University of São Paulo Luiz de Queiroz College of Agriculture

Carbon dioxide assimilation, light use efficiency, growth and population dynamics in current soybean and maize genotypes

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Thesis presented to obtain the degree of Doctor in Science. Area: Crop Science.

Piracicaba 2020 Jackellyne Bruna Sousa Agronomist

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RESUMO

Assimilação de dióxido de carbono, eficiência de uso da luz, dinâmica do crescimento e população em genótipos atuais de soja e milho

Um fator que afeta a produtividade potencial é a condição ambiental do ambiente de produção, principalmente a radiação solar disponível e, consequentemente a temperatura do ar. Avaliou-se o aproveitamento da radiação solar nas culturas de soja e de milho, em especial no arranjo espacial em milho (capítulo I) e assimilação de carbono nas culturas de milho e de soja (capítulo II). No capítulo I observou-se que: (i), para o híbrido de milho BM 812PRO2 de maturidade precoce e alto potencial de rendimento, o aumento da população de plantas de 65.000 para 85.000 plantas ha⁻¹ (+30,1%) resultou em uma queda na produção de 12,4 para 11,3 Mg ha⁻¹ (-8,9%), indicando que o aumento das plantas ha⁻¹ em relação à a população recomendada de 65.000 plantas ha-1 não é uma prática viável; (ii) o espaçamento entre linhas de 0,45 a 0,90 m não interferiu no rendimento, mas 0,90 apresentou maior acúmulo de matéria seca; (iii) uma maior área do solo ocupada por uma planta aumentou o crescimento da planta principalmente durante o período reprodutivo, melhor absorção da radiação solar através de um maior coeficiente de extinção da luz; e (iv). o início do período reprodutivo não foi afetado pela densidade das plantas e espaçamento entre linhas. No capítulo II, observou-se que: (i) o método proposto para a determinação da assimilação de dióxido de carbono deu bons resultados para as culturas de soja e milho. O método é mais completo e sólido porque é baseado no padrão de crescimento completo do ciclo das culturas; (ii) a soja apresentou maior eficiência no uso de luz e carbono em relação ao milho, cultivado nas mesmas condições solo-água-clima.

Palavras-chave: *Glycine max*, *Zea mays*, Índice de colheita, Partição de biomassa, Simbiose

ABSTRACT

Carbon dioxide assimilation, light use efficiency, growth and population dynamics in current soybean and maize genotypes

A factor that affects potential productivity is the environmental condition of the production environment, mainly the available solar radiation and, consequently, the air temperature. The use of solar radiation in soybean and maize crops was evaluated, especially in the spatial arrangement in maize (chapter I) and carbon assimilation in maize and soybean crops (chapter II). In chapter I was observed that: (i). for the maize hybrid BM 812PRO2 of early maturity and high yield potential, the increase of the plant population from 65,000 to 85,000 plants ha⁻¹ (+30.1 %) resulted in a yield decrease from 12.4 to 11.3 Mg ha⁻¹ ¹ (-8.9 %), indicating that the increase of plants ha⁻¹ in relation to the recommended population of 65,000 plants ha⁻¹ is not a viable practice; (ii). row spacings varying from 0.45 to 0.90 m did not interfere with yield, but 0.90 had a higher dry matter accumulation; (iii). a greater soil area occupied by one plant increased plant growth mainly during the reproductive growth period, improved solar radiation absorption through greater light extinction coefficient; and (iv). the beginning of the reproductive period was not affected by plant density and row spacing. In chapter II was observed that: (i). the proposed method for the determination of the Carbon Dioxide Assimilation gave good results for soybean and maize crops. The method is more complete and solid because it is based on the complete growth pattern of the cycle of the crops; (ii). soybean presented a greater efficiency of light and carbon use in relation to maize, grown under the same soil-water-climate conditions.

Keywords: Glycine max, Zea mays, Harvest index, Biomass partition, Symbiosis

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1. INTRODUCTION

The good performance of a crop depends on management practices, adapted genotypes, and integrated management of pests, diseases and weeds. Brazil, in the last 30 years, has become one of the largest producers of soybean (*Glycine max*) and maize (*Zea mays*) as a result of technological advances and technical knowledge available to growers (PARRA, 2006).

Over time, the production system of these crops has changed. currently there are crops with higher plant density, reduced row spacing, pesticides and improved genotypes (BENDER et al., 2013). These modifications may have altered their dynamics of light absorption, photosynthetic efficiency and photoassimilate partitioning.

For conversion of solar radiation energy into plant biomass, the plant depends on factors such as photosynthesis and respiration efficiencies, environment, temperature, water availability, and nutrients required by the crop. To develop crop growth models, Penning de Vries (1989) determined conversion coefficients of solar radiation to biomass in relation to plant biochemical composition.

Know the efficiency of conversion of photosynthetically active radiation (solar energy) to chemical energy (photosynthesis and respiration) for the genotypes currently used, as a function of dry matter accumulation and composition (lipid, lignin, protein, carbohydrate, organic acid and minerals) and carbohydrate partition, will guide the definition of the maize hybrids, or soybean cultivars to be selected by geneticists in the future. This knowledge will also assist in choosing the most suited sowing season for the production environment in order to optimize the use of natural resources such as light (solar radiation), temperature, carbon dioxide and water.

Solar radiation is a resource that directly influences crop productivity (TAIZ et al., 2017). Understand how solar radiation interacts with cultivation is essential to improve the efficiency of its use. Only a fraction of solar radiation can be absorbed by plants and converted to carbohydrates by photosynthesis process.

1.1 Importance of the soybean and maize crops

Brazil is the second largest soybean producer in the world, and in the 2016/2017 crop produced 113.9 million tons in an area of 33.9 million hectares, with an average yield of 3,362 kg ha⁻¹ (EMBRAPA, 2019, CONAB, 2019abc). During the 2019/2020 crop year, nearly 37 million hectares were planted with soy in Brazil, up from almost 36 million hectares in the

preceding crop year. The area planted with soy in the country has increased by more than 52 percent in comparison to 2010. Soybean production is forecasted to reach more than 122 million metric tons in crop year 2019/2020 (ALVES, 2020).

The importance of this commodity is at least partially related to its versatility in use. Soybean can be used as an alternative source of protein in plant-based diets and as a base for processed products such as soy milk. Soybean meal is used as feed for animals. In addition, soybean oil is the main raw material used in the production of biodiesel in Brazil (ALVES, 2020).

Maize is considered the third most common cereal species important in the world (O'KEEFFE, 2009). 1.13 billion are produced worldwide tons, with the United States, China and Brazil the largest producers representing 32.7, 22.8 and 8.6% of the world total (FAOSTAT, 2017). In Brazil, in the 2018/2019 harvest, 241.9 million tons of maize were produced, of which the states of Mato Grosso, Paraná, Rio Grande do Sul and Goiás the largest producers, with approximately 28, 15, 15 and 10% of national production (CONAB, 2019abc).

Brazil produced a record 101 million tons of maize in 2018-2019 and is forecast to equal that total in 2019-2020, according to an October 10 Global Agricultural Information Network report from the U.S. Department of Agriculture (USDA, 2020).

The USDA reported the 2018-2019 crop with an increase of 23% compared to the previous crop, with record area and yields.

Over time yield increased and this was due to improved agricultural practices, including weed control, pest and disease control, plant nutrition, crop genetic improvement, soil correction and fertilization, mainly (CIAMPITTI et al., 2013; BENDER et al., 2013, STEWART et al., 2005).

Maize is the most cultivated cereal in the world. It is used for human consumption and industrial purposes. The industrial and pharmaceutical destination have been increasing, having as examples of byproducts the starch, ethanol, plastic and as base to produce antibiotics, mainly (EDWARDS, 2009).

The cultivation of soybean is one of the largest vegetable sources of protein and oil (CIAMPITTI; SALVAGIOTTI, 2018). In the composition of the grain there is about 40% protein, which is used mainly in animal feed. In addition, soybeans have all the essential amino acids for humans and have a low cost when compared to other protein sources (CARRERA et al., 2011).

1.2 Phenology

Phenology is the study of the stages of crop development and allows the optimization of management and decision-making strategies (FANCELLI; DOURADO-NETO, 2004).

Maize plant development (Table 1) can be divided into the following steps: (i) plant growth and root development (between the stages $V_0 \in V_7$), (ii) definition of productive potential (between the stages $V_4 \in V_6$), (iii) definition of number of rows in spike (between the stages $V_7 \in V_9$), (iv) definition of spike number and size (between the stages $V_{12} \in V_{14}$) e (v) effective grain filling (between the stages R_1 and R_2) (FANCELLI, 2013).

Soybean plant development (Table 2) is also divided into vegetative growth and reproductive growth. Critical periods for crop yield determination are the onset of flowering and pod establishment (R₁-R₃) as well as grain filling R₅-R₆ (FEHR; CAVINESS, 1977).

