# MAKING THE INVISIBLE VISIBLE:

Optimizing Laser Speckle Imaging by using machine learning and targeted recoloring

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# **Abstract**

aser Speckle (LS) Imaging (LSI) is an important technique within the ✓ field of medical imaging, allowing medical professionals to acquire semiquantitative, anatomical, and physiological data about blood vessels and blood flow, which is essential in minimally invasive surgical procedures, providing spatiotemporal resolution and real-time imaging of blood vessels. However, the monochromatic images produced by LS can be difficult for the human eye to interpret due to low contrast and noise from the statistical computation. As a result, LSI can benefit from color mapping to emphasize relevant information (related to blood vessels anatomy). Despite the fact that color mapping for LSI is useful, color maps currently used in many medical imaging procedures are often misleading and obscure details, as they are perceived as non-uniform by the human eye.

In this research we developed a novel interface that allows users to view a montage of different lookup tables (LUT) applied to their image. Additionally, it includes an overlay of the sinusoidal function applied to each LUT, for easy comparison of the uniformity of the colors in them. This is useful as color maps vary in efficiency and utility based on project requirements and the medical professional's personal perception of colors. In addition, we present a pilot application of probability maps (PM), generated by supervised machine learning, to modify hue

in the HSI color model, which has the potential to enhance monochromatic LS images. As a result, the developed original user interface is intended to save time and increase efficiency in differentiating between blood vessels and surrounding tissue.

#### Introduction

Medical optical imaging of blood flow and blood vessels is crucial for both clinical and experimental applications <sup>[6]</sup>. Challenges in detecting hidden blood vessels during surgery have emerged with the increased use of minimally invasive surgical procedures. Therefore, there is a need to find new tools to supplement existing techniques of medical professionals for visualizing hidden blood vessels that must be avoided or strategically cut during surgery <sup>[4]</sup>.

Many biomedical applications use LSI for visualizing vasculature, blood flow and perfusion [6]. Laser speckles are produced in an area illuminated by a coherent scattered laser light [5]. When the scattered light is captured by a camera, a randomly varying interference pattern is acquired. Due to the instability of the scattering particles, the interference fluctuates, causing the image's change. The statistical intensity to computation of speckle patterns provides information about the movement of the scattered particles.  $K = \frac{\sigma}{\langle t \rangle}$  quantifies the blurring of the laser speckles. In the equation, K defines the laser speckle contrast while  $\sigma$  is the standard deviation over time of pixel intensity fluctuations, and *I* is the intensity of a single pixel <sup>[3]</sup>. Despite the benefits and the valuable information LSI contributes, the monochromatic images produced can be misleading for medical professionals due to high levels of statistical noise and low contrast levels. A possible solution for the enhancement of LSI is to increase the contrast and apply color maps accuracy and improve perception of blood vessels in the image.

Color maps are based on individual color perception and are dependent on color contrast, image structure, and intensity. While the information monochromatic images convey is insufficient, color maps can emphasize contours and lines within the image [2]. The 255 shades of grey in a typical 8-bit LS image will be separated into 2553 different shades of red, green and blue. Unfortunately, the color maps used in medical imaging procedures, many especially in microsurgeries, are difficult to interpret, as they can inadvertently conceal relevant information and blur fine details of body tissue and blood vessels. For instance, the rainbow color map dominates the scientific visualization world, yet the colors change un-uniformly, are sensitive to deficiencies in vision [10], and do not follow any natural ordering perception of the human eye. When designing a color map, it is important to ensure that the magnitude of the incremental change in perceptual lightness of the colors is uniform. Sinusoidal wave functions can be used to ensure that the contrast level of lookup tables (LUT) changes uniformly. The spatial frequency of the sine wave can be set within the range that is most sensitive to the human eye. Since the sine wave is uniformly visible across the full width of the image, the color map should have uniform perceptual contrast, ensuring that important details within the image are not obscured [9].

In order to fulfil our goals Fiji/ImageJ was chosen as a convenient software platform for image processing and analysis [12].

