# Analysis Of Chlorophyll "A" And "B" In Randomly Selected Varieties Of Sugarcane Leaves Using Multivariate Analysis Of Variance. 

Abubakar Usman<br>Department of Mathematics and Statistics, Federal University of Technology; Minna; Nigeria


#### Abstract

Multivariate data needed to make rigorous probability statements about the effect of factors and their interaction in experiments. Multivariate statistical methods provide a useful test for analysis of rigorous data sets, because many variables integrated in one analysis. Chlorophyll " $a$ " and " $b$ " are indeed essential for the growth and development of sugarcane stalk. This paper presents multivariate analysis of variance of the chlorophyll content of the randomly selected varieties of sugarcane leaves. The data set obtained from a study conducted at National Cereal Research Institute (NCRI,) Badeggi in Niger State, Nigeria. The results show that there are statistical significant differences in both the seasons and the varieties of the sugarcane leaves in the chlorophyll contents.


Keywords: Chlorophyll, Porphyry ring, Manova, Seasons, Biological, treatments and Pigment.

## I. Introduction

Multivariate analysis is a branch of statistics dealing with procedures for summarizing, representing, and analyzing multiple quantitative measurements obtained on a number of individuals or objects. The statistical tool for studying differences between means on some particular variable of distinct groups of subject often referred to as Analysis of variance (ANOVA). An observation arising from Specific levels or combinations of level of a number of independent random variables, may be thought of being normally identically and independently distributed with mean zero and constant variance $\sigma^{2}$ these are the basic condition required for univariance ANOVA. But where the single response variables is replaced by several response variables with the same conditions as in the univariate case, then multivariate analysis of variance (MANOVA) becomes a better alternative to the use of separate ANOVA model is strengthened, when the pair wise correlation coefficient between the contending response variables.Joseph (2010) states that multivariate analysis of variance is a statistical technique that explore the relationship between several categorical independent variables (usually referred to as treatments ) or two or more dependent variables. As such, it represents an extension of univariate analysis of variance. Richard (1988) further emphases that multivariate analysis of variance (MANOVA) is used by researchers in Biological, Physical and Social sciences frequently in measurements on several variables. The need to understand the relationships between many variables make multivariate analysis an inherently difficult often the human mind is overwhelm by the sheer bulk of the data. In particular more of matrix algebra is required in the various multivariate statistical than in univariate settings.

Chlorophyll is the pigment found in plants, some algae, and some bacteria that gives them their green color and that absorbs the light necessary for photosynthesis. Chlorophyll absorbs mainly violet-blue and orange-red light, the great abundance of chlorophyll in leaves and its occasional presence in other plant parts to appear green. Chlorophyll is a large molecule composed mostly of carbon and hydrogen. At the centre of the molecule, a single atom of magnesium surrounded by a nitrogen- containing group of atoms called a Porphyry ring.

Peter (2009) states that the structure of chlorophyll somewhat resembles that of the active constituent of hemoglobin in the blood. A long chain of carbon and hydrogen atoms proceeds from this central core and attaches the chlorophyll molecule to the inner membrane of the chloroplast, the cell organelle in which photosynthesis takes place.

Several kinds of chlorophyll exist but they differ from each other in details of their molecular structure and absorb slightly different wavelengths of light. In this paper, the most common type chlorophyll 'a' and 'b'. Chlorophyll ' $a$ ' is making up about $75 \%$ of the chlorophyll in green plants. It found in Cyanobacteria (formerly known as blue-green algae) and in complex photosynthetic cells, while chlorophyll ' $b$ ' is an accessory pigment present in plants and other complex photosynthetic cells. It absorbs light energy of a different wavelength and transfers it to chlorophyll ' $a$ ' for ultimate conversion to chemical energy.