Phase	Stadium	Description
	V ₀	Emergency
V V Vegetative V V V	V ₂	2 ^a . fully developed leaf
	V4	4 ^a . fully developed leaf
	V ₆	6 ^a . fully developed leaf
	V ₈	8 ^a . fully developed leaf
	V ₁₂	12 ^a . fully developed leaf
	V ₁₄	14 ^a . fully developed leaf
	VT	Tassel emission and opening of male flowers
	R1	Full Bloom
	R ₂	Pasty Grains
Reproductive	R₃	Milky Grains
	R4	Farinaceous Grains
	R5	Hard Farinaceous Grains
	R ₆	Physiological maturity point

Table 1. Phenological stage of maize plants when 50% of the plants have the characteristic described in the table. The classification divides the cycle of growth in two stages, the vegetative development (V_n) and the reproductive development (R_n) stage.

Adapted from Ritchie et al. (1996).

Table 2. Phenological stage of soybean plants when 50% of the plants have the characteristic described in the table. The classification divides the cycle of growth in two stages, the vegetative development (V_n) and the reproductive development (R_n) stage.

Phase	Stadium	Description
	VE	From emergency to open cotyledons
	V ₁	First node, open unifoliate leaves
Vegetative	V2	Second node, the first open trifoliate leaf
	V ₃	Third node, the second trifoliate leaf open
	Vn	Nth node with open trifoliate leaf, before flowering
	R₁	Beginning of flowering: up to 50% of plants with at least one flower
	R ₂	Full bloom: Most racemes with open flowers
	R₃	End of flowering pods up to 1.5 cm
	R ₄	Most pods of the upper third from 2 to 4 cm
	R5.1	Grãos com início de formação a 10% da granação
Poproductivo	R5.2	Grain filling between 10 - 25% of the grain in most pods
Reproductive	R _{5.3}	Grain filling between 25 to 50% of the grain in most pods
	R5.4	Grain filling between 50 to 75% of the grain in most pods
	R5.5	Grain filling between 75 to 100% of the grain in most pods
	R ₆	Most pods with 100% grain filled and green leaves
	R ₇	0 to 50% yellowing of leaves and pods
	R ₈	Field maturity (harvest point)

Adapted from Fehr and Caviness (1977).

1.3 Potential yield

Potential yield of a crop for a given site is the dry matter (DM) mass produced by a standard crop, fully covering the ground, with solar radiation, photoperiod and temperature as limiting factors. It is also determined by several factors, among which are: genetic (variety), the degree of adaptation to the environment (which opens the possibility of varying the number of plants per hectare), availability of water and nutrients, pest control and diseases, considering all the developmental periods until the crop matures (CAMARGO, 1984).

1.4 Attainable yield

It is determined as the maximum yield of a crop in a given site, influenced by the following limiting factors: soil water and nutrient availability.

1.5 Biomass accumulation

Nutritional requirements of crops vary over the crop cycle and have peaks of maximum and minimum nutrient uptake (MARTINS et al., 2017a; 2017b). Knowing the accumulation and partitioning of nutrients in the phenological stages contributes to the definition of fertilization strategies and replacement of nutrients exported by the crop, and

thus maintaining the nutritional balance of the production system (SOUZA, 2006; VON PINHO et al., 2009).

High yields will be achieved through highly productive plants; however, management strategies that minimize stress conditions for the plant provide better use of the crop's genetic potential (SOUZA, 2006).

1.6 Crop growth rate

Growth analyzes are useful for verifying physiological adaptations and carbohydrate partitioning between plant organs. Maize crop growth rate has been well studied in the past (PENNING DE VRIES, 1989; VON PINHO et al., 2009). However, these studies need to be updated due to the genetic modifications of cultivars over time. This information will help in understanding plant performance and updating mathematical growth models.

1.7 Solar energy conversion

The source of energy for plants is the sun, through the ionizing radiation that reaches the earth, which is called incident radiation at the top of the atmosphere (Q₀). By traversing the atmosphere until it reaches the leaf surface, the radiation is attenuated and called global radiation (Qg), and its intensity depends on weather conditions, time of year, time of day, latitude of the place and solar declination (HARGREAVES; SAMANI, 1982). Part of this radiation between the wavelengths of 400 to 700 nm is absorbed by the plant and converted into sugars (CHO)_n by the photosynthesis process. This energy, already being in the chemical form, is then used for plant growth and development, respiration, substance transport, and conversion processes to lignin, protein, fatty acids, carbohydrates (HARGREAVES; SAMANI, 1982). Penning de Vries (1989) defined conversion factors and energy costs of processing and transport of the above-mentioned substances.

1.8 Hypothesis

The conversion efficiency of photosynthetically active radiation to chemical energy defines the amount and composition of dry matter (where photosynthesis and respiration directly account for 96% of dry matter, since C and O are supplied by carbon dioxide, and H is water) and carbohydrate partitioning, which determines the yield and quality of soybean and maize. Maximizing the efficiency of light transformation into dry matter is an important strategy to guide management actions, adapting the genotype to the production environment.

1.9 Objectives

The main objective of this research is to: i. better understands the relation between available solar radiation and maize yield, considering crop light interactions in different plant populations, in an environment of no soil and water restriction; and ii. propose a new methodology to derive the carbon dioxide assimilation curve from the crop growth analysis (soybean and maize), using a new definition and a new procedure for calculating the efficiency of carbohydrate conversion into dry matter, as a function of their dry matter composition.

1.10 Specific objectives

The specific objectives of this research are: *i*. to determine the CO₂ assimilation curve and the efficiency of carbon and light use in soybean and maize genotypes; *ii*. to characterize the total dry matter production and composition of the different organs (root, stem, leaf and reproductive organs) in maize and soybean crops, to explore the possibility of increasing their energy conversion efficiencies; *iii*. to evaluate the carbohydrate, lignin, protein and oil contents in the dry matter of both crops; *iv*. to evaluate influence of leaf area on energy conversion of soybean and maize crops; *v*. to evaluate biometrics, yield and its components in soybean and maize crops; and *vi*. to determine conversion factors of solar radiation to dry matter mass.

2. CHAPTER I – SEEDING RATE AND ROW SPACING ON THE DEVELOPMENT AND YIELD OF MAIZE IN HIGH YIELD TROPICAL ENVIRONMENT

Abstract

Maize seeding rate and row spacing are commonly subject to genotype versus environment interaction, and the characterization of this interaction is necessary to improve yield potential of future cultivars. To improve maize yield, farmers usually increase the seeding rate that is greater than the recommended rate. However, whether the increasing seeding rate could improve maize yield is unclear. Additionally, the interaction of seeding rate and row spacing on maize development, yield, and yield composition needs further study. Therefore, a field experiment was conducted varying seeding rate and row spacing under a management of high fertility and water availability in Piracicaba, SP, Brazil. For the maize hybrid of early maturity and high yield potential, it was observed that: (i) the increase of the seeding rate from 65,000 to 85,000 seeds ha⁻¹ resulted in a yield decrease of 8.9% from 12.4 to 11.3 Mg ha⁻¹, indicating that the increase of seeds ha⁻¹ in relation to the recommended seeding rate for this hybrid of 65,000 seeds ha⁻¹ is not a viable practice; (ii) row spacings varying from 0.45 to 0.90 m did not interfere with yield, but 0.90 had a higher dry matter accumulation; (iii) a greater soil area occupied by one plant increased plant growth mainly during the reproductive growth period, improved solar radiation absorption through greater light extinction coefficient; and (iv) the beginning of the reproductive period was not affected by seeding rate and row spacing.

Keywords: Plant population, dry matter accumulation, leaf area index, radiation use efficiency.

2.1 Introduction

Maize is one of the poaceae species that offers the highest grain yield potential and is sensitive to seeding rate variations. Modern hybrids have higher yield potential and can generally tolerate higher seeding rates, because they can support better intraspecific competition (GRASSINI et al., 2015; TOLLENAAR; LEE, 2002).

As in modern hybrids increasing seeding rate is raising grain yield, there is a renewed interest in investigating the interaction between row spacing and seeding rate (LICHT; HUFFMAN, 2017).

Refining agricultural practices such as narrower row spacing and higher seeding rate, coupled with more stress-tolerant modern maize hybrids may have changed the yield potential and biomass partitioning of the maize crop.

The main purpose of increasing the seeding rate is to enhance maize yield in terms of grain mass per area, thus making the crop system more efficient and competitive per unit area. In the absence of biotic or abiotic stresses, grain yield is related to the amount of intercepted solar radiation by the crop, and the use of a higher seeding rate, with an earlier canopy closure, could maximize the solar global radiation interception.

The crop potential yield highly depends on the interaction between the external environment and the genetic background of the maize hybrid (BENDER et al., 2015). Expansion to new cropping areas is restricted in most countries since the best areas are already under cultivation, and in many cases this expansion has a considerable effect related to environmental issues (GRASSINI et al., 2015). Therefore, yield increase is mainly sought through management practices that lead to a better use of the local soil and climate resources. One aspect to be explored, mainly in cases of newly introduced hybrids, is seeding rate and row spacing. These parameters are related to a better soil exploitation and a more efficient use of the available solar energy to be transformed into yield (PENNING DE VRIES, 1989).

