

## Investigation of Chlorophyll and Nitrogen Contents Using Non-Invasive Method in Common Hazel (*Corylus avellana* L.) Leaves

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**Abstract:** Over the last decade, technological developments have it possible to quickly and non-destructively, the Chlorophyll (CHL) status of plants more recently, non-destructive optical methods, based on the absorbance and/or reflectance of light by the intact leaf have been developed. The objective of this study was to evaluate the ability of a portable chlorophyll meter to estimate chlorophyll and nitrogen in common hazel (*Corylus avellana* L.). Significant correlations were observed between (CCI) and (N) ( $p \leq 0.001$ ,  $r^2 = 60\%$ ). Total chlorophyll content ranged from 0.10-0.66  $\mu\text{g mm}^{-2}$  and Nitrogen ( $N_{\text{dw}}$ ) values ranged from 1.1-3.4%.

**Key words:** *Corylus avellana*, nitrogen, portable chlorophyll meter, Fandogluo forest, Iran

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### INTRODUCTION

Traditionally, wet chemical methods have required chlorophyll extraction in a solvent followed by the spectrophotometric determination of absorbance by chlorophyll solution and conversion from absorbance to concentration using standard published equations. more recently, non-destructive optical methods based on the absorbance and/or reflectance of light by the intact leaf have been developed.

Estimation of total chlorophyll and nitrogen content is a potentially important application for both forest managers and researchers. Significant correlations between total foliar extractable chlorophyll and Chlorophyll Content Index values (CCI) obtained with portable chlorophyll meters have been reported for a number of agricultural species including cabbage, cotton and pea (Marquard and Tipton, 1987), sorghum and pigeonpea (Yamamoto *et al.*, 2002) muskmelon (Fahrurozi *et al.*, 2001), several fruit tree species (Schaper and Chacko, 1991) and several forest tree such as paper birch (Richardson *et al.*, 2002) and apple (*Malus domestica* Brorhck.) (Campbell *et al.*, 1990), red maple (*Acer rubrum* L.) (Sibley *et al.*, 1996) and sugar maple (Van den Berg and Perkins, 2004).

### MATERIALS AND METHODS

**Site study:** The fandoughluo forest was selected in order to estimate chlorophyll and nitrogen contents in common hazel leaves. This site study was located in Namin forest in Northwestern of Iran. With 38°26'55" Northern latitude and 47°36'18" East longitude located in from 1380-1440 m.a.s.l. the study area experienced with annual

precipitation is 338.9 mm and the average temperature during the coldest duration is 3.6°C as well as the average temperature during hottest month is 15.5°C. Relative air humidity ranging from 51.4% in summer and 83.1% in winter (Anonymous, 2008).

**Sampling and variable measurement:** Samples were collected in 2010 with in 0.05 ha of the fandoughluo forest (Elevation 1400 m). About 50 individual, visually-healthy common hazel leaves were taken randomly from both understorey and open-grown saplings. Following collection, samples were placed in plastic bags and stored in a refrigerator until further analyses could be completed.

Five chlorophyll content measurements were collected from each leaf with the CCM-200 portable chlorophyll meter (Opti-Sciences, Tyngsboro, Ma) which calculates a unitless Chlorophyll Content Index (CCI) value from the ratio of optical absorbance at 655 nm to that at 940 nm. These data yielded an average CCI for each leaf sample. Immediately following collection of (CCI) values, leaf samples were dried in an oven at 70°C (Van den Berg and Perkins, 2004). Nitrogen content at a percentage of dry weight ( $N_{\text{dw}}$ ) was determined and simple linear regression was used to determine the relationship between  $N_{\text{dw}}$  and CCI. Research examining the utility of chlorophyll meters to assess North American forest tree species has been somewhat limited. Significant correlations have been reported for CCI and total chlorophyll in paper birch (*Betula papyrifera* Marsh.) (Richardson *et al.*, 2002) red maple (*Acer rubrum* L.) (Sibley *et al.*, 1996) and sugar maple (*Acer Saccharum* Marsh.) (Van den Berg and Perkins, 2004). Cate and Perkins (2004) reported significant correlations between CCI and total chlorophyll in sugar maple leaves collected during the fall coloration period.

(Van den Berg and Perkins, 2004) were observed significant correlation between CCI and N. Data indicate that the meter tested can be an effective tool for estimating relative chlorophyll and nitrogen content in sugar maple leaves. Thus, the objective of this study was to establish the ability of a portable chlorophyll meter to estimate total chlorophyll and nitrogen content in large heterogeneous samples of common hazel (*Corylus avellana* L.).

