

# Characterizing Leaf N with Digital Images and the Association of "Greenness" with Yield

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## Introduction

The environmental implications of nitrate pollution coupled with the cost of N fertilizers have compelled agronomists to develop quick and accurate methods of determining plant N. Our objective was to use a digital camera and image-analysis software to quantify the "greenness" of corn (*Zea mays* L.) leaves that could serve as a relative indicator of plant N status. In addition to Digital Color Analysis (DCA) of leaves, we also evaluated relationships among SPAD (which has been found closely related to corn yield, Zhang et al., 2008), as well as total leaf N, and the use of internal standards for camera calibration.

## Materials and Methods

### Field trial

- Urea was hand applied at 6 rates to 3-leaf corn (0, 67, 135, 200, 270, 335 kg N/ha) and replicated 4 times at Rohwer, Arkansas.
  - At tasseling, the uppermost collared leaf was sampled, chlorophyll meter readings were taken, and leaves were photographed against a neutral color felt background under normal fluorescent lighting.
  - Images were processed through Sigma Scan Pro (V. 5.0, SPSS Science, Chicago, Il.) analysis software to determine Hue, Saturation, and Brightness.
  - Hue, Saturation, and Brightness were used to calculate a dark green color index (DGCI, Karcher and Richardson, 2003).
  - Internal standards with known values of hue, saturation, and brightness were included in each image.
- ### Greenhouse
- Six N treatments (0, 168, 336, 500, 675, and 840 mg N/25cm pot) with two replications were made from Hoagland's solution and saucer applied.
  - The uppermost collared leaf was sampled at various developmental stages and subjected to SPAD and DCA.
  - DCA was recorded with three cameras of varying quality to determine the efficacy of calibration standards.

