

# A Benchmark Study of DeepLabV3+, U-Net++, and Attention U-Net for Blood Cell Segmentation

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**ABSTRACT:** Cell segmentation is a critical process in biomedical image analysis. This study evaluated the performance of three state-of-the-art deep learning models-DeepLabV3+, U-Net++, and Attention U-Net-using the Blood Cell Count and Detection (BCCD) dataset, which contains annotated images of blood cells. The models were rigorously analyzed through qualitative and quantitative evaluations, employing accuracy, precision, recall, and F1 score metrics. The results demonstrated that all three models achieved high segmentation performance, with U-Net++ excelling in accuracy (0.9740), precision (0.9511), and F1 score (0.9576), Attention U-Net achieving the highest recall (0.9692), and DeepLabV3+ providing a balanced performance across all metrics. Qualitative analyses revealed that U-Net++ delivered superior segmentation of complex and overlapping cell structures, while Attention U-Net exhibited exceptional sensitivity to dense cell clusters. Training and validation curves of the models confirmed their stability and generalizability, indicating efficient convergence without overfitting. By highlighting the unique strengths of each model, this study emphasized the importance of selecting architectures tailored to specific tasks. Future research will expand the application of these models to diverse biomedical datasets to further advance automated image analysis and its impact on healthcare outcomes.

**KEYWORDS:** Cell segmentation; DeepLabV3+; U-Net++; attention U-Net; biomedical image analysis; b30lood cell

#### **1. Introduction**

Recent advancements in biomedical research have emphasized the importance of precise and efficient cell segmentation, which plays a crucial role in disease diagnosis, drug discovery, and cellular behavior monitoring. Accurate segmentation provides critical insights into cell morphology, facilitating the early detection of diseases such as cancer, assessing drug efficacy, and supporting personalized medicine [1]. Furthermore, high-throughput experiments require automated image analysis to process vast amounts of data efficiently and extract meaningful conclusions [2]. The importance of improving segmentation techniques is underscored by their applications in quantifying immune responses in infectious diseases, tracking tumor

progression, and evaluating treatment efficacy. In regenerative medicine and tissue engineering, segmentation enables the monitoring of stem cell differentiation and the success of therapeutic interventions [3].

Traditional segmentation methods, such as manual annotations and classical image processing techniques, have often been time-consuming, error-prone, and dependent on predefined rules [4]. These limitations are exacerbated when handling large datasets, where manual efforts become subjective and non-scalable. Additionally, classical methods have struggled with overlapping cells, irregular cell structures, and variations in imaging conditions, reinforcing the necessity for automated approaches [5]. Deep learning has emerged as a revolutionary tool in biomedical image analysis, offering hierarchical feature learning capabilities that significantly enhance segmentation accuracy and efficiency.

One of the foundational architectures in biomedical segmentation was U-Net, which introduced a fully convolutional encoder-decoder structure with skip connections, enabling precise segmentation while preserving spatial information [6]. This model laid the groundwork for more advanced architectures such as DeepLabV3+, U-Net++, and Attention U-Net. DeepLabV3+ utilizes Atrous Spatial Pyramid Pooling (ASPP) to extract multi-scale contextual features, making it highly effective for overlapping and irregularly shaped cells [7]. U-Net++ improves skip connections through a nested architecture, refining segmentation boundaries and enhancing detail preservation [8]. Attention U-Net integrates attention gates to selectively focus on critical regions, suppressing background noise and improving segmentation in complex cellular environments [9]. Despite these advancements, a comprehensive comparative analysis of these models for biomedical cell segmentation u-Net using the Blood Cell Count and Detection (BCCD) dataset, evaluating their segmentation performance in terms of accuracy, sensitivity, specificity, and F1 score. This research provides an in-depth performance comparison to guide the selection of optimal architectures for biomedical applications.

On the other hand, traditional segmentation techniques, such as Otsu's Thresholding [10] and the Watershed Algorithm [11], have long been used in biomedical imaging. Otsu's method, a histogram-based global thresholding technique, is effective for simple, high-contrast images but fails in overlapping cell segmentation [12]. Watershed segmentation, a region-based approach, effectively separates touching objects but often suffers from over-segmentation in noisy images [13]. While these classical methods are computationally efficient, they lack the adaptability required for complex biomedical images.

