Nuclei Image Segmentation using Deep Learning

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Abstract—**Medical image segmentation is an important step in medical image analysis, especially as a crucial prerequisite for efficient disease diagnosis and treatment. In this project we have used VM-UNet, a novel medical image segmentation architecture that integrates Vision Mamba blocks within a U-Net framework for enhanced feature extraction in nuclei segmentation tasks. The proposed model addresses the limitations of traditional convolutional neural networks by incorporating selective state space models for better long-range dependency modeling. We evaluate VM-UNet on the MonuSeg-2018 KMMS (Karyotype Microscopy Medical Segmentation) dataset, comparing it against baseline segmentation approaches. The architecture includes specialized components such as VMBlocks with residual connections, comprehensive data augmentation strategies, and a combined loss function integrating Dice and Focal losses. Experimental results show that VM-UNet effectively balances computational efficiency with segmentation accuracy, making it suitable for medical imaging applications where precise boundary detection is critical..**

Index Terms—**Medical Image Segmentation, Vision Mamba, U-Net, Nuclei Segmentation, Deep Learning, Computer Vision**

1. Introduction

With the continuous development of medical imaging technology, medical images have become essential for disease diagnosis and treatment planning. Medical image segmentation remains a fundamental and critical challenge in computer-aided diagnosis, particularly in cytology and histopathology, where accurate identification and segmentation of cell nuclei are essential for reliable disease diagnosis and prognosis. Precise nuclei segmentation directly impacts treatment planning and patient outcomes, as it enables quantification and analysis of cellular structures such as nuclear morphology, size, and spatial distribution, which are key biomarkers for conditions ranging from inflammation to cancer. This process is foundational for subsequent tasks like cell classification, grading of tumors, and tracking disease progression. Despite advances in imaging technologies and computational methods, medical images often present heterogeneous backgrounds, overlapping nuclei, variable staining intensities, and low-contrast boundaries, which make automated segmentation difficult. These challenges are compounded by the vast diversity of tissue types, imaging modalities, and staining protocols used across different clinical and research settings. Consequently, the manual segmentation performed by pathologists is not only time-consuming and labor-intensive but also subject to inter-observer variability, highlighting the pressing need for consistent and efficient automated solutions. Developing robust and efficient deep learning-based segmentation techniques is therefore crucial to enhance accuracy, reproducibility, and efficiency in clinical decision-making, paving the way for more personalized and data-driven healthcare. Traditional segmentation methods, such as thresholding, region-growing, and watershed algorithms, often struggle with the inherent complexities of medical images, as they rely on handcrafted features and simplistic assumptions about intensity and texture. Recent advances in deep learning, particularly convolutional neural networks (CNNs), have significantly improved segmentation accuracy by learning hierarchical feature representations directly from data. Among these, U-Net [1] has emerged as a standard architecture for biomedical image segmentation, renowned for its symmetric encoder-decoder structure and skip connections that effectively capture both local features and broader contextual information. Its success has spurred a wave of innovations and architectural variants designed to further tackle the nuanced difficulties of medical image analysis.

Despite their success, CNN-based approaches face inherent limitations in capturing long-range dependencies due to their reliance on local receptive fields and fixed-size convolutional kernels, which restrict the ability to model global contextual relationships across an image. Although deeper networks and larger kernels can partially mitigate this issue, they often introduce higher computational costs and still struggle to effectively capture long-range spatial dependencies, a challenge that becomes especially critical in complex image segmentation tasks such as medical image segmentation where global anatomical consistency is essential. To overcome these limitations, recent developments in state space models, particularly Mamba [2], have introduced architectures capable of linear-time sequence modeling with global receptive fields, enabling efficient handling of long sequences without the quadratic complexity associated with attention-based methods. These models allow selective propagation and retention of relevant information across long spatial sequences, leading to improved contextual understanding and more stable learning. Vision Mamba further adapts these sequence modeling capabilities specifically for visual data by transforming spatial features into sequential representations while preserving important local structures, thereby enabling efficient and scalable processing of high-resolution images. By integrating such models into segmentation frameworks, it becomes possible to effectively address the combined challenges of local feature extraction and global context modeling, ultimately improving accuracy and robustness in medical image segmentation.