Commercial maize hybrids have a plant with single stalk, so that the number of seeds sown drives the number of stalks per area. Thus, the only way that maize can moderately compensate for lower seeding rate changes on the final yield is through an adaptation of ear development. A flex ear hybrid adjusts its ear growth according to the conditions that prevail in the field, by modifying the number of grains per ear that reach full maturity. This feature allows, for example, the plant to better compensate for plant mortality or bad plant stand formation, and consequently preserve the yield when harsh field conditions occur. On the other hand, a fixed ear development keeps the total number of grains per ear relatively stable, regardless of the environmental conditions (SHARIOT-ULLAH et al., 2013).

Recently interest has arisen on the effects of row spacing on maize grain yield and yield components. Decreasing the row spacing from 0.762 m to 0.508 m increased yield in 3% (LICHT; HUFFMAN, 2017).

The objective of this study was to evaluate how seeding rate and row spacing affect yield, yield components and radiation use efficiency of a modern maize hybrid released to the Brazilian market, grown in a high yield tropical environment with respect to nutrient and water availabilities.

2.2 Materials and methods

2.2.1 Experimental design

A field experiment was carried out in Piracicaba, SP, Brazil (22°41'S, 47°38' W, 546 m altitude) during the 2017/2018 growing season, under humid subtropical climate – Cfa (ALVARES et al., 2013). The soil was classified as Oxisol, based on the Brazilian system of

soil classification (EMBRAPA, 2019), or a Haplustox according to Soil Survey (USDA, 2020). Soil properties for 0-0.2 and 0.2-0.4 m depth were as follows: pH (CaCl₂) of 5.7 and 5.5 units, a soil organic matter (Walkley-Black) of 9 and 7 g dm⁻³, P (resin as extractor) of 30 and 11 mg dm⁻³; K of 5.3 and 3.2 mmol_c dm⁻³, respectively.

The experimental design consisted of completely randomized blocks with five replications, composed of two seeding rates (6.5 and 8.5 plants m⁻², SR_{6.5} and SR_{8.5}, respectively) and two row spacings (0.45 and 0.90 m, RS₄₅ and RS₉₀, respectively). The treatments were nominated as follow: T₁ as RS₄₅ and SR_{6.5}; T₂ as RS₄₅ and SR_{8.5}; T₃ as RS₉₀ and SR_{6.5}; T₄ as RS₉₀ and SR_{8.5}. Plots had an area of 63 m² (10 x 6.3 m), and the useful area consisted of the 5 central lines.

The maize hybrid was BM 812PRO2, early maturity group and high yield potential. The crop was sown on April 4th, 2018 in succession to soybean (first season) and sorghum (winter season) in a till cultivation system.

Basic fertilization consisted of 70 kg $[P_2O_5]$ ha⁻¹ as triple superphosphate, 40 kg $[K_2O]$ ha⁻¹ as potassium chloride, and 20 kg [N] ha⁻¹ as urea. Top dressing fertilization of 100 kg [N] ha⁻¹ as urea was carried out at the V₄ growth stage and 100 kg [N] ha⁻¹ as urea at R₁. Pests, weeds and diseases were properly controlled.

2.2.2 Plant measurements

Plant evaluations consisted of sampling one whole plant per replicate, carried out approximately every 14 days according to phenological stages, which totalized 10 samplings (V₂, V₄, V₆, V₁₀, R₁, R₂, R₃, R₄, R₅ and R₆) (RITCHIE et al., 1996) until harvest. Plants were separated in root, stem, leaf and reproductive organs. Roots were collected only from soil area of $0.3 \times 0.3 \times 0.3 m (0.09 m^3)$, to avoid great disturbance in the field.

The plant material was dried at 65°C until constant weight to evaluate dry matter (DM). To analyze possible differences in growth patterns of the different treatments, DM evolution in time was adjusted to sigmoidal models for further calculation of growth rates d(DM)/dt. The specific sigmoidal model for the evolution of the DM (Mg ha⁻¹) accumulation as a function of time t (RG) was:

$$DM = \frac{a}{1 + e^{-\frac{(t-b)}{c}}}$$
(1)

The adjustment of the original DM data to the model was done using the program TableCurve 2D^{©,} which minimizes the deviations and calculates the empirical parameters a, b and c. DM being a sigmoidal equation, its first derivative is bell shaped, indicating that the

rate of DM accumulation passes through a maximum. Therefore, the rate of DM accumulation (dDM/dt, kg ha⁻¹ d⁻¹) is given by:

$$\frac{dDM}{dt} = \frac{a \cdot e^{-\frac{(t-b)}{c}}}{c \cdot \left[1 + e^{-\frac{(t-b)}{c}}\right]^2}$$
(2)

The coordinates of the maximum can be obtained by making the second derivative equal to zero. The second derivative is given by:

$$\frac{d^2 DM}{dt^2} = \frac{a.e^{-\frac{(t-b)}{c}} \cdot \left\{2.e^{-\frac{(t-b)}{c}} - \left[1 + e^{-\frac{(t-b)}{c}}\right]\right\}}{c^2 \cdot \left[1 + e^{-\frac{(t-b)}{c}}\right]^3}$$
(3)

Making $d^2DM/dt^2 = 0$ we verify that the moment t (relative growth) in which we have the maximum rate is:

$$t = b \tag{4}$$

and that the maximum rate $(dDM/dt)_{max}$ at the moment t = b is:

$$\frac{dDM}{dt}\left(\frac{d^2Y}{dt^2} = 0\right) = \frac{a}{4.c}$$
(5)

and also that the value of DM at t = b (inflexion point of the sigmoid) is:

$$DM\left(\frac{d^2Y}{dt^2} = 0\right) = \frac{a}{2} \tag{6}$$

and that the maximum dry mass (DM_{max}) at the end of the growth cycle tends to:

$$DMmax = a \tag{7}$$

2.2.3 Leaf Area Index (LAI)

The leaf area (LA) of each plant was measured using a Li-3100C equipment, for all treatments at each evaluation. The leaf area index (LAI), was calculated by the ratio of the LA of one plant by the area occupied by one plant. LAI was calculated for only for the main treatments T_1 , T_2 , T_3 and T_4 .

2.2.4 Light interception and radiation-use efficiency

Solar radiation was continuously measured as photosynthetically active radiation (PAR, MJ day⁻¹) with three PAR sensors (radiometers model SQ-326-SS Apogee instruments); two were installed above the plant canopy, one facing upwards for the incoming radiation (PAR_I) and the other facing downwards to measure the reflected radiation (PAR_R) and finally one was placed on the soil surface to measure the transmitted

radiation (PAR_T). The radiation absorbed by the plant canopy (PAR_A) was calculated by difference: $PAR_A = PAR_I - PAR_R - PAR_T$, or in relative terms $PAR_a = 1 - PAR_R - PAR_T$.

According to the theory of Penning de Vries (1989):

$$PAR_{A} = 1 - [EXP - (k \times LAI)]$$
(8)

where k is the light extinction coefficient. The coefficient k was evaluated making the regression of Equation (8) in the logarithmic form: $ln(1 - PAR_A) = -k \times LAI$. This regression was made with PAR_A and LAI values of treatments T₁ to T₄. The regression was performed minimizing the deviations.

2.2.5 Radiation use efficiency

According to Monteith and Moss (1977), in healthy plants with adequate water and nutrient supply, the DM production (g m⁻²) is a function of accumulated PAR_A and can be represented by the equation:

$$DM = RUE \times PAR_A \tag{9}$$

Thus, Pearson's correlations were made between accumulated DM for each sampling date and PAR_A in order to calculate RUE (g MJ⁻¹) from the respective slopes. The trend line of this linear regression was forced to pass through the origin ($\beta_0 = 0$), generating only the angular coefficient that corresponds to the radiation use efficiency (RUE) (MONTEITH, 1994).

2.2.6 Air temperature, degree-days and water balance

Air temperature was measured in the field together with PAR measurements during all Julian days t_i of the experiment. As time variables we used the Julian day (t_i), Days after Emergence (DAE) and the relative growth (RG) based on the degree day (DD, °C day) concept (REICHARDT; TIMM, 2019). Table 3 shows the correspondence of the sampling dates with the respective DAE, RG and vegetative stages.

Table 3. Experimental timetable showing the correspondence among number of samplings, days after emergence (DAE), relative growth (RG) and phenological stages (PS) of the crop during its development.

DAE	0	9	23	37	51	65	79	93	108	122	136	140
RG	0	0.07	0.17	0.27	0.38	0.48	0.59	0.69	0.8	0.9	1	1
PS	Vc	V ₂	V_4	V_6	V ₁₀	R ₁	R ₂	R ₃	R_4	R_5	R ₆	R ₇

The DD_i were taken for DAE_{ii} as the daily mean air temperature (T_{air}, $^{\circ}$ C) minus 10 $^{\circ}$ C, taken as the lower basal temperature for the maize plant. The accumulated DD (ADD) or ADD_i at DAE_i is the sum of the DD_i from DAE 1 to DAE_{ii}. The RG_i, also at DAE_i is given by the ratio ADD_i/ADD_{tot}, where ADD_{tot} refers to the sum of ADD over the growth cycle, varying from 0 to 1. Figure 1 shows the daily variation of the maximum temperature (T_{max}), minimum temperature (T_{min}) and average temperature (T_{ave}) during crop development.



