

# Estimated Photosynthetic Activity from Its Electrical Impedance Spectroscopy

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## Abstract

We study the photosynthetic activity from the electrical impedance spectroscopy in darkness and in light, using the Cole's model. The used lights active photosynthetic are red, blue and yellow. This analysis shows that the photochemical phase with active photosynthetic lights are characterized by a decrease of the extra chlorophyll space resistance. The non-photochemical phase is characterized by an almost constant or increase of the extra chlorophyll space resistance. We also observe a vertical offset at the spectrometric curves of bio impedance, implying a difference in electrical activity in darkness and inactive photosynthetic lights respectively. However, the green light is found to be inactive, since the photosynthetic activity is the same as in darkness.

**Keywords:** active photosynthetic lights; chlorophyll pigment; electrical impedance spectroscopy; inactive photosynthetic lights; model of Cole; Photosynthesis; non-photochemical phase; photochemical phase.

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## 1. Introduction

Since 1873, John Burdon Sanderson identified the first electrical signals in plants [1]. Many scientists worked on the possibility of using electrical signals to diagnose the state or physiological activity of plants [2, 3]. However, photosynthesis is one of the main physiological activities of plants causing the production of organic matter, growth and productivity of plants. During decades, several methods have been proposed to estimate the photosynthetic activity of living plants. These methods include chlorophyll fluorescence, the amount of  $O_2$  emitted or the amount of  $CO_2$  assimilated; unfortunately, none of the above directly gives us information about the electric activity taking place within photosynthesis. It has been proven that the measurement of chlorophyll fluorescence is the most method used to study the photosynthetic efficiency [4, 5]. The measurement of fluorescence pigments allows for quick information on the light conversion, transfer and dissipation in photosystem II (PSII) [6, 7]. Without some data on the fluorescence, investigations on the photosynthetic efficiency seem incomplete [8, 9]. However, when industrial applications of photosynthesis are considered, that is to say artificial photosynthesis from which solar energy is transformed into electrical energy [10], it is appropriate to develop new photosynthesis investigation methods from the electrical properties of the chlorophyll solution.

Several scientists nowadays use electrical, electric and electronic properties like dielectric behaviors, ac electrical conductivity, to characterize or to differentiate inorganic matter [11-15].

The electrical impedance spectrum (EIS) is a method of studying inorganic and organic structures [16-19], allowing us to have an idea about the physiological activities of plants [20] on the phases of ripening bananas, also to differentiate the types of apples [21].

The main concern is to study the behavior of extra chlorophyll space resistance and the impedance spectroscopy of a solution of chlorophyll in darkness and in light. We intend to use the Cole's model.

The rest of the paper is therefore presented as follows: In section 2, we present the materials, explain the methods and the model that we use to obtain the results that are presented and discussed in section 3. Section 4 is devoting to some concluding remarks the paper.

## 2. Materials and methods

Experiments in this paper were conducted at the Laboratory of Biophysics of the University of Yaoundé I, and were repeated seven times under the same condition at temperature of ( $23^{\circ}C \pm 2^{\circ}C$ ).

### 2.1 Sampling and preparation

Our chlorophyll solution was obtained from 20 g of *Carica papaya* leaves; properly ground in a mortar with 50 ml of acetone solution and then filtered through a filter paper and funnel [22]. 30ml of crude chlorophyll solution was obtained and introduced into an 8.5 cm diameter dish.

Cole's model [23] was used for the variation of the extra chlorophyll space resistance as a function of the light intensity. In fact, the solution of crude chlorophyll was assumed to be particles of crude chlorophyll in the acetone. Two spaces were considered; the first one called extra chlorophyll space and the second, an intra-chlorophyll space ( Fig.1).

The model is made up of two- branched parallel circuit where R represents the extra chlorophyll space resistance, R' represents intra chlorophyll space resistance, and C represents chlorophyll capacitance (Fig.2).

The impedance of this circuit is expressed as:

$$Z = \frac{R+RR'(R+R')(C\omega)^2 - jCR^2\omega}{1+[C\omega(R+R')]^2} \quad (1)$$

Where the real part and imaginary parts are given by

$$Zr = \frac{R+RR'(R+R')(C\omega)^2}{1+[C\omega(R+R')]^2} \quad (2)$$

$$Zi = \frac{-CR^2\omega}{1+[C\omega(R+R')]^2} \quad (3)$$

the resistive and capacitive parts of the impedance respectively

$$j = \sqrt{-1}, \omega = 2\pi f$$

At zero frequency, i.e  $\omega = 0$ ;

$$Z = R; \quad (4)$$

While at infinite frequency, i.e.  $\omega = \infty$ ;

$$Z = \frac{RR'}{R+R'} \quad (5)$$

Having to the intra chlorophyll space resistance

$$R' = \frac{RZ}{R+Z} \quad (6)$$

We further write:

$Rt = R+R'$ ; and  $\alpha = RR'$ ; and we deduce the expression of  $\omega$  from (2). In replacing the latter into (3), the following relation between  $Zr$  and  $Zi$  can be written:

$$Zi = \frac{1}{Rt} (-Rt^2 Zr^2 + Rt(2\alpha + R^2)Zr - \alpha RRt)^{1/2} \quad (7)$$