This paper introduces VM-UNet, a novel hybrid architecture that integrates Vision Mamba blocks within the traditional U-Net framework, aiming to simultaneously leverage the strengths of local feature extraction and global context modeling for medical image segmentation tasks. By combining convolutional operations with selective state-space modeling, VM-UNet is designed to capture both fine-grained spatial details and long-range dependencies, which are often challenging for standard CNN-based segmentation networks. The primary contributions of this work are threefold. First, we propose a novel VM-UNet architecture that incorporates Vision Mamba blocks at multiple encoding and decoding stages, enhancing the network's ability to learn complex representations of nuclei structures in histopathology images. Second, we develop a comprehensive data augmentation pipeline, including geometric transformations, intensity adjustments, and random cropping, to improve the model's robustness and generalization on limited and heterogeneous datasets. Third, we provide a thorough evaluation of VM-UNet on the KMMS dataset, employing balanced splitting strategies and a combination of Dice and Focal loss functions, to demonstrate the model's effectiveness and stability compared to baseline segmentation approaches.

2. Background and Related Work

2.1. Medical Image Segmentation

Medical image segmentation has progressively evolved from traditional image processing approaches, such as thresholding, edge detection, and region-growing methods, toward advanced deep learning–based techniques that are capable of automatically learning hierarchical and task-specific feature representations directly from data. Among these methods, U-Net [1] marked a major breakthrough in biomedical image segmentation by introducing an encoder–decoder architecture with symmetric skip connections, allowing the network to effectively combine high-level semantic information from deeper layers with fine-grained spatial details from shallower layers, thereby enabling accurate localization and boundary delineation. Building upon this foundational architecture, subsequent variants such as Attention U-Net [3] and UNet++ [4] further enhanced segmentation performance by incorporating attention mechanisms to selectively emphasize anatomically relevant regions and by introducing nested and densely connected architectures to better capture multi-scale contextual information. Collectively, these developments have significantly improved the ability of deep learning models to handle complex and heterogeneous medical images, including cases involving overlapping structures, variable contrast, and significant inter-sample variability, thus establishing deep learning as a dominant paradigm in modern medical image segmentation.

2.2. Vision Transformers and State Space Models

Vision Transformers (ViTs) [5] introduced self-attention mechanisms to computer vision, enabling explicit modeling of global dependencies across an image and achieving impressive performance on a wide range of vision tasks. However, this global attention mechanism comes with quadratic computational and memory complexity with respect to input size, which significantly limits scalability, especially for high-resolution images and dense prediction tasks. In contrast, State Space Models (SSMs), particularly Mamba [2], provide an alternative approach by enabling linear-time sequence modeling with selective information propagation, allowing efficient handling of long sequences while maintaining expressive power. These models dynamically control the flow of information across sequence elements, making them well-suited for capturing long-range dependencies without excessive computational overhead. Building on these advantages, Vision Mamba adapts SSM-based architectures specifically for visual tasks by restructuring spatial features into sequential representations, offering a computationally efficient and scalable alternative to transformer-based models for image analysis while preserving both global context and important local information.

2.3. Hybrid Architectures

Recent research has increasingly focused on hybrid architectures that aim to combine the strong local feature extraction capabilities of convolutional neural networks (CNNs) with the global context modeling power offered by attention mechanisms or transformer-based modules. Such hybrid designs seek to overcome the inherent locality of CNNs while avoiding the high computational cost typically associated with full self-attention. For instance, TransUNet [6] integrates transformer blocks into the widely used U-Net architecture, enabling effective modeling of long-range dependencies and significantly improving segmentation accuracy in medical imaging tasks. Similarly, Swin-UNet [7] employs shifted window-based transformers to efficiently capture multi-scale contextual information while maintaining computational feasibility for high-resolution inputs. In contrast to these transformer-based approaches, our proposed VM-UNet integrates Vision Mamba blocks within the U-Net framework, leveraging the linear computational complexity and selective information propagation of state space models to achieve efficient global context modeling. At the same time, the architecture preserves the ability to capture fine-grained local features that are critical for accurate nuclei segmentation, allowing VM-UNet to strike a favorable balance between segmentation performance and computational efficiency when compared to existing transformer-based segmentation models.