Figure 1. Air temperature (A), rainfall and irrigation (B), and hydric balance during the cropping cycle (C).

A water balance was made according to Thornthwaite & Matter's approach for the complete growth period to show that water deficits were minimized. During periods of drought, a central pivot system provided 7 mm every second working day (Figure 1C).

2.2.7 Harvest and yield components

At complete maturity the final harvest of the experiment was carried out.

For the evaluation of the yield components, five ears were collected from each treatment, for the following evaluations: grains per area, lines per ear, number of grains per line, number of grains per ear, and mass of hundred grains.

An area of 4.0 m² per plot was hand-clipped at full maturity to evaluate the final yield (kg ha⁻¹).

2.2.8 Statistical analysis

The Shapiro–Wilk statistic test indicated normality for all data. Plant measurements and yield components were submitted to analysis of variance, and when significant, were classified by the Tukey's test at 5% probability. The program TableCurve 2D[©] was employed to adjust experimental data to models. In order to calculate the Radiation Use Efficiency (RUE), dry matter data were plotted as a function of PAR_A and fitted to a linear regression model.

2.3 Results

The variation of dry matter of the different plant components is shown in Figure 3, as a function of the relative growth (RG). Because during the reproductive period all treatments presented decreases in DM of root (Figure 2A), leaf (Figure 2B), stem (Figure 2C), it was assumed that such decreases represent dead organs, mass transfer of these organs to reproductive organs (Figure 2D) and senescence. Therefore, the highest values of root, stem and leaf were taken as maximum growth, and the differences accounted for losses.

During the vegetative growth period up to about RG 0.4, the development differences between plant components in treatments were very small, and for RG between 0.4 and 1.0 (reproductive growth period) distinct growth patterns appeared distinguishing treatments. At the end of the growth cycle (RG = 1.0) T_3 (RS₉₀ and SR_{6.5}) presented the highest DM accumulation (Figure 2E) for all organs but stem, and T_2 (RS₄₅ and SR_{8.5}) presented the lowest for all organs.



Figure 2. Root (**A**), leaf (**B**), stem (**C**), reproductive organs (**D**) and total dry matter (e) by relative growth through maize cycle evaluated in 2017/2018 growing season in Piracicaba, São Paulo, Brazil.

For the total DM accumulation, the models of Equations 1 and 2 were applied to measured data and the result is presented in Figure 3. For the final DM_{max} only T_2 (RS₄₅ and SR_{8.5}) was lower in relation to the other treatments, while the others didn't differ between them. The maximum growth rates (dDM/dt) presented distinct behavior indicating a clear effect of treatments.



Figure 3. Sigmoid models for total dry matter (DM) accumulation (solid lines) (for T₁ as RS₄₅ and SR_{6.5}; T₂ as RS₄₅ and SR_{8.5}; T₃ as RS₉₀ and SR_{6.5}; T₄ as RS₉₀ and SR_{8.5}) and rates of DM accumulation (dY/dt) for all treatments (dashed lines) as a function of relative development (RD) through maize cycle evaluated in 2017/2018 growing season in Piracicaba, São Paulo, Brazil. (dDM/dt)_{max}, RD_{max}, and DM_{max} refers to maximum rate of DR accumulation, relative development of maximum rate of DM accumulation, and maximum accumulation of dry matter, respectively.

The DM partition into organs was very similar for all main treatments, therefore only shown for treatment T₁ (RS₄₅ and SR_{6.5}) in Figure 4.



Figure 4. Dry matter partition as a function of the relative growth through maize cycle with row spacing of 0.45 m and seeding rate of 6.5 seeds m⁻² evaluated in 2017/2018 growing season in Piracicaba, São Paulo, Brazil.

The leaf area over time shows that for each sampling date it did not differ very much among treatments (Figure 5a). There is, however, a tendency of T_1 (RS_{0.45} and SR_{6.5}) and T_3 (RS_{0.9} and SR_{6.5}) presenting higher values in relation to the other, mainly during the vegetative stage.

The Leaf Area Index LAI was calculated using the final area occupied by one plant (T₁: 0.16 m², T₂: 0.17 m², T₃: 0.12 m², and T₄: 0.14m²). T₁ (RS₄₅ and SR_{8.5}) occupying the largest soil area, presented the lowest values of LAI (Figure 5B). Since its calculation includes the area used by one plant, the behavior of LAI differs somewhat from that of LA. Treatments with SR_{8.5} had a higher IAF, and there was a tendency for a higher IAF with RS₉₀ in both SR.



Figure 5. Leaf area (LA, **A**) and leaf area index (LAI, **B**) as a function of days after emergence through maize cycle evaluated in 2017/2018 growing season in Piracicaba, São Paulo, Brazil.

The light extinction coefficient (k) and radiation use efficiency (RUE) indicate a trend of higher k for lower seeding rates (SR_{6.5}) and narrow row spacing (RS₄₅). RUE tends to be higher in greater seeding rate (SR_{8.5}) and larger row spacing (RS₉₀) (Table 4).

	S	Seeding rate (seeds	m⁻²)			
Row spacing (m)	6.5	8.5	Avg.			
	Light extinction coefficient (k)					
0.45	0.52	0.47	0.5			
0.90	0.5	0.41	0.46			
Avg.	0.51 A	0.44 B				
	Radiation use efficiency					
0.45	2.82	2.75	2.79 B			
0.90	3.01	3.42	3.22 A			
Avg.	2.92	3.09				
Variance analysis	k	RUE				
Seeding rate (SR)	0.0008	0.6091				
Row spacing (R)	0.109	0.0131				
R x SR	0.5745	0.8226				

Table 4. Light extinction coefficient and radiation use efficiency of maize plants in two seeding rates and two rows spacing in Piracicaba, SP, Brazil.

The cumulative dry matter as a function of absorbed photosynthetic active radiation (PAR_A) all treatments had significant values of R^2 and above 0.9, indicating a good adjustment (Figure 6).



Figure 6. Cumulative dry matter as a function of absorbed photosynthetic active radiation (PAR_A) and respective regressions in two row spacings and two seeding rates in Piracicaba, SP, Brazil.

For the evaluated yield components, there was interaction only for 100-grains mass. Seeding rate affected yield, grains per ear, and grains per line. Row spacing did not affect any of the evaluated yield components (Table 5).

The 100-grain mass within the SR_{6.5} did not differ between the two row spacings, however within the SR_{8.5} the 100-grain mass in the RS₉₀ was 10.5% higher. 100-grain mass within RS₄₅ was 15.5% higher at the SR_{6.5}, and within RS₉₀ had no differ between seeding rates.

Yield was 9.7% higher at the SR_{6.5}. The number of grains per ear was 29.5% higher at the SR_{6.5}. The number of grains per line was 24% higher at the SR_{6.5}.

No effect of row spacing and seeding rate treatments was found on the number of grains per area and lines per ear.

	Seeding rate (seeds m ⁻²)) Seeding rate (seeds m ⁻²)		
Row spacing (m)	6.5	8.5	Avg.	6.5	8.5	Avg.
	Yield (Mg ha ⁻¹) 100-grain mass (100-)-GM, g)
0.45	12.6(T1)	11.2 (T2)	11.9	394 aA	341 bB	368
0.90	12.2(T3)	11.4 (T4)	11.8	382 aA	377 aA	379
Avg.	12.4 a	11.3 b	11.8	388	359	374
-	Grains	s per area (G	A, nº)	Grain	s per ear (G	E, nº)
0.45	3204	3286	3245	524	394	459
0.90	3209	3038	3124	545	431	488
Avg.	3207	3162	3185	535 a	413 b	474
-	Lines per ear (LE, n°)			Grain	s per line (G	L, nº)
0.45	15.8	15.8	15.8	34.6	26.6	30.6
0.90	15.9	15.8	15.8	34.6	29.1	31.9
Avg.	15.9	15.8	15.8	34.6 a	27.9 b	31.2
Variance analysis	Yield	100-GM	GA	GE	LE	GL
Seeding rate (SR)	0.0056	0.0002	0.6681	0.0000	0.8657	0.0001
Row spacing (R)	0.8067	0.0518	0.2465	0.0798	0.9550	0.2757
R x SR	0.3141	0.0008	0.2513	0.6079	0.8657	0.2902

Table 5. Grain yield, 100-grain mass, grains per area, grains per ear, lines per ear, and grains per line of maize plants for two seeding rates and two row spacings in Piracicaba, SP, Brazil.

Lower case letters in the line and upper-case letters in the column rank the averages by the Tukey's test at 5% probability.