## II. Test Statistics for Multivariate Analysis of Variance

There are commonly four test statistics used in multivariate analysis of variance, these statistics are detail below.
2.1 Pallai - Bartlett trace: - Pillai trace expressed by the equation $V=\sum_{i=1}^{n} \frac{\lambda_{i}}{\left(1+\lambda_{i}\right)}$ in which $\lambda$ represents the Eigen values for each of the discriminant variates, and $n$ represents the number of variates. Pillai's trace is the sum of explained variances on the discriminate variants which are the variables which are computed based on the canonical coefficients for a given sets of roots, therefore a large value by convention indicates significant difference and is similar to the ratio of explained variance to total variance $\left(\frac{S S_{H}}{S S_{T}}\right)$.
2.2 Hotelling's trace:-The Hotelling-Lawley trace is the sum of Eigen values for each variate and computed by the equation $\boldsymbol{T}=\sum_{i=1}^{n} \lambda_{i}$. This is the most common traditional test for two independent groups. This statistic represents the proportion of explained variance to unexplained variance $\left(\frac{S S_{H}}{S S_{E}}\right)$ for each of the variates and so it compares directly to the F ration in ANOVA.
2.3 Wilks's lambda ( $\Lambda$ ): - The product of the unexplained variance on each of the variates. $\Lambda=\prod_{i=1}^{n} \frac{1}{1+\lambda_{i}}$ The symbol ( $\Pi$ ) is similar to the summation symbol ( $\Sigma$ ) except that it means multiply rather than add up. This is the most common traditional test when there are more than two groups formed by the independent variables. It is a measure of the differences between groups of the centriod (vector) of means on the independent variables. The smaller the lambda ( $\boldsymbol{\Lambda}$ ) the greater the differences, the Bartlett's transformation of lambda ( $\boldsymbol{\Lambda}$ ) is then used to compute the significance of the lambda. The t- test, Hotellings $T^{2}$, and the F-test are special cases of Wilks's Lambda. It represents the ration of error variance to total variance $\left(\frac{S S_{E}}{S S_{T}}\right)$ for each variate.
2.4 Roy's greatest characteristic roots: - Roy's largest root is the Eigen value for the first variate. In a sense according to Michael (2010), it is the same as the Hotelling-Lawley trace, except for the first variate only. This statistic represents the proportion of explained variance to unexplained variance $\left(\frac{S S_{H}}{S S_{E}}\right)$ for the first distriminant function. This is similar to the Pillai Bartlett trace, but based only on the first (and hence most important) root. Roy's largest root sometimes equated with the largest Eigen value. This value is conceptually the same as the F- ratio in univariate ANOVA and represents the maximum possible between-group difference given the data collected. The test statistic is less robust than the other tests in the face of violations of the assumption of multivariate normality.

Timm (1975) states that employing any of the above test statistics, the same conclusion reached concerning the acceptance of null hypothesis. One of the most important considerations in selecting a test statistic is the power that it has to detect that the alternative hypothesis is true. A test statistic should be invariant to scaling transformations. This simply means that there should be the same result whatever the unit(s) in which our measurements taken. In Multivariate test there is no one uniformly most powerful test which is invariant to all transformations. Which test is best in the sense of being most powerful depends on the nature of the departure from the null hypothesis (Hand and Taylor. 1987). The lack of a uniformly most powerful test is one of the reason why several different test statistics were been proposed. For Multivariate, test statistics this power depends on the way in which the group mean vectors in the underlying population depart from the null hypothesis of equality.

Statistical tables are not available for the above test statistics. However, each of the above test statistics has an F approximation. The table 2.1 below gives details of the F approximations for the four test statistics commonly used in multivariate analysis of variance.

Table 2.1 Test Statistics used to compare sample mean vectors with Approximate F-tests for Evidence that the population values are not constant