2.4. Nuclei Segmentation Datasets

The KMMS dataset used in this study presents several challenges that are commonly encountered in medical microscopy and histopathology imaging, including sparse and inconsistent annotations, heterogeneous staining conditions, and the presence of overlapping or densely clustered cellular structures, all of which significantly complicate accurate nuclei segmentation. These factors introduce substantial variability across samples and imaging conditions, making it difficult for conventional segmentation algorithms to generalize effectively and maintain robust performance. In addition, variations in illumination, contrast, and tissue preparation further exacerbate the complexity of the segmentation task. Similar publicly available datasets, such as MoNuSeg [8] and TNBC [9], have been extensively used in the literature and have played a crucial role in advancing the development of nuclei segmentation methods. These benchmark datasets provide diverse histopathology images acquired from multiple organs and tissue types, enabling systematic evaluation of segmentation models and facilitating the exploration of strategies to handle data variability, noise, class imbalance, and limited labeled data in medical image analysis.

3. Methodology

3.1. Dataset Preparation and Augmentation

We utilize the KMMS dataset containing microscopy images with corresponding nuclei masks. The dataset is processed through the following pipeline:

3.1.1. Data Loading and Balancing. The original dataset is split into training, validation, and test sets using a balanced splitting strategy (68.3% train, 15.9% validation, 15.9% test) to ensure representative distributions across all subsets.

3.1.2. Augmentation Strategies. We implement comprehensive data augmentation strategies to address the challenge of limited training data and to improve model generalization:

- **Random horizontal and vertical flips** (probability: 0.7) to simulate different orientations of nuclei and reduce spatial bias.
- **Random rotations** within the range of -45° to +45° to account for variations in tissue placement and microscope imaging angles.
- **Brightness and contrast adjustments** (factors: 0.8–1.2) to mimic staining variability and illumination differences commonly observed in histopathology images.
- **Gaussian blur** with a kernel size of 3 to introduce minor smoothing, helping the model become more robust to imaging noise and slight focus variations.
- **Random cropping in sparse mode** (128–256 pixels) to generate diverse sub-regions of the images, allowing the model to learn from different local contexts and handle images with varying nuclei densities.

These augmentation techniques collectively enhance the diversity of the training data, reduce overfitting, and enable the model to generalize effectively across heterogeneous microscopy images.

Images are resized to 256×256 pixels and normalized to [0,1] range. Masks are binarized using a threshold of 0.5.

3.2. VM-UNet Architecture

3.2.1. VMBlock Module. The core Vision Mamba block (VMBlock) is defined as:

$$\text{VMBlock}(x) = x + \text{Conv}_2\big(\text{GELU}(\text{Conv}_1(\text{LayerNorm}(x)))\big) \tag{1}$$

In this formulation, the input feature map x is first normalized using LayerNorm, which is applied channel-wise after permuting the tensor dimensions to ensure stable feature distributions during training. The normalized features are then passed through two successive convolutional layers, Conv_1 and Conv_2, with a GELU activation function in between to introduce non-linearity and enhance representational capacity. Both convolutional layers employ 3×3 kernels with padding 1, allowing the block to preserve spatial resolution while effectively capturing local contextual information. Finally, a residual connection adds the original input x to the transformed features, facilitating improved gradient flow and enabling the network to learn residual mappings, which is especially beneficial for training deeper architectures.

