

Visualizations for Glomeruli Detection Algorithm Assessment

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Abstract

The Human BioMolecular Atlas Program (HuBMAP) aims to create an open, global atlas of the human body at the cellular level. One component of this overarching goal is to identify glomeruli, functional tissue units (FTUs) consisting of capillaries that facilitate filtration of blood, within whole slide images of kidney tissue. Once these glomeruli are detected in the microscopy images, information on size and location within the kidney samples can be used to build a spatially accurate model of human kidneys for the HuBMAP. The Faster RCNN and Mask RCNN machine learning (ML) algorithms were applied to detect glomeruli as a step towards automating this process. Properly formulated visualizations assisted the designers of these ML algorithms in better understanding the training data being used to create the glomeruli detection algorithms, the structure or outliers within the output data of the Faster RCNN algorithm, and the weaknesses of the Mask RCNN algorithm.

I. INTRODUCTION

The Human BioMolecular Atlas Program (HuBMAP) aims to create an open, global atlas of the human body at the cellular level [6]. One component of this overarching goal is to identify glomeruli, functional tissue units (FTUs) consisting of capillaries that facilitate filtration of blood, within whole slide images of kidney tissue. Once these glomeruli are detected in the microscopy images, information on size and location within the kidney samples can be used to build a spatially accurate model of human kidneys for the HuBMAP.

Manual identification and classification of FTUs from microscopy images requires highly trained experts and is labor intensive [7]. Many machine learning (ML) algorithms have been previously applied to automate detection of glomeruli [1], [3], [5], [7], [9]. For this work, the Faster RCNN and Mask RCNN ML algorithms were utilized to detect glomeruli in Periodic acid-Schiff (PAS) stained whole slide images of kidney tissue samples. Faster RCNN and Mask RCNN are both convolutional neural network (CNN) algorithms designed for object detection in images. The Faster RCNN algorithm outputs bounding boxes around identified glomeruli, which are described by x-min, x-max, y-min, and y-max measures within the context of the original slide image. The Mask RCNN algorithm takes this a step further, outputting a pixel-wise map of glomeruli vs. non-glomeruli sections of arbitrary shape from the input image.

Properly formulated visualizations can assist the designers of ML algorithms in several ways. If the goal is to make high caliber ML algorithms, then the data set used to train said algorithms must be thoroughly explored and understood. This can be achieved through calculation and visualization of metrics that investigate variation within the dataset. In addition, ML algorithms often employ high-dimensional vectors, which can be very difficult to visualize. To translate these vectors into a visualization that can be more easily understood by the human eye, embedding spaces are created. Essentially, high-dimensional graph representations are translated into an optimized low-dimensional representation that is as structurally similar as possible to the original [2]. This translation onto a 2-dimensional space is what allows visual identification of hidden structure or outliers within data. Additionally, investigating incorrect predictions generated by an algorithm may give insight into how to optimize it for the future. One approach to this is to visually examine false positives, which in the context of this work means areas that were detected as glomeruli but are not included in the ground truth. Although humans and ML algorithms “view” images in very different ways and tune into very different patterns, inspection of false positives may help identify what caused the error. This report details the creation of visualizations to provide better understanding of the training data used to create the glomeruli detection algorithms, any structure or outliers within the output data of the Faster RCNN algorithm, and any weaknesses of the Mask RCNN algorithm to the designers of the Faster RCNN and Mask RCNN glomeruli detection algorithms.

II. DATA

This work utilized a dataset of human kidney whole slide images provided by the HuBMAP Consortium, which will be available at <https://hubmapconsortium.org/hubmap-timeline-to-data/> once the data has been released. The raw kidney image data was generated by the BIOmolecular Multimodal Imaging Center (BIOMIC) at Vanderbilt University. (See <https://medschool.vanderbilt.edu/biomic/> for more information.) This dataset is composed of 13 samples, each with multiple images that correspond to different microscopy types. All samples include PAS stain images, and many include imaging mass spectrometry as well. In addition, this dataset provides information on where the sample was positioned within the kidney and whether it was taken from a left or right kidney, which is vital information for the HuBMAP objective.