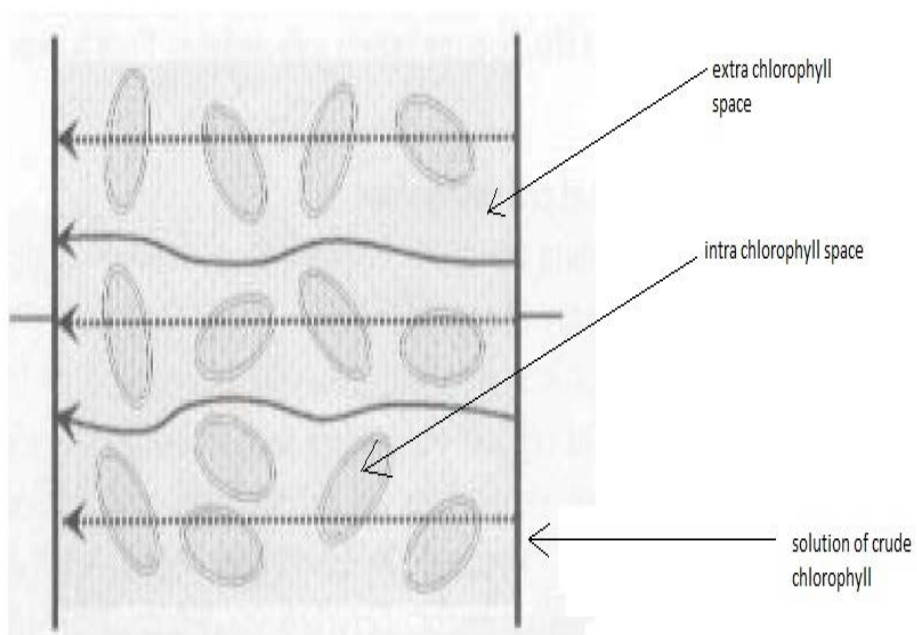
We should however stress that equation (7) does not explicitly depend on  $\omega$ . Furthermore, equation (7) is a second-order polynomial equation in  $Z_r$ , and its discriminant is:

$$\Delta = [Rt(\alpha - RRt)]^2$$

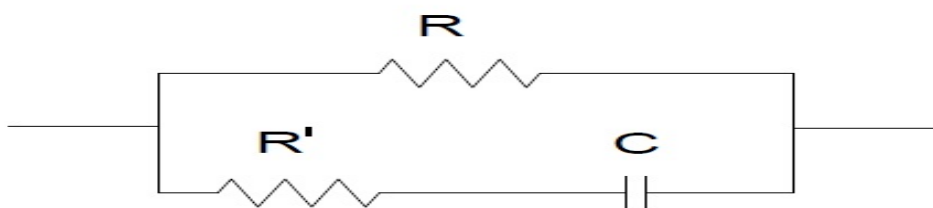
It is obvious that  $\Delta > 0$ ; which had to the solution

$$Z_{r1} = R \quad \text{and} \quad Z_{r2} = RR' / (R + R')$$

Which are indeed the impedance at zero and infinite frequency respectively.



**Figure 1:** Low and high frequency current paths in crude chlorophyll solution. The dashed lines represents current path of high frequency and the solid lines represents current path of low frequency

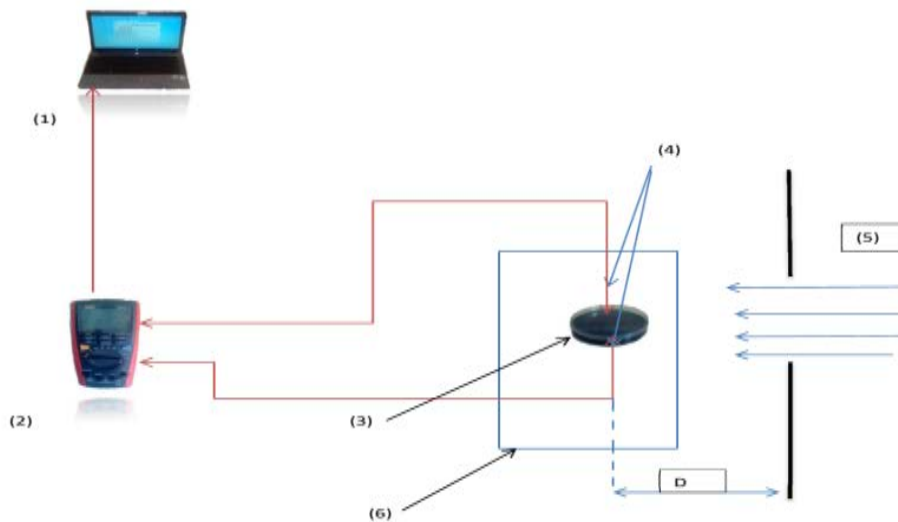


**Figure 2:** Electrical model of crude chlorophyll solution. R is the extra chlorophyll space resistance, R' is the intra chlorophyll space resistance and C is the capacitance of the pigments.

## 2.2 Measurement of electrical parameters

At zero frequency, the digital multimeter smart "U.T.71A." was programmed to automatically detect the extra chlorophyll space resistance ( $R$ ) of the solution in the dark for 10 minutes and for the next 10 minutes in the presence of light (white, red, blue, yellow, and green), produced by lamps of 1000W placed 1,5 m apart from the chlorophyll solution ( Fig.3). An IR filters were used to reduce artefactual heating effect in the specimen. All values measured by the multimeter were automatically transferred to a laptop for analysis. To capture the electrical signal from the crude chlorophyll solution, self-adhesive electrodes for electro cardiology were used [24]. The intra chlorophyll space resistance( $R'$ ) was obtained by measuring the impedance( $Z$ ) of the solution at high frequency(20,83KHz), for a sinusoidal voltage of 0,5V produce by a low frequency generator (L.F.G) and by using Eq(6) (Fig.4).

