# Assessment of Digital Image Focus Quality using Heat Map Visualization

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## Abstract

Assessing the focus quality of pathology samples for clinicians, researchers, and industry is an ongoing challenge. Whole Slide Imagers can scan >95% of the slide in focus but there will be small sample areas that require additional quality control. Providing a focus heat map of scanned tissue allows clinicians to quickly quality check the image for adequate focus. In addition to the visual indication, additional focus metrics are computed for threshold analytics.

## Objectives

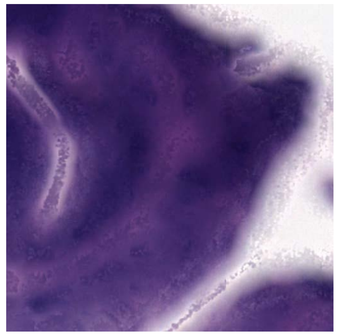
- Show the evolution of focus quality control (QC) on cytology sample scans.
- Demonstrate the advantage of the focus QC heat map as an additional tool for quality evaluation.
- Discuss the limitations of this technique.

## Focus Quality Control Evolution

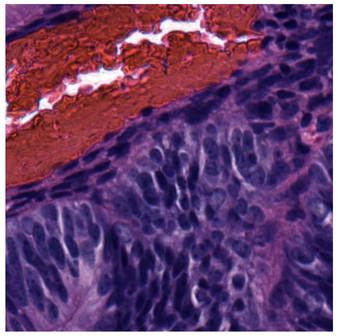
Hologic implemented a focus score method to allow for efficient quality control of the whole slide image. The image is divided into a fine grid of tiles (512x512 pixels, or about 130 microns per side). The image tiles are processed via standard image processing techniques (saturation, brightness, morphologic features, etc). The first step evaluates the amount of foreground content. If the tile's foreground content is too low the tile is scored as blank. Non-blank tiles are then scored using a normalized Brenner Score[1,2].

$$F_{Brenner} = \sum_{Height} \sum_{Width} (i(x+2,y) - i(x,y))^2 \qquad F_{Normalized} = F_{Brenner} / \text{foreground pixel count}$$

Tiles that score too low (100 or less) demonstrate a lack of contrast and are scored as out-of-focus. Tiles that score over 100 demonstrate strong contrast and are scored as in-focus.



Normalized Brenner Score of 36.3

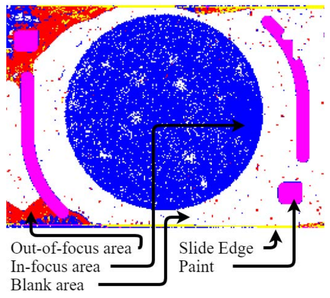


Normalized Brenner Score of 287.9

Once all the tiles are scored the overall in-focus tile counts and out-of-focus tile counts are used to determine if the focus quality of the entire image is acceptable or unacceptable. The percent of out-of-focus tiles must be below a threshold, the absolute number of out-of-focus tiles must be below a threshold, and the number of contiguous out-of-focus tiles must be below a threshold for the focus to be acceptable.

This technique works very well in general but artifacts on the slide may score as out-of-focus tiles. These may cause a slide scan that has good focus quality to fail. To improve yield, two filters are placed after the blank tile detector but before the tiles are Brenner scored. The first filter checks for the presence of a vertical slide edge and scores the tile as a slide edge. The second filter checks for opaque areas on the slide and scores the tile as paint. Thus, the focus score of a tile can be one of: 0 - out-of-focus; 1 - in-focus; 2 - blank; 3 - slide-edge; or 4 - paint.

The focus score values can be assigned individual colors to produce an image that has one pixel for each 512x512 tile in the whole slide image. This image is the focus quality heat map. The heat maps for an entire batch of slides can be easily generated and reviewed for patterns of in-focus and out-of-focus areas.




## References

1. L. Firestone et al., "Comparison of Autofocus Methods for Automated Microscopy" Cytometry, 12:195-206, 1991.
2. Q Wu, F Merchant, K. Castleman, "Microscope Image Processing" Academic Press, 2008, pp. 444-448