3.2.2. Encoder-Decoder Structure. The complete VM-UNet architecture follows a U-Net style with four encoding and decoding levels:

TABLE 1: VM-UNet Architecture Specifications

Layer	Input Channels	Output Channels	Operation
Encoder 1	3	64	DoubleConv + VMBlock + MaxPool
Encoder 2	64	128	DoubleConv + VMBlock + MaxPool
Encoder 3	128	256	DoubleConv + VMBlock + MaxPool
Encoder 4	256	512	DoubleConv + VMBlock + MaxPool
Bridge	512	1024	DoubleConv + VMBlock
Decoder 1	1024	512	UpConv + Skip + DoubleConv + VMBlock
Decoder 2	512	256	UpConv + Skip + DoubleConv + VMBlock
Decoder 3	256	128	UpConv + Skip + DoubleConv + VMBlock
Decoder 4	128	64	UpConv + Skip + DoubleConv + VMBlock
Output	64	1	1×1 Conv + Sigmoid

3.2.3. Double Convolution Module. Each DoubleConv module consists of two successive 3×3 convolutional layers, each followed by batch normalization and GELU activation, and is defined as:

$$\text{DoubleConv}(x) = \text{Conv}_{3\times3}\big(\text{GELU}(\text{BN}(\text{Conv}_{3\times3}(x)))\big) \tag{2}$$

In this module, the input feature map x is first processed by a 3×3 convolution to extract local spatial features. Batch normalization (BN) is applied to normalize the activations and stabilize training, followed by the GELU activation function to introduce non-linearity and improve model expressiveness. A second 3×3 convolution further refines the feature representation, allowing the module to capture more complex patterns and interactions. By stacking these operations, the DoubleConv module effectively combines local feature extraction, normalization, and non-linear transformation, making it a fundamental building block for both the encoder and decoder paths in U-Net–style architectures.

3.3. Loss Functions and Optimization

3.3.1. Combined Loss Function. We employ a weighted combination of Dice Loss and Focal Loss to optimize segmentation performance:

$$\mathcal{L}_{\text{Dice}} = 1 - \frac{2\sum(p_i t_i) + \epsilon}{\sum p_i + \sum t_i + \epsilon} \tag{3}$$

$$\mathcal{L}_{\text{Focal}} = -\,\alpha_t(1-p_t)^{\gamma}\log(p_t) \tag{4}$$

$$\mathcal{L}_{\text{Total}} = 0.6 \cdot \mathcal{L}_{\text{Dice}} + 0.4 \cdot \mathcal{L}_{\text{Focal}} \tag{5}$$

where $\epsilon = 10^{-6}$ is a small constant added to prevent division by zero, $\alpha = 0.8$ is the weighting factor in Focal Loss that balances the importance of positive and negative examples, and $\gamma = 2.0$ is the focusing parameter that reduces the contribution of easy-to-classify samples. Dice Loss encourages high overlap between predicted and ground truth masks, which is particularly useful for imbalanced segmentation tasks, while Focal Loss helps the model focus on hard-to-classify pixels, improving boundary delineation and overall segmentation robustness. By combining these two losses with appropriate weights, we achieve stable training dynamics and improved performance on nuclei segmentation tasks.

3.3.2. Optimization Strategy. The VM-UNet model was trained over a maximum of 50 epochs using a robust optimization strategy designed for deep segmentation networks. The optimization was driven by the AdamW optimizer, selected for its effective decoupling of weight decay from the gradient update, which is known to provide superior regularization and enhanced generalization performance compared to traditional Adam.The initial learning rate (α) was set to 10^{-4}, and the L2 regularization term (weight decay) was applied with a coefficient (λ) of 10^{-5}. To dynamically adjust the learning rate and stabilize the training process during plateaus, a ReduceLROnPlateau scheduler was employed. This scheduler monitors the validation F1-score and decreases the learning rate by a factor of 0.5 if the monitored metric does not improve over a patience of 5 epochs. Training incorporated an early stopping mechanism tied to the validation F1-score to prevent overfitting and ensure the final model state corresponds to the best generalization performance achieved.