Deep learning-based segmentation models, such as SegNet [14] and Mask R-CNN [15], have advanced the field significantly. SegNet, an encoder-decoder model, effectively extracts spatial features but lacks skip connections, which limits its ability to retain fine details. Mask R-CNN, developed for instance segmentation, provides precise object delineation but is computationally expensive, making it less practical for large-scale biomedical applications [16]. More recently, Transformer-based models, such as TransUNet [17], have integrated CNN feature extraction with self-attention mechanisms, demonstrating superior segmentation capabilities. However, these models require large datasets for training and have high computational complexity, limiting their real-world feasibility in medical imaging [18].

Given these considerations, DeepLabV3+, U-Net++, and Attention U-Net were selected for this study due to their proven effectiveness in biomedical segmentation. DeepLabV3+, with its multi-scale feature extraction capabilities, excels in capturing complex cellular structures [7]. U-Net++, with nested skip connections, enhances spatial information retention, making it ideal for precise boundary segmentation [8]. Attention U-Net employs attention gates to enhance regional focus, making it particularly effective in dense cellular environments [9]. These models provide an optimal balance of segmentation accuracy, computational efficiency, and adaptability, making them suitable for biomedical image analysis.

Deep learning-based segmentation models have also demonstrated strong performance across multiple domains, including biomedical and non-medical applications. In non-medical fields, DeepLabV3+ has been applied in autonomous driving for lane detection, achieving 94.5% accuracy and demonstrating its capability in handling complex spatial layouts [19]. U-Net++ has been used in environmental monitoring, particularly in flood mapping, where it achieved an F1 score of 92.3%, highlighting its ability to preserve fine details [20]. Attention U-Net has demonstrated strong performance in wildfire boundary detection, achieving 95.8% sensitivity, showcasing its effectiveness in detecting irregular spatial patterns [21].

In biomedical applications, U-Net++ has been widely employed for melanoma segmentation, achieving a Dice coefficient of 89.7%, outperforming conventional segmentation methods [22]. DeepLabV3+ has been applied in brain tumor segmentation, obtaining an accuracy of 93.1% on the BraTS dataset [23]. Attention U-Net has shown strong performance in pancreas segmentation, achieving an average Dice score of 87.6%, proving its effectiveness in small and complex anatomical structures [24].

Comparative studies have analyzed the effectiveness of these models in different biomedical applications. For instance, a study on liver segmentation using CT images found that Attention U-Net achieved the highest sensitivity (94.8%), while U-Net++ had the highest specificity (92.7%) [25]. Similarly, in skin lesion segmentation, DeepLabV3+ achieved an IoU of 86.5%, while U-Net++ and Attention U-Net reached 88.1% and 90.2%, respectively [26]. These findings highlight the strengths of each model, with U-Net++ excelling in boundary refinement, Attention U-Net performing well in dense cellular regions, and DeepLabV3+ offering robust multi-scale analysis [27].

The literature supports the growing role of deep learning in biomedical segmentation and highlights the limitations of traditional methods. While classical techniques such as Otsu's Thresholding and Watershed struggle with overlapping structures, deep learning models overcome these limitations by leveraging feature extraction and advanced network architectures [28]. Among them, DeepLabV3+, U-Net++, and Attention U-Net provide the best balance of accuracy, computational efficiency, and generalizability, making them the optimal candidates for biomedical segmentation tasks [29].

Despite these advancements, a comprehensive comparison of these models on cell segmentation tasks has remained limited, mainly using the Blood Cell Count and Detection (BCCD) dataset. The BCCD dataset, a widely used benchmark for blood cell segmentation, provides annotated images of blood cells. This study aims to evaluate the performance of DeepLabV3+, U-Net++, and Attention U-Net on the BCCD dataset, analyzing their effectiveness in terms of accuracy, sensitivity, specificity, and F1 score. This research seeks to guide the selection of appropriate architectures for specific biomedical applications.

## 3. Methodology

This study employed a structured approach for cell segmentation using three advanced deep learning models: DeepLabV3+, U-Net++, and Attention U-Net. The methodology consisted of

several sequential stages, including data pre-processing, training, validation, testing, and performance evaluation. Figure 1 provides a system overview of the steps involved in this study. The first stage began with data pre-processing, which included initiating the dataset, reshaping the data, and splitting it into training, validation, and testing subsets. Pre-processing ensured that the input data was normalized and formatted correctly to improve the performance and stability of the models during training. The data reshaping technique ensured that all the images were already the same size for training.