The data was analyzed using XLSTAT and Sine qua non software.



**Figure 3:** Measure of the extra chlorophyll space resistance ( $R$ ) of the solution in the darkness for 10 minutes followed by the next 10 minutes in the presence of light, produced by a lamp of 1000W placed at 1.5 m apart from the chlorophyll solution. 1 - Laptop; 2 - multi-meter; 3 - petri-dish; 4- electrodes; 5 - source of light; 6 - curve; D is the distance between the source of light and the petri-dish.

## 3. Results

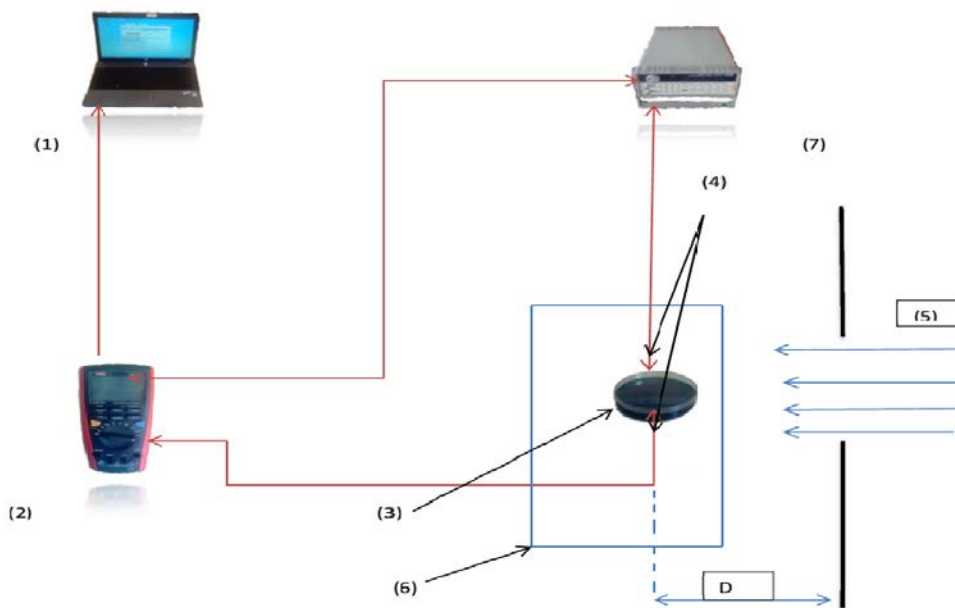
Different measures have been made, where resistances have been obtained in darkness and in lights for different colors. For each case, we look experimentally for values of  $\alpha$ ,  $R_t$  and we plot the impedance  $Z_i$  versus  $Z_r$ , from the equation (7).

### 3.1 Experiment 1: in white light

The extra chlorophyll resistance ( $R$ ) varies very little and an average value for the first 10 minutes is 0. 441M  $\Omega$  in darkness(coefficient of variation is 0.090); and 0; 352M  $\Omega$  for the next 10 minutes in white light,

(coefficient of variation is 0.037), showing a difference of  $89000\Omega$  ( Fig.5).

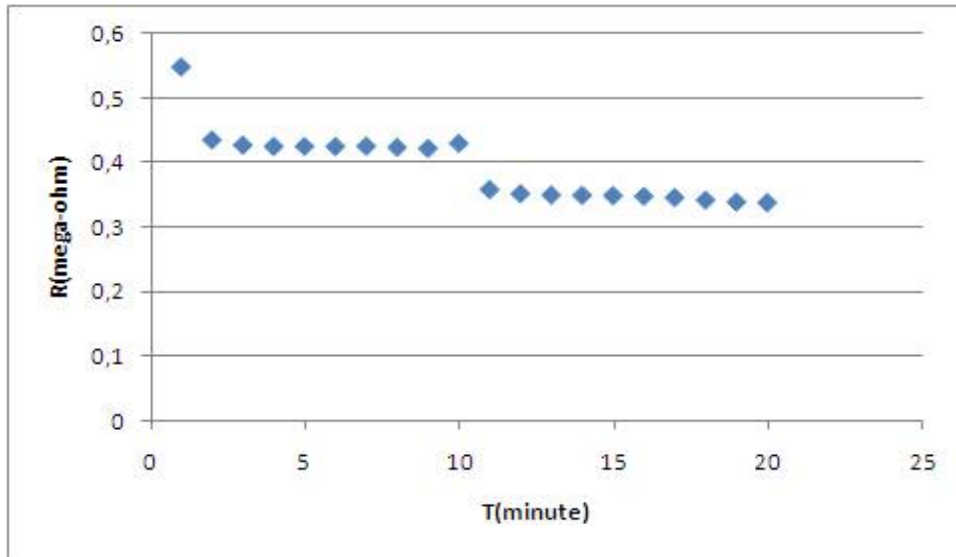
This variation is observed from the 10<sup>th</sup> minute, at which the chlorophyll solution is lighted with white light. The electrical impedance spectroscopy of Fig.6 also reveals two electrical states of a spectrum corresponding to the electric impedance of the organic chlorophyll solution in white light and the other in darkness. These (EIS) differ in the capacitive part of the impedance; it is approximately  $0.08M\ \Omega$  in the presence of white light and  $0.16M\ \Omega$  in the presence of darkness. The width ( $Z\ r1 - Z\ r2$ ) of curves are  $0.32$  and  $0.17M\ \Omega$  in darkness and in presence of white light respectively. The parameters used to plot Fig.6 are given by the Table1.



