## INTRODUCTION

It is well known within the forensic science community that hair in the telogen growth phase is the most common type of hair found at crime scenes due to the periodic shedding of hair from a person’s body. The telogen phase is the mature stage of the growth phase in which the follicle is dormant or resting. Due to the relatively simple nature of telogen hair follicles, they tend to “contain very little quantity of DNA such that most nuclear material cannot be detected”. As a result, the North Carolina State Crime Laboratory (NCSCL), telogen hair root suitability for DNA analysis has historically been determined by the presence or absence of tissue around the root in the form of follicular tags. If tissue was present, the hair root was a sample suitable for DNA analysis; a destructive process to remove the tissue when the presence of nuclei was undetermined. It is unknown if the quantity of nuclei needed to generate a DNA profile was present in the tissue. Recent advancements in the Forensic Biology Section’s detection limits prompted the Trace Evidence hair examiners to research new staining techniques to improve this process suggested two methods of staining the tissue around hair roots to reveal the presence or absence of nuclei. These staining techniques coupled with the recent advancements in the NCSCL Forensic Biology Section’s detection limits prompted the Trace Evidence hair examiners to research ways to improve the current procedures for determining hair root DNA suitability.

### METHODS

**Hair Root Staining Protocol:**
1. Soak the root in Modified Harris Hematoxylin for 2 minutes.
2. Rinse the root with deionized water, followed by absolute ethanol.
3. Soak the root in Tannic Acid and Alum for 30 seconds.
4. Place the hair on a microscope slide and temporarily mount in xylene or xylene substitute.
5. Use the nuclear rich hair root with a transmissional light microscope and record the presence of any nuclei. The nuclei are dark red or purple in color and usually oval in shape.

To modify the modified Harris Hematoxylin, we performed as expected, a freshly plucked anagen hair root was stained. Numerous dark red or purple oval nuclei indicative of nuclei were noted as shown below (taken at 250x magnification).

### RESULTS AND CONCLUSIONS

The Trace Evidence Hair examiners decided to validate the method of hematoxylin staining for use in screening roots in the telogen growth phase in order to obtain a DNA profile from the North Carolina State Crime Laboratory in 11 or more nuclei. The implementation of hematoxylin staining into casework has resulted in a 26% increase in quantification cut-off pass rate and a 33% reduction in the number of hair roots sent for DNA analysis.

#### Quantitative Data

<table>
<thead>
<tr>
<th>Groupings</th>
<th>No. of roots</th>
<th>No. of nuclei</th>
<th>% Nuclei</th>
<th>% Profiles Obtained</th>
</tr>
</thead>
<tbody>
<tr>
<td>without Hematoxylin Staining</td>
<td>36</td>
<td>0</td>
<td>0%</td>
<td>85%</td>
</tr>
<tr>
<td>With Hematoxylin Staining</td>
<td>45</td>
<td>9</td>
<td>100%</td>
<td>85%</td>
</tr>
</tbody>
</table>

The data showed that 36% of Group I passed the quantification cut-off versus 88% of Group II. The quantitative data from the Hair Comparison casework simulation using 20 telogen growth phase hair roots in Group II (11 to 20 nuclei) showed that 50% were above the quantification threshold and would have proceeded to DNA amplification. Therefore, a root must contain 11 or more nuclei in order to be considered suitable for DNA analysis at the NCSCL.

#### Conclusions

- The data showed that the presence of nuclei was undetermined in the tissue around the hair roots.
- The telogen phase is the mature stage of the growth phase in which the follicle is dormant or resting.
- The nuclei were dark red or purple in color and usually oval in shape.
- The implementation of hematoxylin staining into casework has resulted in a 26% increase in quantification cut-off pass rate and a 33% reduction in the number of hair roots sent for DNA analysis.

### ACKNOWLEDGEMENTS

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**REFERENCES**