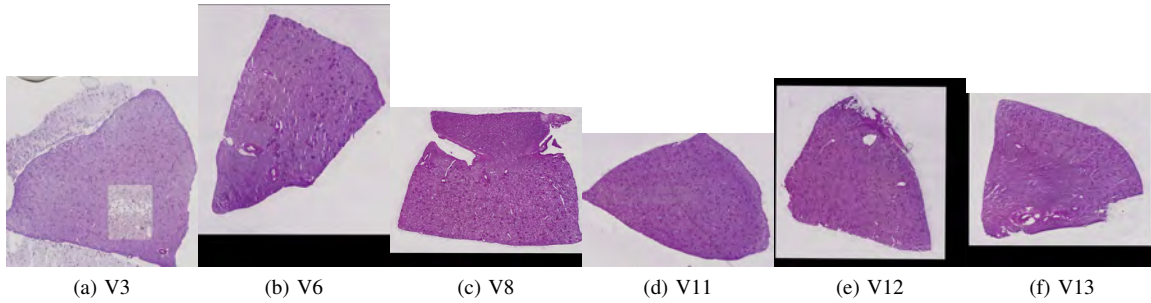


Fig. 1: Raw kidney image data.

From these 13 samples, a subset of six PAS stain images were used to generate the ML training dataset. See Figure 1 for the six whole slide images and their abbreviated names, which reference the Vanderbilt team and unique sample identification numbers. This glomeruli detection training dataset of manually generated glomeruli annotations was created by four users who marked areas corresponding to glomeruli on the six images, and it will be available at <https://github.com/cns-iu> once the data is released. All users performed their annotations on laptops that included Windows 10 OS, a trackpad, and 1920x1080 screen resolution. Half of the users made annotations with the software QuPath and its brush tool, while the other half utilized ImageJ with its oval tool. The users also reported the order in which the images were annotated and the total number of glomeruli detected per image. The annotation data was gathered and cleaned manually in Excel, and basic summary statistics were calculated, including total count of annotations by user, count of annotations by user per image, mean annotation count per image, and root mean squared error (RMSE) for annotation counts by image. The ground truth was determined by finding the areas of an image that were agreed to be glomeruli by all four individuals who provided manual annotations of the training data images. Intersection over Union (IoU) values were calculated by dividing the area of intersection between each user's annotations and the ground truth by the area of union between the annotations and ground truth.

III. METHODS

A. Insight Needs

The designers of the glomeruli detection ML algorithms, who are working towards the eventual HuBMAP goal, need actionable insights to optimize their Faster RCNN and Mask RCNN models. They need to know more about the training data they are utilizing, look for structure within the output of the algorithm, and inspect errors made by the algorithm. These needs were addressed by three visualizations: the training dataset infographic, the Faster RCNN UMAP Embedding Space, and the Mask RCNN False Positive Overlay.

B. Faster RCNN Algorithm

The Faster RCNN algorithm is one type of CNN used for object detection in images. CNNs employ neural networks for deep learning and allow unsupervised feature generation [3]. See Figure 2 for a diagram representing the structure of the Faster RCNN algorithm. This algorithm takes an image as input, which it then divides into smaller rectangular regions. From then on, it considers each region to be a separate image. Next, these regions are passed to the CNN, which provides classes and bounding boxes for detected objects. In the case of this work, the classes are "Glomeruli" or "Non-Glomeruli". After this is complete for all regions, they are combined to make the original image with glomeruli detected in rectangular boxes. The algorithm outputs the data describing these detection boxes as separate rows in a CSV file which describes each annotation prediction as a single row of data. Fields include "filename", "xmin", "xmax", "ymin", and "ymax". The "filename" refers to the unique number given to the region of the original image where the annotation was detected.

