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## Nitrogen and phosphate fertilizers in elephant-grass for energy use

José Augusto de Almeida Sant'Ana<sup>1</sup>, Rogério Figueiredo Daher<sup>2</sup>, Niraldo José Ponciano<sup>2</sup>, Marcia Maria Paes Santos<sup>1</sup>, Alexandre Pio Viana<sup>1</sup>, Erik da Silva Oliveira<sup>1</sup>, Francisco José da Silva Ledo<sup>3</sup>, Bruna Rafaela da Silva Menezes<sup>4\*</sup>, Carlos Lacy Santos<sup>1</sup> and Wallace Luís de Lima<sup>1</sup>

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Nitrogen is the mineral element that plants most require. Its deficiency quickly inhibits plant growth. Excessive nitrogen fertilization raises the cost of biomass production. Therefore, there is a need to find the appropriate dose of nitrogen fertilizer that provides greater efficiency in dry matter production. The objective of this study was to evaluate the effects of nitrogen and phosphate fertilizers on the chemical composition of the biomass of elephant-grass for energy use. A randomized-blocks experimental design in a split-plot arrangement was adopted. Three elephant-grass genotypes were evaluated (Guaçu/IZ.2, Cameroon-Piracicaba, and Capim Cana D'África) with five levels of nitrogen fertilization (0, 250, 500, 1,000 and 2,000 kg ha<sup>-1</sup>), as urea, and four levels of phosphate fertilization (50, 100, 200 and 400 kg ha<sup>-1</sup>), as single superphosphate. Results showed that the chemical composition, determined by the neutral detergent fiber (NDF), acid detergent fiber (ADF), cellulose (CEL), and lignin (LIG), contents displayed values that qualified the researched genotypes for energy use. Nitrogen applications had a quadratic effect on NDF and LIG contents in genotype Guaçu/IZ.2. A maximum dose of nitrogen fertilizer that will provide greater efficiency in the use of nitrogen for energy use varies among elephant-grass genotypes.

**Key words:** acid detergent fiber, cellulose, lignin, neutral detergent fiber.

### INTRODUCTION

Finite energy sources have put the world system on alert. Fossil fuels (oil, coal, and natural gas) are part of this scenario, and these are highly pollutant and affect the

planet's balance, causing the greenhouse effect. Thus, the search for alternative sources to fossil energies has become a challenge to the world scientific and

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governmental communities.

Elephant-grass is a species of the family Poaceae with great photosynthetic (C4 metabolism) and dry-matter accumulation abilities that can be compared to sugarcane. Research has shown its tremendous advantages compared with other sources energy. The species has great productivity in smaller areas, a shorter productive cycle with better cash flow, the possibility of total mechanization, renewable energy, and higher carbon assimilation (Quesada, 2005; Mazzarella, 2007).

In the search for sustainable biomass production for energy generation, the Poaceae species, especially sugarcane (*Saccharum officinarum*) and elephant-grass (*Pennisetum purpureum* Schum) stand out for their qualitative features. Fiber represents over 30% of the composition of these plants, which qualifies them for energy production (Urquiaga et al., 1992; Boddey, 1995; Samson et al., 2005; Kauter et al., 2006). When in its fresh state, elephant-grass can be used in direct combustion in ovens, substituting wood and coal, often going through a step of densification and structuring into pellets, cubes, or briquettes. At the energy balance, a ratio of 21.3 units of renewable energy per unit of fossil energy was found in the agricultural production process (Samson et al., 2005).

According to Taiz and Zeiger (2009), nitrogen is the mineral element that plants most require. Its deficiency quickly inhibits plant growth. Phosphorus, on the other hand, is an integral component of important plant cell compounds; it is used in the energy metabolism of plants. Phosphorus is of the macronutrients less required by the plant, but the Brazilian soils are poor in this element. In addition, phosphate fertilization stimulates the nitrogen absorption by the plant (Moreira et al., 2006). The increasing application of nitrogen fertilization significantly increases dry matter production (Flores et al., 2012). And dry matter production is positively correlated with fiber production in elephant-grass (Rossi et al., 2014).

Van Soest (1994) stated that the cell wall is composed of low-solubility structural carbohydrates, which correspond to the crude fiber fraction of the forage. The cell wall can be separated into neutral detergent fiber (NDF), which determines its concentration in the plant and expresses the cellulose and hemicellulose; and acid detergent fiber (ADF), which determines the cell wall quality and expresses the lignin, silica, and cutin. NDF and ADF are directly proportional to the calorific value of the material. Lignin is a component of the plant cell wall and an inhibitor of the digestibility of forage plants whose activity increases as the plant matures (Fukushima et al., 2000). Cellulose is the polysaccharide formed by  $\beta$ -glucose units, with high molecular weight (300,000 to 500,000 g mol<sup>-1</sup>). Having a linear structure, it is the main component of the cell wall (Penedo, 1980).

Excessive nitrogen fertilization raises the cost of biomass production. This is because ammonia synthesis requires high investment in fossil energy (Robertson and

Grace, 2004). Therefore, there is a need to find the appropriate dose of nitrogen fertilizer that provides greater efficiency in dry matter production.

Thus, the objective of this study was to evaluate the effect of nitrogen and phosphate fertilizers on the chemical composition of the biomass of elephant-grass for energy use.

## MATERIALS AND METHODS

The trial was conducted in the facilities of the Instituto Federal do Espírito Santo, Alegre Campus, in Espírito Santo State, Brazil ("20°45'57.9" S latitude, "41°27'23.93" W longitude, and 126 m altitude). According to Köppen's international classification, the climate of the planting region is a Cwa type, that is, a tropical hot and wet type, with cold and dry winters, an average annual temperature of 23.1°C, and total average annual precipitation of 1,341 mm (Lima et al., 2008). The experiment was developed on a soil classified as a Eutrophic Udox (Embrapa, 2006).

Soil samples were collected from the 0 to 20 cm layer for an analysis of particle size, whose results were: sand, 76.25%; silt, 2.51%; and clay, 21.23%. The results of the chemical analyses of the soil sampled from the 0 to 20 cm were: pH in water, 5.96; hydrogen + aluminum, calcium, and magnesium, CEC (pH 7.0), 1.93, 1.5, 0.53, and 4.16 cmol dm<sup>-3</sup>, respectively; phosphorus, 19 mg dm<sup>-3</sup>; and potassium, 67 mg dm<sup>-3</sup>.

A randomized-blocks experimental design, in a split-plot arrangement, was adopted. Three elephant-grass genotypes (Guaçu/IZ.2, Cameroon-Piracicaba, and Capim Cana D'África) were evaluated, with five levels of nitrogen fertilization and four levels of phosphate fertilization applied. Three replicates were utilized per treatment. The experimental area was formed by thirty-six 12-m rows, and 3.0 m of the extremities of each planting row served as border. Rows were spaced by 1.5 m, and each block was formed by 60 experimental units with 2.40 m of linear extension. Stems were placed aligned in a row, into furrows, arranged with the base of a plant touching the apex of another plant. Stems, with 50 and 60 cm in length, were covered with 3 cm of soil. A cutting height of 10 cm. Five levels of nitrogen fertilization (0, 250, 500, 1,000, and 2,000 kg ha<sup>-1</sup> of urea) were applied in installments during the cycle of the crop (March 05, 2011; October 3, 2011; January 31, 2012; March 17, 2012; and April 21, 2012). Four doses of phosphate fertilizer (50, 100, 200, and 400 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub>) were applied in the sub-plot, using single superphosphate.

The experiment was established on April 22, 2010. Two plot-leveling cuts were made due to climatic factors and flaws encountered at planting. The first plot-leveling cut occurred on October 19, 2010 (180 days after planting), and the second was made on March 02, 2011. The first evaluation harvest was performed on August 29, 2010 (180 days after the last plot-leveling cut), and the second on June 25, 2011 (300 days after the first harvest). The chemical composition of the biomass was evaluated in the third harvest, made on December 22, 2011 (180 days after the second harvest). The material was weighed, chopped, and placed separately in paper bags, which were immediately dried in a forced-air oven at 65°C for 72 h. Subsequently, samples were weighed and ground in a Wiley mill with 1 mm sieves and conditioned in an airtight container to be analyzed later.

The biomass chemical composition characteristics percentage of neutral detergent fiber (NDF), percentage of acid detergent fiber (ADF), percentage of cellulose (CEL), and percentage of lignin (LIG) were evaluated in the Laboratory of Food Analysis of Embrapa Dairy Cattle, in Coronel Pacheco - MG, Brazil, by near infrared reflectance spectroscopy (NIRS), using a Perstorp analytical spectrometer (Silver Spring, MD, model 5000) coupled to



**Table 1.** Summary of the analysis of variance of the characteristics NDF, ADF, LIG, and CEL in the three elephant-grass genotypes evaluated in Alegre - ES, Brazil, 2011/2012.

Source of variation	Mean square (MS)			
	NDF	ADF	LIG	CEL
Block	254.9047	66.3207	15.54766	97.95584
Genotype (G)	0.05716 <sup>ns</sup>	7.6019 <sup>ns</sup>	2.8187 <sup>ns</sup>	0.7085 <sup>ns</sup>
**Error (a)	2.94576	2.90129	2.54953	1.15259
Phosphate (P)	1.4922 <sup>ns</sup>	1.5133 <sup>ns</sup>	0.0660 <sup>ns</sup>	0.3369 <sup>ns</sup>
G×P	0.3898 <sup>ns</sup>	0.6149 <sup>ns</sup>	0.5563 <sup>ns</sup>	1.0119 <sup>ns</sup>
**Error (b)	2.28027	3.57994	0.30223	3.18345
Nitrogen (N)	1.9640 <sup>ns</sup>	0.5189 <sup>ns</sup>	1.0665 <sup>ns</sup>	4.6015 <sup>ns</sup>
G×N	2.9240 <sup>ns</sup>	3.4473 <sup>ns</sup>	0.5829 <sup>ns</sup>	3.7262 <sup>ns</sup>
P×N	3.1080 <sup>ns</sup>	1.7703 <sup>ns</sup>	0.2504 <sup>ns</sup>	2.7645 <sup>ns</sup>
G×P×N	1.6207 <sup>ns</sup>	2.2501 <sup>ns</sup>	0.4112 <sup>ns</sup>	1.8646 <sup>ns</sup>
**Error(c)	2.33397	2.72142	0.40814	2.12657
Mean	77.21	48.86	9.65	37.40
CV (%)	1.9785	3.3762	6.6159	3.8984

NDF - percentage of neutral detergent fiber; ADF - percentage of acid detergent fiber; LIG - percentage of lignin; and CEL - percentage of cellulose.

\*\* Significant at 1% probability level according to the F test; \* Significant at 5% probability level according to the F test; ns = not significant.

a microcomputer equipped with ISI software version 4.1 (Infrasoft International, University, Park, PA). The material was read at the wavelengths of 1,100 to 2,500 nm.

Statistical analysis was performed using GENES software (Cruz, 2013). An analysis of variance was initially run based on the average of the sub-subplots, utilizing the following statistical model (split-split-plot):

$$Y_{ijkl} = \mu + B_i + G_j + \varepsilon_a + P_j + (GP)_{ij} + \varepsilon_b + N_k + (GN)_{ik} + (PN)_{jk} + (GPN)_{ijk} + \varepsilon_c \quad (1)$$

where:

$Y_{ijkl}$  = observed value relative to genotype i, at phosphorus level j and nitrogen level k, in block l;  $\mu$  = overall mean of the trial;  $G_i$  = effect of genotype i;  $B_l$  = effect of block l;  $\varepsilon_a$  = effect of the error associated with genotype i in block l ~NID (0,  $\sigma_{\varepsilon_a}^2$ );  $P_j$  = effect of phosphorus level j;  $(GP)_{ij}$  = effect of the interaction between genotype i and phosphorus level j;  $\varepsilon_b$  = effect of error b associated with genotype i at phosphorus level j, in block l ~NID (0,  $\sigma_{\varepsilon_b}^2$ );  $N_k$  = effect of nitrogen level k;  $(GN)_{ik}$  = effect of the interaction between genotype i and nitrogen level k;  $(PN)_{jk}$  = effect of the interaction between phosphorus level j and nitrogen level k;  $(GPN)_{ijk}$  = effect of the interaction among genotype i, phosphorus level j, and nitrogen level k;  $\varepsilon_c$  = effect of error c associated with genotype i at phosphorus level j and nitrogen level k, in block l ~NID (0,  $\sigma_{\varepsilon_c}^2$ ).  $\varepsilon_a$ ,  $\varepsilon_b$  and  $\varepsilon_c$  ~NID (0,  $\sigma_{\varepsilon_{a,b,c}}^2$ ).

Tukey's test of means was used for the evaluated characteristics, at a significance level of 5%. For the case of a significant effect involving the source of variation nitrogen levels, a combined polynomial regression analysis was used for the 1st- and 2nd-degree linear models, with the respective analyses of variance of regression, testing the significances of the sources of variation due to regression and due to regression deviations.

## RESULTS AND DISCUSSION

The observed coefficients of variation (CV) for NDF, ADF, LIG, and CEL were 1.97, 3.37, 6.61, and 3.89, respectively (Table 1). They were considered low, which indicates good experimental quality for the evaluated characteristics (Cargnelutti and Storck, 2007).

A non-significant effect ( $p > 0.05$ ) was detected for all characteristics evaluated in all sources of variation related to the main effects. The statistical analyses showed that there was no significant interaction involving the different factors for the evaluated chemical composition characteristics. Because of the lack of significance for the sources of variation containing the phosphorus factor isolated (main effect) or interacting with the other factors genotype and nitrogen (genotype × phosphorus, nitrogen × phosphorus, and nitrogen × phosphorus × genotype), the levels of this factor were considered restrictions to the procedures of the other analyses (Table 1). The mean values for the characteristics of the biomass chemical composition (NDF, ADF, LIG, and CEL) resulting from the statistical analyses, for every genotype evaluated as a function of the nitrogen-fertilization levels, are described in Table 2.

**Table 2.** Mean values for the chemical composition characteristics of each group related to the five levels of nitrogen in Alegre - ES, Brazil, 2011/2012.

Trait	Genotype	Doses of nitrogen (kg ha <sup>-1</sup> )				
		0	500	1000	1500	2000
NDF	Guaçu/IZ.2	76.60 <sup>a</sup>	77.55 <sup>a</sup>	77.65 <sup>a</sup>	77.79 <sup>a</sup>	75.40 <sup>a</sup>
	Cameroon	77.61 <sup>a</sup>	77.28 <sup>a</sup>	77.23 <sup>a</sup>	76.54 <sup>a</sup>	77.30 <sup>a</sup>
	Capim Cana D'África	77.17 <sup>a</sup>	77.72 <sup>a</sup>	77.14 <sup>a</sup>	77.22 <sup>a</sup>	76.99 <sup>a</sup>
ADF	Guaçu/IZ.2	47.80 <sup>b</sup>	48.70 <sup>a</sup>	49.03 <sup>a</sup>	49.05 <sup>a</sup>	48.46 <sup>a</sup>
	Cameroon	50.07 <sup>a</sup>	49.05 <sup>a</sup>	49.38 <sup>a</sup>	48.83 <sup>a</sup>	48.99 <sup>a</sup>
	Capim Cana D'África	48.22 <sup>b</sup>	48.86 <sup>a</sup>	48.49 <sup>a</sup>	49.04 <sup>a</sup>	48.88 <sup>a</sup>
LIG	Guaçu/IZ.2	9.11 <sup>b</sup>	9.48 <sup>a</sup>	9.82 <sup>a</sup>	9.76 <sup>a</sup>	9.20 <sup>b</sup>
	Cameroon	9.93 <sup>a</sup>	9.73 <sup>a</sup>	9.72 <sup>a</sup>	10.12 <sup>a</sup>	9.97 <sup>a</sup>
	Capim Cana D'África	9.17 <sup>b</sup>	9.65 <sup>a</sup>	9.68 <sup>a</sup>	9.73 <sup>a</sup>	9.70 <sup>ab</sup>
CEL	Guaçu/IZ.2	37.18 <sup>a</sup>	37.55 <sup>a</sup>	37.74 <sup>a</sup>	37.76 <sup>a</sup>	36.72 <sup>a</sup>
	Cameroon	37.49 <sup>a</sup>	37.29 <sup>a</sup>	37.44 <sup>a</sup>	36.36 <sup>b</sup>	36.93 <sup>a</sup>
	Capim Cana D'África	37.98 <sup>a</sup>	37.74 <sup>a</sup>	37.04 <sup>a</sup>	37.77 <sup>a</sup>	37.05 <sup>a</sup>

NDF - percentage of neutral detergent fiber; ADF - percentage of acid detergent fiber; LIG - percentage of lignin; and CEL - percentage of cellulose. Means followed by common letters vertically do not differ statistically at 5% probability level according to Tukey's test.

Neutral detergent fiber (NDF) values ranging from 75.40 to 77.79% were found, with nitrogen not affecting the evaluated contents. These values were similar to those obtained by Magalhães et al. (2009), in studies on the effect of nitrogen levels on three elephant-grass genotypes. These authors observed average NDF contents around 70%, with no significant effects of nitrogen fertilization on this characteristic. These values were considered acceptable for the use of elephant-grass as an alternative energy source.

Santos (2011) in an experiment with cultivation of elephant-grass found NDF contents between 73.72% (with mineral gypsum) and 76.56% (without gypsum) for Cameroon; 76.71% (with mineral gypsum) and 78.42% (without gypsum) for Gramafante; and 77.26% (with mineral gypsum) and 77.88% (without gypsum) for Roxo. Flores et al. (2013) evaluated the effects of nitrogen fertilization and harvesting age on the quality of elephant-grass biomass and observed mean NDF values of 69 and 58% in the stem and in the leaf, respectively, and that nitrogen fertilization did not influence these concentrations.

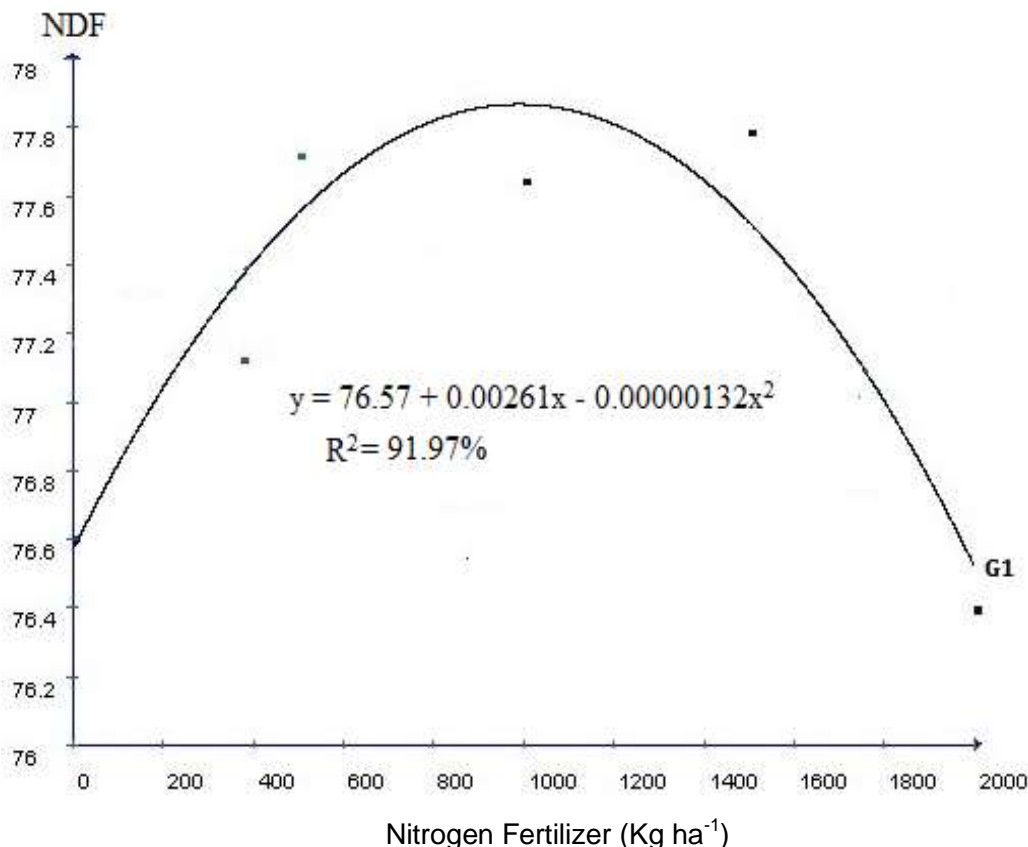
The mean values for the characteristic percentage of ADF between the analyzed genotypes varied from 47.80 to 50.07% (Table 2). A significant difference was observed between the means for the lack of nitrogen fertilization, in which the mean of genotype Cameroon stood out among the means of the other genotypes. These values agree with those found by Filho et al. (2000), who, after 100 days of cultivating elephant-grass cultivar Roxo, obtained 48% ADF.