2.4 Discussion

This study compares maize crops grown under two seeding rates and two row spacings. The design involves treatments T_1 (RS₄₅ and SR_{6.5}) and T_3 (RS₉₀ and SR_{6.5}) with 65,000 plants ha⁻¹, each plant occupying 0.153 m² but areas of different shapes; T_2 (RS₄₅ and SR_{8.5}) and T₄ (RS₉₀ and SR_{8.5}) with 85,000 plants ha⁻¹, occupying 0.117 m², also of different shapes. During the vegetative period (emergence to about 60 DAE or V₁₀, Table 1), plants of all treatments grew at very similar rates (Figs. 3 and 4), apparently not reacting to seeding rate and row spacing. For the reproductive period (from 60 to 136 DAE or R₆), growth patterns differentiated very much among treatments, with T₃ (RS₉₀ and SR_{6.5}) presenting the highest values of DM accumulation in root, leaf, reproductive organs and total DM. This explains a better use of soil resources by T₃ (RS₉₀ and SR_{6.5}) and of T₁ (RS₄₅ and SR_{6.5}) plants due to the greater available soil volume. For the reproductive period, a marked lower growth was found on T₂ (RS₄₅ and SR_{8.5}), due to the higher plant density and, consequently, lower available soil volume (Figures 3 and 4).

The application of the sigmoid model, equation 1, to the total accumulated DM (Figure 3), indicates no difference in DM_{max} between T_1 (RS₄₅ and SR_{6.5}) and T_4 (RS₉₀ and SR_{8.5}), and a higher value was found for T_3 (RS₉₀ and SR_{8.5}), and a marked reduction was found in T_2 (RS₄₅ and SR_{8.5}) in relation to the other treatments.

The growth rate curves (Figure 3) show very little difference among the relative development of maximum rate of DM accumulation (RD_{max}) at which the maximum growth rates (dDM/dt)_{max} occur. This means that the beginning of the reproductive period was not affected by plant density and row spacing. These maximum growth rates, however, present distinct values, again with a superiority of T₃ (RS_{90} and $SR_{6.5}$) and a lowest value for T₂ (RS_{45} and $SR_{8.5}$) (Figure 3).

The DM partition did not differ among treatments, therefore only shown for T_1 in Figure 4.

For the variation of the leaf area (LA) there was no significant difference among the treatments during the complete growth cycle, however, T₃ (RS₉₀ and SR_{8.5}) indicates a tendency of higher values, in accordance with the temporary variation of DM, as expected (Figure 5). Larger values of LA represent a larger area for solar energy capture. This evidences that T₁ (RS₄₅ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}), although with lower population, presented larger or more leaves (which were not counted). Despite of this fact, due to the larger soil area per plant, they presented the lowest values of the LAI (Figure 5). According to Equations 8 and 9, LAI affects the absorbed photosynthetic radiation PAR_A which is responsible for the conversion of solar energy into DM. Therefore, the lower LAI values of treatments T₁ (RS₄₅ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) in relation to T₂ (RS₄₅ and SR_{8.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and SR_{6.5}) in relation to T₂ (RS₄₅ and SR_{6.5}) do indicate a better use of the solar RS_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₃ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) in relation to T₂ (RS₄₅ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) in relation to T₂ (RS₄₅ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) in relation to T₂ (RS₄₅ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and F₆ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and F₆ (RS₉₀ and SR_{6.5}) and T₄ (RS₉₀ and SR_{6.5}) and T₆ (RS₉₀ and SR_{6.5}) and T₆

Despite of the differences in the radiation parameters discussed above, RUE did not differ among treatments (Table 4).

In relation to yield components, the trait grains per ear is one of the determinants of the final yield, together with the number of grain rows and grains per row and these traits were best for T_3 (RS₉₀ and SR_{6.5}) and somewhat for T_1 (RS₄₅ and SR_{6.5}), indicating that the greater area used by one plant was beneficial for soil and water exploitation. The smaller seeding rate also enhanced solar radiation absorption in T_1 (RS₄₅ and SR_{6.5}) and T_3 (RS₉₀ and SR_{6.5}), as discussed above. The trait 100-grain mass also showed a primacy of T_1 (RS₄₅ and SR_{6.5}) and T_3 (RS₉₀ and SR_{6.5}), which certainly contributed to the final grain yield of T_1 and T_3 .

2.5 Conclusions

For the maize hybrid BM 812PRO2 of early maturity and high yield potential, it was observed that: i. the increase of the plant population from 65,000 to 85,000 plants ha⁻¹ (+30.1 %) resulted in a yield decrease from 12.4 to 11.3 Mg ha⁻¹ (-8.9 %), indicating that the increase of plants ha⁻¹ in relation to the recommended population of 65,000 plants ha⁻¹ is not a viable practice; ii. row spacings varying from 0.45 to 0.90 m did not interfere with yield, but 0.90 had a higher dry matter accumulation; iii. a greater soil area occupied by one plant increased plant growth mainly during the reproductive growth period, improved solar radiation absorption through greater light extinction coefficient; and iv. the beginning of the reproductive period was not affected by plant density and row spacing.

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3. CHAPTER II – METHODOLOGY FOR THE DETERMINATION OF THE CARBON DIOXIDE ASSIMILATION CURVE AND OF THE CARBON AND LIGHT USE EFFICIENCY IN RECENT SOYBEAN AND MAIZE GENOTYPES

Abstract

The estimation of the ability of agricultural crops in fixing atmospheric carbon dioxide is still not dominated. This study contributes to the measurement of the assimilation of carbon dioxide by crops through photosynthesis, using two major crops: the maize and the soybean. A novel methodology for this measurement is presented, based on the observation of the development of the plant, and on the measurement of the available solar radiation. Relations between crop growth and development, and the conversion of solar radiation into dry matter (DM) are shown. Special emphasis is given to DM accumulation and its rates along the development of the crop; to the partition of DM, to leaf area indexes, to light extinction coefficients and to the efficiency of light use by the two crops. As a result, the proposed method for the determination of the Carbon Dioxide Assimilation presented good results for soybean and maize crops and indicates a great potential for the estimation of carbon sequestration by crops, over large territory extensions. The study also concluded that soybean presented a greater efficiency of light and carbon use in relation to maize, grown under the same soil-water-climate conditions.

Keywords: Energy conversion by crops, photosynthesis, crop growth and development.

3.1 Introduction

The performance of a crop is a consequence of several factors, such as the development of technologies specific to the region, adapted genotypes; the use of integrated pest, disease and weed management, the adequacy of the application techniques, fertilization and the use of selective products; precision agriculture connected to harvesting techniques, storage logistics and transportation. All these factors combined made Brazil over the last 30 years an agricultural powerhouse, aiming to produce around 105 million tons of grain in 2020 (PARRA, 2006).

Presently, soybean has been considered as a strategic crop for the viability of increasing productivity and agricultural production in the Brazilian territory. According to the Ministry of Agriculture (MAPA, 2016), the growth of soybean cultivation in Brazil has been very accelerated over the last four decades, not so much by the expansion of the production area, but by the increased productivity in the field, as the result of an intensive use of more efficient technologies.

Maize, in turn, also occupies a prominent place in the Brazilian agriculture. The economic importance of maize is characterized by the various forms of its use ranging from animal feed to the high technology industry (FERREIRA, 2015). It is a widely used crop and

one of the most widely distributed agricultural products in the world, both in production and consumption.

Recently the production system has also changed, currently there are crops with higher plant density, reduced spacing, use of agrochemicals for crop protection and transgenic genotypes have also increased crop productivity (BENDER et al., 2013). Improving agronomic practices, and the increasing use of high technology in crops, may have altered the dynamics of nutrient absorption and carbon partition in crops.

One of the main factors affecting the potential yield of a cultivar of a certain species is the climatic condition of the planting site, especially the available solar radiation. In studies of solar radiation conversion to dry matter mass (productivity), it is necessary to know several factors that come into the calculation of this conversion.

The conversion of solar energy into dry matter mass (yield) of a crop is currently studied using empirical plant parameters obtained elsewhere and long time ago. This project aims to develop a methodology to directly determine the carbon assimilation rate for soybean and maize crops, from the march of dry matter accumulation, dry matter composition and partition in the different organs of the plant.

Knowledge of the conversion efficiency of the photosynthetically active radiation PAR (part of the solar energy) to chemical energy (photosynthesis and respiration) for the genotypes currently used, as a function of dry matter composition (lipids, protein, fibrous carbohydrates, soluble carbohydrates and minerals) and of the carbon partition, will guide the definition of the maize hybrid, or soybean cultivar to be cultivated. It will also assist in choosing the most appropriate sowing season for each particular production environment in order to optimize the use of natural resources such as light (solar radiation), temperature and carbon dioxide. In addition, this knowledge will be of great value to crop breeders of these two crops. The conversion efficiency of PAR to chemical energy is dependent on the dry matter, as C and O are supplied by the carbon dioxide, and H is provided by the carbohydrate partition, which defines productivity and quality of soybean and maize. Maximizing the efficiency of light to dry matter conversion is the best strategy to guide management actions, as it currently is done by adapting genotypes to each production environment (PENNING DE VRIES, 1989).

On the other hand, a key parameter in studies of climate change is carbon sequestration. Therefore, we present here a new methodology to calculate the carbon

dioxide accumulation by agricultural crops, using soybean and maize as study cases. It is shown that this methodology can be used to evaluate carbon sequestration over large areas.