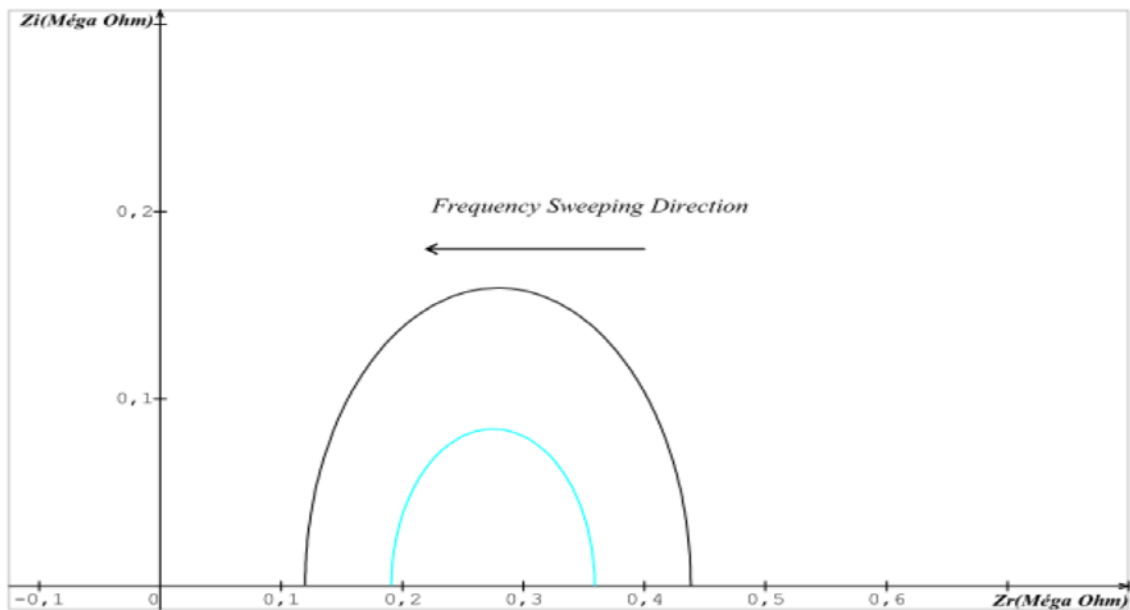
**Figure 4:** The intra chlorophyll space resistance ( $R'$ ) was obtained by measuring the impedance ( $Z$ ) of the solution at high frequency (20.83KHz), for a sinusoidal voltage of 0.5V produced by a L.F.G. and by using equation (6). 1 - laptop; 2 - multi-meter; 3 - petri-dish; 4 - electrodes; 5 - source of light; 6 - curve; 7- low frequency generator; D is the distance between the source of light and the petri-dish.

**Table1:** Parameters in presence of white light

Parameters	$R_t$	$\alpha$	R
Darkness	1.801	0.600	0.441
White light	0.801	0.158	0.352



**Figure 5:** extra chlorophyll space resistance of the solution in the darkness for 10 minutes followed by another 10 minutes in white light. The extra chlorophyll resistance (R) varies very little and an average value for the first 10 minutes is  $0.441M \Omega$  in darkness; and  $0.352M \Omega$  for the next 10 minutes in white light, showing a difference of  $89000\Omega$ . This variation is observed from the 10th minute, at which the chlorophyll solution is lighted with white light



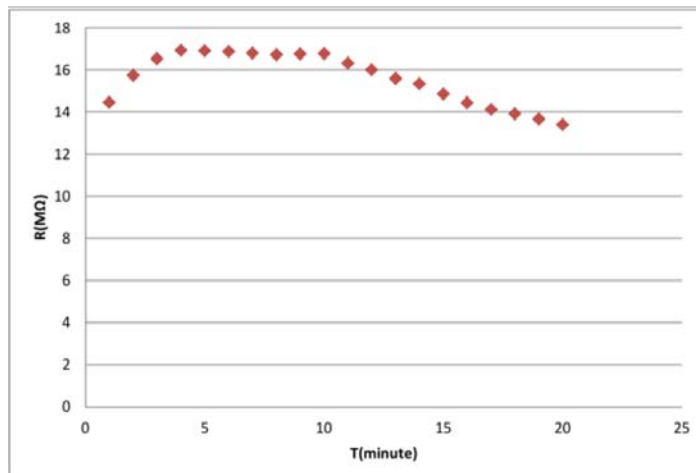
**Figure 6:** Impedance spectra of chlorophyll solution in white light (pink curve) and darkness (black curve). The frequency increases from right (0Hz) to left (20.83 KHz).  $Z_r$  is the resistive part of impedance or the resistance and  $Z_i$  the capacitive part or reactance. These (CSB) differ in the capacitive part of the impedance; it is approximately  $0.08M \Omega$  in the presence of white light and  $0.16M \Omega$  in the presence of darkness. The width ( $Z_{r1} - Z_{r2}$ ) of curves are  $0.32$  and  $0.17M \Omega$  in darkness and in presence of white light respectively.