Santos et al. (2003) found, in tropical grasses, ADF

values of 36 and 38%, in varieties Mott and Pioneiro, respectively. Martins-Costa et al. (2008) were evaluating the nutritive value of elephant-grass at different harvest ages, found percentage values ranging from 41.37 and 49.40%. Rossi (2010) in an experiment with the elephant-grass genotypes Guaçu/IZ.2, Cameroon, and Capim Cana D'África, found values varying between 43.14 and 46.47%. Parente et al. (2012) found ADF contents of 40.53 and 46.40% in elephant-grass subjected to four nitrogen fertilization levels (0, 100, 200, and 300 kg ha<sup>-1</sup>).

Lignin contents (LIG) were found to range from 9.11 to 10.12% (Table 2). A significant difference was observed between the means for the lack of nitrogen fertilization, in which the mean of genotype Cameroon stood out among the genotypes. Genotype Cameroon displayed a significant difference compared with the mean of Guaçu/IZ.2 at the fertilization level of 2,000 kg ha<sup>-1</sup>. Zanetti et al. (2009) evaluated five elephant-grass genotypes and found LIG values of 9.7 and 13.3% for genotype Cameroon in soils classified as Red-Yellow Ultisol and Haplic Ultisol, respectively. In an experiment with five elephant-grass genotypes, with plants at 180 days of age, Morais et al. (2009) obtained 9 to 9.7% for this characteristic.

Cellulose values were found to range from 36.72 to 37.98% (Table 2). A significant difference was observed between the means for the level of nitrogen fertilization (1500 kg ha<sup>-1</sup>), with the mean of genotype Cameroon standing out among the others for its lowest value. Zanetti et al. (2009), evaluating elephant-grass genotypes in Red-Yellow Ultisol and Haplic Ultisol soils, found CEL values of 28.7 and 26.8% for genotype



**Figure 1.** 2nd degree growth curve for the characteristic percentage of neutral detergent fiber (NDF) according to levels of nitrogen fertilizer for genotype Guaçu/IZ.2 (G1).

Cameroon, respectively. Quesada et al. (2004) found values above 40%. Morais et al. (2009) found CEL values of 33.8 to 35.8%.

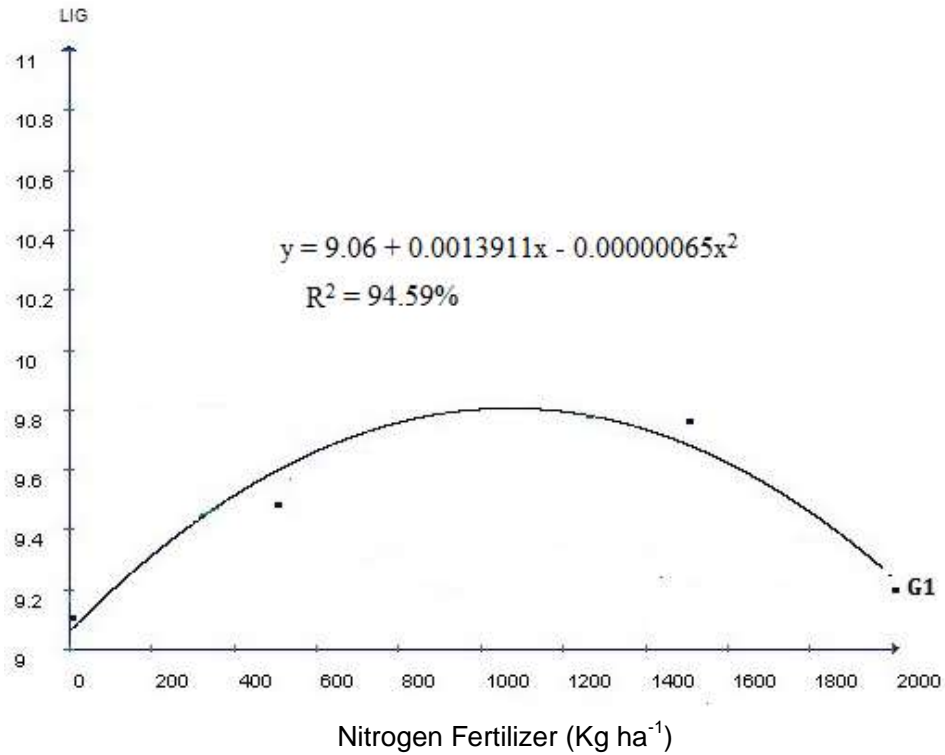
Results revealed  $R^2 = 91.97\%$ , that is, that 91.97% of the variation in the response NDF are explained by the regression equation  $y = 76.57 + 0.00261x - 0.00000132x^2$ , with the maximum value estimated at 988.63 kg ha<sup>-1</sup> of nitrogen fertilization (Figure 1). For genotypes Cameroon and Capim Cana D'África, no significant effect ( $p > 0.05$ ) of nitrogen fertilizer influenced the NDF response. The estimate of the mean square for the 2-nd degree linear model for NDF according to the levels of N (fertilizer) for genotype Guaçu/IZ.2 (G1) was significant at 1% probability level, but not at 5%, by the F test.

For genotype Guaçu/IZ.2 (G1), the estimates of the mean square due to regression for the 2-nd degree linear model for NDF according to the N (fertilizer) levels were significant at  $p < 0.01$ , but not at  $p > 0.05$  by the F test. Results demonstrated an  $R^2 = 94.59\%$ , that is, 94.59% of the variation in the response are explained by the regression equation  $y = 9.06 + 0.0013911x - 0.00000065x^2$ , with maximum value estimated at 1,070.07 kg ha<sup>-1</sup> of nitrogen fertilization (Figure 2).

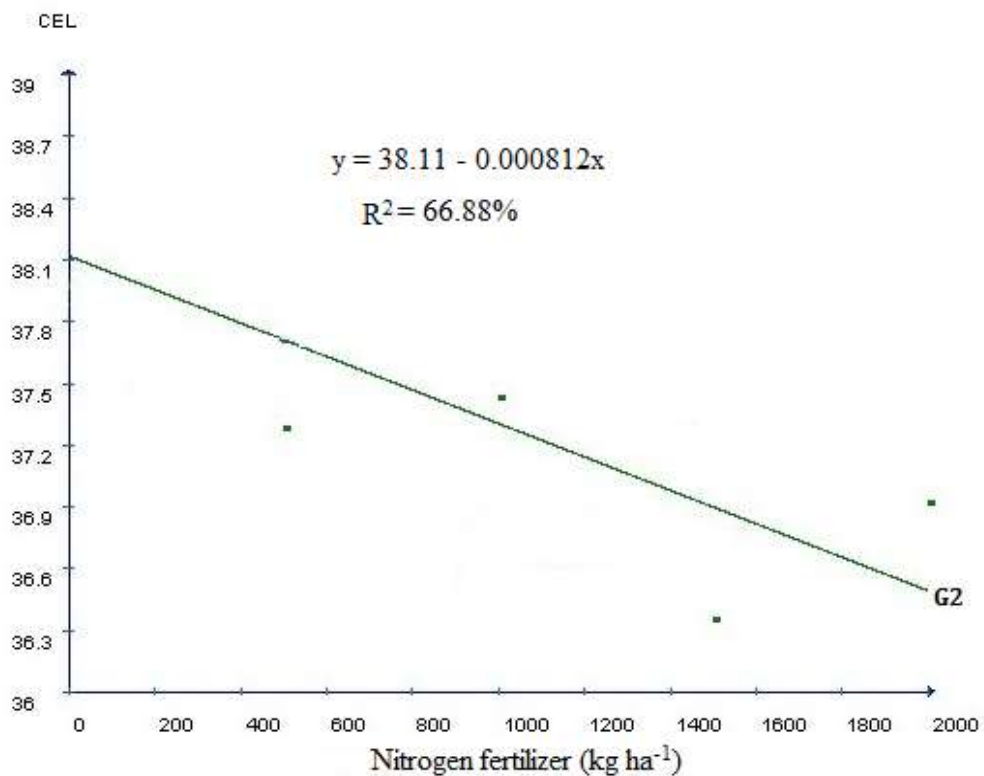
Beyond this value, LIG will have a significant linear effect for genotype Capim Cana D'África (G3), the estimates of the mean square due to regression for the 1st-degree linear model for the characteristic LIG as a function of N (fertilizer) levels were significant at  $p < 0.01$ . However, at 5% probability, these estimates were not significant. Results showed  $R^2 = 59.18\%$ , that is, 59.18% of the variation in the response are explained by the regression equation  $y = 9.36 + 0.000227x$  (Figure 4). Downward trend as the nitrogen fertilization is increased, in this genotype.

However, the increase of the nitrogen fertilization does not result in a significant increase in the LIG content from 1500 kg ha<sup>-1</sup>.

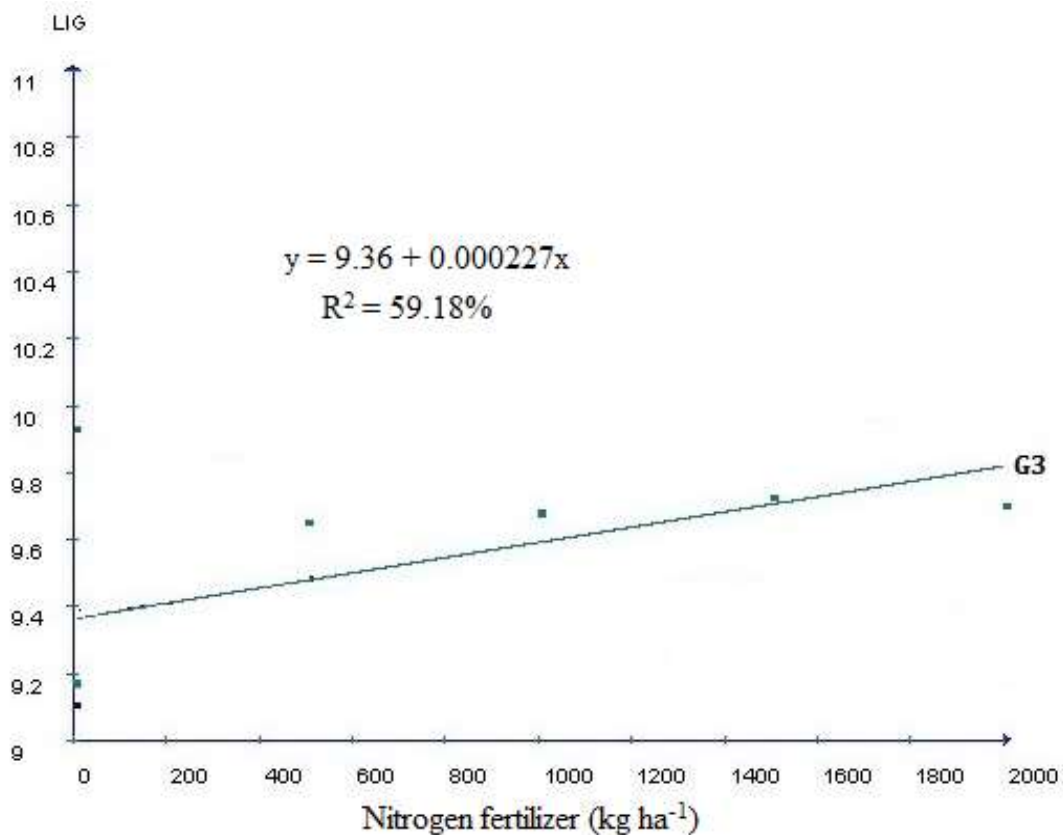
The estimates of the mean square for the 1-st degree linear model for CEL according to the N (fertilizer) levels for genotype Cameroon (G2) were significant at  $p < 0.01$  and not significant at  $p > 0.05$ , by the F test. These results indicate a reduction of cellulose with an increase in nitrogen fertilization (Figure 3). A similar result was found by Magalhães et al. (2015), who observed a decrease in the fiber content of palisade grass with the increase in the nitrogen fertilization level. In this case, the energy potential was reduced as nitrogen fertilization was



**Figure 2.** 2nd degree growth curve for the characteristic lignin (LIG) according to five levels of nitrogen fertilizer for genotype Guaçu/IZ.2 (G1).



**Figure 3.** 1st-degree growth curve for the characteristic cellulose (CEL) according to five levels of nitrogen fertilizer for genotype Cameroon (G2).



**Figure 4.** 1st-degree growth curve for the characteristic lignin (LIG) according to five levels of nitrogen fertilizer for genotype Capim Cana D'África (G3).

increased.

## Conclusions

The NDF, ADF, and LIG characteristics showed desirable values for energy use. A maximum dose of nitrogen fertilizer that will provide greater efficiency in the use of nitrogen for energy use varies among elephant-grass genotypes. According to the results presented, these doses would be 1000 kg ha<sup>-1</sup> for the Guaçu / IZ.2 genotype and 1500 kg ha<sup>-1</sup> dose of nitrogen for the Cameroon and Capim Cana D'África genotypes.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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*Full Length Research Paper*

## Thermal comfort analysis of agricultural machinery operators with thermography

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Heat stress appears as a strong candidate for physical wear and tear, leading to lower yields and high risks of accidents due to lack of attention. In this context, this work aims to study the possible influence of the internal temperature of metal roof tractor, skin temperature of face operator, relating it to possible physiological responses connected with environmental variables such as relative humidity air and wind speed. An insulating composite panel based on residual materials (cement bag) was prepared and placed under the tractor roof for thermal isolation purpose. The tractor used in the experiments is manufactured in 1997, with engine power of 173 hp at 2200 rpm; it works for 8200 h and has original factory roof. The experiment was carried out in the open field for five days, following the same start time (11 h 40 min) and ending time (13 h 40 min); it corresponds with the maximum sun intensity, where in the first hour there was panel and after 12 h 40 min, there was no panel. After analyzing the data, there was a reduction in the average temperature inside the cabin with the presence of the insulating panel. The physical workload, based on heart rate, remained in "light" range, even with no physical requirements for the operator. Regarding the average temperature facial skin, it showed an increase of 1.1°C after the panel removal. According to the classification of temperature level and humidity, despite a steady increase in value during the 2 h of the experiment, the index remained in the position of little discomfort. Based on the data, it is clear that simple measures can be taken to improve the thermal environment in tractors with roof control platform, in addition to stimulating the study for improving cooling in tractor cabin.

**Key words:** Instrumentation, sustainability, welfare, agribusiness.

### INTRODUCTION

A major difficulty faced by operators of agricultural machinery in hot environment is the harsh working conditions which can cause great discomfort and low income.

According to Jay and Kanye (2010), a high exposure time, combined with variations in temperature and

humidity in the workplace can put workers at risk. Actually, even with all the ergonomic evolution, targeting the security and welfare of the operator, agricultural tractors in circulation in the country, for most part, present problems of comfort and safety for operators (Santos et al., 2010).

According to Costa (2012), environmental risks are classified as physical, chemical and biological hazards. The authors comment on the importance of studying the problems caused by heat stress, for example heat rash, heat cramps, heat exhaustion and heat stroke.

Although there are tractors with acclimatized cabs, most of the fleet of the small and medium properties is composed of tractors that have only roofs; these may be of different materials. The use of roofs to avoid direct contact with heat is not enough to protect the operators from exposure to high temperature; it may lead to, according to Leite (2002), the same health problems, interfering with productivity and increase possibility of occurrence of errors that can lead to accidents.

Today, with the advancement in safety standards at work, there is a search for better conditions of ergonomics and operator safety, significantly reducing the incidence of illness linked to the steady work (Alves et al., 2011). Thus, it becomes more significant for the development of agricultural machine. However, recent studies point to the need for improvement and development in the ergonomic design of agricultural tractors (Baesso et al., 2014).

The human thermoregulation is essential for the maintenance of the physiological responses of production, absorption and heat loss. During exercise or even at rest, the human body needs instruments for achieving homeostasis and to ensure temperature of about 37.0°C, to avoid hyperthermia and hypothermia (Costa, 2012). Heat exchange between the human body and environment is done by an organic system called thermoregulation (Sousa, 2014).

The surface skin temperature and heart rate can act as important indicators for checking the thermal stress in machine operators. Therefore, instruments used for measuring chemical temperature and different types of contact thermal sensors have been used (Lim et al., 2008). However, this type of specific measure directly on the skin can infer misinterpretations, since heating does not occur uniformly on the skin (Vainer, 2005; Sousa, 2014). Thus, it is necessary to use a more appropriate method for measuring the surface temperature of the skin, such as the use of thermographic camera.

The use of infrared thermographic camera is considered, a non-destructive character of technical and non-invasive, since it does not have direct contact with the analyzed surface. The image is constituted from thermal infrared radiation emitted by different surface materials (Castanedo, 2005; Maldague, 2001; Tavares, 2006).

From these studies, it is feasible to do a comprehensive assessment of some factors that can

influence the thermal comfort of the operator tractors with metal cowling and may modify the conditions to which it is exposed through simple measures, such as the coupling of a panel composed of recycled materials.

## MATERIALS AND METHODS

The experiment was conducted at the Faculty of Animal Science and Food Engineering (FZEA), University of São Paulo (USP), Campus Fernando Costa, Pirassununga. The campus geographical location is latitude 21° 59'S and longitude 47° 26'W; it has an average height of 634 m. The climate is subtropical, Cwa, with dry winter and hot and rainy summer, according to Köppen classification (Oliveira and Prado, 1984).

For uniformity of data, the study area remained the same throughout the experiment; it was strategically chosen: it is an open area without trees and buildings that can cause shadows.

A tractor, manufactured in 1997, with engine power of 173 hp at 2200 rpm, was used; it works for 8200 h. The structural component of the control stage (the area where the tractor driver operates) follows the factory originality even though it is a machine already widely used.

A TESTO thermographic camera model 875-2 as well as their respective software, TESTO IRSoft version 3.7 was used to evaluate the temperature of human skin. Air temperature, relative humidity and wind speed were taken by two portable thermo-hygro-anemometer model THAL-300.

As proposed by Sousa (2014), experimental images were obtained at 10 min at a distance of 0.5 to 1.5 m in three different regions of interest: outer surface of the roof, inner surface of the roof and the facial skin surface operator.

After the image acquisition, it was treated by the respective software, in order to obtain data concerning the average temperature and humidity in a specific area of interest. This delimitation was carried out carefully to exclude not interesting objects that could adversely influence the measurements. In addition to the thermal images, the software allows selection of Palette "moisture image", providing a moisture image captured area.

For comparative purposes and changes in the thermal environment under the metal hood, a thermal insulating material board between the head of the operator and the tractor hood (roof) was employed. This thermal insulating board was supplied from Buildings Laboratory and Ambience (CONSTRAMBI). Four boards were produced with recycling material of empty cement bags and milk cartons. The panels have a size of 50 cm x 50 cm x 1.5 cm. The four plates were fixed inside the hood together in a single module with the aid of an MDF (Medium Density Fibreboard) with dimensions of 1,00 x 1,30 m. To fill empty areas and further reduce the heat transfer, polystyrene was used and covered with aluminium foil.

The acquisition rate for heart beat was given by a portable heart monitor DLK, model HRM-E001. The heart rate data were collected at intervals of 10 min for a period of 2 h of the experiment, thereby generating a database of 20 samples per experiment. Couto (1995) classified the physical workload in six levels, relating them to heart beats, as shown in Table 1.

The ITU is defined, according to Costa et al. (2009), using Equation 1:

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**Table 1.** Physical load classification work by heart beat rate.

Physical workload	Heart rate (bpm)
Very lightweight	<75
lightweight	75-100
Moderately heavy	100-125
Heavy	125-150
Very heavy	150-175
Extremely heavy	>175

Couto (1995). Ergonomia aplicada ao trabalho: o manual técnico da máquina humana. Belo Horizonte: Ergo.