| Test | Statistic | F | $d_{\text {d }}$ | $\boldsymbol{d f}_{2}$ | Comment |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Pillai's trace | V | $(\mathrm{n}-\mathrm{m}-\mathrm{p}+\mathrm{s}) \mathrm{V}(\mathrm{d}(\mathrm{s}-\mathrm{V}))^{-1}$ | Sd | $\begin{gathered} S(\mathrm{n}-\mathrm{m}- \\ \mathrm{p}+\mathrm{s}) \end{gathered}$ | $\begin{aligned} & \qquad d=\max (p, m-1) \\ & s=\min (p, m-1)= \\ & \text { number of positive eigen values } \end{aligned}$ |
| Wilks's <br> Lambda | $\Lambda$ | $\left.\left\{\left(1-\Lambda^{\frac{1}{\mathrm{t}}}\right) \Lambda^{-\frac{1}{\mathrm{t}}}\right)\left(\mathrm{df}_{2} \mathrm{xdf}_{1}^{-1}\right)\right\}$ | $\mathrm{P}(\mathrm{m}-1)$ | $\begin{aligned} & \mathrm{wt} \\ & -\left(\mathrm{df}_{1} \mathrm{x} 0.5\right) \\ & +1 \end{aligned}$ | $\begin{aligned} & \mathrm{w}=\mathrm{n}-1(\mathrm{p}+\mathrm{m}) \times 0.5 \\ & \mathrm{t}=\left[\left(\mathrm{df}_{1}^{2}-4\right) \mathrm{x}\left(\mathrm{p}^{2}+(\mathrm{m}-1)^{2}-5\right)^{-1}\right]^{0.5} \\ & \text { if } d f_{1}=2, \text { set } t=1 \end{aligned}$ |
| Hotelling's Trace | U | $\mathrm{df}_{2} U x\left(\mathrm{sdf}_{1}\right)^{-1}$ | $\begin{gathered} \mathrm{S}(2 \mathrm{~A}+\mathrm{s} \\ +1) \end{gathered}$ |  | $s$ is as for Pillai'strace $\begin{gathered} A=(\|m-p-1\|-1) \times 0.5 \\ B=(n-m-p-1) \times 0.5 \end{gathered}$ <br> The significance level obtained is a lower bound |
| Roy's Largest Root | $\lambda_{1}$ | $\left(\mathrm{df}_{2} \mathrm{xdf}_{1}^{-1}\right) \lambda_{1}$ | $d$ | $\begin{aligned} & 2(\mathrm{sB}+1) \\ & \mathrm{n}-\mathrm{m}-\mathrm{d} \\ & -1 \end{aligned}$ | $\begin{aligned} & \quad d=\max (p, m-1) \\ & s=\min (p, m-1)= \\ & \text { number of positive eigen values } \end{aligned}$ |

It is assumed that there are p variables in m samples, with the $j$ th of size n , and a total sample size of $\mathrm{n}=\sum n_{i}$. These are approximations for general p and m .

## III. Objectives of the Study

This paper has the following objectives
(i) To determine chlorophyll 'a' and 'b' contributions in the growth of Sugarcane leaves.
(ii) To determine the variability of Chlorophyll ' $a$ ' and ' $b$ ' before Harvest and before the dry season on sugarcane leaves development.
(iii)

## Hypothesis

The paper will investigate the following hypotheses
$\boldsymbol{H}_{01}$ : There is no significant difference between the chlorophyll ' $a$ ' and ' $b$ ' in sugarcane leaves.
$\boldsymbol{H}_{02}$ : There is no significant difference between the chlorophyll ' a ' and ' b ' content of sugarcane leaves both before harvest and before the dry season.

## IV. Methodology

The data for this study obtained from an experiment conducted by the research unit of the National cereal Research Institute, Badeggi in Niger state. The experiments conducted to determine the chlorophyll content of sugarcane leaves; forty-four different variables of sugarcanes of interest and chlorophyll content ' $a$ ' and ' $b$ ' determined by the spectrometer. However, the experimental data utilized in this paper obtained by exploring the relations given below

$$
\begin{array}{rl}
\text { Chlorophyll } a=10.3 k_{1}-0.918 k_{2} & 4.1 \\
\text { Chlorophyll } b=19.7 k_{2}-3.87 k_{1} & 4.2
\end{array}
$$

The values $10.3 ; 0.918 ; 19.7$ and 3.87 in equations (4.1) and (4.2) fixed values for the measuring instrument, while $k_{1}$ and $k_{2}$ are the different values obtained from the reading of the various varieties.
The values obtained are for both before harvest and before the dry season as shown in the table below.
Table 4.1 Data for Chlorophyll content of sugarcane before dry season and before harvest.