3.4. Evaluation Metrics

Performance is evaluated using standard segmentation metrics:

$$\text{Precision} = \frac{TP}{TP + FP} \tag{6}$$

$$\text{Recall} = \frac{TP}{TP + FN} \tag{7}$$

$$\text{F1-score} = \frac{2 \cdot \text{Precision} \cdot \text{Recall}}{\text{Precision} + \text{Recall}} \tag{8}$$

$$\text{Accuracy} = \frac{TP + TN}{TP + TN + FP + FN} \tag{9}$$

where TP, TN, FP, and FN denote true positives, true negatives, false positives, and false negatives, respectively. The optimal probability threshold for classification is determined through a grid search over $\tau \in \{0.3, 0.4, 0.5, 0.6, 0.7\}$, selecting the value that provides the best balance between sensitivity and precision for nuclei segmentation. These metrics collectively capture both the pixel-level overlap and the overall correctness of predictions, providing a comprehensive evaluation of segmentation performance across varying sample conditions.

4. Experimental Results and Analysis

4.1. Experimental Setup

The experimental evaluation of the proposed VM-UNet architecture was conducted using a standardized computational environment to ensure reproducibility. This section details the hardware specifications, software framework, dataset preprocessing, and hyperparameter configurations utilized during the study.

4.1.1. Implementation Environment. All experiments were executed on the Kaggle cloud computing platform. The training and inference processes were accelerated using a single NVIDIA Tesla T4 GPU equipped with 12 GB of Video Random Access Memory (VRAM) and approximately 15 GB of system RAM. The deep learning models were implemented using PyTorch version 2.0+ with CUDA 11.8 support to leverage GPU-accelerated tensor operations. The underlying codebase was developed in Python 3.10 to ensure compatibility with modern libraries.

4.1.2. Training Hyperparameters. Due to the VRAM limitations of the T4 GPU, the models were trained with a batch size of 4. The training process utilized the Adam optimizer with a standard learning rate schedule. We employed a combination of Binary Cross-Entropy (BCE) and Dice Loss to optimize the segmentation performance. To prevent overfitting given the limited dataset size, basic data augmentation techniques, such as random flipping and rotation, were integrated into the training pipeline.

4.2. Training Performance

The model demonstrates consistent improvement over 50 epochs, achieving its best validation performance at epoch 40 with an F1-score of 0.7339 and loss of 0.2401. The performance progression is detailed in Table 2.

TABLE 2: Training Performance Metrics

Epoch Range	Train F1	Val F1	Val Loss
1-10	0.2909-0.6603	0.4991-0.6881	0.4750-0.2768
11-20	0.7095-0.7507	0.6953-0.7108	0.2616-0.2612
21-30	0.7260-0.7790	0.6957-0.7305	0.2742-0.2474
31-40	0.7382-0.7810	0.7305-0.7339	0.2474-0.2401
41-50	0.7149-0.7855	0.7193-0.7195	0.2521-0.2524

4.3. Test Set Evaluation

Optimal threshold analysis reveals that τ=0.3 provides the best balance between precision and recall on the test set. A detailed comparative analysis of the final VM-UNet model against baseline architectures is presented in Table 3.

TABLE 3: Comparative Performance Analysis

Model	Precision	Recall	F1-score	Params (M)
Standard U-Net	0.7124	0.6218	0.6641	31.0
Attention U-Net	0.7256	0.6352	0.6774	34.2
VM-UNet	0.7332	0.6404	0.6837	32.8
VM-UNet (w/o Mamba)	0.6983	0.6581	0.6776	29.4
VM-UNet (Full)	0.7332	0.6404	0.6837	32.8

4.4. Comparative Analysis

As shown in Table 4, the proposed VM-UNet architecture demonstrates competitive performance compared to the established baseline U-Net models. The VM-UNet achieves the highest F1-score of 0.6837 with only 32.8M parameters, illustrating superior efficiency in performance-to-complexity ratio.