**Table 2.** Classification of heat stress range according to the ITU.

Rage (°C)	Caloric stress
<20	Cold
15 - 20	Comfortable
20 - 25	Little discomfort
>25	Discomfort

Costa et al. (2009). Conforto térmico na cidade de Natal e Ceará Mirim/RN utilizando os métodos de ITU e WCI 3: 223-347.

$$ITU = T - 0.55 (1 - UR) (T - 14) \quad (1)$$

where ITU is the temperature and humidity index, °C; T is the Air temperature, °C; and UR is the relative humidity in decimal fraction. According to Costa et al. (2009), it is still possible to classify the comfort range according to the value of the ITU (Table 2).

This study was done with one operator; he is 23 years, 1.83 m tall, weighs 96 kg, healthy with no smoking or alcohol addictions. The works have adequate potable water close to the work area. According to Sousa (2014), the following caution should be taken:

- (1) Avoid any physical activity that could affect body temperature and blood flow on the recorded locations for at least 1 h before performing the operation;
- (2) Avoid drinking alcohol, coffee or tea prior to operation;
- (3) Avoid sun burning;
- (4) Avoid smoking during the operation;
- (5) Avoid using sunscreen and/or other cosmetics such as cream, gel or spray;
- (6) Avoid exposure to very hot water and bathe before the operation;
- (7) Avoid wearing tight clothes, avoiding any grip on the skin;
- (8) Avoid wearing rings, earrings, bracelets and chains during operation;
- (9) Avoid intercourse in the last 12 h;
- (10) Avoid the use of drugs;
- (11) Avoid eating 2 h before the experiment

The results were analyzed using simple linear regression, Pearson correlation coefficient and comparison of means by Tukey test at 5% probability.

## RESULTS AND DISCUSSION

The measured results are shown in graphics (Figures 1

to 6). Figures 1, 2, 4, and 6 show a red segmented vertical line oriented at 12 h and 40 min; in the first period (11 h 40 min to 12 h 40 min) the measurements were made with the panel attached but in the second period (12 h 40 min to 13 h 40 min), there was no panel.

The average values of the indoor and outdoor roof temperatures are represented respectively by  $T_{MIC}$  and  $T_{MEC}$  abbreviations for the two stages of the study, which means, with and without the insulating panel in the 2 h experiment (from 11:40 to 13:40 h) (Figure 1).

At the beginning of the experiment (11:40 to 12:00) an increase in temperature on either side of the roof is as shown in Figure 1. This is because the first samples were collected at the beginning of exposure to the sun. Thus, from the moment when the tractor is placed in the experimental area, both temperatures rise and in 20 min, stabilization is achieved. From the point of stabilization at 12.00 to the panel subtraction point at 12 h/40 min, the temperature difference between the inner surface of the panel in relation to the outer surface of the roof was inferior (12.8°C).

The last sampling performed with the panel was at 12 h and 40 min where an elevation of 10.8°C was found at an interval of 10 min prior to the next collection (12 h 50 min). At that moment, there is an inversion temperature of superiorities where  $T_{MIC}$  becomes an average of 2.4°C higher than  $T_{MEC}$ .

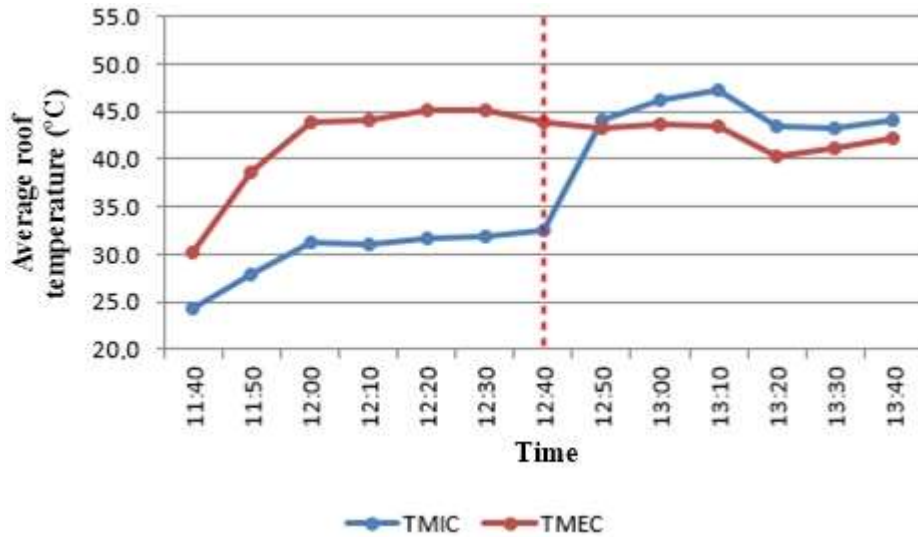
The panel showed good thermal insulation, as the average roof temperature was 31.62°C with the panel, rising to an average of 44.88°C without the same. It is also possible to check the heat reflection on the inner surface of the panel influencing the temperature outside the roof, since the average external temperature of the hood ranged from 44.4°C coupled with the panel to 42.58°C with the removal of the same. The panel can be considered to simulate a "greenhouse effect" with the metal tractor roof and interfered significantly in  $T_{MEC}$  and  $T_{MIC}$ , as shown in Table 3.

The mean values of heart rate were determined based on the maximum rate in each data collection interval. The average data of heartbeats are as shown in Figure 2.

It can be seen that before the start of the collections in full sun (11 h 40 min), the operator is in a range of 75 to 100 bpm, which is classified as "light work of physical load", keeping this category throughout the experiment. This result is normal, since no physical nature of activity that requires a significant change during the five experimental days has been established. Thus, the variation in heart rate may be limited to more functions connected exclusively to the thermoregulatory response.

Beginning a review of the beats with the tractor at 10 min in the environment (11 h/50 min), it appears that the average rate before the panel removal was 88.67 bpm; it rises to 92.57 bpm after 12 h 40 min, leading to a significant change.

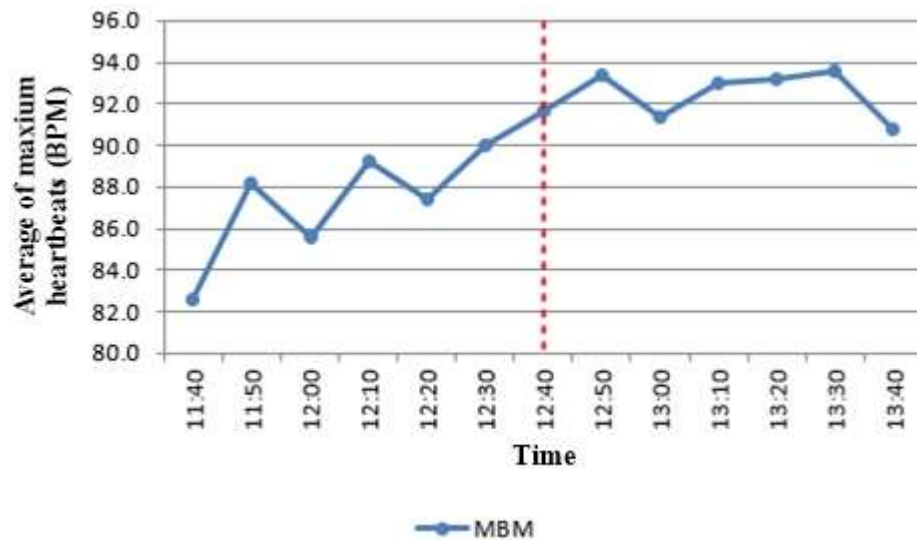
Initially, there is an adjustment of the human thermoregulatory mechanism in relation to the



**Figure 1.** Average temperature inside of the roof ( $T_{MIC}$ ) and average temperature outside of the roof ( $T_{MEC}$ ) during the experiment.

**Table 3.** Pearson correlations between the facial skin moisture and the variables: average wind speed, average internal temperature of the roof, cutaneous average temperature and average maximum heart rate.

Correlation of facial skin moisture with:	Pearson's coefficient	Classification
Average wind speed	-0.7	Strong negative linear correlation
Average internal temperature roof	0.7	Strong positive linear correlation
Cutaneous average temperature	-0.8	Strong negative linear correlation
Average maximum heart rate	0.5	Moderate positive linear correlation



**Figure 2.** Average of maximum heartbeats during the experiment.

environment in which the operator is inserted. Applying the method of Pearson correlation coefficients (Callegari-

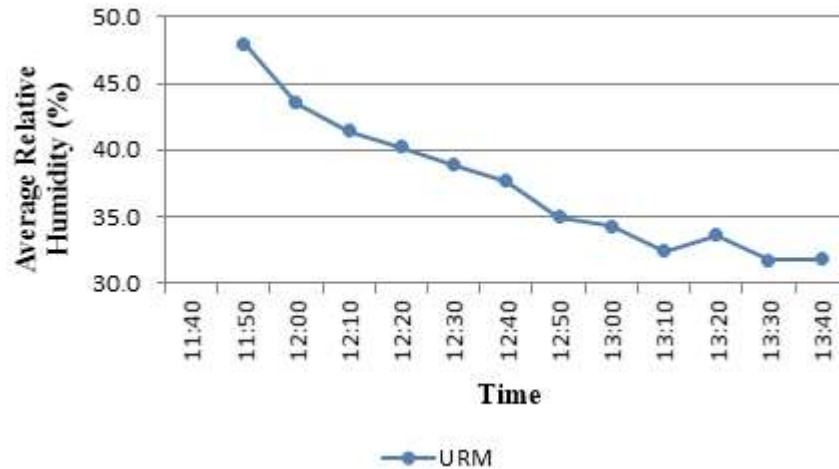


Figure 3. Average relative humidity during the experiment.

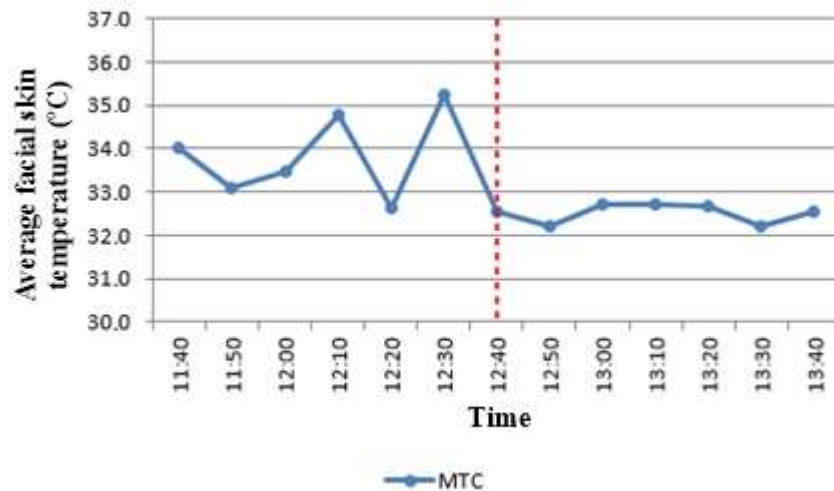


Figure 4. Average facial skin temperature during the experiment.

Jacques, 2003), there is a moderate positive linear correlation of 0.5 between the variables "average relative humidity" (Figure 3) and "average facial skin temperature" (Figure 4). This correlation may indicate that there was a more effective face heat exchange because of sweat evaporation (Widmaier et al., 2006) in the period after the panel subtraction.

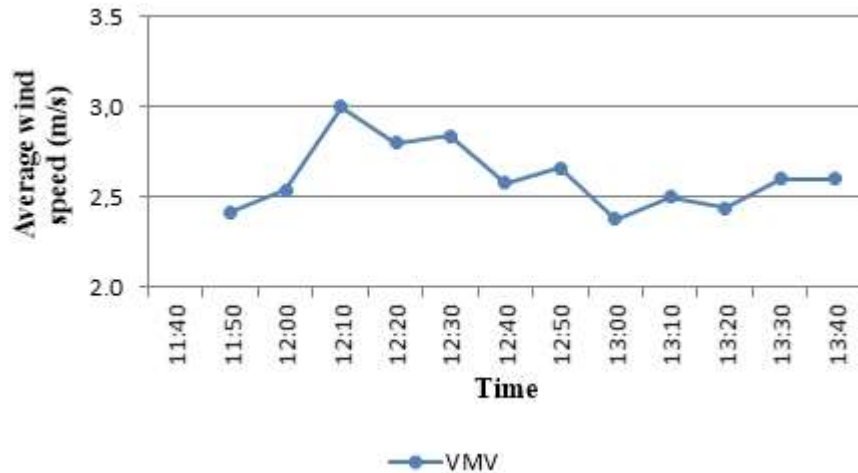
In the initial period (11 h 50 min to 12 h 40 min), there was a near 10% reduction in relative humidity, with an average of 41.6% relative humidity (12 h 40 min to 13 h 40 min); there was less variation range, close to 6%, with an average of 33.1% relative humidity. Therefore, this heat exchange (skin - environment) may have occurred in higher intensity after the removal of the panel, because of the reduction of the relative humidity experienced during the experiment. This is due to the fact that the lower the relative humidity, the easier would be the evaporation of

sweat; the higher the relative humidity, the more difficult would be the evaporation of sweat produced (McArdle et al., 1992)

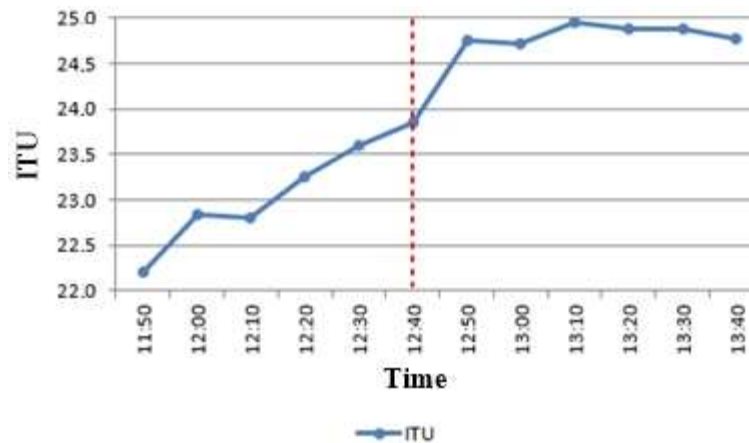
According to the data expressed in Figure 5, the average facial skin temperatures remained very close to that stipulated by Sousa (2014) (33.7°C). The skin temperature maintained at 33.6°C for the period with the panel and 32.5°C for the period without the panel indicate a significant difference.

For the calculation of ITU using the method developed by Costa et al. (2009), it is important that the average wind speed does not exceed  $3.0 \text{ m s}^{-1}$ . Thus, it becomes a feasible calculation using Equation 1, because as shown in Figure 6, all the averages remained in the range between  $2.0$  and  $3.0 \text{ m s}^{-1}$ .

The ITU was calculated based on the relative humidity in the control platform and the average air temperature.



**Figure 5.** Average wind speed ( $\text{m s}^{-1}$ ) during the experiment.



**Figure 6.** Temperature and humidity index (ITU) during the experiment.

These results are as shown in Figure 6, for all the periods collected. Before the data a short rise could be noticed in the respective index during the experiment, with a stabilization in an average range of 24.85 after the removal of the panel. It is important to note that the average ITU during the time that the panel remained attached to the roof was 23.43, lower than the average ITU when without the panel.

Based on this methodology, the ITU remained throughout the experiment in a range between 20 and 25, classified based on heat stress, as "little discomfort." However, there is a high ITU proximity for the last time to experiment with the limit of 25, which when exceeded would form part of the "uncomfortable" rating.

The correlation between the maximum average heart rate and temperature and moisture content did not exceed the range of "little discomfort"; it had a considerable rise, as shown in Figure 6, especially

subsequent to the panel subtraction (12 h 40 min). After the referred panel subtraction, there is the biggest jump of index between the measured intervals, rising to 0.9 units. Thus, a very strong linear correlation was obtained (Callegari-Jacques, 2003), equal to 0.9. Making it plausible to say that there is a strong positive correlation between the ITU and the heartbeat; it can be inferred that with increasing discomfort, there is an increase in heart rate.

Applying the method of Pearson correlations between the facial skin moisture and average wind speed, average internal temperature of the roof, cutaneous average temperature and average of heartbeats, there were positive and negative correlations, classified according to Callegari-Jacques (2003) (Table 3). It is evident that the average wind speed has a strong correlation, but reverses with facial skin moisture. This means that with an increase in wind speed, there is a decrease in skin

temperature by convection (Guyton and Hall, 2006; Widmaier et al., 2006).

The internal temperature of the roof has a strong positive correlation with the facial skin moisture, with a coefficient of 0.7. These data allow an understanding that there may be an influence of the inner roof temperature, regardless of the panel presence panel or facial moisture. This means that there is a thermoregulatory response of the body to increase the sweating rate to perform a loss of heat, when an increase in the internal temperature of the roof occurs.

The facial skin average temperature showed a strong negative correlation with facial moisture. These data suggest that there is a reduction of facial skin temperature when there is a higher transpiration rate.

The correlation data between heartbeats and facial moisture indicate an increased rate relative to the increase in facial skin moisture. The increase in heartbeat, consequently blood flow, can be related to the loss of water (sweat) to refresh the skin and reduce body heat (Pascoe et al., 2008; Sousa, 2014).

In conclusion, there was a reduction of 13.3°C average temperature of the inner roof with the presence of the insulating panel for the period without the panel and facial skin average temperature increased by 1.1 °C after the panel removal.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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*Full Length Research Paper*

# Towards being equal to them: Impact of organic certified production systems on women empowerment in agriculture

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The objective of the study was to determine the degree of women empowerment in agriculture as well as examine the effect of organic certification and other socio-economic and cultural factors on women empowerment in agriculture in Kenya. This objective was achieved using data from peri-urban vegetable and rural honey producing households. It followed the innovative multidimensional measurement of women empowerment in agriculture, and the univariate and multivariate two limit Tobit models was used to assess the determinants of women empowerment. The two limit Tobit models results affirmed the hypothesis that organic certification opens up knowledge space for women empowerment in agriculture in some domains but had more impact among women in vegetable producing households. However, the variation of the degree of women empowerment in agriculture was also influenced by men, women and household socioeconomic and cultural characteristics. Policies geared towards enhancing women's social capital and ownership of assets will improve the women household bargaining power and subsequently women empowerment in Agriculture.

**Key words:** Women empowerment in agriculture, organic certification, peri-urban, rural, knowledge space.

## INTRODUCTION

Recently, developing countries have experienced proliferation of local market oriented organic production systems due to growing local demand for organic products resulting from the increasing income and urbanization (Hattam et al., 2012; Probst et al., 2012).

The consumers in this market niche are concerned with the food attributes thus willing to pay premium prices for organic products (Hattam et al., 2012). This trend has

shifted the marketers focus from promotion of food products to the promotion of food attributes among consumers and potential consumers (Stolzenback et al., 2013; Costanigro et al., 2014) through certification of the processes along the agricultural value chains. On the producers end, certified organic production is achieved through adoption of sustainable production and produce handling techniques with the aim of reaping economic,

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social and cultural benefits from the certified processes (UNCTAD, 2006; Blackman and Naranjo, 2012; Hattam et al., 2012).

The potential of organic agricultural systems has made it an attractive business model in developing countries aimed at improving the livelihood of smallholder agricultural producers through farmer led organization. Consequently, governmental agencies, local and international non-governmental organizations, international donors and other development partners in the Sub-Saharan Africa have been involved in encouraging the adoption of organic production systems as a developmental pathway for smallholder agriculture (Hattam et al., 2012). One of the important outcomes of certified organic agriculture is women empowerment in agriculture (WEIA, hereafter)<sup>1</sup> which has gained much interest among policy makers and researchers (Farnworth and Hutchings, 2009; Arestoff and Djemai, 2013). This is as a result of women constituting up to 75% of agricultural producers in the Sub-Saharan Africa and yet their role is unrecognized (World Bank, 2008).

Several studies (Doss, 2006; Ellis et al., 2007; Fantahun et al., 2007; World Bank, 2008; Chhay, 2011; Swaminathan et al., 2012) have highlighted the importance of empowering women in the society. Swaminathan et al. (2012) found that empowering women in India through home and land ownership increased mobility of women and their decision making ability regarding their own expenditure, health and work.