In the training stage, the three models underwent hyperparameter tuning, pooling layer adjustments, and layer configuration optimization to achieve optimal performance. This iterative process ensured that each model was tailored to effectively handle the complexities of the dataset. Once the training was complete, the testing phase was carried out. During this stage, the trained models were evaluated using the test dataset to predict segmentation outputs. The predictions were compared against the ground truth to assess the segmentation accuracy. Finally, the performance evaluation stage involved computing various metrics, including accuracy, sensitivity, specificity, and F1 score. These metrics were used to compare the effectiveness of DeepLabV3+, U-Net++, and Attention U-Net in segmenting cells. The model with the best overall performance was identified based on the evaluation results. This methodology provided a comprehensive framework for analyzing the performance of state-of-the-art deep learning models in cell segmentation, contributing to the advancement of automated biomedical image analysis.



Figure 1. System overview of the proposed research.

### 3.1. Dataset.

The BCCD dataset was a publicly available dataset designed for training and evaluating machine learning models for blood cell segmentation and classification. This dataset served as an essential benchmark for tasks such as cell counting, detection, and morphometry, providing high-quality labeled data for biomedical image analysis [25]. The dataset was originally sourced from Kaggle [25], and it contained microscopic images of blood cells and the corresponding binary masks that segmented individual cells from the background. A rich diversity of cellular shapes and configurations, including overlapping cells, irregularly shaped cells, and cells captured under varying lighting conditions, was collected in these datasets.

For this study, the dataset was split into training (80%) and testing (20%) subsets. The BCCD dataset suffered from class imbalance. To tackle this problem, we applied weighted binary cross-entropy (WBCE). WBCE assigned a higher weight to positive pixels (cells) to ensure they contributed more to the loss function, helping the model learn better segmentation in imbalanced conditions. For the testing phase, unseen images and their masks were used to evaluate model performance, ensuring that the results reflected real-world generalization capabilities. The diversity of the BCCD dataset, combined with its structured annotations, made it particularly suitable for benchmarking state-of-the-art deep learning models such as DeepLabV3+, U-Net++, and Attention U-Net in biomedical applications.

## 3.2. Model architecture.

DeepLabV3+ was a powerful semantic segmentation model that leveraged atrous spatial pyramid pooling (ASPP) to capture contextual information at multiple scales. It featured a unique encoder-decoder structure where the encoder extracted rich feature representations, and the decoder refined these features to produce detailed segmentation maps. U-Net++ was an enhanced version of the original U-Net architecture, designed to bridge the semantic gap between the encoder and decoder sub-networks. This was achieved through nested and dense skip connections, which allowed for better feature propagation and localization accuracy.

Meanwhile, Attention U-Net incorporated attention gates within the standard U-Net framework to focus on relevant regions of the input image while suppressing irrelevant background information. This mechanism enabled the model to effectively segment intricate and small structures in biomedical images. Table 1 summarized the parameters and total number of layers for each model. The comparison highlighted the trade-offs between model complexity, computational cost, and segmentation performance. While DeepLabV3+ had the highest number of parameters and layers, providing exceptional performance in challenging tasks, its computational demands were not ideal for resource-constrained environments. Conversely, U-Net++ and Attention U-Net offered lightweight alternatives with excellent segmentation capabilities, making them suitable for a wider range of applications. DeepLabV3+ exhibited the highest inference time, which was approximately 61% slower than U-Net++. This increased computational cost was attributed to its deeper architecture. In contrast, U-Net++ and Attention U-Net demonstrated faster inference times, highlighting their potential for deployment in time-sensitive biomedical imaging tasks.

Table 1. Model's parameters.						
Model's name Total parameters		Total number of layers	Inference Time			
DeepLabV3+	27,585,857	200	0.0263 sec/image			
Attention U-Net	8,127,620	52	0.0181 sec/image			
U-Net++	7,781,761	25	0.0163 sec/image			

To determine whether the performance differences among the models were statistically
significant, we conducted paired T-tests and Wilcoxon Signed-Rank tests. Table 2 summarized
he statistical results across the three models.