**3.2 Experiment 2: in red light (wavelength: 650 nm)**

The same experiment is now carried out with red light. The average value of the extra chlorophyll resistance (R) is 16.461M  $\Omega$  for the first 10 minutes in darkness; and 14.779M  $\Omega$ , for the next 10 minutes in red light, showing a difference of 1682000 $\Omega$  (see Fig7). This variation is observed from the 10<sup>th</sup> minute, at which the chlorophyll solution is lighted with red light. . The electrical impedance spectroscopy of Fig.8 also reveals two electrical states of a spectrum corresponding to the electrical impedance of the organic chlorophyll solution in red light and the other in darkness. These CSB differ in the capacitive part of the impedance; it is approximately 7.34M  $\Omega$  in the presence of red light and 8.32M  $\Omega$  in darkness. The diameters ( $Z_{r1} - Z_{r2}$ ) of curves are 16.19 and 14.46M  $\Omega$  in darkness and in the presence of red light respectively. The corresponding parameters values used to plot Fig.8 are summarized in table 2.

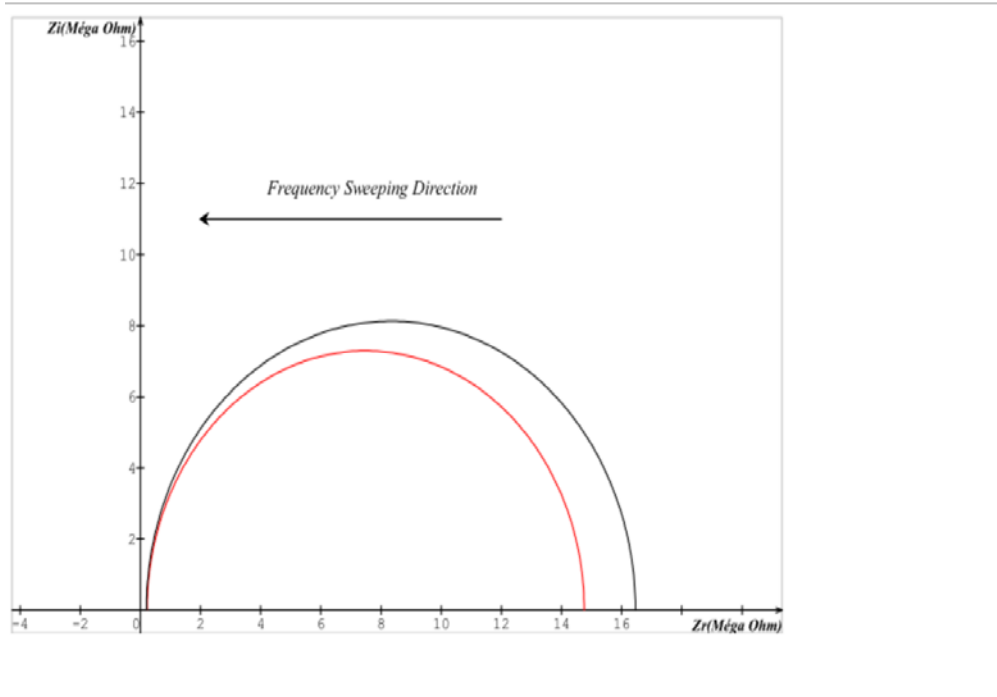
**Table 2:** Parameters in presence of red light

Parameters	Rt	$\alpha$	R
Darkness	16.662	3.326	16.461
White light	14.981	2.99	14.799



**Figure7:** extra chlorophyll space resistance of the solution in the darkness for 10 minutes followed by another 10 minutes in red light. The average value of the extra chlorophyll resistance (R) is 16.461M  $\Omega$  for the first 10 minutes in darkness; and 14.779M  $\Omega$ , for the next 10 minutes in red light, showing a difference of 1682000 $\Omega$  . This variation is observed from the 10<sup>th</sup> minute, at which the chlorophyll solution is lighted with red light.





**Figure 8:** Impedance spectra of chlorophyll solution in red light (red curve) and darkness (black curve). The frequency increases from right (0Hz) to left (20.83 KHz).  $Z_r$  is the resistive part of impedance or the resistance and  $Z_i$  the capacitive part or reactance. These CSB differ in the capacitive part of the impedance; it is approximately  $7.34M \Omega$  in the presence of red light and  $8.32M \Omega$  in darkness. The diameters ( $Z_{r1} - Z_{r2}$ ) of curves are  $16.19$  and  $14.46M \Omega$  in darkness and in the presence of red light respectively.

**3.3 Experiment 3: in blue light (wavelength: 470 nm)**

The same experiment is now carried out with blue light. The average value of the extra chlorophyll resistance ( $R$ ) is  $3.041M \Omega$  for the first 10 minutes in darkness; and  $3.479M \Omega$  for the next 10 minutes in blue light, showing a difference of  $438000\Omega$  (see Fig.9). This variation is still observed from the 10<sup>th</sup> minute, at which the chlorophyll solution is lighted with blue light. The electrical impedance spectroscopy of Fig.10 also reveals two electrical states of a spectrum corresponding to the electrical impedance of the organic chlorophyll solution in blue light and the other in darkness. These CSB differ in the capacitive part of the impedance; it is approximately  $1.64M \Omega$  in blue light and  $1.43M \Omega$  in darkness. The diameters ( $Z_{r1} - Z_{r2}$ ) of curves are  $2.83$  and  $3.28M \Omega$  in darkness and in blue light respectively. The results of such behaviors are summarized in Fig.10, with the parameter values given in table 3.