## RESULTS AND DISCUSSION

Nitrogen ( $N_{dw}$ ) values ranged from 1.1-3.4% (Fig. 1). The relationship between CCI and ( $N_{dw}$ ) was significantly linear ( $p < 0.001$ ). Correlation analysis indicates 60% of the variation in N was predicted by CCI (Fig. 1). Relationships between CCI and N were found to be explained by differences in leaf thickness, relationships between CCI

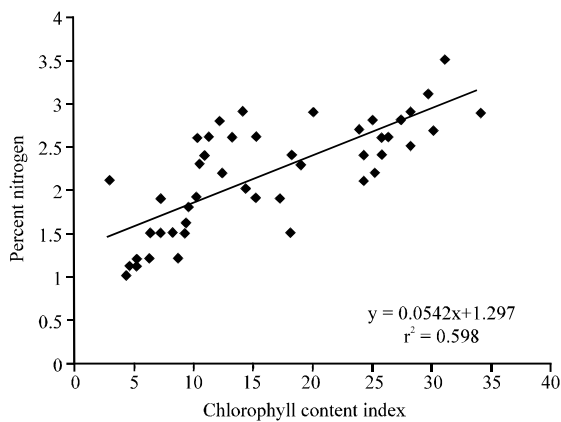


Fig. 1: Linear regression of Chlorophyll Content Index (CCI) with total percent nitrogen (by dry weight) in 50 common hazel leaves

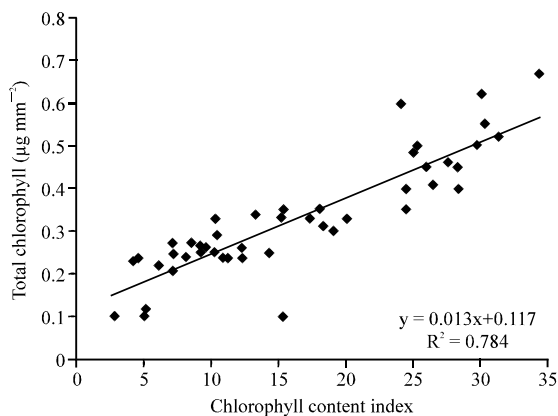


Fig. 2: Linear regression of Chlorophyll Content Index (CCI) with total extractable chlorophyll ( $\mu\text{g mm}^{-2}$ ) in 50 common hazel leaves

and N were found to be explained by differences in leaf thickness, Chong and Robison (2003) also found that examining N on an area basis improved correlation coefficients both within and across dates in sweetgum (*Liquidambar styraciflua* L.).

Total extractable chlorophyll values from 50 samples ranged from 0.10-0.66  $\mu\text{g mm}^{-2}$  with a mean of 0.33 ( $\pm 0.31$ ). These values are within the published range for sugar maple leaves (Van den Berg and Perkins, 2004) CCI ranged from 1.4-2.6%. The relationship between total extractable chlorophyll and CCI was significantly linear with  $r^2$  indicating that 78% ( $p < 0.001$ ) of variation was explained by a linear model (Fig. 2).

## CONCLUSION

These data showed that the noninvasive optical method all provided reliable estimates of relative leaf chlorophyll.

## ACKNOWLEDGEMENT

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## REFERENCES

- Anonymous, 2008. Climatological data. Climatology station of Namin Area, Ardabil province, Iran.
- Campbell, R.J., K.N. Mobley, R.P. Marini and D.G. Pfeiffer, 1990. Growing conditions alter the relationship between SPAD-501 values and apple leaf chlorophyll. Hortscience, 25: 330-331.
- Cate, T.M. and T.D. Perkins, 2004. Chlorophyll content monitoring in sugar maple (*Acer saccharum*). Tree Physiol., 23: 1077-1079.
- Chong, S.X. and D.S. Robison, 2003. Nondestructive and rapid estimation of hardwood foliar nitrogen status using the SPAD-502 chlorophyll meter. Forest Ecol. Manage., 181: 331-338.
- Fahrurrozi, A., A. Katrine and Stewart, 2001. Relationships between extractable chlorophyll and SPAD values in muskmelon leaves. J. Plant Nutr., 24: 961-966.
- Marquard, R.D. and J.I. Tipton, 1987. Relationship between extractable chlorophyll meters: a comparison of two types of meters. Hort Sci., 27: 69-71.
- Richardson, A.D., S.P. Duigan and G.P. Berlyn, 2002. An evaluation of noninvasive methods to estimate foliar chlorophyll content. New Phytologist, 153: 185-194.

- Schaper. H. and E.K. Chacko, 1991. Relation between extractable chlorophyll and portable chlorophyll meter readings in leaves of eight tropical and subtropical fruit-tree species. *J. Plant Physiol.*, 138: 674-677.
- Sibley, J.L., D.J. Eakes, C.H. Gilliam, G.J. Keever, W.A. Dozier and D.G. Himelrick, 1996. Foliar SPAD-502 meter values, nitrogen levels and extractable chlorophyll for red maple selections. *HortScience*, 31: 468-470.
- Van den Berg, A.K. and T.D. Perkins, 2004. Evaluation of a portable chlorophyll meter to estimate chlorophyll and nitrogen contents in sugar maple (*Acer saccharum* Marsh). *Leaves. Forest Ecol. Manage.*, 200: 113-117.
- Yamamoto, A., T. Nakamura, J.J. Adu-Gyamfi and M. Saigusa, 2002. Relationship between chlorophyll content in Leaves of sorghum and pigeonpea determined by extraction methods and by chlorophyll meter (SPAD-502). *J. Plant Nutr.*, 25: 2295-2301.