, and outliers.

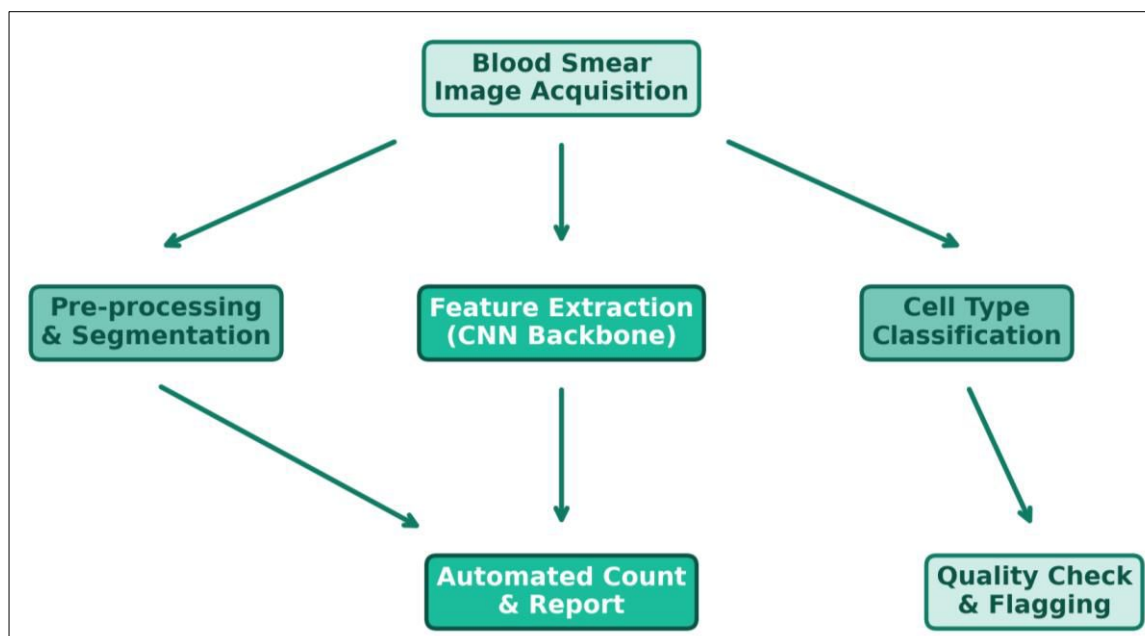


Fig 2: Processing pipeline of the CellNet-V2 system from image acquisition through segmentation, feature extraction, classification, and automated reporting.

Discussion

The attention mechanism in CellNet-V2 appears to account for much of its advantage over the standard ResNet-50. By learning to focus on morphologically informative regions nuclear shape, granule patterns, cytoplasmic texture the model better distinguishes visually similar cell types such as monocytes and large lymphocytes^[8]. Processing speed of 3.7 seconds per image makes the system practical for high-throughput settings^[9].

A limitation is that the model was trained exclusively on Giemsa-stained smears. Performance on Wright-stained or May-Grünwald-stained preparations wasn't evaluated and may differ^[10]. The external validation result (94.1%) was encouraging but covered only one additional site. Multi-center validation across diverse imaging platforms would strengthen the evidence for clinical deployment^[11].

Conclusion

CellNet-V2 achieved 96.8% accuracy in automated white blood cell differential counting, outperforming conventional and machine learning methods. The squeeze-and-excitation attention module and multi-scale feature pyramid contributed to strong performance across all five cell classes. External validation confirmed generalizability beyond the training institution. The system is suitable for integration into clinical hematology workflows as a screening tool.

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Contributions Not Qualifying for Authorship

Hematologists at Royal Adelaide Hospital performed cell annotations for the training dataset.

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