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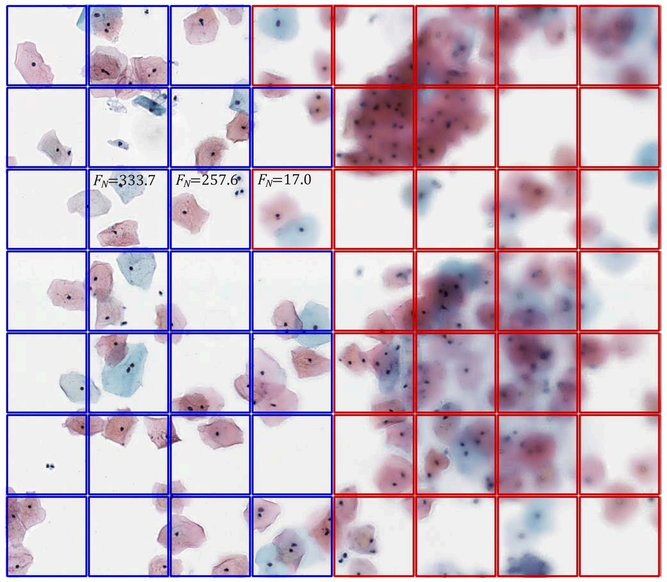
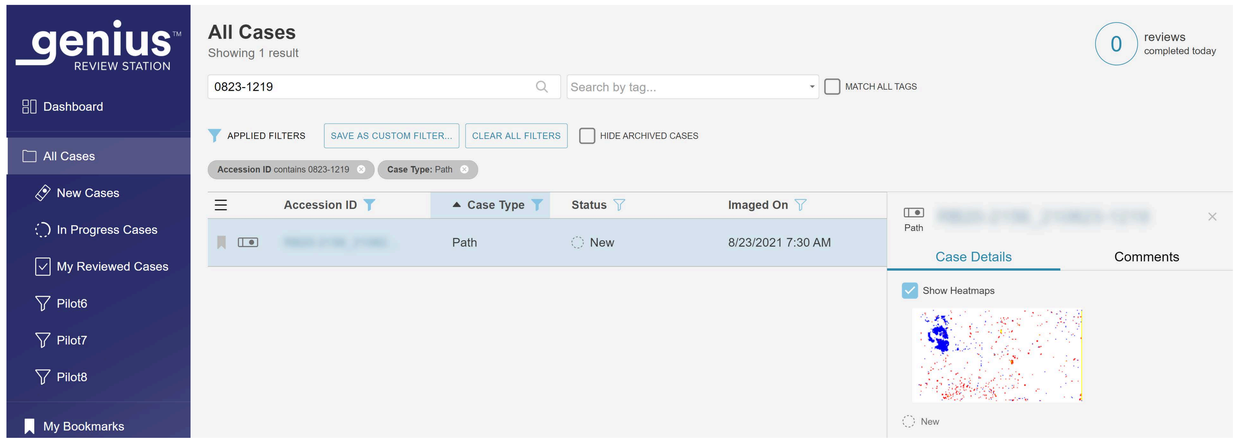
## Results

The validation of the focus quality scoring was done on multiple whole slide images. For each whole slide image, the focus score of each tile was overlaid over the whole slide image as a color-coded grid - one tile per grid box. The entire whole slide image was scanned and the count of incorrectly scored in-focus and the count of incorrectly scored out-of-focus tiles were tallied. These counts along with the in-focus tile total and out-of-focus tile total yielded the sensitivity and specificity of the focus quality algorithm. The sensitivity was 95.5% with a confidence interval of 95.2% to 95.7%. The specificity was 99.73% with a confidence interval of 99.70% to 99.75%.




## The Focus Heat Map in Pathology

Across the spectrum of pathology sample types, challenges to focus quality control abound. While new methods are investigated to address these challenges, presenting the focus quality heat map to the end users allows their expert judgment to determine the utility of the slide scan. For example, the heat map displayed in the case below shows a pathology slide scan that would typically fail focus quality control due to the percent of out-of-focus tiles. The heat map reveals that a full scan of the slide was performed, and the slide is dirty (red pixels) with very little actual sample (blue pixels). That sample scored as in-focus. So, the end user may decide to review the slide.




## Discussion

The tile scoring technique works very well on a large variety of pathology samples. However, there are sample types that challenge the technique. For example, large areas of low contrast tissue may result in a low Brenner score even when the tissue is in focus. Dirty slides are troublesome if the sample content is sparse. Even though in the majority of cases, cleaning the slide will resolve the issue, the long-term goal is to handle this case and other troublesome cases using image processing techniques. While the heat map is a nice aid for research where the need for in depth analysis may trump throughput, fully automated focus quality control is necessary in production.

## Conclusions

Focus heat maps are an effective method for end users to quality control the focus of specimens on the review station. In addition to visual quality control, the focus data may be used to generate acceptable thresholds and "reject" samples from entering the database if too much area is out of focus. These rejected samples can then be triaged on the Whole Slide Imager using other scanning modes to properly image the sample type. These focus identification methods provide tremendous value to the end user and lab organization. The clinician can be more efficient and spend more time on accurate diagnosis.