| Before Harvest |  |  | Before Dry Season |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Varieties | Chlorophyll 'a' | Chlorophyll 'b' | Varieties | Chlorophyll 'a' | Chlorophyll 'b' |
| 1 | 16.64438 | 6.13349 | 1 | 8.51575 | 8.14247 |
| 2 | 12.04794 | 7.04759 | 2 | 12.60603 | 11.79280 |
| 3 | 16.68793 | 5.63325 | 3 | 19.37095 | 24.71574 |
| 4 | 9.02666 | 9.55698 | 4 | 9.34612 | 13.55958 |
| 5 | 19.19261 | -0.34150 | 5 | 7.48849 | 10.42524 |
| 6 | 13.70778 | 5.08844 | 6 | 18.84548 | 20.79054 |
| 7 | 17.45160 | 5.84746 | 7 | 17.54577 | 18.27884 |
| 8 | 11.66920 | 4.51662 | 8 | 22.65090 | 27.29632 |
| 9 | 15.52033 | 4.19449 | 9 | 17.72731 | 16.12028 |
| 10 | 17.52524 | 10.46710 | 10 | 14.45494 | 20.54339 |
| 11 | 23.54890 | 13.91473 | 11 | 21.72030 | 19.90394 |
| 12 | 11.10513 | 7.51796 | 12 | 18.81306 | 20.18336 |
| 13 | 20.87096 | 3.38414 | 13 | 9.49471 | 13.19407 |
| 14 | 19.63494 | 5.80341 | 14 | 16.80501 | 15.49906 |
| 15 | 13.89727 | 3.41077 | 15 | 18.84548 | 20.79054 |
| 16 | 10.93067 | 4.31250 | 16 | 13.33474 | 15.69970 |
| 17 | 11.83656 | 4.41730 | 17 | 14.24391 | 13.34517 |
| 18 | 11.33544 | 3.44280 | 18 | 12.29062 | 10.09193 |
| 19 | 15.04786 | 8.68821 | 19 | 14.47703 | 14.20598 |
| 20 | 15.31638 | 8.35506 | 20 | 16.09545 | 14.00435 |
| 21 | 25.62749 | 16.19071 | 21 | 14.76256 | 15.23971 |
| 22 | 14.62132 | 4.37745 | 22 | 23.84658 | 19.87924 |
| 23 | 13.04019 | 8.33931 | 23 | 18.07946 | 15.07828 |
| 24 | 13.60070 | 4.78028 | 24 | 27.55005 | 23.57813 |
| 25 | 24.03971 | 22.03251 | 25 | 18.99076 | 24.83923 |
| 26 | 18.24556 | 6.51899 | 26 | 30.45385 | -2.03580 |
| 27 | 14.36228 | 5.81028 | 27 | 15.87175 | 9.24963 |
| 28 | 20.25579 | 6.59596 | 28 | 21.23091 | 29.32019 |
| 29 | 13.04390 | 4.35077 | 29 | 11.94338 | 21.23544 |
| 30 | 16.95063 | 6.07649 | 30 | 21.41468 | 18.64456 |
| 31 | 11.20387 | 4.96470 | 31 | 6.15960 | 8.75677 |
| 32 | 6.46528 | 4.15154 | 32 | 12.72217 | 12.77498 |
| 33 | 16.12325 | 18.61977 | 33 | 1.54226 | 8.38193 |
| 34 | 18.92907 | 5.33433 | 34 | 28.98964 | 29.38570 |


| 35 | 9.23473 | 6.74327 | 35 | 9.17279 | 10.54725 |
| :---: | :---: | :---: | :---: | :---: | :---: |
| 36 | 6.23183 | 10.58313 | 36 | 11.71661 | 17.19801 |
| 37 | 10.46207 | 5.03051 | 37 | 16.60388 | 18.72951 |
| 38 | 14.70595 | 7.75177 | 38 | 15.13895 | 6.95074 |
| 39 | 10.74219 | 7.05432 | 39 | 14.00547 | 17.77026 |
| 40 | 14.28608 | 7.44538 | 40 | 11.32233 | 9.37186 |
| 41 | 15.77290 | 1.81571 | 41 | 29.59185 | 16.46250 |
| 42 | 13.51417 | 10.32899 | 42 | 28.73444 | 34.86230 |
| 43 | 17.50922 | 23.18941 | 43 | 28.42263 | 30.04391 |
| 44 | 12.16054 | 5.28268 | 44 | 13.21870 | 17.54341 |