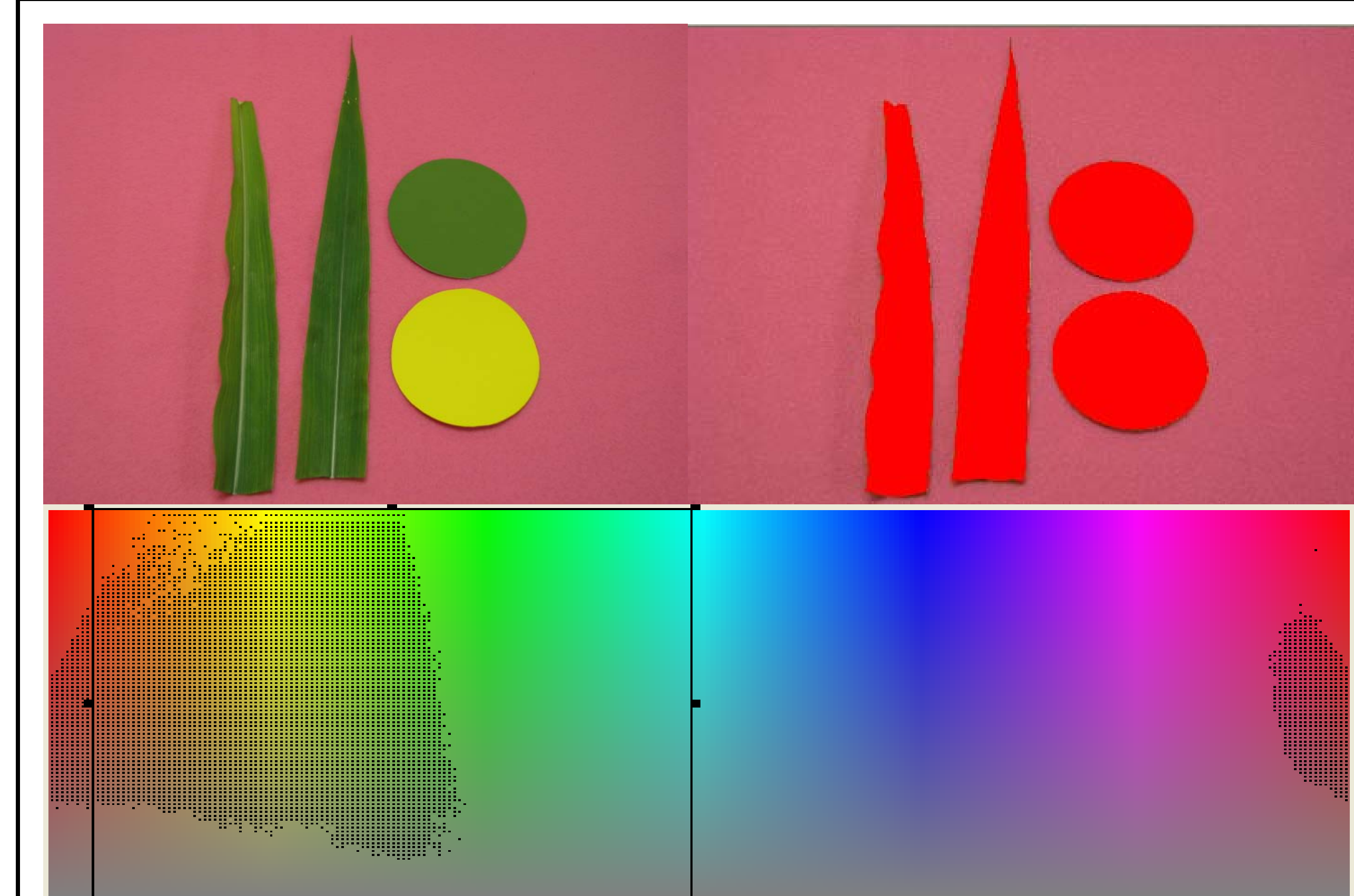


Figure 1. The color threshold in the lower image dictates the pixels to be analyzed in the upper image. Analysis recognizes standards for calibration based on shape.