Table 2. Model's statistical comparison.						
Comparison	T-test p-value	Wilcoxon test p-value				
DeepLabV3+ vs. U-Net++	0.0031	0.1250				
U-Net++ vs. Attention U-Net	0.8083	0.6250				
DeepLabV3+ vs. Attention U-Net	0.1143	0.1250				

The T-test confirmed a significant difference between DeepLabV3+ and U-Net++ (p = 0.0031), indicating that U-Net++ significantly outperformed DeepLabV3+ in segmentation accuracy. However, no significant differences were found between U-Net++ and Attention U-Net (p = 0.8083) or between DeepLabV3+ and Attention U-Net (p = 0.1143), suggesting comparable segmentation performance between these models.

#### 3.3. Evaluation Metrics.

To comprehensively evaluate the performance of the segmentation models, multiple metrics were employed to ensure a robust assessment of the model segmentation capabilities, such as accuracy, sensitivity, specificity, F1 score, Dice coefficient, and Intersection over Union (IoU). Accuracy measures the proportion of correctly classified pixels, providing a general measure of the model's effectiveness:

$$Accuracy = \frac{(TP + TN)}{(TP + FP + TN + FN)}$$
(1)

where TP is True Positives (correctly segmented positive pixels), TN is True Negatives (correctly segmented negative pixels), FP is False Positives (pixels incorrectly classified as positive), and FN is False Negatives (pixels incorrectly classified as negative). This metric is useful for understanding the overall performance but may not distinguish well in cases of imbalanced datasets.

Precision quantifies the proportion of true positive pixels among all predicted positive pixels:

$$Precision = \frac{TP}{TP + FP}$$
(2)

This metric emphasizes the model's ability to avoid false positives, making it particularly valuable in applications where over-segmentation could lead to erroneous conclusions. In Recall, it measures the ability of the model to correctly identify all true positive pixels:

$$Recall = \frac{TP}{TP + FN}$$
(3)

High recall ensures that the model captures the majority of relevant structures, such as cells, without missing important details. For F1 Score is the harmonic mean of precision and recall, providing a balanced measure of performance in scenarios with class imbalances:

$$F1 Score = \frac{2 \times Precision \times Recall}{Precision + Recall}$$
(4)

This metric highlights the trade-off between precision and recall, ensuring that the model performs well in detecting and segmenting relevant structures without excessive false positives or negatives. Dice coefficient quantifies how well the predicted segmentation matches the ground truth:

$$Dice = \frac{Area \ of \ Overlap}{Total \ Area} \tag{5}$$

This metric is more sensitive to small objects, making it useful for medical image segmentation. It will calculate how well the alignment of your ground truth and predicted result. Meanwhile, IoU is defined as the ratio of intersection to union between prediction and ground truth:

$$IoU = \frac{Area \ of \ Overlap}{Area \ of \ Union} \tag{6}$$

Higher values of IoU and Dice indicated better segmentation performance. These metrics collectively ensured a robust evaluation of each model, capturing its strengths and weaknesses in terms of overall performance, the ability to detect structures of interest, and the balance between false positives and false negatives. By focusing on accuracy, precision, recall, F1 score, Dice coefficient, and IoU, this study provided a comprehensive assessment of the segmentation models' effectiveness for biomedical image analysis.



Figure 1. Training and validation accuracy curves for (a) DeepLabV3+ (b) U-Net++ (c) Attention U-Net.

## 4. Results and Discussion

This section presented the experimental results of DeepLabV3+, U-Net++, and Attention U-Net on the BCCD dataset. The models were evaluated based on accuracy, precision, recall, and F1 score. Additionally, training loss and accuracy graphs were provided to demonstrate model convergence and performance over 100 epochs. The models were implemented in Python using TensorFlow and Keras libraries. The training was conducted on an NVIDIA RTX 3090 GPU with a batch size of 16, a learning rate of 1e-3, and the Adam optimizer. Fine-tuning of all parameters was applied to prevent overfitting.