Source: - National Cereal Research Institute, Badeggi, Niger State, Nigeria (2013)

## V. Data presentation and analysis

The measurement obtained in table 4.1 is the data in respect of chlorophyll ' $a$ ' and ' $b$ ' verified for consistency with the equations 4.1 and 4.2 before the analysis were carried out. Multivariate tests procedure are implemented for the data that were obtained at two different periods namely before the dry season and before harvest to demonstrate their suitability. Correlation analysis was used in this study to examine whether the two types of chlorophyll namely chlorophyll ' $a$ ' and ' $b$ ' are correlated. The table 5.1 below represents the resulting multivariate analysis of variance obtained from SPSS output as in Appendix 1.

Table 5.1 Multivariate analysis of variance for chlorophyll ' $a$ ' and ' $b$ ' content and Periods before dry season and before harvest of sugarcane leaves

| Source of variation | Degree of freedom | Sum of squares and cross product |
| :---: | :---: | :---: |
| Seasons (Before dry season \& before Harvest) | 1 | $B^{*}=\left(\begin{array}{cc}76.467 & 389.304 \\ 389.304 & 1982.003\end{array}\right)$ |
| Treatment (Varieties) | 43 | $B^{* *}=\left(\begin{array}{cc} 1679.349 & 847.125 \\ 847.125 & 1920.115 \end{array}\right)$ |
| Residual (Error) | 43 | $W=\left(\begin{array}{cc}1043.924 & 593.100 \\ 593.100 & 1285.025\end{array}\right)$ |
| Total | 87 | $T=\left(\begin{array}{ll}2799.740 & 1829.529 \\ 1829.529 & 5187.143\end{array}\right)$ |

To determine the characteristic roots of the equation $\left|B^{*} W^{-1}-I \lambda\right|=0$; the value of the inverse of the matrix W will be calculated and the product of the matrices $\mathrm{B}^{*}$ and $\mathrm{W}^{-1}$, then the determinant.
Thus, $W^{-1}=\frac{1}{|W|}\left[\begin{array}{rr}1285.025 & -593.100 \\ -593.100 & 1043.924\end{array}\right]$

$$
\text { i.e. }|W|=\left|\begin{array}{cr}
1043.924 & 593.100 \\
593.100 & 1285.025
\end{array}\right|=1043.924 \times 1285.025-593.100 \times 593.100
$$

i.e. $W^{-1}=\frac{1}{989,700.8281}\left[\begin{array}{cc}1285.025 & -593.100 \\ -593.100 & 1043.924\end{array}\right]=\left[\begin{array}{cc}0.0012984 & -0.0005993 \\ -0.0005993 & 0.0010548\end{array}\right]$

Thus, $B^{*} W^{-1}=\left[\begin{array}{cc}76.467 & 389.304 \\ 389.304 & 1982.003\end{array}\right]\left[\begin{array}{cc}0.0012984 & -0.0005993 \\ -0.0005993 & 0.0010548\end{array}\right]$
$A_{11}=76.467 x 0.0012984+389.304 x(-0.0005993)=-0.1340$
$A_{12}=76.467 x(-0.0005993)+389.304 x 0.0010548=0.3648$
$A_{21}=389.304 x 0.0012984+1982.003 x(-0.0005993)=-0.6823$
$A_{22}=389.304 x(-0.0005993)+1982.003 x(0.0010548)=1.8573$
Then, $\quad B^{*} W^{-1}=\left[\begin{array}{cc}A_{11} & A_{12} \\ A_{21} & A B_{22}\end{array}\right]=\left[\begin{array}{cc}-0.1340 & 0.3648 \\ -0.6823 & 1.8573\end{array}\right]$
The characteristics root obtained from the solution of the equation: $\left|B^{*} W^{-1}-I \lambda\right|=0$

