"Spectral Reflectance of Carotenoid-based Plumage Patches and Individual Fitness in Northern Michigan American Restarts (*Setophaga ruticilla*)"

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Introduction:

Brightly colored organisms usually posses this attribute either as a means of aposematic coloration or for sexual selection. In birds, the significance of bright plumage in mate choice is well documented (Inouye, Hill, Stradi, & Montgomerie, 2001) and recent research suggests that aspects (brightness, saturation and hue) of plumage coloration may also be mechanisms by which birds select mates (Siefferman & Hill, (2005). Further, because pigment deposition during feather growth is costly (McGraw, Hill, & Parker, 2005). it is hypothesized that plumage coloration not only is affected by age but also is also indicative of a bird's individual quality (McGraw & Hill, 2000). Birds of higher quality with certain plumage coloration may be selected for mating more often than those who do not display certain plumage coloration. Recent studies suggest that carotenoid-based feather pigmentation may be indicative of individual quality and may be used in mate-choice decisions in birds. American redstarts (*Setophaga ruticilla*) have carotenoid-based pigment patches in both their remiges and rectrices, and there is suggestion that these patches may play a role in mate choice. However, this question remains to be adequately studied in this species. The purpose of our study was to look for detectable variation in color attributes of these patches by age and sex, and to see if this variation could be related to individual fitness, as measured by various correlates of reproductive success.

Methods and Materials:

Feather collection- We collected the third right retrix from male and female American Redstarts captured in northern Michigan using mist-nets from 1999 to 2001(Figure 1.1a and b). The birds were then observed through the breeding season and reproductive success variables recorded. These included date of first egg laid, egg number and mass, the number of nestlings, and mass of eggs five days after being laid.

Feather analysis- We used an Ocean Optics USB4000 spectrometer to obtain reflectance spectra for each feather. *Spectrasuite* software was used in concert with the spectrometer to collect and process the reflectance data (Ocean Optics, 2007). Using this software, we smoothed each spectral curve using a "scans to average" value equal to five, and a smoothing boxcar width of 10. A halogen light source was used and reflectance values were taken using an integration time of 100ms. The sampling probe was placed in a holder that held it 1cm away from the sample at all times and at an incidence angle of 45 degrees. We collected four to five spectra for each feather patch, depending on the size of the pigmented area, in order to obtain an average patch coloration used later in analyses of color variables. Figure 1.2 and 1.3 are representative spectra for one female and one male spectral scan, respectively.

Spectrum analysis- After obtaining visible spectra they were processed using the Java based software CLR: Color Analysis Program v1.02 (Montgomerie R, 2008) to obtain measurements of hue, brightness, and saturation. Equations used to calculate each are presented in Table 1.1

Statistical analysis- We used general linear models (GLMs) on ranked data (Zar 1996) to assess reflectance differences by age and sex and Spearman's correlations to examine relationships between feather attributes and reproductive performance.

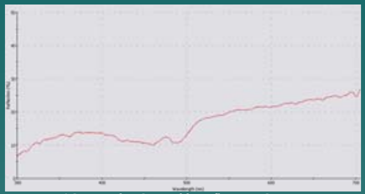

Figure 1.2. ASY Female R3R Visible Reflectance Spectrum obtained with *Spectrasuite* software

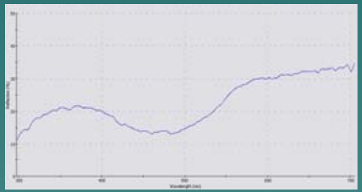

Figure 1.3. ASY Male R3R Visible Reflectance Spectrum obtained with *Spectrasuite* software

Table 1.1. Equations for Brightness, Saturation and Hue used by CLR v1.02

$$B_1 = B_T = \int_{\lambda \min}^{\lambda \max} R_i = \sum_{\lambda \min}^{\lambda \max} R_i$$

$$S_1 = \sum_{\lambda a}^{\lambda b} R_i \Big/ \sum_{\lambda \min}^{\lambda \max} R_i = \sum_{\lambda a}^{\lambda b} R_i \Big/ B_1$$

$$H_1 = \lambda_{R \max}$$

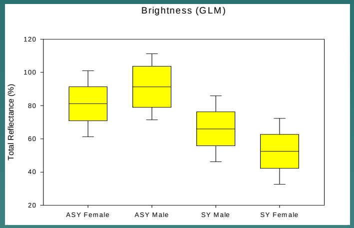

Figure 1.4.Rectrix brightness by age and sex according to Total Reflectance. Lines represent mean, boxes encompass plus/minus 1 S.E. and whiskers represent plus/minus 1 S.D.