# **Materials and Methods**

ImageJ Macro Code (IJM) automates FIJI functions and is derived from Java [13]. Code development is done using Macro on FIJI, with which tools were developed to assist professionals in distinguishing blood vessels on LS images. Trainable WEKA Segmentation is a FIJI plugin for supervised machine learning algorithms, which produces an 8-bit pixel-based binary segmentation of selected images. It does so by letting users add traces to two or more training classes. After training, the program creates a PM of each pixel belonging to its

class based on the current classifier [1]. In a Hue Saturation Intensity (HSI) color space, hue defines the attribute of a pure color, distinguishing among different colors. Saturation characterizes the shade of color [12]. By replacing the hue in an HSI image with a PM, without changing the intensity values, the perception of the image will be improved, without creating false data. Additionally, the montage overlays a bar with a sinusoidal function to ensure the uniformity of the colors in various LUTs. The research developed a novel interface allows for fast and efficient identification of the best colormap during surgical or other procedures so as to better distinguish between blood vessels and tissues.

Original code is developed on FIJI to enhance LS images previously acquired from Dr. Kalchenko's laboratory.

The first aspect of this research is creating a montage of enhanced LS images, using IJM, with various LUTs, including multiple guides to help professionals choose the best LUT for their image as color perception differs by scenario. In the montage, the chosen LUTs are saved in an array. By iterating through the array, different color maps are applied to the original image, and a scale can be overlaid. Moreover, the user can specify the image enhancement with auto-contrast and gamma correction with a gamma value of 1.5 or 2, depending on the contrast of the original image.



Figure 1: User options on the novel user interface

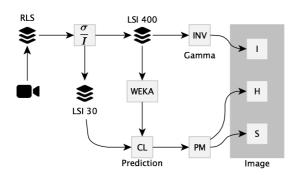


Figure 2: Image enhancement workflow. CL: Classifier, H: Hue, INV: Invert, I: Intensity, LSI: Laser Speckle Image Stack, PM: Probability Map, RLS: Raw Laser Speckle Image, S: Saturation

The second aspect of this research is to enhance the monochromatic and noisy images acquired from LSI with machine learning. A workflow that color codes the image without distorting and potentially losing important details is developed using supervised machine learning and image hue modification.

As LSI is intended for use in real-time surgical procedures, the number of images taken must be reduced. The typical number of pictures taken for a good noise to signal ratio is 400. For real-time LSI, the stack size must decrease to a maximum of 30 images. However, this reduction will negatively affect image quality, as the images become noisier and have less contrast.

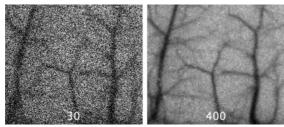


Figure 3: An example of two rendered LS images. 30 frames (left), 400 frames (right)

The WEKA binary segmentation plugin on FIJI is used to classify blood vessels and surrounding tissue. To increase the visibility of blood vessels, different approaches for training the classifiers were applied. The best enhancement for lower-quality LS test images is achieved by classifying a higher-quality image, and applying the classifier on the test image to train the model. The trained classifier then generates a PM, which illustrates the

probability of a pixel of the image being a part of a blood vessel, that we utilized to color code the original image. First, the raw data of the original image is separated into hue, saturation and intensity values. The PM is applied to the saturation and the hue. Replacing the hue and saturation of an RGB image with the PM of a LS image creates a colored version of the same image with details that are more differentiable to the human eye. The original rendered LS image is used as the intensity, so the image is not distorted and details of the blood vessels are preserved. Gamma correction and FIJI's auto contrast function are applied to the intensity values to further increase the contrast and improve perception of the image. Additionally, inversion of the hue and a bandpass filter are applied to increase the contrast and perceptual uniformity of the image.

Finally, sinusoidal wave functions were applied to compare the perceptual uniformity of the color contrast of the varying LUTs on the images within the montage.

$$I_{new} = I_{old} + \sin(f * \pi * I_{old}) * a$$

Equation 1: Sinusoidal function applied for any given pixel. The intensity I is modified by using the parameters f (frequency) and a (amplitude).

Based on this equation, Macro code was developed to display a bar of the sinusoidal gradient of the LUTs underneath each image in the montage to illustrate the uniformity of the various LUTs.