## Results and Discussion

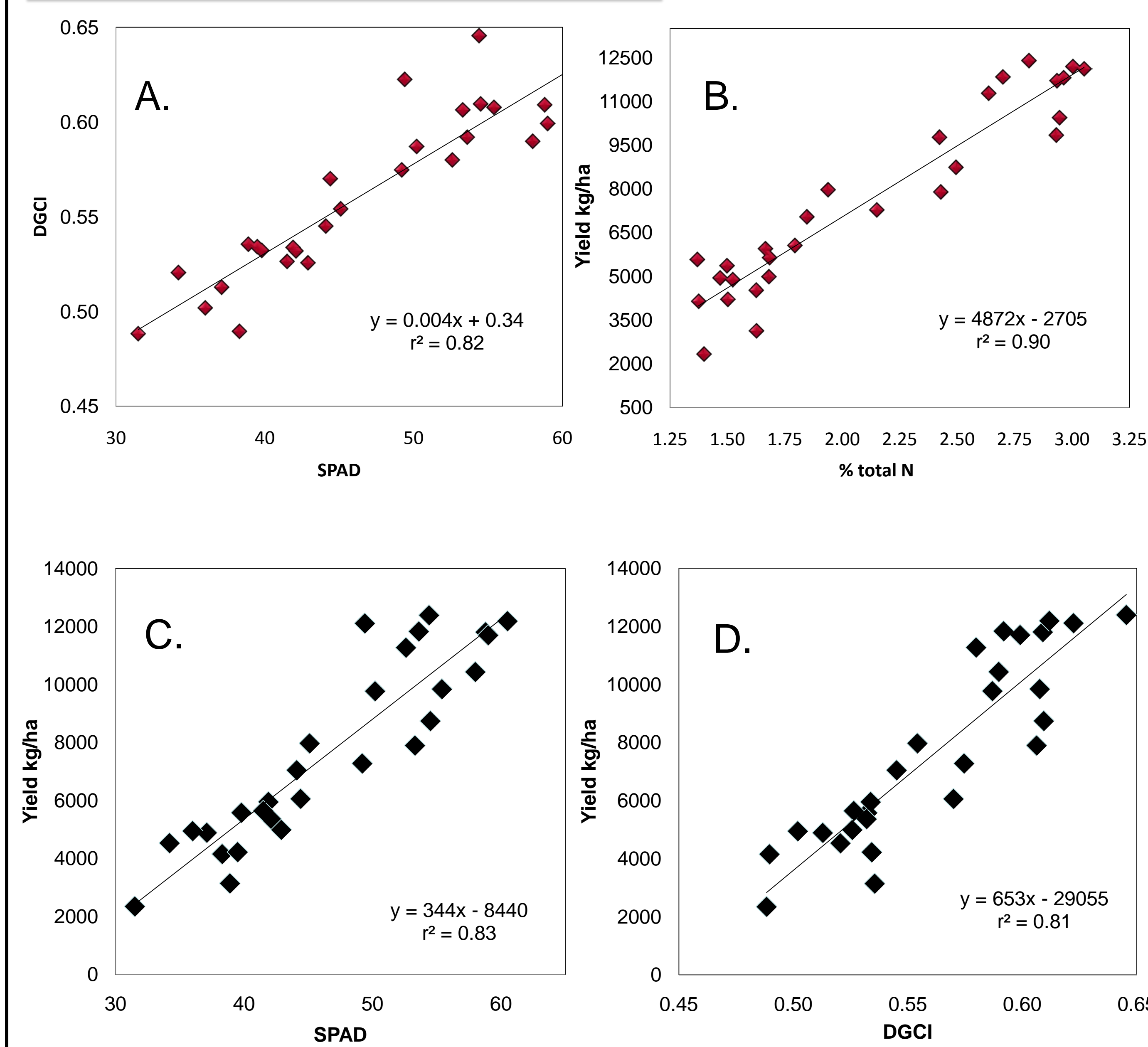


Figure 2. The above graphs show agreement among SPAD and DGCI (A), as well as % total N and overall grain yield (B). Graphs (C) and (D) show the relationship among SPAD and DGCI measurements taken at tasseling to yield.

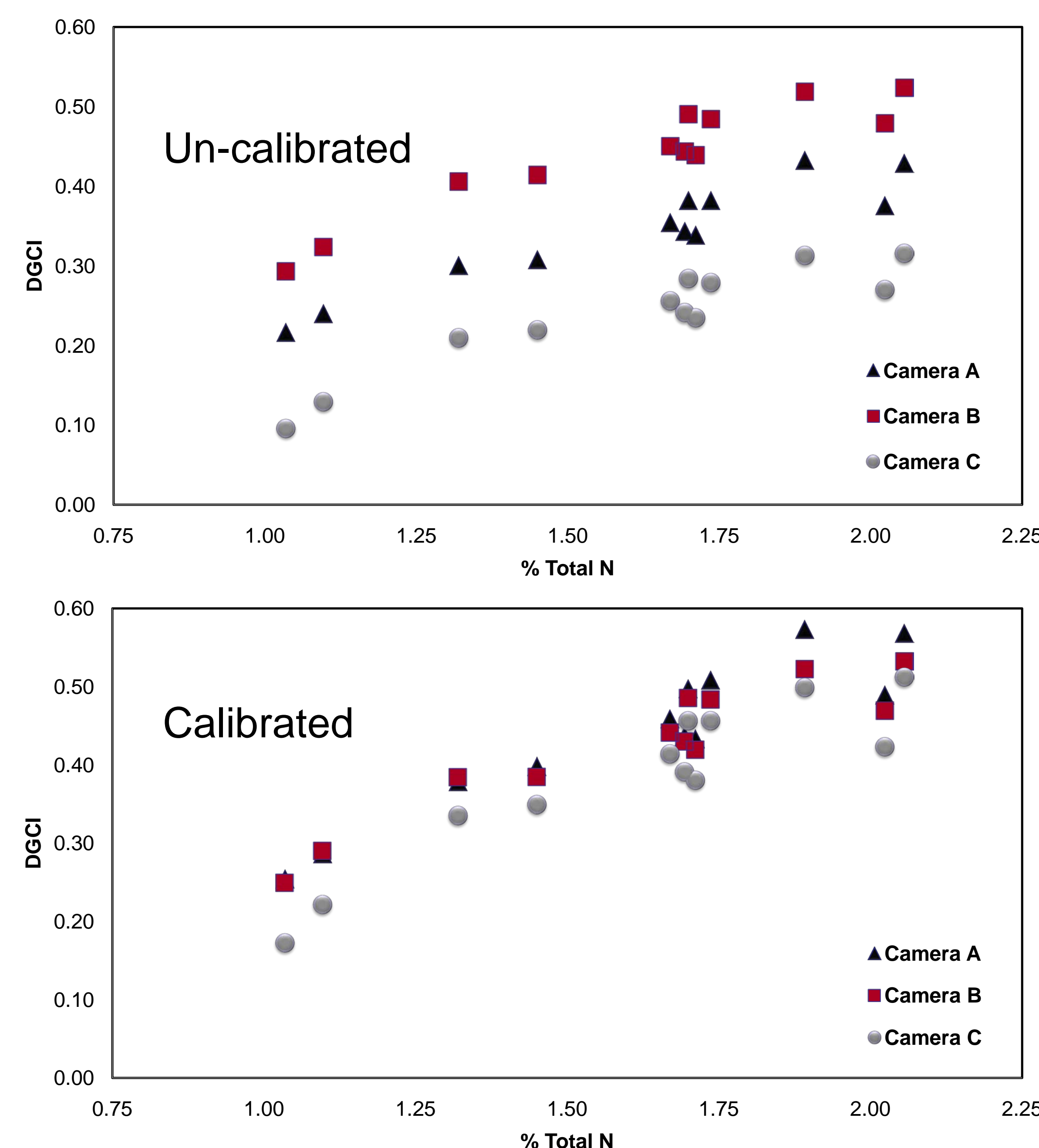


Figure 3. Internal standards with known Hue, Saturation, and Brightness values (seen in fig 1.) are included in images and can be used for camera calibration. Three different cameras were used in the experiment. The graphs above show the difference in un-calibrated and calibrated images.

