

**DECLARATION**

**DEVELOPING A CALIBRATION CURVE TO  
QUANTIFY CHLOROPHYLL CONTENT IN  
*Hevea brasiliensis***

A dissertation submitted to the  
Faculty of Animal Science and Export Agriculture  
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## ABSTRACT

Chlorophyll meter, SPAD-502 is widely used in determination of leaf chlorophyll content in field plants as it is non-destructive and less time consuming. Variations in chlorophyll content among varieties of different species have been identified. However, investigations of any genotypic variations related to rubber have not been reported so far. In order to convert relative SPAD meter values into units of absolute chlorophyll concentration, it is necessary to employ a calibration curve. Leaf characteristics such as leaf thickness, epicuticular wax content, water content and leaf biomass varies among the genotypes thus it could affect the readings of the SPAD-502 chlorophyll meter. Therefore, this study was carried out to develop a calibration curve to quantify chlorophyll content and effect of leaf thickness, epicuticular wax content, water content and leaf biomass on the readings of SPAD-502 meter.

Most commonly grown four rubber genotypes i.e. RRIC 121, RRIC 100, RRISL 203 and RRISL 2001 were selected for the study. Leaf samples of each genotype that in the similar growth stages were collected from the budwood nurseries in Dartonfield estate, Agalawatta. SPAD values of each leaf sample were taken and used to analyse the actual chlorophyll content by acetone extraction method. Leaf thickness, water content, epicuticular wax content and leaf biomass of each leaf sample were determined by using standard test methods.

Standard calibration curves were developed separately for each genotype on per leaf biomass (mg/g) and per leaf area (mg/cm<sup>2</sup>) basis by regressing with correspondent SPAD values. Most appropriate calibration model for all tested genotypes was the second order polynomial with above 95 % accuracy ( $R^2 = 0.95$ ). Highest leaf thickness and epicuticular wax content were observed in RRIC 100 genotype whilst RRISL 203 genotype gave the lowest values. Highest water content and leaf biomass were observed in RRIC 121 and RRISL 2001, respectively.

Leaf chlorophyll content of RRIC 121 genotype was comparatively lower with averages of 3.598 mg/g and 0.036 mg/cm<sup>2</sup>. However, highest leaf chlorophyll content was observed in RRISL 203 genotype and the average values were 5.238 mg/g and 0.054 mg/cm<sup>2</sup>. Despite of the genotype, average leaf epicuticular wax content, water content and leaf biomass was 0.079 mg/cm<sup>2</sup> and 7.998 mg/cm<sup>2</sup>, 10.889 mg/cm<sup>2</sup>, respectively whilst leaf thickness was 0.126 mm.

Correlation between leaf thickness, epicuticular wax content, leaf biomass and water content with the reading of SPAD-502 chlorophyll meter were not significant ( $p=0.05$ ) in all four genotypes and revealed that these four parameters did not affect the reading of SPAD-502 chlorophyll meter in determination of leaf chlorophyll content rubber.

*Key Words:* Rubber, Chlorophyll content, Epicuticular wax, Leaf biomass, Leaf thickness, SPAD-502 Chlorophyll meter