In Ghana, Doss (2006) found that increasing resources in the hands of the women had positive effects on the children welfare while Chhay (2011) finds that increased women control on at least part of household income, is associated with better household nutrition, health, and education levels. Women empowerment was found to reduce under-five mortality (the fourth Millennium Development Goal) in Ethiopia, a common problem in most developing countries (Fantahun et al., 2007).

Food and Agriculture Organization (FAO) (2006) and World Bank (2008) also strongly link WEIA with food improved food security in developing countries. In the sub-Saharan region, specifically Kenya, Tanzania and Ethiopia, World Bank (2005) reports on 10 to 20% increase in output if women entrepreneurs and producers are given the same education and inputs as men. Further, findings by Ellis et al. (2007) in Kenya are that elimination of the gender-based inequalities in accessing agricultural inputs and education had the potential of increasing the country's Gross Domestic Product (GDP) by 4.3% points, and sustained increase of year-on-year in GDP growth of 2-3.5percent points.

Proponents of women empowerment argue that certified organic production systems provides one of the

vivacious ways of addressing social inequities, exploitative relationship and dependencies which conventional production systems is propagating (Farnworth and Hutchings, 2009). This theory is further advanced by Trauger (2004), that sustainable production system provides spaces of knowledge to marginalized women unlike the conventional production systems. However, this hypothesis is still not clear in empirical literature. Further, Farnworth and Hutchings (2009) reports on existence of anecdotal empirical evidence on the impact of organic certification on WEIA in literature. Thus, this suggests a fundamental need to explore in greater depth the relationship between organic certification and women empowerment which is important for project planners and policy analysts.

In Kenya, like other countries in the Sub-Saharan region, studies have reported on low levels WEIA attributed to gender-related constraints and vulnerabilities of women compared to men (World Bank, 2008; FAO, 2011). However, the "low" level in WEIA is also not clear in empirical literature. Further, if it is low, the interest of the policy makers, developmental planners and partners would be to understand how the socio-economic and cultural characteristics influence the level of women empowerment in order to make decisive interventions. However such information is scarce in empirical literature, partly attributed to lack of a clear tool to measure WEIA in the face of renewed interest in agriculture as the engine for growth and development (FAO, 2011; Alkire et al., 2013).

It is on this background that this study aims to fill this knowledge gaps with an exploratory study on local oriented market consisting of certified organic and noncertified vegetable and honey producers in Kenya. Therefore, the objective of the study was to provide micro level empirical evidence on the degree of women empowerment and to examine the impact of organic certification and other socioeconomic and cultural factors that influence of WEIA. The study provides a test of the hypothesis that organic certification positively influence WEIA on the premise that it provides spaces of knowledge to women compared to the conventional systems. This was within a context of acknowledgement that WEIA is multidimensional. Thus, the measurement of WEIA was through the innovative methodology proposed by Alkire *et al.* (2013), with some modification in the methodology and using econometric tools. The study contributes to literature in three aspects:

- (1) It provides a micro level evidence of degree of WEIA in two production systems consisting of peri-urban and rural area producers;
- (2) It makes a methodological contribution to the Alkire et al. (2013) on the measurement of the dimensions of empowerment. In particular, the study borrows from the business world on how to measure women empowerment in leadership domain by adapting the authentic leadership measurement by Walumbwa et al. (2008) as opposed to

<sup>1</sup> In this study empowerment follows Kabeer (2001) definition that it is "the expansion of people's ability to make strategic life choices in a context where this ability was previously denied to them". The study limits itself to women

the group and public speaking methodology proposed by Alkire et al. (2013);

(3) It uses the two limit multivariate modeling approach used mostly in demand estimation to determine the impact of organic certification after controlling for potential endogeneity and other socio-economic and cultural factors among vegetable and honey producers on the different domains of WEIA thus allowing for domain interactions of the unobservables.

## METHODOLOGY

### Study area

This study used two local market oriented case sites in Kenya; vegetables production in Ongata Rongai district located in peri-urban area and organic honey production in Mwingi district in the rural areas. Ongata Rongai district has both the conventional and certified vegetable farmers and lies on coordinates 1°21'34"S 36°39'44"E bordering Nairobi the capital city of Kenya (KCIDP, 2013).

Farmers supply of organic vegetables to hotels and restaurants, supermarkets, several organic shops and others are sold directly to organic consumers in flea market in Nairobi (Kamau et al., 2018). Community Sustainable Agriculture and Healthy Environmental Program (CSHEP) and government extension providing women empowerment integrated extension services to the organic farmers. The production processes and marketing are certified by Encert Kenya. The organic certification project is coordinated by Kenya Organic Agricultural Network (KOAN) and includes components of women empowerment by integrating women in agricultural production and marketing trainings and leadership in the farmer led organizations. Women are also facilitated to access the markets for organic products. For comparison, similar conventional farmers in the area were sampled.

Mwingi district is among the high quality honey producers in Kenya and is located in 0° 56' 0" South, 38° 4' 0" East and is a semi-arid region, a highly food insecure and livelihood of the residents depends on rain fed agro-pastoralism and honey production. About 60% of the population living in area lives below the poverty line (Galu et al., 2010). Honey production in the district is classified as organic because of minimal or no usage of external inorganic inputs for crop or livestock farming and the presence of surrounding forest buffer zones. International Fund for Agricultural Development (IFAD) and The International Centre of Insect Physiology and Ecology (ICIPE) jointly initiated the project of commercializing organic honey production involving over 2000 households. This led to establishment of Mwingi Honey Place and several honey collection centers to undertake value addition and marketing of processed honey and wax as the main by product. Production, processing and marketing activities are certified by Kenya Organic Agriculture Network (KOAN) and Institute of Marketecology (IMO), Switzerland.<sup>2</sup> The woman empowerment component includes involving women production and marketing of honey through intensive and frequent trainings in farmer groups that includes men and women. Women are also involved in the leadership of the marketing group and the individual farmer groups. Some disadvantaged women are given beehives by the project to enable them engage in commercial honey production.

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Data were collected from 237 and 232 households involved in vegetables and honey producing households respectively selected using multi-stage sampling approach. However, the sample reduced to 203 and 207 households among vegetables and honey producers respectively due to unavailability of some women during the survey period in the months of June and July 2013 and the exclusion of single and widowed women from original sample because of the need to include the impact of men characteristics in the analysis. Among the 203 households in vegetable production system, 62% were conventional farmers and 38% were certified organic farmers while for honey producers 49% were noncertified and 51% were certified organic producers. The study used a semi-structured questionnaire through in-depth face-to-face interviews to sampled households by trained enumerators. Contextual data was collected through focus group discussion conducted in each case study sites.

### Measuring WEIA

The study adopted the methodology of measuring WEIA as proposed by Alkire et al. (2013) since it reflects the diverse aspects in empowerment literature<sup>3</sup>. However, some modifications are made to how the different domains are measured. The five domains that constitute WEIA are production, income, resource, leadership and time. The production domain was composed of;

- (1) The woman input in production decisions involving cash and food crop farming, livestock keeping and aqua farming, and
- (2) Autonomy in production involving agricultural and livestock production, type of crops to grow, type of inputs to use, when and who to deliver the produce to the market.

However, instead of the ranked scale and binary variables used to measure the levels of empowerment by Alkire et al. (2013), the study opted for a range between 0 and 10% which was later transformed to 0 and 100%. This was found necessary in getting stated actual level of woman participation in the decision making in the various components and reducing measurement errors involving limited ranked scales. This mode of measurement was used in all the subsequent components of the domains of women empowerment.

In the resource domain, the indicators comprised of ownership of land, other assets, decision on sale purchase and the transfer of land and other assets besides decisions regarding credit. The income domain was used to measure the decision making of the woman involving income generated in the household. The subcomponents of the domain were;

- (1) The woman's participation in decisions on income generated from cash and food crop farming, livestock keeping and aqua farming, and
- (2) The woman feelings on making decisions regarding her salaried or wage employment, major and minor household expenditure if she wanted.

The leadership domain saw a major change in its components. Instead of using the group and speaking in public as proposed by Alkire et al. (2013), the study opted for the authentic leadership measurement by Walumbwa et al. (2008) which captures four important aspects of leadership; self-awareness, relational transparency, internalized moral perspective and balanced

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<sup>2</sup> For more details on the project visit ICIPE website at:

<http://www.icipe.org/index.php/component/content/article/62-commercial-insects-programme/402-wild-silk-and-honey-bee-farming-for-income-generation-and-biodiversity-conservation-through-value-chain-approach.html>. Accessed on 9<sup>th</sup> October 2013.

<sup>3</sup> See the article by Alkire *et al* (2013) for the review of other studies on women empowerment measurement and the details of weights for each subcomponent in different domains for brevity purposes.



processing of the woman.<sup>4</sup> The deviation was that the leadership indicators of belonging to a group proposed by Alkire et al. (2013) according to the study could be inadequate indicator since group membership results more to social capital formation than leadership (Christoforou, 2011; Tumbo et al., 2013). The time domain was measured by the level of satisfaction on the available time for leisure and the work load for the woman. The overall index was computed from the five domains using the equal weights of each domain as proposed by Alkire et al. (2013).

### Modeling the determinants of women empowerment domains

To determine the factors that influence the five domains of women empowerment in agriculture, the study used the multivariate two limit Tobit analysis. The use of the ordinary least square would have been possible but the presence of the zero observation in some domains and the presence of lower and upper limits would lead to biased and inconsistent estimates (Ma et al., 2006).

The Tobit model estimates are consistent because of truncation of the domains at zero. The study opted for the two limits multivariate Tobit model in contrast to individual domain Tobit model because it allows for the unobservables that determine women empowerment in one domain have a likelihood of being correlated to those of other domains. Very few studies have used this methodology in agriculture (Gillespie and Mishra, 2011; Ali et al., 2012) and is mostly used in economics in demand estimation (Ma et al., 2006). Let the 5 domains of WEIA be denoted by  $d$  with  $n$  observation and  $X$  a vector of variables including the organic certified production participation (*ocertprod*) variable hypothesized to be determining  $d$ , then the observed WEIA domains  $we_{ih}$  are determined by;

$$we_{ih}^* = X'_{ih}\beta_i + \varepsilon_{ih}, 1 \leq i \leq d, 1 \leq h \leq n \quad (1)$$

$$we_{ih} = \begin{cases} we_{ih}^* & \text{if } we_{ih}^* > 0 \\ 0 & \text{if } we_{ih}^* \leq 0 \end{cases} \quad (2)$$

where  $we_{ih}^*$  is the latent variable and  $\varepsilon_h = (\varepsilon_{1h}, \varepsilon_{2h}, \dots, \varepsilon_{dh})' \sim N_d(0, \Omega)$ . The dimensions of the  $\beta_i$  is  $s_i \times 1$  and  $\Omega$  is a  $d \times d$  symmetric positive matrix. The observed value of  $we_{ih}$  equals the true value of if  $we_{ih}^* > 0$ ; otherwise, the observed value of  $we_{ih}$  is left censored to be zero (Ma et al., 2006). The latent women empowerment index for the  $i^{th}$  domain of the  $h^{th}$  woman is denoted by  $we_{ih}^*$  and the observed index of empowerment is  $we_{ih}$ , which is either positive or zero. Huang (2001) expressed the systems of equations as;

$$\begin{bmatrix} we_{1h}^* \\ we_{2h}^* \\ \vdots \\ we_{dh}^* \end{bmatrix} = \begin{bmatrix} X'_{1h} & 0 & \cdots & 0 \\ 0 & X'_{2h} & \cdots & 0 \\ \vdots & \vdots & \ddots & \vdots \\ 0 & 0 & \cdots & X'_{dh} \end{bmatrix} \otimes \begin{bmatrix} \beta_1 \\ \beta_2 \\ \vdots \\ \beta_h \end{bmatrix} + \begin{bmatrix} \varepsilon_{1h} \\ \varepsilon_{2h} \\ \vdots \\ \varepsilon_{dh} \end{bmatrix} \quad (3)$$

This can be rewritten as;

$$we_h^* = X_h\beta + \varepsilon_h, h = 1, 2, \dots, n \quad (4)$$

Where  $we_h^* = (we_{1h}^*, we_{2h}^*, \dots, we_{dh}^*)'$ ,

$X_h = \text{diagram}(X'_{1h}, X'_{2h}, \dots, X'_{dh})$ , and

$\beta = (\beta_1', \beta_2', \dots, \beta_d')$  is a  $s \times 1$  vector with  $s = \sum_{i=1}^d h_i$ .

However, it should be noted that in all this specification that some women might not be having zeros level of empowerment in any domain specific is in the domains which implies there will be censoring points at point zero. Therefore, possible combination of the WEIA at censoring points is  $2^d$  represented by a  $2^d \times 1$  vector  $C_s$ ,  $c = 1, 2, \dots, 2^d$ . The likelihood function is accounting for all the censoring combination of all observations is specified as;

$$L(WE; \beta, \Omega) = \prod_{h=1}^n L_h^{C_s}(we_h; \beta, \Omega) \quad (5)$$

where  $WE = (we_1', we_2', \dots, we_n)'$  and  $L_h^{C_s}$  shows the likelihood combination that the domain specific women empowerment index of woman  $h$  falls in regime  $s$ .

However, the inclusion of the dummy variable organic certification (*ocertprod*) as an explanatory variable in equations 3 or 4 would be a potential source of endogeneity leading to errors in the estimated parameters. To control endogeneity problem in the study, the probit model for participation in certified organic farming was estimated for vegetable and honey producers. This was followed by the prediction of the propensity scores for participation variable that was later used in equation 3 or 4 in estimating the impact of organic certified farming on each of the domains of WEIA (Ma et al., 2006; Grootendorst, 2007). However, it should be noted that since the overall index is derived from all the five domain of WEIA, it would be erroneous to estimate its determinants together with domain specific determinants. Its estimation is modeled in the next section.

### Modeling the determinants of the overall WEIA index

To determine the relationship between overall WEIA and organic certification and other socioeconomic and cultural variables, the study used the univariate Two-Limit Tobit model and its structural equation is written as;

$$owe_i^* = X_i\alpha + v_i \quad (6)$$

where,  $owe_i^*$  is a latent variable of overall WEIA for the  $i^{th}$  woman,

$X$  is a vector of independent variables postulated to be determining the intensity of women empowerment including the

<sup>4</sup> See attached questionnaire on how the study measured the leadership domain in appendix 1. Questionnaire modified from the sample in the National University blog by Walumbwa and associates.

**Table 1.** Variables for determinants of women empowerment in agriculture.

Variable	Description of the variable	Mean for vegetable producers	Mean for honey producers
Offarm_man	Off-farm activity participation by the husband , 1=Yes 0=No	0.62 (0.49)	0.58 (0.50)
Offarm_fem	Off-farm activity participation by the wife, 1=Yes 0=No	0.40 (0 .49)	0.43 (0.50)
Educ_ man <sup>a</sup>	Education level of the husband	2.78 (1.11)	2.51 (0.94)
Educ_ fem <sup>a</sup>	Education level of the wife	2.25 (1.05)	1.85 (0.95)
Female age	Age of the household head (years)	45.40 (13.56)	48.41 (12.27)
Head _ fem	Whether the wife is the household head, 1=Yes 0=No	0.23 (0.42)	0.11 (0.16)
Marry age	Age the wife was married in years	27.71 (4.58)	20.42 (4.30)
Age gap	Spousal age gap in years (husband age-wife age)	4.45 (7.69)	9.13 (7.78)
Wgroup_het <sup>bc</sup>	Group heterogeneity index	0.68 (0.15)	0.12 (0.30)
Wmeet index <sup>b</sup>	Meeting attendance index (meetings attended/ scheduled meetings)	0.73 (0 .30)	0.79 (0.34)
Wdensity <sup>b</sup>	Number of active groups household involved in 2012	1.28 (0.94)	1.69 (0.83)
Wtrust <sup>b</sup>	Level of trust in groups (0-100%)	6.60 (3.53)	8.15 (1.68)
ocertprod	Propensity to be organic certified producer	0.43 (0 .38)	0.53 (0.38)

Figures in parenthesis are standard errors of the respective means.

<sup>a</sup>Education measured in terms of 1=not gone to school 2=primary 3=secondary 4= tertiary 5= university; <sup>b</sup>Women level social capital dimensions; <sup>c</sup>The woman heterogeneity index derived from questions of whether members in women groups were from the same neighborhood, occupation, kingroup, economic status, religion, gender, education level and age group.

organic certified propensity scores estimated by probit model in section 2.3 as the participation variable.

The  $\alpha$ 's are parameters of the independent variables to be estimated and  $\varepsilon$  is the error term and independently distributed error term assumed to be normally distributed with a mean of zero and a constant variance. The univariate two limit Tobit model takes into account the censoring both from below and above. The observed is defined by the following generic measurement equation:

$$\begin{aligned} owe_i &= owe^* \text{ if } owe^* > \tau \\ owe_i &= \tau_{owe} \text{ if } owe^* \leq \tau \end{aligned} \tag{7}$$

Typically, the two Limit Tobit model assumes that  $\tau = 0$  which means the data is censored at zero. However, the overall WEIA for farmers range between 0% and 100% (Tobin, 1958). Thus, substitute  $\tau$  in equation 7 results into:

$$\begin{aligned} owe_i &= owe^* \text{ if } 0 < owe^* < 1 \\ owe_i &= 0 \text{ if } owe^* \leq 0 \\ owe_i &= 1 \text{ if } owe^* \geq 1 \end{aligned} \tag{8}$$

Therefore, the model assumes that there is an underlying women empowerment index equal to  $x_i\alpha + v_i$  which was observed only when it is some number between 0 and 100%; otherwise  $owe_i^*$  qualifies as an unobserved latent variable (Green, 2002). The empirical univariate two limit Tobit model was estimated among vegetable and honey producers and took the form of:

$$owe_i^* = \alpha_0 + \sum_{n=1}^n \alpha_n X_i + v_i \tag{9}$$

The independent variables  $X$  included in univariate and multivariate two limits Tobit models are described in the next section.

### Variables included in the models

As described earlier, the probit model was used to derive the propensity scores to be used in the univariate and the multivariate Tobit models in determining the impact of organic certification on domain specific and overall WEIA. This was for the purposes of correcting for potential endogeneity. The variables included in the probit model, their measurement and the descriptive statistics for vegetable and honey producers are reported in Appendix 2. However, the main interest of the study was on measuring the degree of WEIA and on impact of organic certification and other socio-economic and cultural factors that determines the degree of women empowerment in agriculture, thus, much focus was given to this variables. The variables and their descriptive statistics are presented in Table 1.

The husband characteristics in a marriage could play role in the women empowerment process (Anderson and Eswaran, 2009). The husband's characteristic included were education level and the spousal age gap (the difference between the age of the woman from that of the man) and participation in off-farm activities. The study hypothesis that husbands participation in off-farm and higher education could result to higher levels of empowerment. This was on the premise that they facilitate exposure to information and knowledge which could reduce the subjective opinion on incapability of women involvement in agricultural decision making. Higher spousal age gap is associated in literature with hegemony on the younger spouse (Guilbert, 2013).

**Table 2.** Mean of dimensions of WEIA (0-100%).

Dimensions	Vegetable producers			Honey producers		
	Noncertified	Certified	Overall	Noncertified	Certified	Overall
Production	40.52 (19.21)	39.15 (23.12)	39.46 (20.32)	37.23 (18.28)	41.97** (22.13)	39.63 (18.92)
Income	35.21 (25.23)	36.13 (19.35)	35.86 (21.10)	27.23 (21.01)	27.78 (20.02)	27.25 (21.05)
Resources	38.35 (24.21)	42.91** (18.23)	40.56 (19.46)	30.96 (20.99)	30.31 (21.11)	30.85 (20.96)
Leadership	37.29 (41.19)	42.28*** (42.84)	39.72 (44.01)	36.34 (29.97)	42.23*** (39.88)	37.54 (36.17)
Time	36.71 (41.23)	31.28*** (39.54)	34.83 (40.82)	39.11 (27.36)	43.21** (28.29)	41.81 (30.82)
Overall women empowerment index	36.41 (16.17)	41.12** (28.36)	38.08 (25.36)	35.43 (23.28)	0.37.51 (32.19)	35.41 (23.26)

Figures in parenthesis are standard deviations. \*\* and \*\*\* indicates that the mean values are significantly different from the noncertified producing households in each product type at 5% and 1% level respectively.