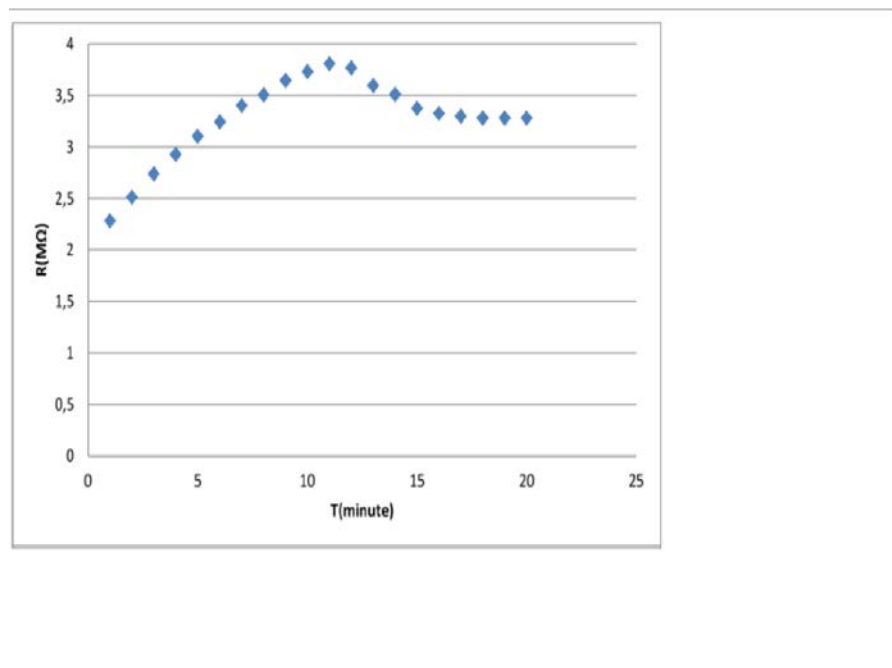
**3.4 Experiment 4: in yellow light (wavelength: 580nm)**

The same experiment is now carried out with yellow light. It is therefore found that, the average value of the extra chlorophyll resistance ( $R$ ) is  $10.551M \Omega$  for the first 10 minutes in darkness; while it is  $10.50M \Omega$  for the next 10 minutes in yellow light, showing a difference of  $51000\Omega$  (see Fig.11).

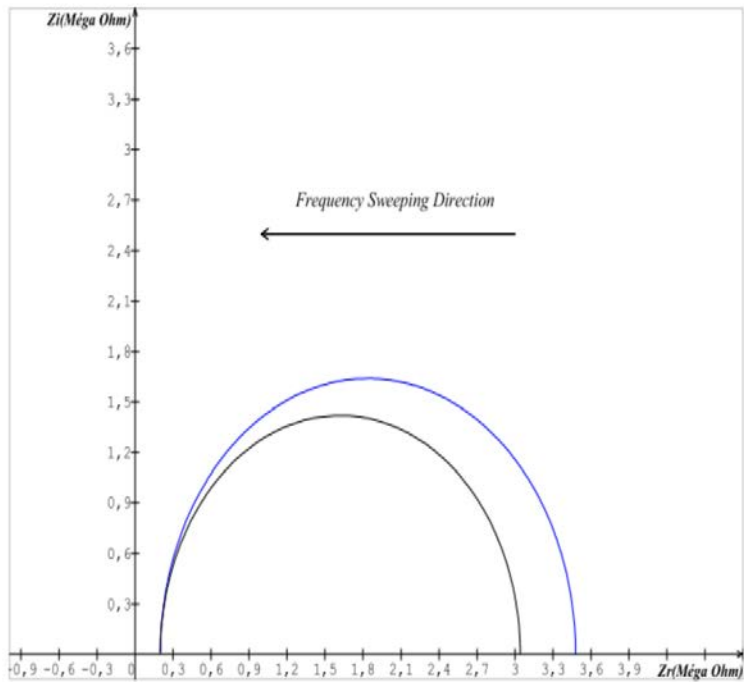
From the 11<sup>th</sup> minute, a small variation is observed. Consequently, the EIS of Fig.12 reveals two electrical states which respectively correspond to the electrical impedance of the organic chlorophyll solution in yellow light and the other in darkness. These CSB don't differ more in the capacitive part of the impedance like white, red and blue light. This is approximately  $5.12M \Omega$  in the presence of yellow light and  $5.17M \Omega$  in the presence of darkness. The widths ( $Z r1 - Z r2$ ) of curves are equal and worth  $10; 37M \Omega$  in darkness and in the presence yellow light. For instance, the results of Fig.12 are based on the parameter values given by table 4.

**Table 3:** Parameters in presence of blue light

Parameters	Rt	$\alpha$	R
Darkness	3.254	0.65	3.041
White light	3.690	0.737	3.479



**Figure 9:** extra chlorophyll space resistance of the solution in darkness for 10 minutes followed by another 10 minutes in blue light. The average value of the extra chlorophyll resistance (R) is  $3.041 M \Omega$  for the first 10 minutes in darkness; and  $3.479M \Omega$  for the next 10 minutes in blue light, showing a difference of  $438000\Omega$ . This variation is observed from the 10<sup>th</sup> minute, at which the chlorophyll solution is lighted with blue light.