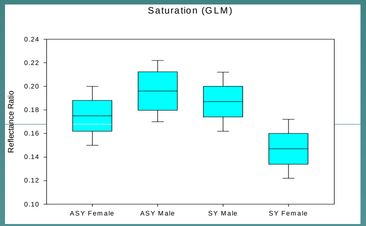

Figure 1.5. Rectrix saturation by age and sex according to Reflectance ratio. Lines represent mean, boxes encompass plus/minus 1 S.E. and whiskers represent plus/minus 1 S.D.

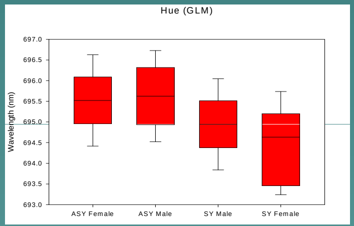

Figure 1.6. Rectrix hue by age and sex according to Wavelength. Lines represent mean, boxes encompass plus/minus 1 S.E. and whiskers represent plus/minus 1 S.D.

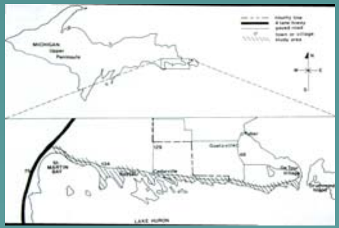

Figure 1.1a and b. Map of American Redstart capture site and photo of Lake Huron shoreline, respectively

Results:

Brightness – We found age effect ($F_{1,24}$=11.83, P = 0.002) but no sex effect ($F_{1,24}$=2.286, P = 0.144). Older birds were brighter than younger birds (Figure 1.4).

Saturation – We found a significant sex effect ($F_{1,24}$=9.289, P = 0.006) and the age effect approached significance ($F_{1,24}$=3.538, P = 0.072). Males were more saturated than females (Figure 1.5).

Hue – Our results suggest that there was no sex ($F_{1,24}$=0.228, P = 0.637) though an age effect approached significance ($F_{1,24}$=3.242, P = 0.084; see Figure 1.6).

Reproductive performance – There were significant relationships between female hue and nestling number (r = 0.708, n = 10, P = 0.022) as well as female saturation and 5-day old nestling mass (r = 0.624, n = 10, P = 0.054).

Discussion:

1. To our knowledge this is the first study to use a spectrometer to assess sex and age related variation in carotenoid-based plumage coloration and to look for relationships between color attributes and reproductive performance in American Redstarts.

2. The influence of age on brightness is common, and has been documented in numerous avian species. However the lack of a sex effect was unexpected. While more work is necessary, it is possible that brightness of carotenoid-based plumage patches in male redstarts may not be a sexually-selected characteristic.

3. Saturation is a measure of the degree to which a color appears to be pure (Andersson and Prager 2006). The sex-based differences in saturation likely reflect increased carotenoid deposition – males were more saturated than females. Higher levels in males suggests that saturation may be a feature upon which females make mate choice decisions.

4. The relationships between female reproductive performance, hue and saturation are interesting and warrant further examination. We cannot rule out the possibility that these relationships are a consequence of age – both saturation and hue may be influenced by age, as might reproductive performance. Alternatively, there is abundant evidence that carotenoid coloration reflects individual quality [carotenoids are linked to both quality of diet and health (McGraw 2006)]. Consequently, the observed relationships may reflect mate choice decisions or individual female quality, both of which may affect reproductive performance.

Acknowledgements:

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