C. Mask RCNN Algorithm

The Mask RCNN algorithm is built upon the Faster RCNN algorithm, but it employs an instance segmentation extension that allows prediction of segmentation masks for each annotation. See Figure 3 for a diagram of the Mask RCNN structure. Rather than relying on the rectangular regions of the Faster RCNN algorithm for outputting detection boxes, the Mask RCNN provides a classification of "Glomeruli" or "Non-Glomeruli" to each pixel in the original image. This allows the resulting annotations to be any shape describable by pixels and enables the creation of mask overlays for use on the original image.

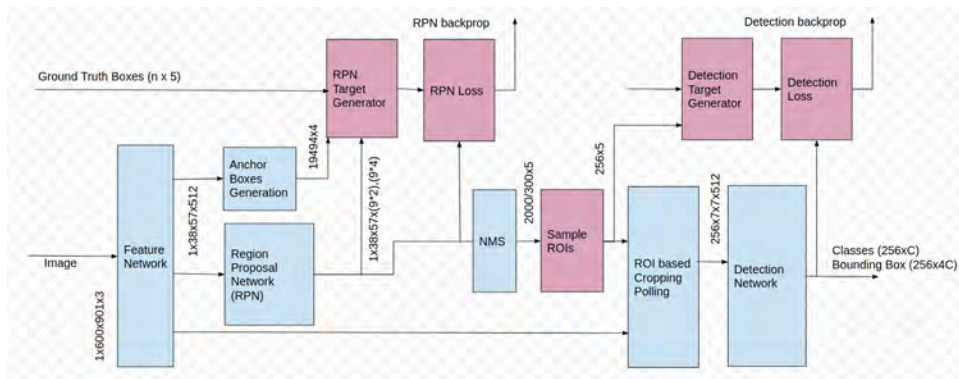


Fig. 2: Faster RCNN Diagram [4]

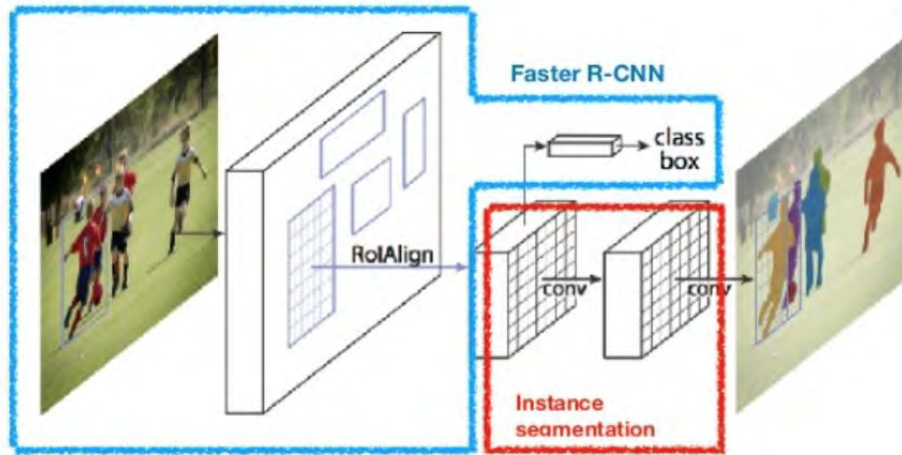


Fig. 3: Mask RCNN Diagram [8]

D. Training Dataset Infographic

The annotation data, including basic summary statistics, was imported into Tableau. This tool was used to create the Counts by User and IoU by Tool graphs. Refer to Table 1 for Data Visualization Literacy (DVL) Framework Details for all visualizations. These graphs were paired with useful tables providing total counts of annotations by user, mean annotation counts by image, RMSE of counts by image, and IoU values. The resulting graphs and tables were then arranged in an infographic using Microsoft PowerPoint. Examples of a PAS stained whole slide image and each user's annotation style were incorporated into the infographic, and a depiction explaining Intersection over Union was included.