3.2 Material and methods

3.2.1 Experimental area

Experiments were conducted in Brazil, at the experimental area of the University of São Paulo (Esalq/USP), in the municipality of Piracicaba-SP, under a center pivot system to ensure available water to the crops. The area is located within a main soybean and maize production areas, at the geographical coordinates: 22° 41' 30" S and 47° 38′ 30" W and 546 m altitude.

The soil is classified as nitric dystrophic Oxisol (HEIFFIG, 2002). Köppen's climate classification of the region is Cwa with rainy summer and winter drought (ALVARES et al., 2013).

3.2.2 Description of the experiments

Three experiments were conducted under field conditions, two during the main cropping season (October 2017 to March 2018) and one during an extended (second) cropping season (April to July 2018), therefore covering a wide range of solar exposition, extending from Spring to Winter. All crops were sown on the same field.

The first experiment was performed with a soybean crop sown on October 10, 2017. The soybean variety was RK7518 IPRO, of indeterminate growth, using a sowing density of 30 plants m⁻² and a row spacing of 0.45 m.

The second experiment was performed at the same time, with a maize crop sown on October 10, 2017, next to the soybean area (the same area). The maize variety used was AG-9045-PRO3, of tall stature and semi-erect leaves, using a sowing density of 65 plants m^{-2} and a row spacing of 0.9 m.

The third experiment was also performed with maize, sown on Apr. 4, 2018. The maize variety used was BM 812PRO2, of tall structure and semi-erect leaves, using a sowing density of 65 plants m⁻² and a row spacing of 0.9 m.

Since the three experiments consisted of direct measurements of the parameters related to the conversion of solar energy into dry matter, each one consisted of a homogeneous plot of about 0.27 ha (2,700 m²), for plant growth samplings every 14 days. To ensure random sampling, each plot was divided into 60 similar subplots, used to sample whole plants at the 10 sampling dates (one for each date), with 6 replicates. Subplots had

an area of 10.0 X 6.3 m, with a useful experimental area of the 10 central lines for soybean and 5 central lines for maize, discounting 1 m from the edges of the plots.

Before soil preparation, soil samples were collected at a depth of 0 to 20 cm for physical and chemical characterization. Crop management was carried out equally in all plots, with soil pH correction, soil fertilization and homogeneous irrigation (pivot system), in order to offer optimum development conditions under the prevailing climatic conditions in the two growing seasons.

3.2.3 Experimental evaluations

3.2.3.1 Phenology

For the characterization of each stage of development, the phenological determinations were made every 14 days during the cycle of each crop according to the phenological scale presented in Tables 1 and 2 (FEHR; CAVINESS, 1977; RITCHIE et al., 1996).

3.2.3.2 Air temperature, degree-days and water balance

The measurement of air temperature T (°C) was made in an automatic weather station, located about 1 km from the field. The time variables used were the Julian day (t_i), days after emergence (DAE) and the relative development (Dr) which is based on the concept of degree day (DD, °C day) (REICHARDT; TIMM, 2019). With T data we calculated: (a) degrees-day (DD, °C day), based on the equation proposed by Arnold (1959):

$$DDi = \frac{Tmax + Tmin}{2} - Tb \tag{1}$$

where DDi refers to the Julian day ti; Tmax to the maximum temperature of day ti; Tmin to the minimum temperature of day ti and Tb to the lower basal temperature of the crop: 10°C, and (b) the accumulated DD, or ADD_i t_i, which is the sum of the DD_i from t1 to t_i. The Dr_i, also at ti is given by the ratio ADD_i/ADD_{tot}, varying from 0 to 1.



Figure 7. Air temperatures (A), solar radiation (B), rainfall (black line) and irrigation (red line) (C) across the growing season.

Figure 7A shows the daily variation of the maximum temperature (T_{max}), minimum temperature (T_{min}) and average temperature (T_{ave}) during the development of the crops; 7B the variation of the solar radiation, 7C rainfall and irrigation.

A water balance was calculated to assure the optimal available water conditions for the crops. Thornthwaite and Matter's (1955) approach was used for the complete growth cycle, showing that water deficits were minimized for the whole period. During dry spell periods, a central pivot system provided 7 mm every second working day (Figure 7B).

3.2.3.3 Plant measurements

Plant evaluations consisted of sampling one whole plant per replicate (fresh matter FM in fluxogram of Figure 8), carried out approximately every 14 days (t_i) according to phenological stages, which totalized 10 samplings until harvest.

Each V or R (reproductive) stage for maize was defined when about half or more of the plants in the crop are in that specific stage. Although each stage of development is important, we focus here on management guidelines only for the VE, V₃, V₅, V₁₂, V₁₈, R₁ and R₂ to R₆ growth stages (Adapted from Pannar, 2016).

In soybean, as it occurs in a very similar way, observing similar parameters. The subdivisions of the vegetative stages were numbered sequentially (V_1 , V_2 , V_3 , V_4 , V_5 , V_6 , ..., V_n , where V_n is the last node, at the top of the plant, with leaf fully developed). In both cultures (soybean and maize) 10 corresponding phenological stages were defined.

Plants were separated in root FMri, stem FMsi, leaf FMli and later reproductive organs FMroi. Roots were not totally collected, they were the root found in soil volumes 0.30 x 0.30 x 0.30 m around one plant, corresponding to an area of 0.09 m², in order not to disturb very much the plant stand. The leaf area LA was immediately measured with aid of a Li-3100C measurement device. The leaf area index LAI was obtained dividing LA by the soil area SA occupied by one plant.

Plant FM right after sampling was dried at 65 °C until constant weight for dry matter (DM) evaluation. DM and LAI evolution in time was adjusted to models to interpolate experimental data and obtain daily data over the whole growth cycle.

The specific sigmoidal model for the evolution of the DM (kg ha⁻¹) accumulation as a function of time t, was:

$$DM = \frac{a}{1 + e^{-\frac{(t-b)}{c}}}$$
(2)

The adjustment of the original DM data to the model can be done using the program TableCurve 2D, which minimizes the deviations and calculates the empirical parameters a, b and c. DM being a sigmoidal equation, its first derivative (dDM/dt, kg ha⁻¹ day⁻¹) is bell shaped, indicating that the rate of DM accumulation passes through a maximum. Therefore, the rate of DM accumulation (dDM/dt, kg ha⁻¹ d⁻¹) is given by:

$$\frac{dDM}{dt} = \frac{a \cdot e^{-\frac{(t-b)}{c}}}{c \cdot \left[1 + e^{-\frac{(t-b)}{c}}\right]^2}$$
(3)

The coordinates of the maximum can be obtained by making the second derivative equal to zero. The second derivative is given by:

$$\frac{d^2 DM}{dt^2} = \frac{a.e^{-\frac{(t-b)}{c}} \cdot \left\{ 2.e^{-\frac{(t-b)}{c}} - \left[1 + e^{-\frac{(t-b)}{c}}\right] \right\}}{c^2 \cdot \left[1 + e^{-\frac{(t-b)}{c}}\right]^3}$$
(4)

Making $d^2DM/dt^2 = 0$ we verify that the moment t (RG) in which we have the maximum rate is:

$$t = b \tag{5}$$

and that the maximum rate $(dDM/dt)_{max}$ at the moment t = b is:

$$\frac{dDM}{dt}\left(\frac{d^2Y}{dt^2} = 0\right) = \frac{a}{4.c} \tag{6}$$

and also that the value of DM at t = b (inflexion point of the sigmoid) is:

$$DM\left(\frac{d^2Y}{dt^2} = 0\right) = \frac{a}{2} \tag{7}$$

and DM_{max} at the end of the growth cycle:

$$\lim_{t \to \infty} DM = a \tag{8}$$

showing that the value tends to **a**.

DM subsamples of root, leaf, stem and reproductive organ were also used for chemical analyses of their contents Ctrc, Ctsc, Ctlc and Ctroc, respectively, in components c:

fat (oil), fibrous carbohydrates (lignin, hemi-cellulose, cellulose), protein, non-fibrous carbohydrates (sugars), and minerals. The obtained data were also adjusted to best models in order to interpolate experimental data over the whole growing cycle.

3.2.3.4 Light interception and radiation-use efficiency

Solar radiation was continuously measured in the field as photosynthetically active radiation (PAR, MJ day⁻¹) using three PAR sensors (radiometers model SQ-326-SS Apogee instruments, installed on a metal bar of 0.3 m). Two were situated above the plant canopy, one facing upwards for the incoming radiation (PAR_{TOT}) and the other facing downwards for the reflected radiation (PAR_R). Finally, one was placed on the soil surface, across one plant row, to measure the transmitted radiation (PAR_T). The absorbed radiation by the canopy (PAR_A) was obtained by difference:

 $PAR_A = PAR_{TOT} - PAR_R - PAR_T$.

In relative terms we have $PAR_{Ar} = 1 - PAR_{Rr} - PAR_{Tr}$.