The social and economic woman characteristics of the women were also included. Higher education level of the woman could positively influence empowerment since education increases the status of the woman in a family unit and the skills critical in decision making (Jayaweera, 1997). The participation of the woman in off-farm income is also important in increasing the bargaining power as it contributes to woman's self-reliance (Jayaweera, 1997). The variable of whether the woman is the head of the family was also included on the premise of understanding what the effect to women empowerment is when the woman is the family head.

Age of the woman could also determine WEIA and the study hypothesizes that the effect could be positive or negative; as older women will tend to be more empowered because of their experience in marriage and younger women could be more empowered because higher tendency being exposed and have higher levels of education. The age at which the woman married may affect her decision making ability (Guilbert, 2013), and the study hypothesizes that because early marriages is prone in rural areas, it could negatively influence WEIA among honey producers. The woman social capital dimensions (Grootaert, 1999), were also included because it could be a source of information and a platform to develop the woman's decision making skills. The value of agricultural assets was included as an indicator of wealth to understand how does larger wealth affects women empowerment. Land size could also determine the level of empowerment as women are greatly involved in providing agricultural labour force (Table 2).

The domains specific and the overall of women empowerment index in agriculture descriptive statistics are presented in Table 2. Among vegetable producing households significant difference in women empowerment was observed in the resources, leadership, time and the overall index. Conversely, there was significant difference in women empowerment in production, leadership and time dimensions among noncertified and certified honey producing households. Vegetable producers had higher levels of empowerment in production (39%), resources (41%) and leadership (40%). This could imply that men are still controlling the household income and expenditure leading also the higher levels of dissatisfaction in the workload and time available for their leisure activities.

On the contrary, women in honey producing households had higher levels empowerment in production (40%), leadership (36%) and time (42%) domains. Men were at the realm in resource and income domains possible implying that men could be willing to engage women in decision making more in domains which they consider as less important. In the overall index, women in honey producing households a 35% involvement in decision making compared to 38% of vegetable producing households. This could be attributed to the socio-economic and cultural impediments existing in rural areas.

## RESULTS AND DISCUSSION

### Determinants of degree of WEIA

Table 3 reports on the Probit models regression used to generate individual propensity scores among vegetable and honey producers to be used in determining the impact of organic certification on women empowerment in agriculture in the Tobit model. The results indicate that younger farmers with higher education level had higher likelihood of participating in certified organic production among vegetable and honey producers.

In honey production systems, larger household size increased the likelihood of participating in certified production probably to cushion their high household expenditure. Further, participating in off farm activities by the household head, higher household assets value and number of agricultural trainings influenced significantly the likelihood of participation in organic vegetable production systems. Shorter distances to produce markets in rural areas significantly increased the likelihood of participating in honey production systems, indicating the importance of market infrastructure in enhancing market participation among smallholder rural farmers.

In general, higher social capitals in the different dimensions (Grootaert, 1999) were also important in increasing the likelihood of participation in organic certified production systems in both production systems. Finally, since livestock manure is important in supplementing soil nutrients in organic production systems, farmers having closed systems of keeping livestock had higher likelihood of participating in certified organic vegetable production. This eases the collection and transportation of manure in the farms.

Tables 4 and 5 presents the results on the determinants of degree of WEIA among vegetable and honey producers, respectively. The multivariate two limit Tobit estimation was used for the WEIA domains and the univariate two limit Tobit used for the overall index. The significant chi square in both tables indicated that the multivariate technique produces efficient estimates of the

**Table 3.** Determinants of participation in certified organic production systems.

Variable	Vegetable producers		Honey producers	
	Coeff.	Std. err.	Coeff.	Std. err.
Head_age	-0.026*	0.011	-0.044**	0.015
Gender_he	-1.846	0.419	0.849	0.345
Educ_head	0.323**	0.140	0.409**	0.186
Hh size	0.173	0.104	0.350***	0.079
Offfarm_he	0.708*	0.329	0.094	0.042
Agric_assets	0.898*	0.150	0.170	0.149
Farm size	0.208	0.439	0.047	0.204
Extetim	0.014	0.016	-0.016	0.019
Trainnum	0.617**	0.325	-0.215	0.363
Mktkm	-0.149	0.055	0.050***	0.056
Credit	0.519	0.378	0.573	0.506
<b>Household social capital</b>				
Density	0.043**	0.166	-0.117	0.063
Meet_index	0.165	0.375	1.216**	0.495
Group_het	0.144***	0.092	0.840**	0.179
Decision	0.189**	0.069	0.273*	0.076
Trust	-0.067	0.066	0.236	0.284
System	0.859**	0.309	-	
Intercept	-8.900***	1.906	-6.099 **	2.177

Figures in parenthesis are standard errors.

\*, \*\*, \*\*\* correspond to 1, 5 and 10% levels of significance.

domains for WEIA than the univariate estimation. A cursory examination of the results depicts varied effects of the factors that influence the degree of WEIA dimensions and the overall index among vegetable and honey production systems. Husband's participation in off-farm activities was associated with increasing the degree of empowerment in leadership and time domains in vegetable production systems.

In contrast, it significantly influenced positively the degree of women empowerment in resource dimension and income dimension as well as the overall WEIA index among honey producers. This implies that characteristic of the husband do not consistently affect women empowerment in the domains. Note that honey producing households are in rural food insecure region and most of their husbands are in the urban areas engaging in off-farm activities. Hence, women make significant decisions could be as a result of their absence.

Women participation in off-farm activities increases the empowerment in income domain in both production systems and production domain among honey producing households. Off-farm activities provide a source of income which the woman can invest in agriculture and assets thereby increasing her bargaining power in the household. However, the income effect in honey production system is interesting in presence of limited opportunities for off-farm activities in rural areas. This

raises a policy issue on how to open up opportunities for the rural women as a forward gear towards their empowerment. Anderson and Eswaran (2009) reports that direct income in women hands in Bangladesh positively influenced their decision making power. Further, Agarwal (2001) notes that women participation in off-farm activities in India enhances their ownership of assets leading to increased bargaining power. Likewise, Jayaweera (1997) notes that woman's own earning increasing her self-confidence and self-reliance.

Increase in husband education level significantly increased the level of WEIA in leadership and time domain but decrease WEIA in the overall index among vegetable producing households. The implication of this is that highly educated men tend to recognize the need for time and leadership skills of women but still control the income, resource and production activities at the household level. In contrast, increase in the education level of the husband significantly reduced the degree of women empowerment in production, income and resource dimensions among the honey producer. However, comparing the effect of men and woman education level on WEIA, the picture tends to change.

The effect of increasing women education level supersedes the negative effects of increasing the men education level in both production systems. Education is imperative in knowledge development and being able to

**Table 4.** Determinants of women empowerment among vegetable producers.

Variable	Multivariate two limit Tobit model of the dimensions of women empowerment					Univariate Tobit
	Production	Income	Resource	Leadership	Time	Overall index
Offarm_man	-0.032 (0.023)	0.001 (0.036)	0.033 (0.035)	0.048** (0.082)	0.046* (0.037)	0.012 (0.024)
Offarm_fem	0.488 (0.031)	0.115*** (0.035)	-0.017 (0.034)	-0.007 (0.078)	-0.017 (0.035)	0.003 (0.023)
Educ_man	-0.094 (0.016)	-0.018 (0.018)	-0.011 (0.018)	0.067** (0.041)	0.017** (0.018)	-0.013** (0.012)
Educ_fem	0.057 (0.017)	0.019 (0.019)	0.033** (0.018)	0.086 (0.043)	-0.037** (0.019)	0.019** (0.013)
Female age	0.009 (0.001)	0.022*** (0.001)	0.000 (0.001)	0.001 (0.003)	0.003** (0.001)	0.021*** (0.001)
Head_fem	0.185 (0.041)	0.005 (0.047)	-0.050 (0.045)	-0.121 (0.105)	-0.064 (0.048)	0.058 (0.031)
Marry age	0.013 (0.003)	0.003 (0.004)	-0.007** (0.004)	-0.009 (0.009)	-0.002 (0.004)	-0.003 (0.003)
Age gap	-0.041** (0.002)	-0.012 (0.002)	-0.002 (0.002)	-0.002** (0.005)	-0.001 (0.002)	-0.001 (0.001)
Group_het	0.832 (0.095)	0.070** (0.107)	-0.064 (0.104)	0.003 (0.241)	0.012* (0.109)	0.015 (0.072)
Meet index	0.078 (0.046)	0.085** (0.051)	0.018** (0.050)	0.529*** (0.116)	0.100** (0.052)	0.123*** (0.035)
Density	-0.184 (0.015)	-0.013 (0.017)	0.026* (0.017)	0.044 (0.038)	0.006** (0.017)	0.004 (0.011)
Trust	0.075** (0.004)	0.001 (0.005)	0.000 (0.004)	0.068*** (0.010)	0.071** (0.005)	0.029*** (0.003)
Agric_asset	-0.195 (0.017)	-0.006 (0.019)	-0.005 (0.018)	-0.045 (0.042)	-0.034* (0.019)	-0.022* (0.013)
Farm size	0.036 (0.011)	-0.031 (0.012)	0.009 (0.012)	-0.001 (0.028)	0.019** (0.013)	-0.002* (0.008)
ocertprod	0.105 (0.071)	0.054 (0.079)	0.124*** (0.077)	0.314** (0.179)	-0.017*** (0.081)	0.169** (0.053)
Intercept	0.724*** (0.228)	0.169 (0.256)	0.683*** (0.249)	0.307 (0.579)	0.281 (0.261)	0.433 (0.172)
Correlation	-	-	-	-	-	-
Production	1.000	-	-	-	-	-
Income	-0.080	1.000	-	-	-	-
Resource	0.095	-0.156	1.000	-	-	-
Leadership	0.006	0.051	0.142	1.000	-	-
Time	-0.075	0.173	0.078	0.189	1.000	-

Numbers in parenthesis are standard errors of the coefficients. \*, \*\*, \*\*\* correspond to 1, 5 and 10% levels of significance.  $H_0$ : There is no correlation between the error terms LR chi-square (10) = -476.556(p-value = 0.000).

defend one's stance. Hence, educated women tend to be self-confident and assertive which enhances their ability and participation in decision making at the household level. However, Jayaweera (1997) concluded that existing gender ideologies, social and economic constraints concerning women education reduces their degree of empowerment.

Age of the woman had also interesting results. Older woman had higher likelihood of being empowered in income, time and the overall index among vegetable producers and in time dimension among the honey producers. This implies that the decision making in agriculture progressively increases as the woman gets older probably because of the experience and information gained which makes her accustomed to the her role in marriage. This is more so when women empowerment process is visualized as stock that has to be accumulated with time. The results on older women in time dimension depict an impression of them being "contented" with their farming activities as they are highly immobile and having lesser opportunities in off-farm activities compared to the younger women.

The age at marriage significantly influenced positively empowerment of women only among the honey producing

households in production, income, resource and the overall empowerment index. This could be attributed to the lower age in marriage among honey producers which is located in the rural areas compared to the vegetable producers in the peri-urban set up their is breakage of one's cultural beliefs. Engelen and Kok (2003) argue that higher age at marriage in urban areas is associated with inability of the migrants in the new environment to find social connections. The plausible explanation could be that early married women tend to be less self-confident intoning their opinions and experience difficulty in developing their own identity.

Rural areas are associated with early marriages because of the lower education levels and cultural conditioned beliefs. This result provides evidence on the missing link in literature between early marriage and the level of women empowerment in agriculture. Brickell and Chant (2010) notes that young women in marriage tend to have physical and emotional distress and low esteem because of the new environment which negatively affects the decision making at the household level during the initial years in marriage. A delay in the year of marriage in Bangladesh by one year, led to a 6.5% higher likelihood of literacy and 0.3 additional schooling years (Ambrus

**Table 5.** Determinants of women empowerment among honey producers.

Variable	Multivariate two limit Tobit model of the dimensions of women empowerment					Univariate Tobit
	Production	Income	Resource	Leadership	Time	Overall index
Offarm_man	-0.008 (0.033)	0.034* (0.020)	0.033** (-0.210)	0.081 (0.082)	0.034 (0.037)	0.018** (0.027)
Offarm_fem	0.061** (0.031)	0.066** (0.032)	0.035 (0.031)	-0.041 (0.078)	0.010 (0.035)	0.030* (0.026)
Educ_man	-0.033* (0.017)	-0.029** (0.018)	-0.029* (0.017)	-0.002 (0.043)	0.020 (0.020)	-0.018 (0.014)
Educ_fem	0.038** (0.016)	0.045*** (0.016)	0.042*** (0.016)	0.019 (0.039)	0.042** (0.018)	0.043*** (0.013)
Female age	0.000 (0.001)	0.000 (0.001)	0.000 (0.001)	-0.001 (0.003)	0.012* (0.001)	0.000 (0.001)
Head_fem	0.150 (0.033)	0.168 (0.034)	0.119 (0.033)	0.036 (0.082)	0.052 (0.037)	0.106 (0.027)
Marry age	0.008** (0.003)	0.010*** (0.003)	0.007** (0.003)	0.003 (0.008)	-0.005 (0.004)	0.016** (0.003)
Age gap	-0.002 (0.002)	-0.122** (0.002)	-0.012 (0.002)	-0.125** (0.005)	-0.001 (0.002)	-0.021 (0.002)
Group_het	0.001 (0.059)	-0.013 (0.062)	0.031 (0.059)	0.015 (0.148)	0.147 (0.068)	0.073 (0.049)
Meet index	-0.001 (0.049)	-0.018 (0.051)	-0.011 (0.049)	0.055 (0.122)	0.062** (0.056)	0.015 (0.040)
Density	0.007 (0.019)	0.009 (0.020)	0.003 (0.019)	-0.040 (0.047)	0.037* (0.021)	0.003 (0.016)
Trust	-0.001 (0.008)	0.005 (0.009)	-0.009 (0.008)	0.024 (0.021)	-0.006 (0.009)	0.001 (0.007)
Agric_asset	0.011 (0.014)	0.014 (0.015)	0.014 (0.014)	-0.015 (0.035)	-0.004 (0.016)	0.004 (0.012)
Farm size	-0.028 (0.019)	-0.027* (0.020)	-0.014 (0.019)	-0.093** (0.047)	-0.018 (0.022)	-0.029* (0.016)
Ocertprod	0.127** (0.049)	0.016 (0.051)	0.019 (0.049)	0.056*** (0.122)	0.050** (0.056)	0.051 (0.041)
Intercept	0.373** (0.199)	0.318 (0.207)	0.340 (0.200)	0.644 (0.498)	0.367* (0.227)	0.356** (0.165)
Correlation	-	-	-	-	-	-
Production	1.000	-	-	-	-	-
Income	0.958	1.000	-	-	-	-
Resource	0.853	0.878	1.000	-	-	-
Leadership	0.013	0.050	0.088	1.000	-	-
Time	0.164	0.178	0.161	0.188	1.000	-

Numbers in parenthesis are standard errors of the coefficients. \*, \*\*, \*\*\* correspond to 1, 5 and 10% levels of significance.  $H_0$ : There is no correlation between the error terms LR chi-square (10) = -432.174 (p-value = 0.006).

and Field, 2008).

Turning to spousal age gap, increase in spousal age gap led to significant decline in women empowerment in production and leadership dimensions among vegetable producing households and income, leadership and the overall index among the honey producing households. This could be due to larger spouse age gap makes women more vulnerable and reclusive hence they cannot develop and portray their decision making and leadership skills. Thus, this makes the men more dominant in decision making in the family circle. Findings by Carmichael (2011) are that larger spousal age gap in marriage disempowers the younger spouse in decisions in the household and the community at large due to lack of or inadequate self-confidence. Further, Guilbert (2013) notes that negative effect of larger spousal age gap on women empowerment is exacerbated with early marriages which is characterized by low education levels.

Women social capital dimensions measured by density of membership, group heterogeneity index, meeting attendance index and level of trust among the group members was found to have more significant positive effect in empowering women in vegetable producing households than their counterparts. Women in vegetable

producing households benefited most from social networks because the groups are highly heterogeneous in the composition resulting from acculturation in peri-urban areas compared to the honey producers who are in rural areas. This could be the possible explanation for the differences in the effect in the two production systems.

However, most notable was the positive significant effect of the four dimensions of social capital among vegetable producers and meeting attendance index and density of membership among honey producers in influencing leadership domain. This demonstrates the transformative role of social capital in leadership development as it accords women and men a platform for exchanging information, experiences and knowledge spirited to develop leadership and decision making skills in agriculture. Fantahun *et al.* (2007) reports on the importance of higher social capital in Ethiopia in empowering women resulting in reduced under-five mortality and De Silva and Harpham (2007) emphasizes on the importance of maternal social capital in enhancing child nutrition status in developing countries.

Value of agricultural assets was used in the study as a proxy for wealth and the result was surprising. Increasing the value of agricultural assets and farm size led to a

decline in the degree of empowerment on the time domain and the overall index among vegetable producers. The significant results on the time could be attributed to the extra care for the assets is required, which increases the work load and limits her leisure activities. However, this could also show that in wealthy households, the male heroine as a breadwinner could be dominant which limits women involvement in decision making compared to less well-off households where there could be sharing of the breadwinner role between husband and wife. This possible could be the explanation for the insignificant results on the honey producers because of low asset base.

On the contrary, a unit increase in the farm size led to a significant decline in the income and leadership domains and the overall index among honey producers. Possibly, this could be attributed to the higher spousal gap, lower education level of the women and the extra work load in terms of labour which limits women in discovering their abilities. Similar agreement is advanced by Bacon (2010) on woman empowerment being negatively affected by heavy work load with commercialized agriculture and is aggravated further with fulltime domestic chores.

To the link between certified organic production and women empowerment, the study observes mixed result between the two production systems. Among the vegetable producers, organic certification after controlling for potential endogeneity had significant positive impact on the production, resource and leadership domains and the overall index of WEIA. However, participating in certified organic farming reduced significantly the level of empowerment in the time domain possible because of being labour intensive and thus would involve more commitment to the farming activities and delivery of the products to the destined markets. Similar finding were reported by Kabeer (2001), that microfinance increased women's asset ownership and income but also increased women's workload in Bangladesh. Anderson and Eswaran (2009) found that women had no control on the income generated from the farms even though they have contributed to its generation in Bangladesh. Further, Allendorf (2007) and Chhay (2011) argue that income in the hands of women compared to those in men had more positive effects to the welfare of their families, women and the community at large.

In contrast, production leadership and time domains are significantly influenced positively by organic certification among honey producers. This could be explained by the trainings the women are involved in relating to production activities which also builds their leadership skill. Further, the interaction among themselves improves the time dimension of the farming activities as the possible leisure activity in many rural areas. Though insignificant, organic certification had a positive effect in resource and income dimensions implying that the women are reaping some benefits which makes them more satisfied in farming thus explaining the

positive effect of the time dimension. However, in general, the social, cultural and economic constraints in the rural areas seems still limits WEIA in honey producing households because of their location in rural areas compared to their counterparts in peri-urban areas.

## CONCLUSION AND POLICY IMPLICATIONS

The study has provided a micro level evidence of the degree of WEIA and the impact of certified organic agriculture and other social, economic and cultural on the degree of women empowerment in vegetable and honey producing households in Kenya. This was achieved using a modified innovative Alkire et al. (2013) multidimensional methodology of measuring WEIA among vegetable producers in the peri-urban areas and the honey producers in the rural areas in Kenya. Evaluating the organic certification effect on women empowerment was deemed important in the face of proliferation of organic certified schemes meant to commercialize smallholder agriculture and tackle gender related and cultural constraints that thwarts women empowerment in developing countries. The study was keen to understand the level of decision making of women at the household level and the community which eventually determined their level of empowerment in agriculture as well as the determinants of their level of empowerment.

The study empirically determined the "low" empowerment question in empirical literature. On average, women involvement in decision making was about 38% and 35% among vegetable and honey producers respectively. The results of the univariate and multivariate two limit Tobit models confirmed Farnworth and Hutchings (2009) hypothesis that organic certified production systems opens up knowledge spaces for women hence contributing to their empowerment but in some domains of WEIA after controlling for potential endogeneity. The study accentuates the importance of knowledge space in women empowerment process. For public policy and program planners, the importance of information through efficient extension service delivery mechanism and this could be achieved using customized techniques and knowledge areas targeting specific domains of WEIA. However, such social norms changing initiatives should also include the both men and women to demystify the negative subjective opinion of WEIA as a "women affair" but as a step towards better family and community livelihood.