Table 1: Data Visualization Literacy Framework Details

Visualization	Tool	Type	Graphic Symbol(s)	Graphic Variable(s)
Counts by User	Tableau	Bar Graph	Area	Hue
IoU by Tool	Tableau	Bar Graph	Area	Hue
UMAP Embedding Space	UMAP package, Python	Scatter Plot	Point	x- and y-position, Hue
False Positive Mask	Python, Adobe Photoshop	Image Overlay	Area	Position, Hue

E. Faster RCNN UMAP Embedding Space

The output of the Faster RCNN algorithm can be viewed as a vectors of data. These vectors, which each refer to one detection box annotation, were used to make an embedding space visualization. The UMAP Python package (available from <https://umap-learn.readthedocs.io/en/latest/index.html>) is a sophisticated and powerful tool for Uniform Manifold Approximation and Projection (UMAP). This package was implemented through a Python script to reduce the high-dimensional data into a 2-dimensional plot using the default parameters "n_neighbors = 15" and "min_dist = 0.1". The annotation datapoints were colored by their region (or "filename") of origin. The code used to generate the embedding space visualization can be found at <https://github.iu.edu/llschers/CapstoneProject>.

F. Mask RCNN False Positive Overlay

The output of the Mask RCNN was stitched together into a greyscale image that depicted a black background with predicted glomeruli annotations in grey. This was transformed via Adobe Photoshop into a mask that could be overlaid on the original kidney slide image. In addition, the ground truth annotations were filled in on this mask so only the false positives predicted by the algorithm remained. In this way, false positives could be identified and easily explored within their context of the surrounding kidney tissue.

IV. RESULTS

A. Training Dataset Infographic

The Glomeruli Detection Training Dataset infographic, shown in Figure 4, contains a great deal of information, including raw kidney data, individual manual annotation examples, and analysis across users. Examples of each of the four users' annotations are given to demonstrate the diversity within the data set. A Tableau visualization depicts counts of annotations both overall and by image, and an included table provides root mean squared error (RMSE) for annotation counts by image. Finally, it introduces the concept of IoU and compares this measure across users and by software used through another Tableau visualization.

The manual annotations generated with QuPath's brush tool have a completely different shape from those drawn with ImageJ's oval tool. The QuPath annotations exhibit great variety in shape, whereas the oval tool created annotations that were much smoother and uniform in shape. In addition, QuPath users typically annotated a greater number of glomeruli per image than ImageJ users. However, when IoU calculations were considered, software tool seemed to have no influence.

When averaged across users, the number of glomeruli in a single image ranged from 150.8 for image V8 to 449.0 for image V11. When comparing the individual glomeruli counts to the average for that image, RMSE ranged from 5.1 for image V6 to 40.7 for image V3. For future manual annotations, the data could be easily updated in the Tableau workbook to provide continued insight as the training dataset deepens.

B. Faster RCNN UMAP Embedding Space

The Faster RCNN UMAP visualization, shown in Figure 5, depicts 92 predicted annotations from 20 regions of kidney slide images. The data does not seem to be separated by image file of origin, which suggests that differences in tissue slide condition or stain saturation did not influence the algorithm's predictions. There are no easily discernable outliers in the plot, but there does seem to be some minimal clustering of the datapoints, particularly in the top left corner of the scatterplot. Further exploration is needed to determine the cause of this potential clustering.

C. Mask RCNN False Positive Overlay

The images utilized to create the visualization of Mask RCNN false positives are shown in Figure 6 alongside the resulting mask. While there are many false positives to visually assess, the example selected in Figure 7, which is of a size comparable to a glomerulus, seems to include a darker outline similar in appearance to the Bowman's capsule that surround glomeruli. This suggests that the algorithm is using that part of the image to make predictions. Potential solutions to this problem include deepening the training dataset or using imaging mass spectrometry images rather than PAS stain images.