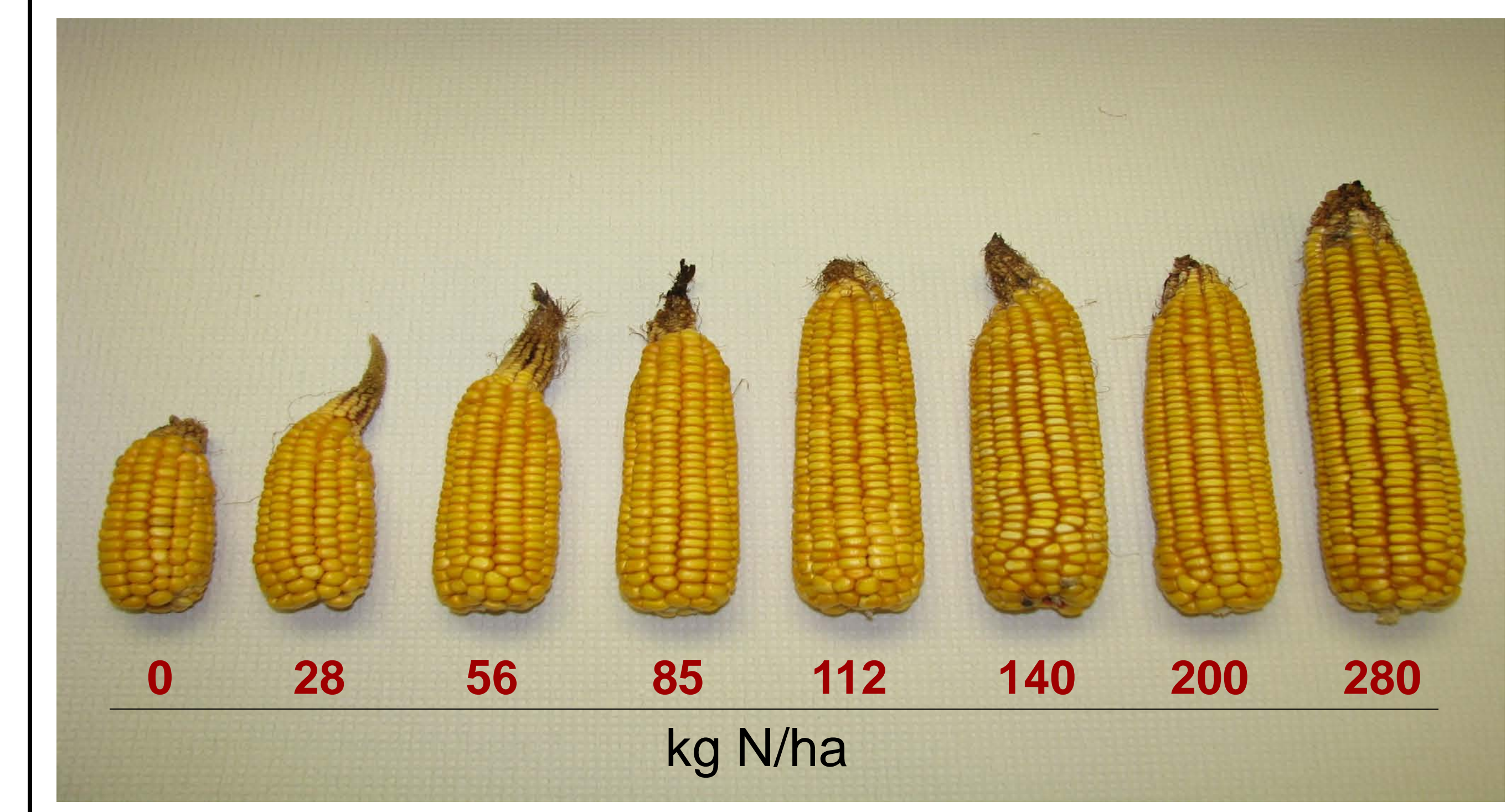


Figure 4. Response of corn ears to increasing N fertilizer.

## Conclusions

- DGCI and SPAD measurements were closely related ( $r^2=0.82$ ).
- Yield increased as leaf N concentration increased.
- Total Leaf N agreed well with DGCI ( $r^2=0.74$ ) and SPAD ( $r^2=0.67$ ).
- Yield was linearly associated with DGCI ( $r^2=0.81$ ) and SPAD ( $r^2=0.83$ ).
- Including standard color disks in images enabled cameras of varying quality to yield nearly identical DGCI values.
- DGCI offers a quick inexpensive method to evaluate corn N status.