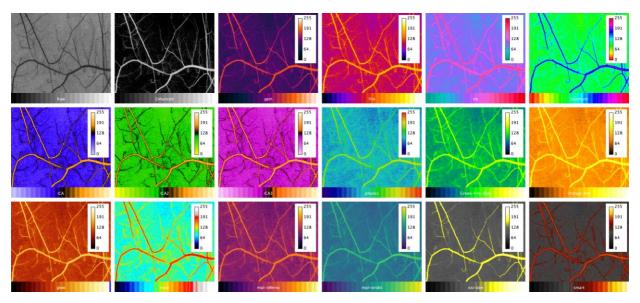


Figure 4: Montage of color maps

## **Results**

A montage of different color maps was developed to help medical professionals choose a color map more efficiently. The montage displays the original LS image, the image after gamma correction and autocontrast, and the images after applying various LUTs, including an original sinusoidal LUT we called "issi-bee". The overlay at the bottom of each image demonstrates the uniformity of each LUT based on sinusoidal functions. The montage can efficiently present valuable information from monochromatic LS images. The variety of colors in the montage is useful because color perception differs based on the LS image and the color perception of individuals.

Macro code was developed on FIJI to turn the probability map of the real time image sinusoidal. Although the sine functions do not enhance the visibility of color coded LS images, they have the potential to improve uniform color contrast.

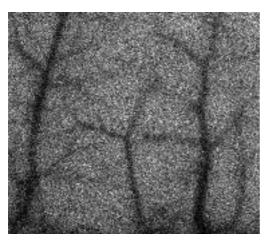


Figure 5: "Red Hot" LUT applied to a sinusoidal function.

The color map is not perceptually uniform as it is hard to differentiate between shades of yellow and red.

To identify whether the color contrast in various LUTs are perceptually uniform, Macro code was developed to apply a sinusoidal wave function to a given image. Running the Macro code (supplementary information required) on a linear monochromatic gradient, then applying a chosen LUT helps identify non-uniform contrast values within a color map.

This research developed a workflow to enhance real time LS images. The "Fast Forest" Random supervised machine learning algorithm was selected among various supervised machine learning models from WEKA for training, because this research discovered that Fast Random Forest yielded the best trained for binary classification of the blood vessels. The probability map, edited with gamma function and gaussian blur, was used to replace the hue of the LS image for color coding. Because the intensity unchanged, all the details in the image were preserved. After color coding monochromatic LS image, the visibility was improved, and the color contrast was more perceptually uniform to the human eye.



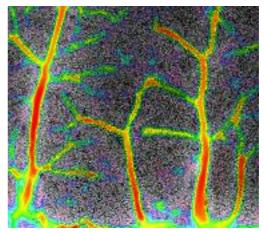


Figure 6: Original Image (left) Image after the workflow (right)

### **Discussion and Conclusion**

The first part of this research created a novel user interface consisting of a montage of color maps. The montage allows medical professionals to choose the most effective color scheme according to their specific need, as the demand for the type of color map varies depending on the LS image and individual color perception. The second part of the research developed a workflow using the probability map from the machine learning model to control the hue of an image. This workflow can potentially be an effective method color to monochromatic LS images, so it can be added to the montage in the future. Additionally, although sinusoidal wave functions were hypothesized to improve the perception of the color contrast in LS images, they showed no effect in the images. In this research, a sinusoidal color bar is overlaid onto each image for the user to compare the uniformity of different LUTs, allowing medical professionals to select the LUT that can be best perceived by the human eye with uniform contrast.

This research is currently limited to applications on LS images as the color maps developed are most useful for them. Moreover, the perception of color varies based on the object of interest and the vision of the user; therefore, the color mapping assessment in this research can be biased. In

the future, a more thorough color perception assessment based on more inputs from experts can improve the quality of the selection of color maps within the montage. Additional work is needed to develop realtime processing for LSI that can assist surgeons in visualizing blood vessels more clearly during surgical procedures. LSI color mapping schemes can also be further improved upon to benefit a wider variety of medical imaging tools that generate monochromatic images. Further statistical analysis of sinusoidal functions can help create a new method to generate sinusoidal LUTs, leading to a more uniform perception of color maps. Last but not least, further research can develop a combination of the probability maps from supervised machine learning and sinusoidal functions to create color maps that have the best uniform color contrast.

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