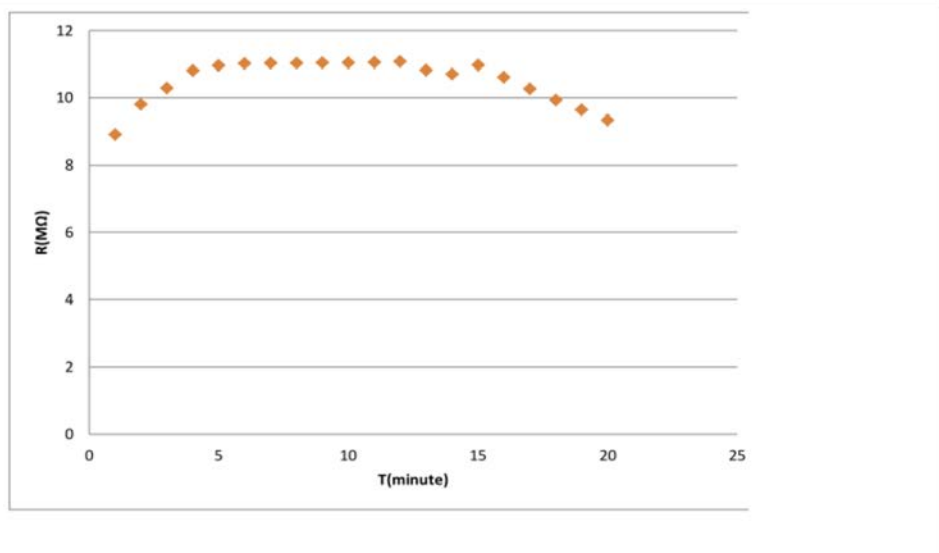
**Figure 10:** Impedance spectra of chlorophyll solution in blue light (blue curve) and darkness (black curve). The frequency increases from right (0Hz) to left (20.83 KHz).  $Z_r$  is the resistive part of impedance or the resistance and  $Z_i$  the capacitive part or reactance. These CSB differ in the capacitive part of the impedance; it is approximately  $1.64M\Omega$  in blue light and  $1.43M\Omega$  in darkness. The diameters ( $Z_{r1} - Z_{r2}$ ) of curves are  $2.83$  and  $3.28M\Omega$  in darkness and in blue light respectively.

**Table 4:** Parameters in presence of yellow light

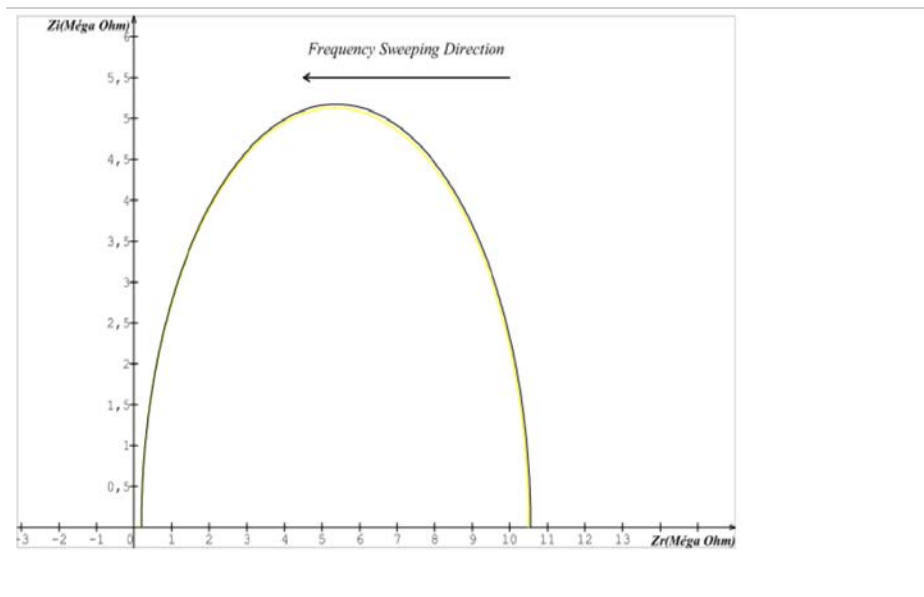
Parameters	$R_t$	$\alpha$	R
Darkness	10.754	2.147	10.551
White light	10.703	2.136	10.5

**3.5 Experiment 5: in green light (wavelength: 520nm)**

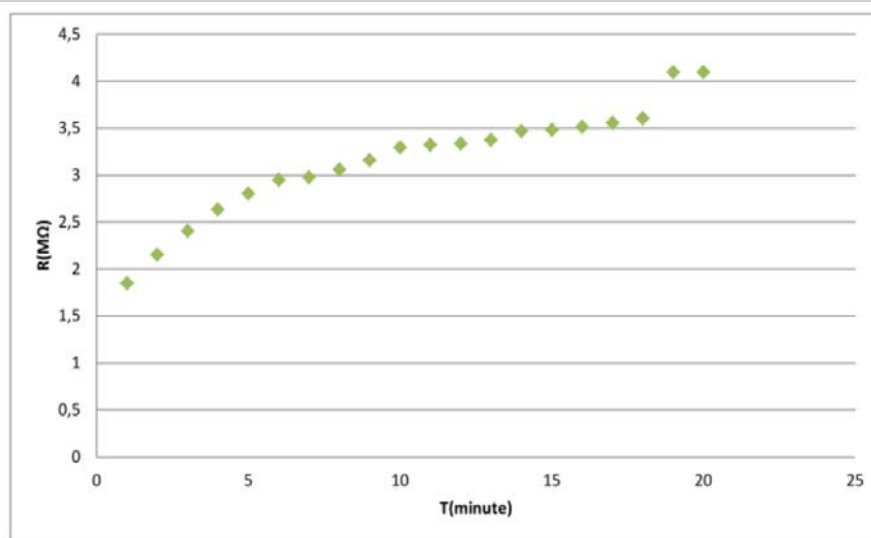
The green light is now used and comparisons are made with darkness. We observe that the values of the extra chlorophyll resistance (R) still increases, from  $1.846M\Omega$  to  $4.095M\Omega$  during 20 minutes as depicted in Fig.13, from darkness to light. However the behaviors observed in the previous experiments, are not really obtained.