V. CONCLUSION

Glomeruli detection can be performed through the application of ML algorithms which are properly trained and optimized. Visualizing the training dataset, algorithm output, and incorrect predictions can aid in this optimization process. Through an array of software tools, visualizations were created that helped meet the insight needs of ML algorithm designers working towards the goal of an atlas of the human body at the cellular level. Variation was uncovered in the manual annotation training set and a workflow was prepared for analyzing future additions to the dataset. The high-dimensional Faster RCNN algorithm output was reduced into a 2-dimensional figure for easier human visual evaluation. The Mask RCNN output was united with the original slide image to explore the root cause of errors. The resulting information from these visualizations will empower the designers to improve the glomeruli detection algorithms' performance.

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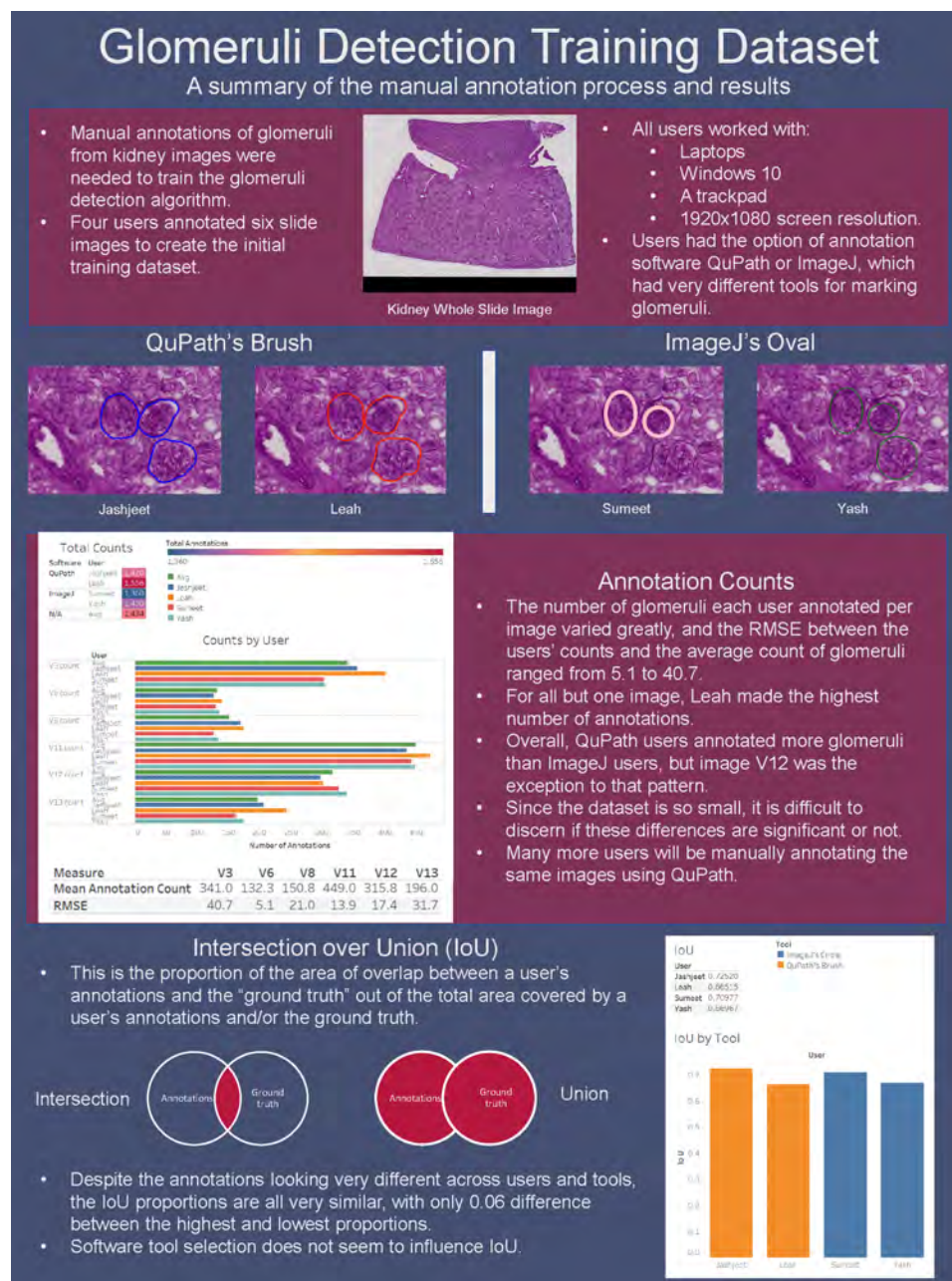