Hyperparameter tuning was performed using a random search approach, which efficiently explored the hyperparameter space by randomly sampling values within pre-defined ranges. The Adam optimizer was selected for its ability to adapt the learning rate dynamically, enhancing convergence stability. The learning rate was randomly sampled from [1e-4, 1e-3, and 1e-2], with a learning rate of 1e-3 being selected based on validation set performance. The batch size was randomly sampled from [8, 16], with 16 being selected as the optimal batch size based on the trade-off between computational efficiency and model performance. The dropout rate was randomly sampled from [0.2, 0.3, 0.4], with 0.3 being selected to prevent overfitting.

After all hyperparameters were carefully selected, the training and validation accuracy and loss graphs were plotted because these graphs were essential for evaluating the effectiveness of the model and the training process. These graphs provided insights into model convergence, with a steady decline in loss and an increase in accuracy, indicating progress toward an optimal solution. They also helped detect overfitting, where training accuracy improved while validation accuracy stagnated or declined, as well as underfitting, where neither curve showed significant improvement. Monitoring these curves ensured training stability and revealed potential issues like an unstable learning rate or noisy data.

Furthermore, they facilitated comparison among models by highlighting differences in convergence rates and learning efficiency. A close alignment between training and validation curves signified good generalization capability, ensuring the model performed well on unseen data. Additionally, these graphs guided improvements in model design by identifying bottlenecks and suggesting adjustments like data augmentation, regularization, or architectural modifications, ultimately contributing to a robust and reliable model.

Figure 2 illustrates all three models' training and validation accuracy across 100 epochs. The graph showed a consistent improvement in both training and validation accuracy. Similarly, Figure 3 presents the training and validation loss curves, demonstrating a significant reduction in loss during the initial epochs. These results indicated that all the models successfully learned to generalize without overfitting.



Figure 2. Training and validation loss curves for (a) DeepLabV3+ (b) U-Net++ (c) Attention U-Net.

To visually evaluate the segmentation quality, Figure 4 illustrates the input image, ground truth mask, and predicted segmentation masks generated by DeepLabV3+, U-Net++, and Attention U-Net for five representative test images. The predictions from all three models closely matched the ground truth, showcasing their ability to perform accurate cell segmentation. Among these, U-Net++ demonstrated the most precise segmentation, particularly in cases of overlapping cells, as evidenced by its well-defined boundaries and minimal false positives.

In addition to the comparative analysis of multiple test images, Figure 5 provides a comprehensive view of one test image with the corresponding overlay of predictions from each model on the ground truth mask. The overlay visualization enabled a clearer assessment of the model segmentation performance, highlighting the success of the models' predictions. For instance, while all models handled isolated cells effectively, U-Net++ consistently achieved

superior performance in detecting and segmenting complex cell clusters, as evident in the overlap regions.



Figure 3. Qualitative comparison of segmentation results.

Furthermore, Table 3 provides a quantitative comparison of the models based on accuracy, precision, recall, and F1 score. Accuracy reflects the overall correctness of the segmentation, measuring the proportion of correctly classified pixels (true positives and true negatives) out of all pixels. In this study, U-Net++ achieved the highest accuracy (0.9740), suggesting its ability to provide reliable predictions across diverse test cases. Precision evaluates the proportion of true positive pixels among all pixels predicted as positive, highlighting the model's ability to avoid over-segmentation. With a precision of 0.9511, U-Net++ minimized false positives effectively, which is critical for tasks where over-segmenting irrelevant structures could lead to misleading results. Recall, on the other hand, measures the model's ability to identify all true positive pixels, indicating sensitivity to target structures. Attention U-Net excelled in this aspect, achieving a recall of 0.9692, demonstrating its effectiveness in capturing true positives, particularly in challenging scenarios with densely overlapping cells. F1 score, the harmonic mean of precision and recall, provides a balanced measure of the trade-off between these metrics. U-Net++ achieved the highest F1 score (0.9576), highlighting its ability to balance precision and recall effectively.



Figure 4. Overlay Comparison of Predicted Segmentation.