**Figure 11:** Extra chlorophyll space resistance of the solution in the darkness for 10 minutes followed by another 10 minutes in the yellow light. The average value of the extra chlorophyll resistance (R) is  $10.551M \Omega$  for the first 10 minutes in darkness; while it is  $10.50M \Omega$  for the next 10 minutes in yellow light, showing a difference of  $51000\Omega$ . From the 11<sup>th</sup> minute, a small variation is observed.



**Figure 12:** Impedance spectra of chlorophyll solution in the presence of yellow light (yellow curve) and darkness (black curve). The frequency increases from right (0Hz) to left (20.83 KHz). Zr is the resistive part of impedance or the resistance and Zi the capacitive part or reactance. These CSB don't differ more in the capacitive part of the impedance like white, red and blue light. This is approximately  $5.12M \Omega$  in the presence of yellow light and  $5.17M \Omega$  in the presence of darkness. The widths ( $Zr1 - Zr2$ ) of curves are equal and worth  $10.37M \Omega$  in darkness and in the presence yellow light.



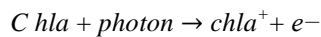
**Figure 13:** Extra chlorophyll space resistance of the solution in darkness for 10 minutes followed another 10 minutes in green light. The values of the extra chlorophyll resistance (R) still increases, from 1.846M Ω to 4.095M Ω during 20 minutes, from darkness to light.

#### 4. Discussion

From all the above results, there is a strong relationship between the measure of electrical parameters and photosynthetic activity. In this framework, the decrease of extra chlorophyll space resistance(R) in the presence of white, red, blue and yellow light, (Fig.5,7,9,11) can be explained by the fact that the absorption spectra of a crude chlorophyll is maximal for red (650-700nm) and blue (<500nm) lights, partial for yellow light. In figure11, the fact that the effect of yellow light begins at the 11<sup>th</sup> minute, and the fact that in Fig.12 it is clearly obvious that there is no difference between the black and yellow curves, which is mainly due to the fact that yellow light is partially absorbed by crude chlorophyll. As a main reason, photosynthetic pigments units form an antenna which collects the emitted light energy from the lamp; when a pigment captures a photon energy corresponding to its absorption capacity, one of its electrons which come from the double conjugates bonds (delocalises electrons) of tetrapyrrole nucleus, goes to the excited state. This energy can be transmitted in four ways:

- Either by reissuing in the form of light photon,
- Either in the form of heat,
- Either by sending energy by resonance,
- Either by performing photochemistry,

The fourth way corresponds to the transfer of energy to an electron, and the production by a particular molecule of chlorophyll 'a' and high electron energy in the reaction center according to the equation



The presence of the charge carriers in the extra chlorophyll space will result in increased conductivity that is to say reducing the extra chlorophyll space resistance, which confirms the work of P. Sellers [25] who studied the relationship between stomatal conductance of the leaves and the light intensity and showed that the conductivity of maize leaves increases as a function of the light intensity, that is to say that the electrical resistance decreases as a function of the light intensity.

In figure 13, the fact that the green light does not create a variation of the behavior of the curve like white, red, blue and yellow light (Fig. 5, 7, 9, 11) can be explained by the fact that the absorption spectra of a crude chlorophyll is null for green light; this implies that there is no release of charge carriers in the extra chlorophyll space.

## 5. Conclusions

The purpose of this study was to examine the photosynthetic activity from the electrical impedance spectroscopy (EIS) in darkness and in light, using the model of Cole. This analysis shows that the photochemical phase is characterized by a decrease of the extra chlorophyll space resistance and non-photochemical phase is characterized by an almost constant or increase of the extra chlorophyll space resistance. We also observed a vertical offset at the (CSB), reflecting a difference in electrical activity, one in the darkness and the other in light (white, red, blue and yellow light). It is clear that, from all the above results, this method gives us more direct information about the electrical behaviors of chlorophyll pigment than fluorescence for example, where we need first to determine parameters like: primary fluorescence ( $F_0$ ), maximal fluorescence ( $F_m$ ), variable fluorescence ( $F_v$ ), the quantum photochemical yield of the photosystem II ( $\Phi_{PSII}$ ), and the assimilation quantum yield of  $CO_2$  ( $\Phi_{CO_2}$ ) before having an information about the electrical behavior of chlorophyll pigment.

## Acknowledgements

We would like to express a special thanks to the Pr Ambang Zachée, Vice chancellor in charge of research and international cooperation in science of education E.N.S. Yaoundé; the Head of nuclear Technology Section of Cameroon Dr Saidou; the Fowo Director, Founder Wwoof Cameroon Peter N. Yuhala.

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