Fig. 4: Training Dataset Infographic.

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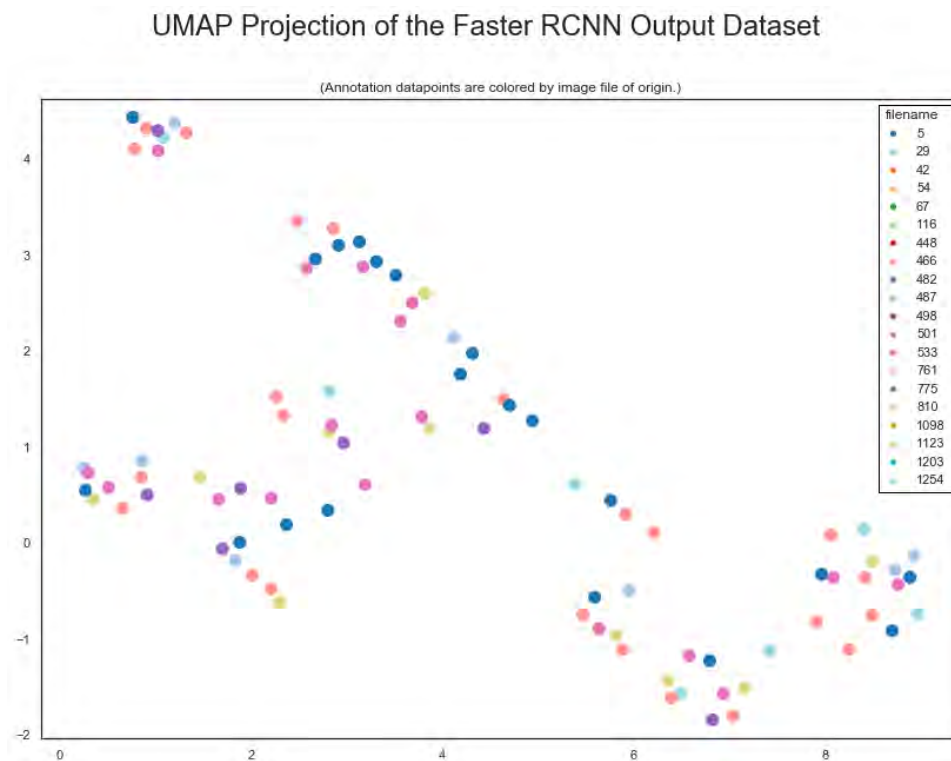