Penning de Vries (1989) estimates PAR_{Ar} using the equation below:

$$\mathsf{PAR}_{\mathsf{Ar}} = 1 - [e^{-(k \times LAI)}] \tag{9}$$

k being the light extinction coefficient, evaluated making the regression of Equation (2) in the logarithmic form (3):

$$\ln(1 - PAR_{Ar}) = -k \times LAI \tag{10}$$

this regression was made with daily PAR_{Ar} and LAI values over the whole growth cycle, minimizing the deviations.

According to Monteith and Moss (1977), in healthy plants with adequate water and nutrient supply, the DM production (kg ha⁻¹) is a function of the accumulated PAR (PAR_A) and can be represented by the equation:

$$DM = RUE \times PAR_A \tag{11}$$

Thus, Pearson's correlations were made between accumulated DM for each sampling date and PAR_A in order to calculate RUE (g MJ⁻¹) from the respective slopes. The trend line of this linear regression was forced to pass through the origin ($\beta_0 = 0$), generating only the slope that corresponds to the RUE (MONTEITH, 1994).

3.2.3.5 Solar energy conversion efficiency into DM

As already mentioned, PAR_A was calculated by difference using data of PAR_{TOT} , PAR_R and PAR_T , all in MJ ha⁻¹ day⁻¹. The conversion of PAR_A into chemical energy has an

efficiency dependent on its intensity, on the plant species, in our case maize as a C₄ plant and soybean as a C₃ plant, and on the plant distribution on the field (population P and row spacing).

As the composition (%) of DM varies among plant organs (Ctr_c, Cts_c, Ctl_c and Ctro_c) in relation to fat, fibrous carbohydrates, protein, soluble carbohydrates and minerals, the conversion factors CB_c and CT_c (kg [DM] kg⁻¹ [CH₂O], Penning de Vries (1999) indicates the different biosynthesis CB and transport CT costs in Table 6. Calculations were made separately for each plant organ and each DM component.

Table 6. Biosynthesis (A) and transport (B) costs; biochemical conversion efficiency (BCE, D) for different DM components (PENNING DE VRIES, 1999).

Component	Biosynthesis costs	Transport costs	Conversion cost	BCE
Component	(A)	(B)	(C = A + B)	(D = 1/C)
Fat (oil)	3.030	0.159	3.189	0.31
Fibrous Carbohydrates	2.119	0.112	2.231	0.45
Protein	1.824	0.096	1.920	0.52
Soluble Carbohydrates	1.211	0.064	1.275	0.78
Minerals	0.000	0.120	0.120	8.3

A, B and C: Kg [glucose] kg⁻¹[component]. D: Kg [component] kg⁻¹[glucose]. Minerals (K, Ca, P and S).

3.2.3.6 Proposed methodology for the calculation of the carbon assimilation rate (Cda)

We propose a way to calculate the daily carbon assimilation rate [Car, kg ha⁻¹[leaf] h⁻¹] of a crop only by following the DM accumulation and measuring the daily PAR (MJ ha⁻¹ day⁻¹) and the average air temperature, through equation (12):

$$Cda_i = 44 \text{ NP}_i LAI_i / (30 \text{ Hi})$$
 (12)

Symbols can be found in Figure 8 and Table 7. The method is based on the analysis of crop growth and leaf area index (LAI). The fluxogram of Figure 8 and Table 7 summarize the calculations made to estimate Cda, stating from DM data. The result of Cda by equation (12) is the daily average of kg of C fixed by the crop, per hectare of leaves, per hour, obtained from the net photosynthesis NP, the leaf area index LAI and the hours of the day Hi. Therefore, these values are specific for the three crops under study.



Figure 8. Fluxogram of the proposed method for the estimation of the Carbon Dioxide Accumulation (Cda). See Table 7 for the respective symbols.

Table 7. Symbols used in the fluxogram of Figure 8.

Index i, used for time, day i
T = air temperature
Index c, used for contents of DM components: oil, protein, fibrous CHO, soluble CHO and minerals
r = root; s = stem; I = leaf; ro = reproductive organs
P = plant population
FMri= fresh root matter (DM) at DAEi; FMsi for stem; FMli for leaf; FMroi for reproductive organs
DMri = root DM at DAEi; DMsi for stem; DMii for leaf; DMroi for reproductive organs
Ctrc = Content of component c (Table 3) in root DM; Ctsi for stem; Ctli for leaf; Ctroi for reproductive organs
CB _c = Biosynthesis cost of component c (Table 2)
CT _c = Transport cost of component j (Table 2)
er = conversion efficiency in root; es in stem; el in leaf; ero in reproductive organs
NP = net photosynthesis $\left(\frac{kg[CHO]}{ha}\right)$
NP'= Derivative of NP with respect to time
$Cda_i = Carbon dioxide accumulation on day i \left(\frac{kg[CO2]}{ha[leaf].hour}\right)$
LA = leaf area of one plant
SA = soil area occupied by one plant
LAI = leaf area index
Factor of Cda = $\left(\frac{44kg[CO2]}{30kg[CHO]}\right)$
H = day length
δ = solar declination
PAR _i = Total photosynthetic radiation (PAR) on day i
PAR _{Ri} = Reflected PAR on day i
PAR⊤i = Transmitted PAR on day i
PAR _{Ai} = Absorbed PAR on day i
Ti = daily air temperature average
CR _{mc} = Coefficient of respiration, maintenance and growth
CRs = Coefficient of solar radiation interception
ECLU _i = Efficiency of carbon and light use on day i
α c t : adjustment coefficients

3.3 Results

3.3.1 Growth and development analysis

3.3.1.1 Soybean

The total DM accumulation of the soybean crop is shown in Figure 9, using the sigmoidal model of equations (2) to (8). Adjustment of experimental data to the model made by the program TableCurve 2D[®], presented a R² of 0.99, indicating that the model very well represents experimental data of DM. The physiological maturity occurred at DAE 133, so that the vegetative period had an extension of 34 days and the reproductive period of 102 days. The partition of DM accumulation to root, stem, leaf and reproductive organs is shown in Figure 10 through the accumulation rates in each organ. The productivity of the crop as grain mass at 13% moisture was 6,470 kg ha⁻¹, which is much higher than the Brazilian average of 3373 kg ha⁻¹.



Figure 9. Soybean total dry matter DM accumulation as a function of time (days after emergence, DAE) (points are experimental and solid line follows equation (2)). DM accumulation rate dDM/dt (bell shaped dotted line). Mmax is the maximum accumulation rate, parameter **a** the limit of total DM accumulation and parameter **b** the moment DAE of the maximum accumulation rate.

The DM partition to root, stem, leaf and reproductive organs for the soybean crop is shown in Figure 10 through their accumulation rates dDM/dt. Through the maximum values the greatest accumulation is for the reproductive organs, with a peak at DAE 100. Well

before that, stem, leaves and roots started with deceasing accumulation rates, indicating a great translocation of DM components from them to the reproductive organs. The relative position of the peak rates, root DAE 55, leaf DAE 60 and stem DAE 66, show that the crop first invested into roots, followed by leaves and then by the stem. Data also show that in terms of DM, this soybean variety invests more in stem than in leaves and roots.



Figure 10. Soybean DM partition shown as shown as DM accumulation rates dDM/dt, for root DMr⁻, stem DMs⁻, leaf DMI⁻ and reproductive organs DMro⁻.

3.3.1.2 Maize

The DM accumulation of the two maize crops is shown in Figure 11 and 12, using the sigmoidal model of equations (2) to (8). Experimental data of the first maize crop, grown in the main season (October to April), also presented a very good adjustment to the model ($R^2 = 0.98$), with a vegetative period of 62 days and a reproductive period of 78 days. The second maize crop grown right after the soybean and on the same field (as commonly done by farmers), presented also a very good adjustment to the model ($R^2 = 0.98$), with a 10 days longer vegetative period of 72 days and a reproductive period of 55 days. During the main growing season (from October 18th 2017 to March 06th 2018) the field received an amount of photosynthetic active radiation PARt of 1,856 MJ ha⁻¹, with an average of 13.5 MJ ha⁻¹ day⁻¹, while during the second growing season (April 11th 2018 to August 21th 2018) 1,256 MJ ha⁻¹, with an average of 9.5 MJ ha⁻¹ day⁻¹. This explains part of the difference between the values of Mmax for the two growing seasons.



Figure 11. Main season maize total dry matter (DM) accumulation as a function of time DAE (points are experimental and solid line follows Equation (2)). DM accumulation rate (dDM/dt) (bell shaped dotted line). M_{max} is the maximum accumulation rate, parameter **a** the limit of total DM accumulation and parameter **b** the moment (days after emergence) of the maximum accumulation rate.



Figure 12. Second season maize total dry matter (DM) accumulation as a function of time (points are experimental and solid line follows equation (2). DM accumulation rate (dDM/dt) (bell shaped dotted line). M_{max} is the maximum accumulation rate, parameter **a** the limit of total DM accumulation and parameter **b** the moment (days after emergence) of the maximum accumulation rate.