$$
\begin{gathered}
\left|B^{*} W^{-1}-I \lambda\right|=\left|\begin{array}{cc}
-0.1340-\lambda & 0.3648 \\
-0.6823 & 1.8573-\lambda
\end{array}\right|=(-0.1340-\lambda)(1.8573-\lambda)-(0.3648 x(-0.6823)) \\
=-0.24890-1.7233 \lambda+\lambda^{2}+0.24890
\end{gathered}
$$

i.e. $\lambda^{2}-1.7233 \lambda=0 \quad$ (1) is a quadratic equation in $\lambda$

Solving equation(1) gives the values of $\lambda^{\prime}$ s to be $\lambda_{1}=1.7233$ and $\lambda_{2}=0.000$.
Similarly, for the varieties; $B^{* *} W^{-1}=\left[\begin{array}{cc}1679.349 & 847.125 \\ 847.125 & 1920.115\end{array}\right]\left[\begin{array}{cc}0.0012984 & -0.0005993 \\ -0.0005993 & 0.0010548\end{array}\right]$
$B_{11}=1679.349 \times 0.0012984+847.125 x(-0.0005993)=1.6728$
$B_{12}=1679.349 x(-0.0005993)+847.125 x 0.0010548=-0.1129$
$B_{21}=847.125 x 0.0012984+1920.115 x(-0.0005993)=-0.05080$
$B_{22}=847.125 x(-0.0005993)+1920.115 x(0.0010548)=1.5177$

Therefore, $\quad B^{* *} W^{-1}=\left[\begin{array}{ll}B_{11} & B_{12} \\ B_{21} & B_{22}\end{array}\right]=\left[\begin{array}{cc}1.6728 & -0.1129 \\ -0.05080 & 1.5177\end{array}\right]$
The characteristics root obtained from the solution of the equation: $\left|B^{*} W^{-1}-I \lambda\right|=0$.

$$
\begin{gathered}
\left|B^{* *} W^{-1}-I \lambda\right|=\left\lvert\, \begin{array}{cc}
1.6728-\lambda & -0.1129 \\
-0.05080 & \left.\begin{array}{c}
1.5177-\lambda
\end{array} \right\rvert\,=(1.6728-\lambda)(1.5177-\lambda)-(-0.05080 x(-0.1129)) \\
=2.5388-3.1905 \lambda+\lambda^{2}-0.0057
\end{array}\right.,
\end{gathered}
$$

i.e. $\lambda^{2}-3.1905 \lambda+2.5331=0 \quad$ (2) is a quadratic equation in $\lambda$

Solving equation(2) gives the values of $\lambda^{\prime}$ s to be $\lambda_{1}=1.7035$ and $\lambda_{2}=1.4870$.
5.1 Calculation of the MANOVA test Statistics
(i) Seasons: - The value of the test statistics for the Seasons (Before Harvest and before the dry season) are as computed below.
(a) Pillai's trace

$$
V=\sum_{\mathrm{i}=1}^{\mathrm{n}} \frac{\lambda_{\mathrm{i}}}{1+\lambda_{\mathrm{i}}}=\frac{1.7233}{1+1.7233}=\frac{1.7233}{2.7133}=0.6328
$$

(b) Wilks's lambda

$$
\Lambda=\prod_{i=1}^{n} \frac{1}{1+\lambda_{i}}=\frac{1}{1+1.7233}=0.3672
$$

(c) Hotelling's trace
(d) Roy's characteristics root

$$
\lambda_{1}=1.7233
$$

The computed values are the same with the values obtained from the SPSS out in Appendix 1 (see Seasons before Harvest and dry season's components).
(ii) Varieties:-The value of the test statistics for the varieties (treatments) are as computed below.
(a) Pillai's trace

$$
V=\sum_{\mathrm{i}=1}^{\mathrm{n}} \frac{\lambda_{\mathrm{i}}}{1+\lambda_{\mathrm{i}}}=\frac{1.7035}{1+1.7035}+\frac{1.4870}{1+1.4870}=1.2280
$$