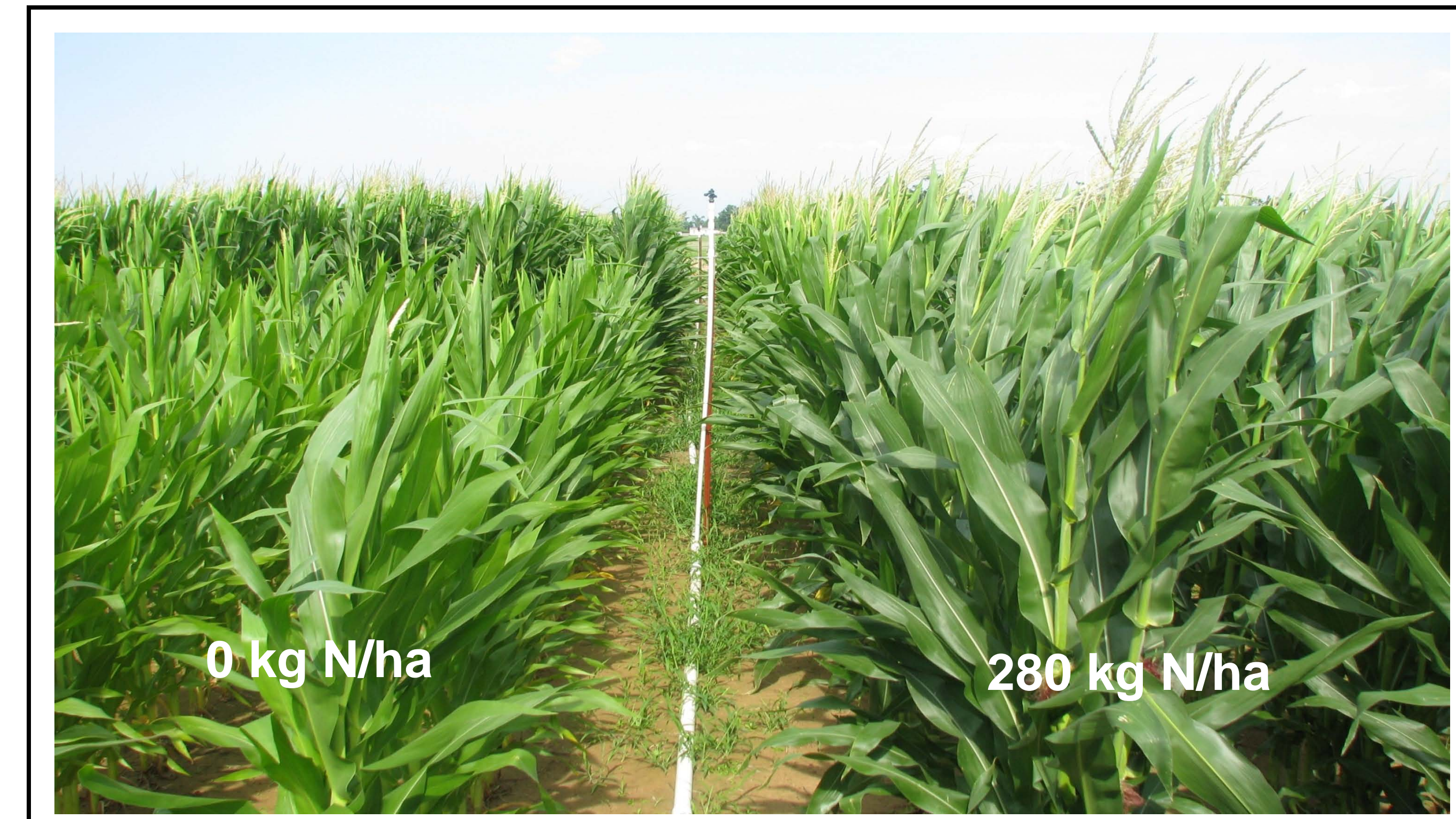


Figure 5. Nitrogen deficient corn on the left is lighter green than the N sufficient corn on the right.

## References

- Karcher, D.E., and M.D. Richardson. 2003. Quantifying turfgrass color using digital image analysis. *Crop Sci.* 43:943-951.
- Zhang, J., A.M. Blackmer, J.W. Ellsworth, and K.J. Koehler. 2008. Sensitivity of chlorophyll meters for diagnosing nitrogen deficiencies of corn in production agriculture. *Agron. J.* 100:543-550.

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