The DM partition to root, stem, leaf and reproductive organs for the main season maize crop is shown in Figure 13 through their accumulation rates dDM/dt. Through the maximum values, the greatest accumulation is for the reproductive organs, with a peak at DAE 78. Well before that, stem, leaves and roots started with deceasing accumulation rates, indicating a great translocation of DM components from them to the reproductive organs. The relative position of the peak rates, leaf DAE 32, root DAE 35 and stem DAE 48, show that the crop first inverted into leaves, followed by roots and then by the stem. Data also show that in terms of DM, this maize variety invests more in stem than in leaves and roots.

The DM partition for the second season maize crop is shown in Figure 14, also through their accumulation rates dDM/dt. Through the maximum values, it can be seen that the greatest accumulation is for the reproductive organs, with a peak at DAE 90. Well before that, stem, leaves and roots started with deceasing accumulation rates, indicating a great translocation of DM components from them to the reproductive organs. The relative position of the peak rates, leaf DAE 35, root DAE 50 and stem DAE 53, show that the crop first

invested into leaves, followed by roots and then by the stem. Data also show that in terms of DM, this maize variety also invests more in stem than in leaves and roots.



Figure 13. Main season maize dry matter (DM) partition shown as shown as DM accumulation rates (dDM/dt), for root (DMr²), stem (DMs²), leaf (DMl²) and reproductive organs (DMro²).



Figure 14. Second season maize dry matter (DM) partition shown as shown as DM accumulation rates (dDM/dt), for root (DMr⁻), stem (DMs⁻), leaf (DMl⁻) and reproductive organs (DMro⁻).

3.3.2 Photosynthesis

The net photosynthesis NP (kg [carbohydrate] ha⁻¹) calculated as indicated in the fluxogram of Figure 8, also had a sigmoidal behavior for the three crops. Soybeans and the main season maize ended up with higher values at the end of the cycle, due to the greater

solar incidence of this season. The second season maize presented the lowest value at the end of the cycle. The maize of the first season started first increasing NP, followed by second season maize and soybean, with respective peaks at DAEs 60, 73 and 86. These results indicate that NP decreased during the reproductive periods for all crops, showing that redistribution from the other organs to the reproductive organs was the main process at this period (Figure 15).



Figure 15. Net Photosynthesis NP and its rates (dNP/dt) for the three crops: soybean, main and second season maize.

The results of Carbon dioxide assimilation Cda, $\left(\frac{kg[CO2]}{ha[leaf].hour}\right)$, presented the highest peak (1200) for soybean, indicating a supremacy of the soybean in relation to maize, with respect to carbon sequestration. The earliest peak of Cda occurred at DAE 60 for the first season of maize (800), however 33% lower than the soybean. The second season maize crop presented an even lower peak (500) occurring at DAE 75 (Figure 16).



Figure 16. Carbon dioxide assimilation (Cda) for the three crops: soybean, main season maize and second season maize.

The efficiency of carbon and light use ECLU is the amount of carbon dioxide assimilation per unit of incident PAR. Figure 17 shows the daily values of ECLU for the three crops and their main values over the cycle. Daily values vary considerably during the cycle due to the great daily variability of solar radiation incidence in relation to the continuous DM accumulation of the plants. The average values show that soybean has the best efficiency, followed by the main season maize and finally second season maize.



Figure 17. ECLU_i = Efficiency of carbon and light use on day i ECLU and average values, for the tree crops: soybean, main season maize and second season maize.

3.4. Discussion

A method is presented for the estimation of the carbon dioxide assimilation Cda using soybean and maize as test plants. Based on the growth pattern of these crops and of local climatic conditions the method followed a calculation of Cda.

The soybean crop DM development shown in Figures 10 and 11 indicate a normal growth pattern that should be a response to the solar radiation input and to the availability of nutrients and water. The sigmoidal model adapted very well to the experimental data collected every 14 days after plant emergence. The total DM production at physiological maturity of 33,685 kg ha⁻¹ can be considered high for this environment, resulting in a grain yield of 6,470 kg ha⁻¹ at 13% moisture, while the Brazilian average soybean productivity is of the order of 3,373 kg ha⁻¹. The DM partition shown in Figure 10 indicates that the plant initially allocated the photosynthetic products first to root, then to leaves and stem. The areas under the bell shaped curves of DM growth rates represent the total amounts of accumulated DM, therefore, during the vegetative period more DM was allocated to the stem, in second place leaves and last to roots. This should be related to the semi-indeterminate characteristic of this soybean variety. The high productivity of this variety is also related to the DM partition to the reproductive organs.

With respect to maize, we present data of two crops grown in sequence, from October 2017 to July 2018, with a wide spectrum of incoming solar radiation. The main growing season has its beginning after the first rains of the wet season and extends to about March-April of the next year. The second season has a lower precipitation and less solar radiation, but very sufficient to conduct a second crop. This second crop, in Brazil called second season, became the most important cropping season in terms of maize production, even surpassing the maize production of the main season. Comparing both crops in Figures 13 and 14, it becomes clear that the second crop has a lower total DM production limit in comparison to the previous main crop. The maximum growth rate also occurs sooner for the main crop (DAE 60) in relation to the second crop (DAE 75). The grain productivity of these crops at 13% moisture were 13,166 kg ha⁻¹ for the main crop and 12,160 kgha⁻¹ for the second crop. It has also to be said that the sigmoidal model of Equation (2) adjusted well to the experimental data. The partition of DM shown in Figures 13 and 14 present a very similar pattern. However, the main crop shows a greater contribution to roots, stem and leaves in relation to the second crop. The allocation to the reproductive organs looks very similar, showing only a delay in the maximum value of 10 days for the second crop. The length of the growth cycle was essentially the same for the two crops.

The net photosynthesis was calculated according to the fluxogram of Figure 8 and is presented in Figure 15. These calculations used the energy conversion factors (Table 7) suggested by Penning de Vries (1999). These factors are being used in the literature since then because no refinement was done on them to date. The NP shown in Figure 15 is cumulative, that is, kg ha⁻¹ of carbohydrate accumulated from DAE 0 to DAE_i. As it can be seen the main season maize assimilated more carbohydrates up to DAE 120, being surpassed by the soybean only at the very end of the cycles. The second maize crop presented lower values of NP in relation to the main maize crop due to the lower incident solar radiation in this season. The NP rates, also presented in Figure 15, of the main season maize show that the maximum value appeared earlier (DAE 60) in relation to the second season maize crop (DAE 73) and soybean (DAE 87). Such results also show a supremacy of the maize with respect to soybean. Even though, the daily carbon assimilation rate Cda shown in Figure 16 shows a supremacy of the soybean in relation to maize, indicating a greater carbon sequestration of this crop. Its magnitude expressed by equation (12) is a direct function of the net photosynthesis rate NP' and of the leaf area index LAI. LAI is included to transform the assimilation rate based on soil area (ha) into leaf area. This is important for the application of this equation to crops of different plant population, consequently with different leaf areas. The calculation also involves the sunshine hours H and the coefficients of respiration, maintenance and growth, and of solar radiation interception, which are crop and local climate parameters. Both maize crops presented a lower Cda with respect to the soybean, and the second maize crop even lower values. Cda is the main output of this methodology and can be extended for other crops and used for carbon sequestration calculations.

In accordance to the results of ECLU (Figure 17), soybean also presented a supremacy over maize, in relation to the efficiency of carbon and light use. The average of soybean was 9099, while for the main season maize it was 6038, and second season maize 2821.

3.5. Conclusions

This study showed that the soybean, a 3C photosynthetic plant is a better carbon fixing crop, in relation to maize, a 4C photosynthetic plant.

The proposed method for the determination of the Carbon Dioxide Assimilation gave good results for soybean and maize crops. The method is more complete and solid because it is based on the complete growth pattern of the cycle of the crops.

Soybean presented a greater efficiency of light and carbon use in relation to maize, grown under the same soil-water-climate conditions.

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4. GENERAL CONCLUSIONS

With respect to plant population and row spacings it was shown that for the early maturity and high yield potential maize hybrid under study that the increase of 65,000 to 85,000 plants ha⁻¹ (+30.1 %) resulted in a yield decrease from 12.4 to 11.3 Mg ha⁻¹ (-8.9 %). Therefore, the increase of plants ha⁻¹ in relation to the recommended population of 65,000 plants ha⁻¹ is shown not to be a viable practice, several times sought by farmers. Row spacings varying from 0.45 to 0.90 m did not interfere with yield, but 0.90 had a higher dry matter accumulation. The greater soil area occupied by one plant increased plant growth mainly during the reproductive growth period, improved solar radiation absorption through greater light extinction coefficient. The beginning of the reproductive period was not affected by plant density and row spacing.

With respect to the proposed method for the determination of the Carbon Dioxide Assimilation, the study presented good results for soybean and maize crops. The method here presented is more complete and solid because it is based on the complete growth pattern of the cycle of the crops. This study also showed that the soybean, a 3C photosynthetic plant, is a better carbon fixing crop, in relation to maize, a 4C photosynthetic plant. As a result, Soybean presented a greater efficiency of light and carbon use in relation to maize, grown under the same soil-water-climate conditions.

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