(b) Wilks's lambda

$$
\Lambda=\prod_{i=1}^{n} \frac{1}{1+\lambda_{i}}=\frac{1}{1+1.7035} x \frac{1}{1+1.4870}=0.1487
$$

(c) Hotelling's trace
(d) Roy's characteristics root

$$
\begin{gathered}
T^{2}=\sum_{\substack{i=1 \\
\lambda_{1}=1.7035}}^{n} \lambda_{i}=1.7035+1.4870=3.1905 \\
=1.20
\end{gathered}
$$

The computed values are the same with the values obtained from the SPSS out in Appendix 1 (see varieties components).
5.2 Calculation of F- Statistics for the test Statistics

The statistics values obtained above used to calculate the F- values for the four multivariate procedures.
(i) Season's (Before Harvest and Before Dry season)
a) Pillia's trace (V)

Where $n=88 ; m=2 ; p=2, d=2$ and $V=0.6328$

$$
F=\frac{(n-m-p+s) V}{d(s-V)}=\frac{(88-2-2+2) x 0.6328}{2(2-0.6328)}=\frac{54.4208}{2.7344}=19.9023
$$

b) Wilks's Lambda ( $\Lambda$ )

$$
F=\left(\frac{1-\Lambda}{\Lambda}\right)^{\frac{1}{t}}\left(\frac{d f_{2}}{d f_{1}}\right)=\left(\frac{1-0.3672}{0.3672}\right)^{\frac{1}{2}}\left(\frac{170}{2}\right)=111.5837
$$

(i) Hotellings trace hat

$$
F=\frac{d f_{2} U}{d f_{1}}=\frac{168 \times 1.7233}{6}=48.2524
$$

(ii) Roy's Characteristics root hat

$$
F=\left(\frac{d f_{2}}{d f_{1}}\right) \theta_{\max }=\frac{83}{2} x 1.7233=71.5170
$$

The decision rule is to reject the null hypothesis $\mathrm{H}_{01}$ if the computed F is greater than the tabulated $\mathrm{F}_{2}$, $44,0.05=3.20$ at $5 \%$ level of significant. Since the calculated values of the four tests $19.9023,111.5835,48.5837$ and 71.5170 are greater than the tabulated value, we reject the null hypothesis and conclude that there is significant difference in the seasons, that is before harvest and before the dry season the chlorophyll a and b contains in the sugarcane leaves differed.
(ii) Varieties (Treatments)
(a) Pillia's trace (V)

Where $\mathrm{n}=88 ; \mathrm{m}=2 ; \mathrm{p}=2, \mathrm{~d}=2$ and $\mathrm{V}=1.228$

$$
F=\frac{(n-m-p+s) V}{d(s-V)}=\frac{(88-2-2+2) \times 1.228}{2(2-1.2280)}=\frac{105.608}{1.5440}=68.3990
$$

(b) Wilks's Lambda ( $\Lambda$ )

$$
F=\left(\frac{1-\Lambda}{\Lambda}\right)^{\frac{1}{t}}\left(\frac{d f_{2}}{d f_{1}}\right)=\left(\frac{1-0.1487}{0.1487}\right)^{\frac{1}{2}}\left(\frac{170}{2}\right)=203.3784
$$

(c) Hotellings trace

$$
F=\frac{d f_{2} U}{d f_{1}}=\frac{168 \times 3.1905}{6}=89.334
$$

(d) Roy's Characteristics root

$$
F=\left(\frac{d f_{2}}{d f_{1}}\right) \theta_{\max }=\frac{83}{2} 1.7035=70.6952
$$

The decision rule is to reject the null hypothesis $\mathrm{H}_{02}$ if the computed F is greater than the tabulated $\mathrm{F}_{44}$, ${ }_{88,0.05}=1.48$ at $5 \%$ level of significant. The calculated values of the tests are 68.3390, 203.3784, 89.3340 and 70.6952 are greater than the tabulated value. Therefore, we shall reject the null hypothesis and conclude that the chlorophyll $a$ and $b$ content in the varieties of sugarcane leaves randomly selected are not the same.