Table 3. Performance comparison across all models.						
Model	Accuracy	Precision	Recall	F1 Score	Dice Coef.	IoU
DeepLabV3+	0.9704	0.9447	0.9589	0.9518	0.9106	0.8988
U-Net++	0.9740	0.9511	0.9642	0.9576	0.9541	0.9298
Attention U-Net	0.9734	0.9450	0.9692	0.9569	0.9415	0.9224

The results demonstrate that all three models are highly effective in segmenting blood cells, with slight variations in their strengths. U-Net++ high precision makes it suitable for applications where false positives must be minimized, while Attention U-Net high recall is advantageous for detecting true positives in dense cell clusters. DeepLabV3+ provides a balanced approach, making it a versatile option for general segmentation tasks. The consistent performance across both qualitative and quantitative analyses emphasizes the reliability of these models. The overlay comparison (Figure 5) provides further evidence of the practical applicability of U-Net++ in complex segmentation tasks, making it the most robust choice for handling overlapping and irregularly shaped cells. These findings underscore the importance of model selection based on the specific requirements of the biomedical application, contributing valuable insights to the advancement of automated cell segmentation.

To further support the claim that U-Net++ performs best in segmenting overlapping cells, we computed Contour Matching Score (CMS), both of which confirmed superior segmentation accuracy for U-Net++. CMS is an objective way to assess segmentation accuracy in overlapping cell regions by comparing predicted and ground-truth contours. Table 4 indicates the comparison of the CMS results for all models.

Table 4. CMS Performance comparison across all models.			
Contour Matching Score (CMS)			
0.3421			
0.5125			
2.3473			

The results indicated that U-Net++ achieved the lowest CMS (0.3421), demonstrating the highest segmentation accuracy. Attention U-Net achieved a moderate CMS (0.5125), while DeepLabV3+ had the highest CMS (2.3473), suggesting difficulty in accurately delineating overlapping cell boundaries. These quantitative results validate the qualitative visual

observations and confirm that U-Net++ outperforms other models in overlapping cell segmentation.

## 5. Conclusions

This benchmark study presented a thorough comparative analysis of three advanced deep learning models, i.e., DeepLabV3+, U-Net++, and Attention U-Net, for cell segmentation using the BCCD dataset. Each model demonstrated exceptional performance across multiple evaluation metrics, with distinct advantages aligning with specific segmentation requirements. U-Net++ emerged as the most effective, with higher results in accuracy, precision, and F1 score, consolidating its benefits in minimizing false positives due to its nested skip connections, which enhanced feature refinement and localization. Attention U-Net showcased its strength in detecting true positives in dense and overlapping cellular regions, yielding higher recall performance. Its integration of attention gates enabled superior focus on relevant regions while suppressing background noise. DeepLabV3+ leveraged its atrous spatial pyramid pooling to handle diverse contextual information, making it a versatile choice for general biomedical segmentation tasks. Trade-offs between model complexity and performance were also considered. DeepLabV3+, while delivering balanced results, had the highest parameter count and the most computationally demanding architecture, potentially limiting its applicability in resource-constrained environments. Conversely, U-Net++ and Attention U-Net were computationally efficient, with significantly lower parameter counts, making them suitable for broader applications without sacrificing segmentation quality. However, U-Net++'s focus on precision may have resulted in slightly reduced sensitivity, while Attention U-Net's emphasis on recall may have led to a marginal increase in false positives. These findings underscored the importance of selecting models based on the specific priorities of a biomedical application, whether it be precision, recall, or computational feasibility. This research provided a valuable framework for guiding such decisions and highlighted the potential for further innovation. Future work can explore ensemble approaches combining the strengths of these models or hybrid architectures integrating attention mechanisms with multiscale feature extraction. Moreover, extending the evaluation to include diverse and noisy datasets will further validate the robustness of these models in real-world scenarios. Future work will also validate the transferability of DeepLabV3+, U-Net++, and Attention U-Net on datasets such as MoNuSeg (nuclei in microscopy segmentation) and ISIC (skin lesion segmentation) to assess their generalization to other biomedical tasks. By addressing these challenges, the insights gained from this study can pave the way for more advanced, reliable, and efficient automated cell segmentation methods, contributing to enhanced healthcare outcomes and accelerating biomedical research.

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## Author Contribution

The authors confirm contribution to the paper as follows: study conception and design: Clara Lavita Angelina; data collection: Ali Rospawan; analysis and interpretation of results: Clara Lavita Angelina, Ali Rospawan; draft manuscript preparation: Clara Lavita Angelina, Ali Rospawan. All authors reviewed the results and approved the final version of the manuscript.

# **Competing Interest**

Authors declare that there is no conflict of interests regarding the publication of the paper.

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