The degree of WEIA was also influenced by several socioeconomic and cultural factors differently in the vegetable and honey production systems. The findings in both production systems on the man and women education on WEIA demonstrated a policy issue on more efforts on girl child education in fighting gender inequality while not neglecting the boy child education. This helps reduces the low age at marriage and the higher spousal

gap increasing their bargaining powers, as education allow for further mental development building self-confidence and self-assertiveness of women. Women participation in off-farm income activities could prove essential in enhancing WEIA particularly in rural areas. The implication to public policy would be on how to open rural areas to create more off-farm activities opportunities for women to induce further their empowerment.

The effect of wealth measured in terms of value of agricultural assets to WEIA was surprising in the vegetable producing households. Increased wealth was associated with decreasing empowerment levels attributed to men commanding ownership of the wealth making them dominant in household decision making. Hence, though assets were not influencing WEIA in rural areas, a lesson has to be learnt from such findings. The study recommends that even if the women are involved in production and the whole income from their production goes to direct consumption; it does not improve the bargaining power of the women at the household level unless part of the income is invested in assets owned jointly or solely by the woman. The importance of social capital through groups was also demonstrated in enhancing the degree of women empowerment particularly in the leadership domain, as gives men and women a better platform to share ideas, knowledge and demonstrate their decision making capabilities important for changing perception.

Despite the limitation of the study by being cross sectional survey it provides a ground breaking empirical evidence and the study proposes need for further comprehensive studies using time series data which will capture the dynamics of WEIA at household level. Though the study has assessed the level and determinants of WEIA, further studies are required to evaluate the effect of WEIA on agricultural productivity and food security at household level.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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## Appendix 1: Leadership

**Instructions to the enumerator:** This section aims at understanding the leadership potential of the woman. Probe to get honest answers on the questions as no answer is correct or wrong. Use the following scale of; 1 = Strongly disagree 2= Disagree 3= Neutral 4=Agree 5=Strongly agree to indicate the scale that accurately describes the woman from the answers she gives. Illustrate some questions with the appropriate community groups and assets that are available to gauge if;

S/N	Question	Rank
1.	She can list her three greatest weaknesses as a person <sup>a</sup>	-
2.	Her actions in the family and the community reflects here core values <sup>b</sup>	-
3.	She seeks other peoples opinion before making up her mind <sup>c</sup>	-
4.	She openly shares her feelings with others in the family and community <sup>d</sup>	-
5.	She can list her greatest three strengths <sup>a</sup>	-
6.	She does not allow group pressure to control her actions <sup>b</sup>	-
7.	She rarely "lie" in front her friends , family and community members <sup>d</sup>	-
8.	She accepts her feelings about herself <sup>a</sup>	-
9.	In controversial family and community issues , people normally know my stand <sup>b</sup>	-
10.	She is guided by her morals in undertaking community and family duties as leader <sup>b</sup>	-
11.	She listens to others in the community and family ideas before making up her mind <sup>c</sup>	-
12.	Once she makes mistakes in the family and community, she admits <sup>d</sup>	-
13.	She listens critically on the ideas of those who disagree with her ideas in the family and community <sup>c</sup>	-
14.	She seeks feedback on what truly she is as a person in the family or community <sup>d</sup>	-
15.	She does not emphasize her point at the expense of others in the family and community <sup>c</sup>	-
16.	She does seek feedback to understand her leadership <sup>a</sup>	-

a, b, c and d relates to questions regarding self-awareness, internalized moral perspective, balanced processing and relational transparency respectively. The figures of the rank are summed and transformed to 100%.

## Appendix 2. Sample statistics for the probit model for organic farming participation.

Variable	Description of the variable	Mean for vegetable producers	Mean for honey producers
Head_age	Age of the household head(Years)	48.85 (13.42)	52.54 (11.70)
Offfarm_he	Off-farm activity participation by the household head , 1=yes 0=no	0.62 (0.49)	0.50 (0.50)
gender_head	Gender of the household head, 1=male 0=female	0.77 (0.42)	0.89 (0.46)
Educ_head <sup>a</sup>	Education level of the household head	3.68 (1.11)	2.41 (0 .94)
Hhsize	Household size, numbers	4.57 (1.60)	6.15 (2.29)
Agric_assets	Value of agricultural assets (KES ,000)	268.42 (245.32)	167.14 (29.24)
Farm_size	Farm size in acres	0.82 (0.86)	3.45 (1.36)
Extetim	Number of contacts with extension service providers in 2012	2.76 (5.38)	1.22 (2.10)
Trainnum	Number of trainings received in the year 2012	6.65 (4.96)	10.60 (1.29)
Mktkm	Distance to the nearest produce market (KMS)	3.41 (2.72)	9.89 (6.11)
Credit	Accessed credit in 2012, 1 accessed credit, 0 no credit	0.18 (0.38)	0.10 (0 .30)
Density <sup>b</sup>	Number of active groups household involved in 2012	1.34 (1.03)	1.74 (1.09)
Meet_index <sup>b</sup>	Meeting attendance index, (meetings attended/ scheduled meetings)	0.89 (0.56)	0.67 (0 .34)
Group_het <sup>bc</sup>	Group heterogeneity index	0.43 (0.15)	0.87 (1.48)
Decision <sup>b</sup>	Household involvement in group decisions making , 0-100%	0.67 (3.41)	0.56 (2.57)
Trust <sup>b</sup>	Level of trust in groups (0-100%)	0.58 (3.53)	0.64 (1.67)
System	System of livestock keeping, 1=closed, 0=open	0.49 (0.49)	2.25 (1.02)

Figures in parenthesis are standard errors of the respective means.

<sup>a</sup>Education measured in terms of 1=not gone to school 2=primary 3=secondary 4= tertiary 5= university; <sup>b</sup>Household level social capital dimensions;

<sup>c</sup>The heterogeneity index derived from questions of whether members were from the same neighborhood, occupation, kingroup, economic status, religion, gender, education level and age group.

## Full Length Research Paper

# Nutrient absorption march and accumulation of nutrients in developing Roxo de Valinhos fig (*Ficus carica* L.) tree, cultivated under different water regimes

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The nutrient absorption march in fig (*Ficus carica* L.) trees and other fruit trees has been studied only at the seedling stage, and little is known about nutrient accumulation in perennial plants when the entire life cycle of the plant is considered. To this end, the present study aimed to evaluate nutrient accumulation in the Roxo de Valinhos fig tree, cultivated with and without supplemental irrigation. The split-plot array in time was adopted, with the main factor arranged in a randomized block design with four replications. The main plot was the use of supplemental irrigation and the subplots were the eight data collection time-points. The fig plants were sampled for 0, 40, 80, 120, 160, 200, 240 and 280 days after pruning (DAP). The maximum accumulation of dry matter mass and nutrients occurred between 160 and 240 DAP in both systems. Plants in the irrigated system showed greater overall accumulation of nutrients in all the organs, more prominently in the leaves, branches, and stem. The dry matter mass and nutrient accumulation was in the order stem > leaves > branches > roots > fruit in the irrigated system and branches > stem > leaves > roots > fruit in the non-irrigated system.

**Key words:** *Ficus carica* L., dry matter mass, nutritional requirement, water irrigation.

## INTRODUCTION

Brazil is the second largest exporter of fresh fig globally, with Turkey being the largest. Fig production in Brazil has increased considerably in recent years, with a fig production area of approximately 2,814 ha, producing

28,253 t of figs with an average yield of 10,040 kg ha<sup>-1</sup> (IBGE, 2014). However, the fig production for both domestic consumption and export is dependent on a single variety, Roxo de Valinhos (Costa et al., 2015). The

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Roxo de Valinhos variety, also called the common fig tree, is known for its high vigor and rusticity, in addition to its ability to adapt well to different environments and tolerance to drastic pruning (Boliari and Corrêa, 1999).

Drastic pruning, an essential practice for fig trees cultivated in Brazil, is carried out to eliminate branches attacked by drills and rusted leaves (Penteado and Franco, 1997). However, with the almost complete removal of the branches, large amounts of nutrients are lost from the rapidly growing parts of the tree. Therefore, the lost nutrients must be sufficiently replenished to meet the nutritional demand of the tree. As for nutritional management of the fig tree, fertilization regimes for the period of seedling plantation and subsequent growth have been recommended exclusively on the basis of soil analysis. Even the analysis of the nutritional status of the plant through leaf diagnosis - although a valuable tool for perennial crops - is still incipient in fig tree culture (Brizola, 2003).

Nutrient absorption march refers to the dynamics of accumulation of nutrients in the plant dry mass at the growth stage (Prado and Nascimento, 2003). Augostinho et al. (2008) emphasized that knowledge of the amount of nutrients accumulated in the plant, at each stage of development, provides valuable information that aids in the designing of a crop fertilization program. According to Malavolta et al. (1997), nutrient absorption by the plant is influenced by the stage of development; it intensifies at the flowering stage, and during formation and growth of the fruit or organ to be harvested. In this context, these authors emphasize the importance of knowing not only the total amount of absorbed nutrients, but also their concentrations at the different stages of development of vegetable crops.

Leonel and Brizola (2011) emphasized that the management practices of drastic pruning of the fig tree makes it difficult to calibrate the dosages of nutrients to be administered to this plant. According to them, it is possible to determine the quantities of such nutrients exported by the production, but it is difficult to assess the nutrient needs for the annual growth of branches, buds, and roots, and for the growth of new leaves. The accumulation of nutrients by plants is a function of the genotypic characteristics of each species, the development stage, and external factors that regulate nutrient absorption by the plants, especially water (Rozane, 2008). In this sense, with the aim of expanding information on the nutritional demand of fig trees, studies on nutrient extraction from branches, leaves, and fruits were carried out by Brizola et al. (2004) and Leonel and Techio (2009). However, studies elucidating the nutrient absorption curve for fig trees grown in field conditions remain scarce. Thus, considering the high amount of nutrients removed during pruning under different crop conditions and the need to replenish these nutrients adequately and at the appropriate phenological stages, the present study aimed to evaluate nutrient

accumulation in the Roxo de Valinhos fig tree, cultivated with and without supplemental irrigation.

## MATERIALS AND METHODS

The study was conducted in the experimental orchard of the Horticulture Department of the Faculty of Agronomic Sciences, FCA/UNESP, Campus de Botucatu-SP. The local geographic coordinates are 22° 52' 47" S latitude, 48° 25' 12" W longitude, and 810 m altitude. The predominant climatic type, based on the Köppen International system, according to the temperate rainy climate - Cfa, characterized as warm temperate (mesothermal) with summer rains and winter drought, with precipitation and average annual temperature of 1530 mm and 21°C, respectively (Cunha et al., 1999). The soil in the area was classified as Red Nitisol (Embrapa, 2006). The results of soil chemical analyses throughout the study are presented in Table 1. Data on rainfall and minimum, average, and maximum daily temperatures were provided by the Meteorological Office of the Natural Resources Department of the Faculty of Agronomic Sciences/UNESP. The climatic data are shown in Figure 1.

Transplantation of the Roxo de Valinhos fig tree seedlings was carried out in December, 2010, adopting a spacing of 2.5 m between rows and 2.5 m between plants. The 30 cm tall seedlings were transplanted into 50 cm × 50 cm × 50 cm pits, which were previously fertilized with 1 L of corral manure, 0.5 kg of limestone, and 0.5 kg of magnesium thermophosphate containing 0.1% boron and 0.25% zinc. Cover fertilization was performed based on the recommendation of Campo Dall'Orto et al. (1996), who advocated the application of fertilizer based on the interpretations from soil chemical analysis and the plant age. Thus, in 2011 and early 2012, each plant was fertilized with 45 g of urea and 35 g of potassium chloride. Fertilization was performed every two months. The fig plants were pruned on June 28, 2011 (6/28/2011) to ensure that each plant contains three productive branches in the first year. Further cuttings were also performed when necessary. For phytosanitary treatment, copper-based products were used to control rust caused by the fungus *Cerotelium fici*. The fungicides tebuconazole (Folicur®) and methyl thiophanate (Cercobin®) were also applied whenever necessary. Control of weeds between the planting lines was performed through periodic mechanical brushing, and crowning of the plants was performed through manual weeding. The soil water retention curves were experimentally obtained with samples of soil in volumetric rings and through Richardsporous plates method of matrix potential and soil moisture values. The soil density obtained was 1.4822 g cm<sup>-3</sup> from 0 to 20 cm, and 1.3593 g cm<sup>-3</sup> from 20 to 40 cm. It was used by the method of minimizing the squares of the deviations being adjusted using the Excel Solver optimization tool and presented coefficients of determination (*r*<sup>2</sup>) of 0.9974 and 0.9930, for the depths of 0 to 20 cm and 20 to 40 cm, respectively. The irrigation system adopted was drip irrigation, using two emitters with a flow of 1.5 L h<sup>-1</sup> per plant. Irrigation management was based on the permanence of the soil matrix potential between the field capacity and a maximum value of 60 kPa, monitored by tensiometers.

$$\theta_{20} = 0.169 + \frac{0.437}{\left[1 + (2.811 \psi_m)^{1.363}\right]^{0.2663}} \quad (1)$$

$$\theta_{40} = 0.210 + \frac{0.641}{\left[1 + (2.77 \psi_m)^{1.5044}\right]^{0.3353}} \quad (2)$$

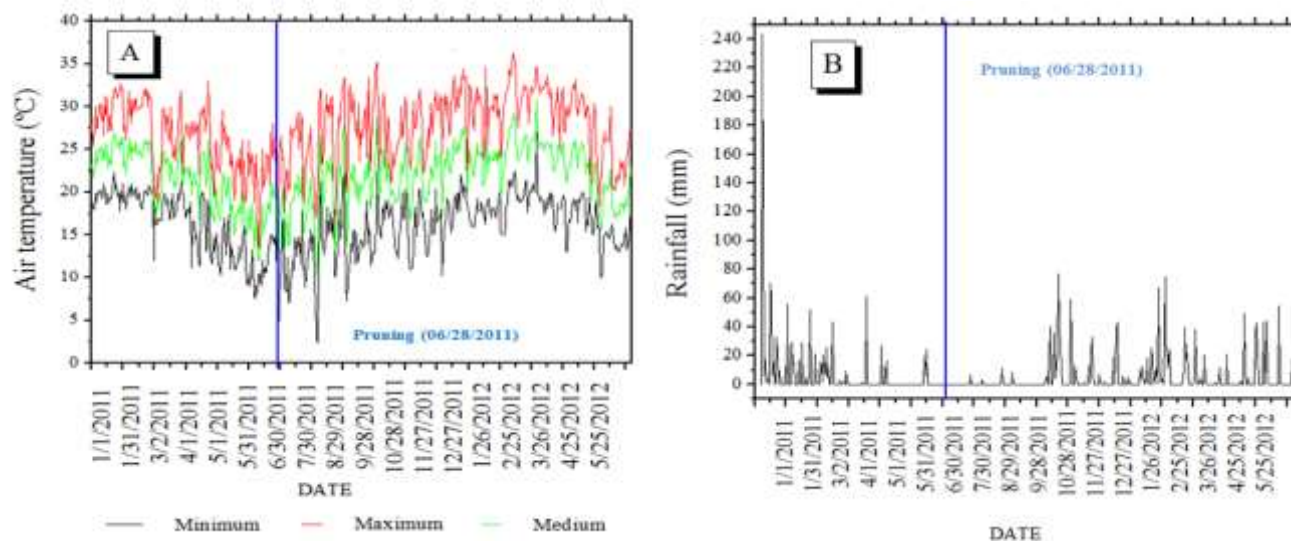
**Table 1.** Chemical characteristics of soil before transplanting.

Sample (cm)	pH	O.M.	Al <sup>3+</sup>	H+Al	K	Ca	Mg	SB	CEC	BS%
	CaCl <sub>2</sub>	g dm <sup>-3</sup>	mmolc dm <sup>-3</sup>							
0-20	5.5	40.0	0.0	24.0	2.0	28.0	11.0	41.0	65.0	64.0
20-40	5.1	40.0	0.0	29.0	2.8	29.0	10.0	42.0	71.0	59.0

	P <sub>resina</sub>	Sulfur	Boron	Copper	Iron	Manganese	Zinc
	mg dm <sup>-3</sup>						
0-20	17.0	3.0	0.22	7.8	77.0	30.2	1.5
20-40	14.0	3.0	0.19	7.3	61.0	25.1	1.1

**Source:** Laboratory of Soil Fertility, Department of Agronomy, Faculty of Agronomic Sciences/UNESP, Botucatu, Brazil. pH in water in a 1:1 ratio; OM: organic matter, determined by the Walkley-Black method; Al<sup>3+</sup>: extracted with 1 mol L<sup>-1</sup> KCl; SB: sum of bases (SB = Ca<sup>2+</sup> + Mg<sup>2+</sup> + K<sup>+</sup> + Na<sup>+</sup>); BS%: base saturation [BS% = (SB/T) × 100], and CEC: cation exchangeable capacity [CEC = SB + (H+Al)].



**Figure 1.** Monthly values of temperature (A) and rainfall (B), between January 01, 2011 (1/1/2011) and May 25, 2012 (5/25/2012), Botucatu, São Paulo, Brazil.

Irrigation management was performed using the tensiometry technique, with batteries of two mercury tensiometers installed in each treatment, in randomly assigned plots. The first tensiometer, installed at a 20 cm depth (relative to the center of the porous capsule of 6 cm length), was considered as the decision tensiometer, and the irrigations were performed based on these readings. The second tensiometer was considered as the control and installed at a 40 cm depth relative to the center of the porous capsule, to control the applied depth of irrigation.

According to Klar (1980), the tensiometer reliably operates up to the -80 kPa range, and the variations of the readings increased when the potential becomes more negative. The tensiometer readings were taken every two days, and values for the mercury column rise were used to obtain the matrix potential. The management was aimed at limiting the soil matrix potential to approximately -30 kPa and/or volumetric moisture equal to 0.2988 cm<sup>3</sup> water cm<sup>-3</sup> soil. The  $\psi_m$  values were verified in the readings, and with these, the respective levels of volumetric humidity were calculated by the difference between both, the need to restore the

water volumes was verified, respectively. The volume of water applied depended on the volume explored by the root system of the plant, which in this case was monitored by the collection of the root systems by the destructive evaluations of the growth analysis, which in this case presented measurements of equatorial and longitudinal lengths having as reference the stem of the plant and the depth reached. The rainfall distribution data and the depths of irrigation used during the experimental period are presented in Table 2.

Eight collections were performed between June, 2011 and April, 2012 (June 28, August 07, September 17, October 25, December 04, 2011; and January 13, February 24, and April 03, 2012). The first collection was performed before pruning in order to characterize the plants before application of the treatments. Eight plants were harvested from the soil using a backhoe, four plants were cultivated with irrigation, and four plants were cultivated without irrigation, totaling 64 plants at the end of the experiment. The interval between two successive collections was 40 days, from formative pruning, which was carried out on June 28, 2011.

**Table 2.** Accumulated monthly values of effective precipitation and depth of irrigation in Roxo de Valinhos fig trees in the early development phase, in Botucatu, São Paulo, Brazil.

Month	Rainfall (mm)	Irrigated water depth (L m <sup>-2</sup> )		Total depth of water received (L m <sup>-2</sup> )	
		CI	SI	CI	SI
January 2011	712.25	–	–	712.25	712.25
February 2011	188.13	–	–	188.13	188.13
March 2011	163.50	–	–	163.50	163.50
April 2011	126.50	–	–	126.50	126.50
May 2011	16.50	–	–	16.50	16.50
June 2011	49.90	–	–	49.90	49.90
July 2011	7.00	16.34	–	23.34	7.00
August 2011	24.75	14.8	–	39.55	24.75
September 2011	0.00	11.98	–	11.98	0.00
October 2011	359.58	47.85	–	407.43	359.58
November 2011	102.50	18.06	–	120.56	102.50
December 2011	143.38	25.27	–	168.65	143.38
January 2012	357.25	23.28	–	380.53	357.25
February 2012	166.75	32.99	–	199.74	166.75
March 2012	58.88	21.93	–	80.81	58.88
Total	2476.85	212.5	–	2689.35	2476.85

Therefore, the collections were performed at 0 (pruning time), 40, 80, 120, 160, 200, 240 and 280 days after pruning (DAP).