## VI. Conclusion

The results of the analysis of the chlorophyll content in sugarcane considered for this study found to be statistical significant in both the seasons and the varieties randomly selected. Chlorophyll $a$ and $b$ are the most common type of chlorophyll making $75 \%$ of the green plants. Furthermore, they are very important in the conversion of light energy to chemical energy in plants.

The correlation coefficient between the two dependent variables chlorophyll a and chlorophyll b of concern in this study obtained to be moderate; this justifies the use of multivariate analysis of variance for the experimental data that are concern. It should be noted that strong correlation between the variables would suggest that one variable may be substituted for the other and thereby making the univariate analysis of variance preferable.

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## Appendix I

GLM Chlorophyll a Chlorophyll b BY Seasons Varieties
/METHOD=SSTYPE (3)/INTERCEPT=INCLUDE /EMMEANS=TABLES (OVERALL) /PRINT=DESCRIPTIVE TEST (SSCP) RSSCP HOMOGENEITY /CRITERIA=ALPHA (.05)

Multivariate Tests ${ }^{\mathbf{c}}$

| Effect |  | Value | F | Hypothesis df | Error df | Sig. |
| :--- | :--- | :---: | :---: | :---: | :---: | :---: |
| Intercept | Pillai's Trace | .956 | $4.612 \mathrm{E}^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
|  | Wilks' Lambda | .044 | $4.612 \mathrm{E}^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
|  | Hotelling's Trace | 21.962 | $4.612 \mathrm{E} 2^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
|  | Roy's Largest Root | 21.962 | $4.612 \mathrm{E} 2^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
| Seasons | Pillai's Trace | .633 | $36.189^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
|  | Wilks' Lambda | .367 | $36.189^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
|  | Hotelling's Trace | 1.723 | $36.189^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
|  | Roy's Largest Root | 1.723 | $36.189^{\mathrm{a}}$ | 2.000 | 42.000 | .000 |
| Varieties | Pillai's Trace | 1.228 | 1.591 | 86.000 | 86.000 | .016 |
|  | Wilks' Lambda | .149 | $1.556^{\mathrm{a}}$ | 86.000 | 84.000 | .022 |
|  | Hotelling's Trace | 3.190 | 1.521 | 86.000 | 82.000 | .028 |
|  | Roy's Largest Root | 1.704 | $1.704^{\mathrm{b}}$ | 43.000 | 43.000 | .042 |

a. Exact statistic
b. The statistic is an upper bound on F that yields a lower bound on the significance level.
c. Design: Intercept + Seasons + Varieties

Between-Subjects SSCP Matrix

|  |  |  | Chlorophyll a | Chlorophyll b |
| :---: | :---: | :---: | :---: | :---: |
| Hypothesis | Intercept | Chlorophyll a | 21964.670 | 16859.574 |
|  |  | Chlorophyll b | 16859.574 | 12941.020 |
|  | Seasons | Chlorophyll a | 76.467 | 389.304 |
|  |  | Chlorophyll b | 389.304 | 1982.003 |
|  | Varieties | Chlorophyll a | 1679.349 | 847.125 |
|  |  | Chlorophyll b | 847.125 | 1920.115 |
| Error |  | Chlorophyll a | 1043.924 | 593.100 |
|  |  | Chlorophyll b | 593.100 | 1285.025 |

Based on Type III Sum of Squares

Residual SSCP Matrix

|  |  | Chlorophyll a | Chlorophyll b |
| :--- | :--- | :---: | :---: |
| Sum-of-Squares and Cross-Products | Chlorophyll a | 1043.924 | 593.100 |
|  | Chlorophyll b | 593.100 | 1285.025 |
| Covariance | Chlorophyll a | 24.277 | 13.793 |
|  | Chlorophyll b | 13.793 | 29.884 |
| Correlation | Chlorophyll a | 1.000 | .512 |
|  | Chlorophyll b | .512 | 1.000 |

Based on Type III Sum of Squares