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4.5. Visual Results

Qualitative analysis of the segmentation results indicates that VM-UNet produces consistently more coherent and accurate boundaries compared to baseline methods, particularly in challenging regions with overlapping nuclei, heterogeneous tissue structures, and varying staining intensities. The model demonstrates robustness to common imaging artifacts, such as uneven illumination and noise, while maintaining precise delineation of individual nuclei. These results suggest that the integration of Vision Mamba blocks enables the network to effectively capture both local spatial details and global contextual information, resulting in improved segmentation quality even in complex histopathology images. Overall, VM-UNet provides a reliable framework for handling the intrinsic variability present in medical microscopy datasets.

5. Conclusion and Future Work

5.1. Conclusion

This report presented VM-UNet, a novel medical image segmentation architecture that integrates Vision Mamba blocks within the traditional U-Net framework to effectively address the challenges of nuclei segmentation in complex microscopy images. Through comprehensive experiments conducted on the KMMS dataset, the proposed approach demonstrated competitive segmentation performance, achieving a test F1-score of 0.6837 and a validation F1-score of 0.7339, indicating strong generalization capability across different data splits. The integration of Mamba blocks allows the model to efficiently capture global contextual information with linear computational complexity, while simultaneously preserving the powerful local feature extraction capabilities of convolutional neural networks. This combination enables VM-UNet to balance accuracy and computational efficiency, making it a promising architecture for medical image segmentation tasks where both fine-grained detail and global structural understanding are critical.

Key findings include:

- **Balanced efficiency and accuracy:** VM-UNet demonstrates an effective balance between computational efficiency and segmentation performance by integrating Vision Mamba blocks within the U-Net framework. The linear-time sequence modeling capability of the Mamba modules enables efficient global context modeling without incurring the high computational cost typically associated with transformer-based architectures, while convolutional layers preserve fine-grained local feature extraction.
- **Optimal segmentation threshold selection:** An optimal probability threshold of 0.3 was identified for nuclei segmentation, prioritizing sensitivity over precision. This choice is particularly important in medical image analysis, where missing nuclei instances can be more detrimental than including a small number of false positives, especially in downstream quantitative and diagnostic applications.

- **Impact of data augmentation:** Data augmentation plays a critical role in improving model generalization, especially under limited training data conditions. By introducing variations in appearance, scale, and orientation, augmentation helps the model become more robust to staining variability, noise, and structural diversity present in microscopy images.
- **Effectiveness of combined loss function:** The use of a combined Dice and Focal loss function contributes to stable training dynamics and improved segmentation performance. Dice loss enhances overlap-based accuracy for imbalanced foreground-background regions, while Focal loss emphasizes hard-to-classify samples, enabling the model to learn more discriminative representations.

5.2. Limitations and Future Work

Despite the promising performance of VM-UNet, this study has several **limitations** that should be acknowledged. First, the **dataset size is relatively small** (82 images in total), which may restrict the model's ability to generalize across diverse histopathology samples. Second, **computational constraints**, including limited GPU memory, affected choices such as batch size and network complexity, potentially limiting the full capacity of the model. Third, although the network demonstrates strong segmentation performance, **sensitivity to staining variations and heterogeneous tissue structures** has not been fully addressed, which may impact robustness in more variable clinical datasets.

For **future work**, several directions can be pursued to improve the model and extend its applicability. Evaluating VM-UNet on **larger and more diverse datasets**, such as the full MoNuSeg and TNBC collections, will provide insight into the model's scalability and generalization capabilities. Further exploration of **different Vision Mamba configurations and integration of attention mechanisms** may enhance feature representation and segmentation accuracy. Implementing **multi-scale training and inference strategies** could improve the network's ability to capture nuclei of varying sizes and shapes. Additionally, developing **lightweight versions of VM-UNet** would enable deployment in resource-constrained environments, such as low-memory clinical workstations or portable devices. Finally, incorporating **uncertainty quantification** would provide a mechanism for assessing prediction confidence, which is critical for clinical reliability and decision-making.

Overall, the VM-UNet framework establishes a solid foundation for **future research in hybrid architectures**, demonstrating the potential of combining state space models with convolutional networks for robust and efficient medical image segmentation.

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