After harvest, the plants were taken to the Fruit Production Laboratory of the Plant Production Department/FCA/UNESP, where they were sectioned into the following parts: root, stem, branches, leaves, and fruits. Each part of the plant was washed with water and detergent, and was eventually washed with distilled water. Subsequently, the plant material was packed in paper bags and dried in a hot-air oven with forced circulation at a temperature of 65°C until it reached constant weight. Finally, it was weighed to determine the mass of dry matter, and then milled in a Willey mill for determination of nutrient concentrations.

The samples were taken to the Department of Natural Resources Laboratory/Soil Science/FCA/UNESP, where the macronutrient concentrations were determined, according to the method recommended by Malavolta et al. (1997). The accumulation of nutrients was determined by multiplying the nutrient concentrations by the dry mass of each organ (root, stem, branches, leaves, and fruits). The experimental design was a split-plot array in time, with the main factor arranged in randomized blocks, with four replications. The main plot was the use of supplementary irrigation, and the subplots were the eight collection time-points. When significant, the regressions (with the independent variable being the evaluation time–DAP) were adjusted by the Sisvar statistical package and the graphical representations made in the Origin 6.0 program. The differences between means were subjected to variance analysis by the F test and compared by the Tukey test at 5% significance, using the Sisvar program (Pimentel Gomes, 2009).

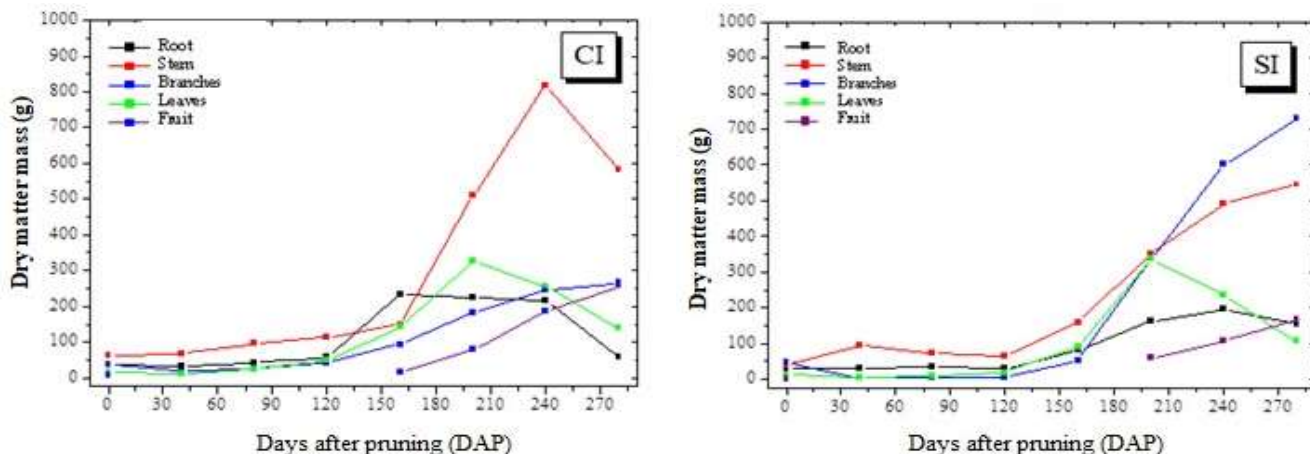
## RESULTS AND DISCUSSION

The cultivation time and water regime had a significant effect on the dry matter mass of Roxo de Valinhos fig plants (Figure 2). In all partitions, a sigmoid behavior for the growth curves was observed in both the systems. The maximum accumulation of the dry matter mass in the

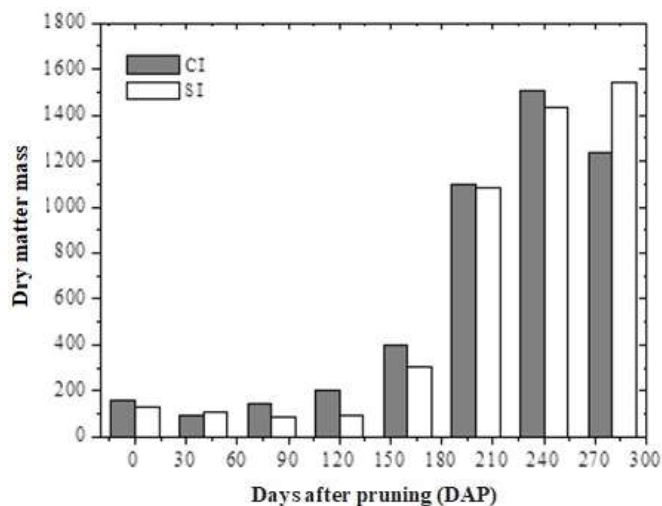
irrigated system was observed between 160 and 240 DAP, with the stem showing the greatest accumulation. In contrast, in the non-irrigated system, the branches showed the greatest accumulation of dry matter mass. Rozane (2008), when studying the dry matter accumulation of two cultivars of starfruit trees grown under different water regimes (irrigated and non-irrigated systems) found that the stem showed the greatest accumulation of dry matter mass in both the water regimes.

Silva et al. (2011) evaluated the dry matter mass of Roxo de Valinhos fig plants in different cropping systems, which combined treatments with or without mulching and treatments with or without irrigation. The results of their study showed that, in all the treatments, the branches showed the greatest accumulation of dry matter mass, which is consistent with the findings of the present study. The findings of Da Costa et al. (2014) also corroborate these previous results and ours. They evaluated the influence of different soil cover management methods on the productivity of fig trees cultivated with and without irrigation, and reported that mulching resulted in a higher average yield of mature fruits and that under these conditions, the branches showed the highest accumulation of dry matter mass.

Figure 3 shows the accumulation of dry matter mass in the whole plant during the entire growth period in the two irrigation systems. In general, irrigation promoted a greater total accumulation of dry matter mass in the 3rd, 4th, and 5th harvests, which occurred at 80, 120, and 160 DAP, respectively. In this period, the third collection was mainly preceded by low rainfall, which explains the higher values for dry matter mass at this time-interval in the



**Figure 2.** Dry matter mass of roots, stem, branches, leaves, and fruits of the Roxo de Valinhos fig tree, according to the cultivation period in the irrigated (IC) and non-irrigated (SI) systems in Botucatu, São Paulo, Brazil, 2013.



**Figure 3.** Dry matter mass of the whole Roxo de Valinhos fig tree according to the cultivation period in the irrigated (IC) and non-irrigated (SI) system in Botucatu, São Paulo, Brazil, 2013.

plants that were irrigated (Figure 1B). However, recovery of dry matter mass production in the non-irrigated plants was observed during the rainy season, even surpassing that of the plants irrigated in the last collection. This behavior may have occurred due to the cycle anticipation of the fig trees that were irrigated.

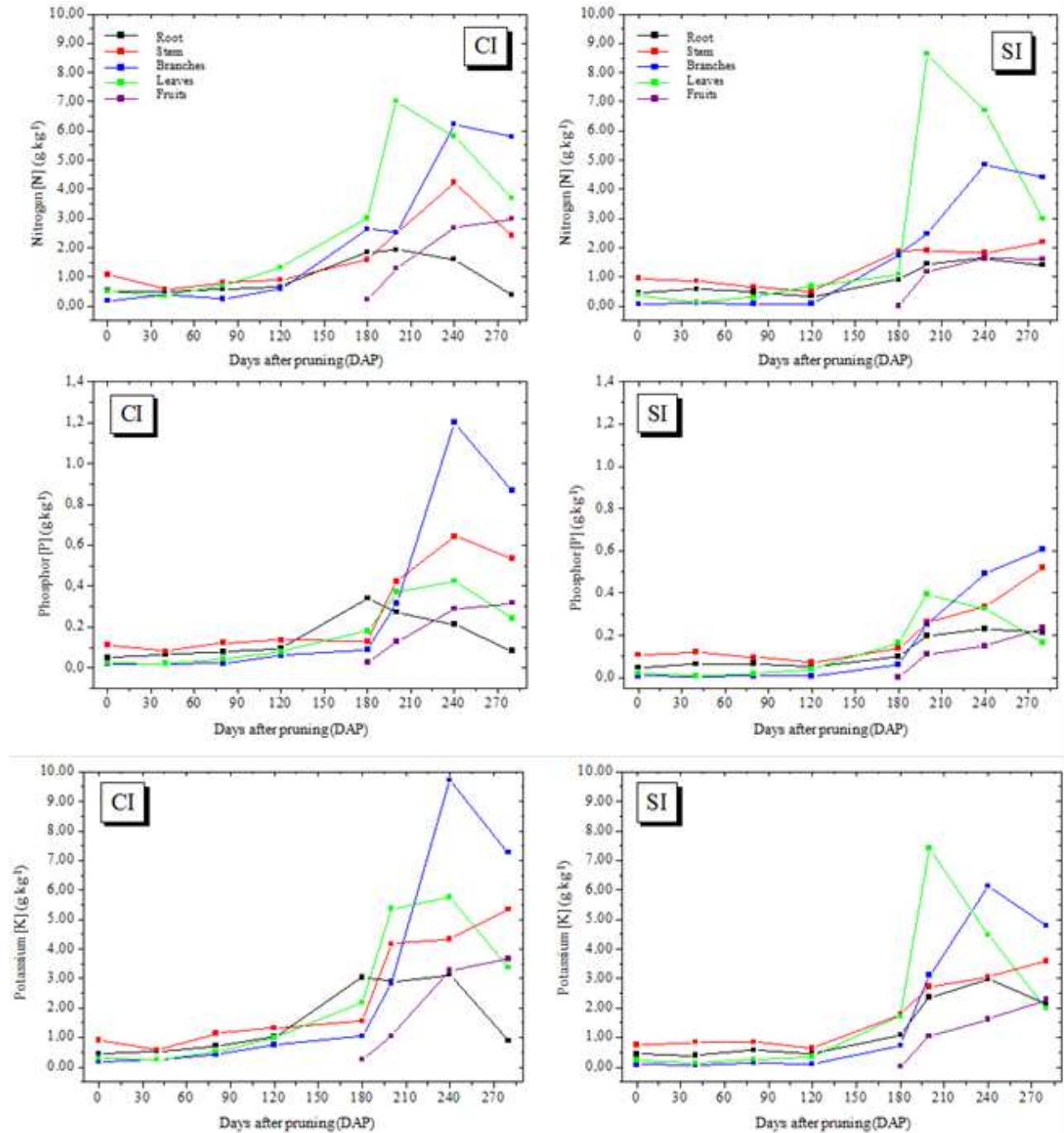
The determination of dry matter mass production is of great importance, because when the nutrient accumulation curve cannot be determined, it provides information that is representative of the nutrient absorption march (Souza and Coelho, 2001). Such a comparison can be made because 5% of plant dry matter mass is composed of mineral nutrients.

The accumulation of macronutrients in the roots,

leaves, stem, branches, and fruits of the Roxo de Valinhos fig tree was influenced by the water regime and the cultivation period (Figures 4 and 5). In general, in plants in both the irrigated and non-irrigated systems, the initial accumulation rate was low, reaching maximum values only between 160 and 240 DAP. The best adjusted regression model for all nutrients was the cubic polynomial. The observed sigmoid behavior for nutrient accumulation is consistent with the accumulation of dry matter mass. The highest accumulation of nitrogen, calcium, and magnesium was observed in leaf dry matter mass under both the irrigated and non-irrigated conditions (Figures 4 and 5). Phosphorus, potassium, and sulfur showed the greatest accumulation in the branch dry matter mass under both the irrigated and non-irrigated conditions.

In general, irrigation favored macronutrient accumulation in most of the analyzed plant organs, with this effect being more evident in the branches and the stem. These results are consistent with those reported by Rozane (2008), who observed a greater accumulation of nutrients in star fruit trees that were irrigated than in those that were not irrigated. It should be noted that the greatest accumulation of macronutrients occurred in the aerial parts (leaves and branches) of the plant. The higher accumulation of N in leaves can be explained by the role of this element in photosynthesis. It is a part of the chlorophyll and the enzymes phosphoenolpyruvate carboxylase and ribulose 1.5 bisphosphate carboxylase/oxygenase, which are responsible for atmospheric CO<sub>2</sub> fixation (Prado, 2008).

Sete et al. (2015) reported that the highest percentages and concentrations (mg plant<sup>-1</sup>) of total N in annual organs (leaves, fruits, and branches) of the fig tree can be attributed to the fast cell division in the plant tissue and, therefore, these organs probably act like an N drain during the vegetative growth and productive cycles of the



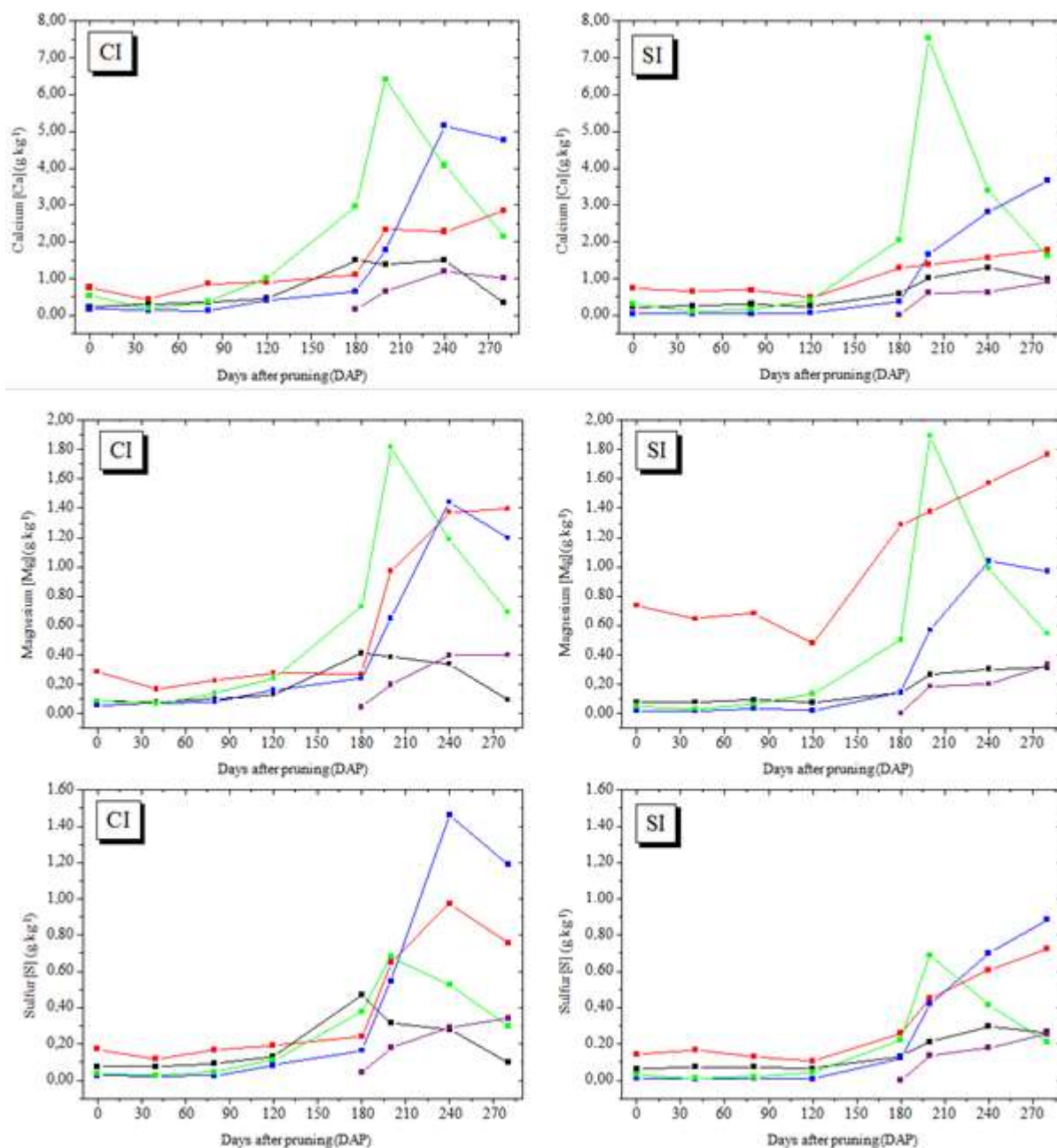
**Figure 4.** Concentrations of nitrogen, phosphorus, and potassium accumulated in the dry mass of root, stem, branches, leaves, and fruits of the Roxo de Valinhos fig tree, according to cultivation period in the irrigated (IC) and non-irrigated (SI) systems in Botucatu, São Paulo, Brazil, 2013.

fig tree.

Celedônio et al. (2016) examined the development of the Roxo de Valinhos fig tree in different cultivation environments and with five levels of fertilization (mineral and biofertilizer). They found that the accumulation of N in fig leaves after fertigation with bovine biofertilizer, was

significantly higher than that after mineral fertilization. According to Grangeiro et al. (2007), accumulation of calcium in the aerial parts is attributable to the fact that this element is absorbed by the roots and translocated to the aerial parts, but is not redistributed to the other parts of the plant owing to its low mobility in the plant.





**Figure 5.** Concentrations of calcium, magnesium, and sulfur accumulated in the dry matter mass of roots, stems, branches, leaves, and fruits of the Roxo de Valinhos fig tree, according to cultivation period in the irrigated (IC) and non-irrigated (SI) systems in Botucatu, São Paulo, Brazil, 2013.

Magnesium was accumulated in the aerial parts, and mainly in the leaves; this finding validates the importance of this element in the constitution of chlorophyll molecule. Depending on the level of magnesium in the plant, between 6 and 25% of the total magnesium is bound to the chlorophyll molecule and another 5 to 10% is firmly attached to pectates in the cell wall or occurs as soluble salt in the vacuole (Marschner, 1995).

In the irrigated system, the branches showed the

greatest accumulation of potassium, but in the non-irrigated system, the greatest accumulation of this element was observed in the leaves. These results agree with those reported by Brizola et al. (2005), which suggest increased accumulation of potassium in the aerial parts (branches and fruits) of fig trees grown under different potassium doses. The authors found that the higher dose resulted in greater accumulation of the total dry matter mass, and consequently, greater potassium

**Table 3.** Regression equations of the macronutrients (g plant<sup>-1</sup>) accumulated over 280 days after pruning of the Roxo de Valinhos fig trees in the irrigated system.