Fig. 5: Faster RCNN UMAP Embedding Space

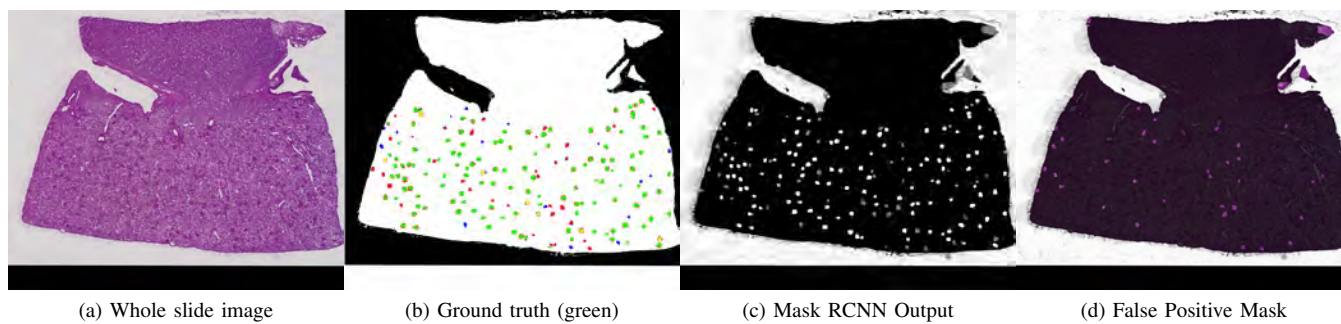


Fig. 6: False Positive Mask Process

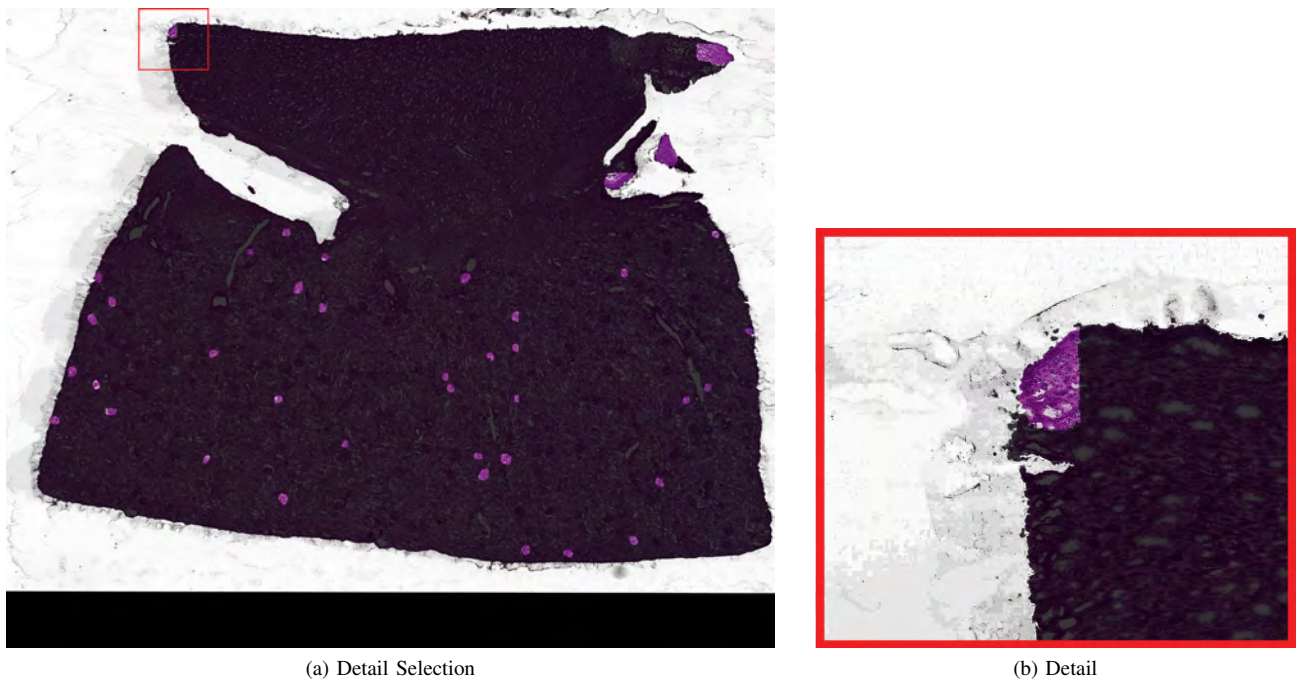


Fig. 7: False Positive Overlay Detail