Organ	Equation	R <sup>2</sup>	P
Nitrogen			
Root	$Y = 0.62877 - 0.01929 \text{ DAP} + 2.78484\text{E-}4 \text{ DAP}^2 - 7.57807\text{E-}7 \text{ DAP}^3$	0.9320	0.0085
Stem	$Y = 1.23409 - 0.03343 \text{ DAP} + 3.5926\text{E-}4 \text{ DAP}^2 - 7.81406\text{E-}7 \text{ DAP}^3$	0.7910	0.0759
Branch	$Y = 0.44626 - 0.02296 \text{ DAP} + 2.67925\text{E-}4 \text{ DAP}^2 - 3.97041\text{E-}7 \text{ DAP}^3$	0.9250	0.1028
Leaf	$Y = 0.86153 - 0.05564 \text{ DAP} + 7.64433\text{E-}4 \text{ DAP}^2 - 1.87765\text{E-}6 \text{ DAP}^3$	0.8482	0.0497
Phosphor			
Root	$Y = 0.06412 - 0.0022 \text{ DAP} + 3.76376\text{E-}5 \text{ DAP}^2 - 1.05789\text{E-}7 \text{ DAP}^3$	0.8684	0.0377
Stem	$Y = 0.13439 - 0.00297 \text{ DAP} + 3.12528\text{E-}5 \text{ DAP}^2 - 5.25176\text{E-}8 \text{ DAP}^3$	0.8206	0.0566
Branch	$Y = 0.07496 - 0.00457 \text{ DAP} + 4.13083\text{E-}5 \text{ DAP}^2 - 4.48419\text{E-}8 \text{ DAP}^3$	0.7706	0.0907
Leaf	$Y = 0.04961 - 0.00335 \text{ DAP} + 4.48925\text{E-}5 \text{ DAP}^2 - 1.07171\text{E-}7 \text{ DAP}^3$	0.8730	0.0289
Potassium			
Root	$Y = 0.66973 - 0.0292 \text{ DAP} + 4.47784\text{E-}4 \text{ DAP}^2 - 1.20848\text{E-}6 \text{ DAP}^3$	0.9312	0.0087
Stem	$Y = 0.93907 - 0.0134 \text{ DAP} + 1.67056\text{E-}4 \text{ DAP}^2 - 2.20513\text{E-}7 \text{ DAP}^3$	0.9063	0.0160
Branch	$Y = 0.61629 - 0.03258 \text{ DAP} + 3.10346\text{E-}4 \text{ DAP}^2 - 3.26977\text{E-}7 \text{ DAP}^3$	0.7868	0.0789
Leaf	$Y = 0.6531 - 0.04686 \text{ DAP} + 6.21756\text{E-}4 \text{ DAP}^2 - 1.47583\text{E-}6 \text{ DAP}^3$	0.8472	0.0415
Calcium			
Root	$Y = 0.34276 - 0.01472 \text{ DAP} + 2.22513\text{E-}4 \text{ DAP}^2 - 6.02589\text{E-}7 \text{ DAP}^3$	0.9095	0.0149
Stem	$Y = 0.72696 - 0.00777 \text{ DAP} + 9.64424\text{E-}5 \text{ DAP}^2 - 1.49271\text{E-}7 \text{ DAP}^3$	0.9051	0.0163
Branch	$Y = 0.34477 - 0.01593 \text{ DAP} + 1.34708\text{E-}4 \text{ DAP}^2 - 4.88292\text{E-}8 \text{ DAP}^3$	0.8777	0.0269
Leaf	$Y = 0.82605 - 0.05924 \text{ DAP} + 7.96088\text{E-}4 \text{ DAP}^2 - 2.02507\text{E-}6 \text{ DAP}^3$	0.8306	0.0507
Magnesium			
Root	$Y = 0.10646 - 0.00392 \text{ DAP} + 5.86091\text{E-}5 \text{ DAP}^2 - 1.59513\text{E-}7 \text{ DAP}^3$	0.9343	0.0079
Stem	$Y = 0.32188 - 0.00679 \text{ DAP} + 6.26643\text{E-}5 \text{ DAP}^2 - 8.34979\text{E-}8 \text{ DAP}^3$	0.8807	0.026
Branch	$Y = 0.11937 - 0.00533 \text{ DAP} + 5.60199\text{E-}5 \text{ DAP}^2 - 7.53361\text{E-}8 \text{ DAP}^3$	0.8567	0.0366
Leaf	$Y = 0.18713 - 0.01474 \text{ DAP} + 2.03224\text{E-}4 \text{ DAP}^2 - 5.13179\text{E-}7 \text{ DAP}^3$	0.7915	0.0756
Sulfur			
Root	$Y = 0.09162 - 0.00318 \text{ DAP} + 5.15842\text{E-}5 \text{ DAP}^2 - 1.43886\text{E-}7 \text{ DAP}^3$	0.8318	0.0501
Stem	$Y = 0.21011 - 0.00548 \text{ DAP} + 5.90493\text{E-}5 \text{ DAP}^2 - 1.11741\text{E-}7 \text{ DAP}^3$	0.8281	0.0521
Branch	$Y = 0.09506 - 0.00603 \text{ DAP} + 5.72701\text{E-}5 \text{ DAP}^2 - 6.91647\text{E-}8 \text{ DAP}^3$	0.8402	0.0453
Leaf	$Y = 0.07414 - 0.00612 \text{ DAP} + 8.49197\text{E-}5 \text{ DAP}^2 - 2.14588\text{E-}7 \text{ DAP}^3$	0.8964	0.0194

accumulation.

The highest levels of phosphorus were accumulated in the branches, with higher values observed for the irrigated system. Brizola et al. (2005), when examining the nutrient content in leaves and branches of the Roxo de Valinhos fig tree under different doses of potassium, reported that in the treatment without potassium fertilization, phosphorus levels were the highest in the branches. The authors observed the same trend in the accumulation of this element when they multiplied its concentration with the dry matter mass of the organ.

The regression equations presented in Tables 3 and 4 represent the accumulation of macronutrients in the fig

trees according to the cultivation period and water regimes. The best-fitted equations were the third-degree or cubic polynomial ones. Prado (2008) reported that for crops in their productive stages, the curve that best describes the extraction nutrients due to time is the sigmoid type, as is the curve for dry matter mass accumulation. Thus, this study suggests that, when the plant is young, the nutrient accumulation is low, and so is the dry matter mass. Thereafter, there is an increase in the dry matter mass accumulation, and absorption of nutrients increases, representing a logarithmic curve. In the final period of physiological maturation, there is a stabilization phase, in which the nutrient absorption is low

**Table 4.** Regression equations of the macronutrients (g plant<sup>-1</sup>) accumulated over 280 days after pruning of the Roxo de Valinhos fig trees in the non-irrigated system.

Organ	Equation	R <sup>2</sup>	P
Nitrogen			
Root	Y = 0.59286 - 0.01052 DAP + 1.18224E-4 DAP <sup>2</sup> - 2.46562E-7 DAP <sup>3</sup>	0.8252	0.0387
Stem	Y = 1.04301 - 0.01653 DAP + 1.70302E-4 DAP <sup>2</sup> - 3.47259E-7 DAP <sup>3</sup>	0.8688	0.0308
Branch	Y = 0.32119 - 0.02611 DAP + 2.87513E-4 DAP <sup>2</sup> - 4.88173E-7 DAP <sup>3</sup>	0.9388	0.0069
Leaf	Y = 1.01319 - 0.08292 DAP + 0.00101 DAP <sup>2</sup> - 2.44048E-6 DAP <sup>3</sup>	0.6432	0.2081
Phosphor			
Root	Y = 0.06247 - 9.6245E-4 DAP + 1.16289E-5 DAP <sup>2</sup> - 2.1604E-8 DAP <sup>3</sup>	0.8684	0.0310
Stem	Y = 0.12006 - 5.90271E-4 DAP + 4.7652E-7 DAP <sup>2</sup> + 2.41778E-8 DAP <sup>3</sup>	0.9728	0.0014
Branch	Y = 0.02495 - 0.00141 DAP + 9.71824E-6 DAP <sup>2</sup> + 1.15002E-8 DAP <sup>3</sup>	0.9526	0.0042
Leaf	Y = 0.04762 - 0.00396 DAP + 5.09479E-5 DAP <sup>2</sup> - 1.25022E-7 DAP <sup>3</sup>	0.8357	0.0478
Potassium			
Root	Y = 0.60082 - 0.01974 DAP + 2.35928E-4 DAP <sup>2</sup> - 5.08721E-7 DAP <sup>3</sup>	0.8430	0.0437
Stem	Y = 0.89488 - 0.01329 DAP + 1.50987E-4 DAP <sup>2</sup> - 2.43926E-7 DAP <sup>3</sup>	0.9501	0.0046
Branch	Y = 0.43606 - 0.03365 DAP + 3.43737E-4 DAP <sup>2</sup> - 5.64571E-7 DAP <sup>3</sup>	0.8419	0.0443
Leaf	Y = 0.74749 - 0.06894 DAP + 8.78672E-4 DAP <sup>2</sup> - 2.18917E-6 DAP <sup>3</sup>	0.6739	0.1792
Calcium			
Root	Y = 0.74749 - 0.06894 DAP + 8.78672E-4 DAP <sup>2</sup> - 2.18917E-6 DAP <sup>3</sup>	0.8759	0.0277
Stem	Y = 0.79221 - 0.00933 DAP + 9.57848E-5 DAP <sup>2</sup> - 1.78424E-7 DAP <sup>3</sup>	0.9345	0.0079
Branch	Y = 0.14564 - 0.00806 DAP + 5.74149E-5 DAP <sup>2</sup> + 6.59375E-8 DAP <sup>3</sup>	0.9583	0.0032
Leaf	Y = 0.73858 - 0.06793 DAP + 8.81258E-4 DAP <sup>2</sup> - 2.24021E-6 DAP <sup>3</sup>	0.6513	0.1994
Magnesium			
Root	Y = 0.09011 - 0.00129 DAP + 1.50093E-5 DAP <sup>2</sup> - 2.62879E-8 DAP <sup>3</sup>	0.9111	0.0144
Stem	Y = 0.79221 - 0.00933 DAP + 9.57848E-5 DAP <sup>2</sup> - 1.78424E-7 DAP <sup>3</sup>	0.9345	0.0079
Branch	Y = 0.06982 - 0.00485 DAP + 4.79346E-5 DAP <sup>2</sup> - 6.32551E-8 DAP <sup>3</sup>	0.8923	0.0209
Leaf	Y = 0.16192 - 0.01577 DAP + 2.09081E-4 DAP <sup>2</sup> - 5.26901E-7 DAP <sup>3</sup>	0.6729	0.1772
Sulfur			
Root	Y = 0.07753 - 0.00132 DAP + 1.52321E-5 DAP <sup>2</sup> - 2.81041E-8 DAP <sup>3</sup>	0.9112	0.0144
Stem	Y = 0.17181 - 0.00235 DAP + 2.10757E-5 DAP <sup>2</sup> - 1.87618E-8 DAP <sup>3</sup>	0.9581	0.0032
Branch	Y = 0.03515 - 0.00231 DAP + 1.90928E-5 DAP <sup>2</sup> + 2.10159E-9 DAP <sup>3</sup>	0.9598	0.0029
Leaf	Y = 0.07174 - 0.00646 DAP + 8.33871E-5 DAP <sup>2</sup> - 2.08321E-7 DAP <sup>3</sup>	0.7397	0.1154

or even negligible.

## Conclusion

The results of the present study suggest that the irrigated system provided greater accumulation of nutrients in all the organs, more prominently so in the leaves, branches, and stem. The maximum accumulation of dry matter mass and nutrients occurred between 160 and 240 DAP. The organ-wise accumulation of dry matter mass in the Roxo de Valinhos fig trees was in the order stem > leaves > branches > roots > fruits for the irrigated system and branches > stem > leaves > roots > fruits for the non-

irrigated system. In conclusion, the stem, branches, and leaves were the organs that showed the greatest accumulation of dry matter mass and nutrients in the Roxo de Valinhos fig tree in the two water regimes. Aiming at the continuity and improvement of this study, it would be relevant in future work to apply the nutrient absorption gait in practice, and thus to determine a fertilization program that would provide satisfactory performance of the fig tree.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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## Full Length Research Paper

# Genotypic variability in some sun flower (*Helianthus annuus* L.) hybrids evaluated in Khordunia under rainfed conditions

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Twenty locally generated hybrids of Sun flower (*Helianthus annuus* L.) hybrids were evaluated in two seasons (2012 and 2013) for yield and yield components at Khordunia area, Blue Nile State under rain fed conditions. A randomized complete block design with six replications was used for laying out the field experiments. The seeds were sown in the second and third week of July in the first and second seasons, respectively in plots 6 × 3 m<sup>2</sup>. Each plot was divided into four ridges 70 cm apart and 6-m long. Three seeds were sown in holes of 20 cm distance along the ridge then thinned into one plant per hole three weeks after sowing. Weeding was practiced three times to control weeds. Rains were recorded during autumn at Khordunia area. Fertilizers were not applied. The heads of the sample were bagged during the seed filling period using paper bags to avoid birds attack. Data were collected on the following characters: Days to 50% flowering, days to maturity, plant height, stem diameter, head diameter (cm), number of seeds/head, percentage of empty seed, 1000-seed weight (g), seed yield/plant (g) and seed yield (t/ha). Phenotypic, genotypic, and environmental variances were determined. The results in season 2012 revealed highly significant differences among the undertaken hybrids for plant height, stem diameter, head diameter, empty seed %, 1000-seed weight, seed yield/pant and seed yield (t/ha), whereas only two characters were significant in 2013. These were empty seed% and 1000-seed weight.

**Key words:** Sunflower, seed yield, genetic variability, genetic advance, heritability.

## INTRODUCTION

The continuous demand for vegetable oils led to the interest in sunflower as a source of good quality oil. It ranks fourth among the world oil crops after palm oil,

rapeseed, and soybean (Abdalla and Abdelnour, 2001). Sun flower (*Helianthus annuus* L.) which belongs to the family *Compositae* was rank the third largest source of oil

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**Table 1.** List of the sunflower hybrids used in the study.

S/N	Parent	Hybrid	Code	Origin
1	R1(Male)	Kh 99 X1	SHA1	UK
2	R5(Male)	Kh99X5 (Salih)*	SHA5	"
3	R6 (Male)	Kh99X6 (SHA 6)*	SHA6	"
4	R7(Male)	Kh 99 X7	SHA7	"
5	R11(Male)	Kh 99 X11	SHA11	"
6	R14(Male)	Kh 99 X13	SHA14	"
7	R15(Male)	Kh 99 X15	SHA15	"
8	R17 (Male)	Kh 99 X17	SHA17	"
9	R18 (Male)	Kh 99 X18	SHA18	"
10	R22 (Male)	Kh 99 X22	SHA22	"
11	R25-1(Male)	Kh 99 X25-1	SHA25-1	"
12	R25-2(Male)	Kh 99 X25-2	SHA25-2	"
13	R29(Male)	Kh 99 X29	SHA29	"
14	R30 (Male)	Kh 99 X30	SHA30	"
15	R32(Male)	Kh 99 X32	SHA32	"
16	R35(Male)	Kh 99 X35	SHA35	"
17	R37(Male)	Kh 99 X37	SHA37	"
18	R41(Male)	Kh 99 X41	SHA41	"
19	R42M(Male)	Kh 99 X42	SHA42-M	"
20	Hysun 33	-	-	Check

\*Released recently as commercial varieties.

crops worldwide, following cotton seed and soybean. *H. annuus* is diploid ( $2n = 2X = 34$ ). The main sunflower producing countries are former USSR, Argentina, France, USA, Romania, former Yugoslavia, Bulgaria, Spain and Turkey. According to FAO (1996), the cultivated area in 1996 all over the world was 21 million hectares, producing 2.5 million metric tons with an average seed yield of 1197 kg/ha. Reports by USDA (2000) noted that world sunflower seed production has increased from an average of 23.5 million tons in the mid 1990's to 26.9 million tons in 2000.

Commercial production of sunflower in the Sudan was initiated in the 1987/1988 season, where 63,000 ha were grown under rain fed conditions by the private sector in Damazine. In the following season (1988/1989) the area was increased to 112,000 ha in Damazine and 34,000 ha in Gedarif State. The average yield was 1.5 t/ ha. Because of increasing demand for vegetable oil and to release more sesame seed and groundnut for export, much attention was focused recently on growing sunflower under the irrigated national schemes as a winter crop.

The climatic conditions and soil requirements for sunflower, generally, indicate that the central clay plain is potentially suitable for sunflower growing. Khidir (1997) reported that the most progressive varieties grown in Sudan are imported hybrids like Hysun 33, Sunbred 281, Tec 1560, Tec 1226, Northrubking, Pioneer 6480 and Dekaln G 100 and few open-pollinated ones, like Polareo,

Rodeo and Hungaria. The economic importance of sunflower is the use of oil and seeds as human food, cake and shoot are used as animal feed. The inner pith of the stem is used for making fine writing paper. The plant is grown as an ornamental, a wind break in vegetable farms and for honey bee husbandry.

Sunflower is a highly cross-pollinated crop, characterized by a high percentage of empty seed in open-pollinated and to a lesser extent in F1 hybrid varieties. In the present changing agriculture scenario and water constraint, area of sun flower production has been increased significantly since 2003. Sun flower hybrids produced contain 39 to 52% oil in the seeds and still have better yield potential (Anonymous, 2006).

The objective of this study is to estimate genetic variability in sunflower hybrids under rainfed conditions.

## MATERIALS AND METHODS

Twenty hybrids of sun flower (*H. annuus* L.) were used to evaluate seed yield and its components in this crop for two consecutive seasons (2012 and 2013) in Khourduonia, Blue Nile State (11° 48' N. Lat. and 34° 19' E. Long.) under rain fed conditions. Rainfalls were recorded during autumn at Khourduonia (Table 2). The total rainfalls were 895 and 848.5 mm in the first and second seasons, respectively (Meteorology Authority, 2013). Nineteen of them were derived from crossing of nineteen locally generated restorer lines with one male sterile line (Kh99). Table 1 show the genetic materials used in this study. The seeds were provided by the Department of Agronomy, Faculty of Agriculture, University of Khartoum.

**Table 2.** Monthly rainfall (mm) during autumn seasons of 2012 and 2013 at Khordunia, Blue Nile State.

Month	2012	2013
May	20.0	48.5
June	80.0	59.0
July	180.0	255.0
August	405.0	236.5
September	200.0	168.5
October	10.0	81.0
<b>Total</b>	<b>895.0</b>	<b>848.5</b>

\* Source: Damazine Agro-metrology Station, Blue Nile State.

### Experimental design and data collection

A randomized complete block design with six replications was used to lay out the field experiments. The seeds of each hybrid were sown in plots 6 x 3 m<sup>2</sup> with four ridges 70 cm apart. Three seeds were sown in holes with spacing of 20 cm along the ridge then thinned into one plant per hole three weeks after sowing. Weeding was practiced three times to control the weeds, and fertilizers were not applied.

The sample plants were randomly selected from middle two ridges, and then their heads were covered during the period of seed filling using paper bags to avoid birds attack. Data were collected on plant height, days to 50% flowering, days to maturity, stem diameter, head diameter (cm), number of seeds/head, empty seed %, 1000-seed weight (g), seed yield/plant (g) and seed yield (t/ha).

### Statistical analysis

The collected data were analyzed according to the standard statistical procedure described by Gomez and Gomez (1984). The estimates obtained from the individual analysis of variance were then used to compute the coefficient of variation (CV%) according to the formula:

$$CV\% = \sqrt{(EMS)/G} \times 100$$

where EMS is the error mean sum squares, G is grand mean, genotypic variance ( $\delta_2g$ ) which estimated as follows:

$$\delta_2g = (M_2 - M_3)/r$$

where  $M_2$ ,  $M_3$  and  $r$  are the mean sum squares for genotype, error and number of replications, respectively, phenotypic variance ( $\delta_2ph$ ) which was calculated according to the following formula:

$$\delta_2ph = \delta_2g + \delta_2e,$$

Environmental ( $\delta_2e$ ) variance was calculated as:

$$\delta_2e = M_3,$$

Genotypic and phenotypic coefficient of variations (GCV and PCV%) which were calculated according to the formula of Burton and Devane (1953) as follows:

$$GCV\% = (\delta_2g / G) \times 100$$

$$PVC\% = (\delta_2ph / G) \times 100$$

where G is the grand mean, heritability estimate ( $h^2$ ) in broad sense was estimated for each character according to the procedure of Johnson et al. (1955) as follows:

$$h^2 = (\delta_2g / \delta_2ph) \times 100$$

Genetic advance (GA) and genetic advance as percentage (GA%) of overall mean which were estimated using the formula of Robinson et al. (1949) as follows:

$$GA = k (\delta_2g / \delta_2ph)$$

$$GA\% = (GA/G) \times 100$$

where G is the grand mean, k is the selection differential (it equals 2.06 at 5% selection intensity) as defined by Lush (1943).

## RESULTS AND DISCUSSION

### Phenotypic and genotypic variability

Days to 50% flowering and days to maturity are characters that represent the reproductive stage. The vegetative stage represents plant height and stem diameter. These characters showed significant differences ( $P \leq 0.05$ ) among the twenty sunflower hybrids in season 2012 and non-significant in season 2013. However, the hybrid SHA5 scored the best values for these characters in both seasons. Head diameter, empty seeds%, number of seeds/head, 1000-seed weight and seed yield/plant represent the yield components. However, most of these characters revealed highly significant differences in both seasons. The hybrids SHA25-2, SHA29 and SHA30 gave the best values if not like to that of the check cultivar Hysun 33 (Tables 3 and 4). These findings are in agreement with those of Asifkhan et al. (2003), Rachid et al. (2004), Zannou et al. (2008), Izquierdo and Aguirrezabal (2008) and Mamta et al. (2017a) who stated significant differences among their respective materials. Moreover, Mamta et al. (2017a) who stated that day to 50% flowering was less affected by environmental conditions.

**Table 3.** Means of 10 characters of 20 sunflower hybrids evaluated at Khordunia in season 2012.

Hybrid	DF	DM	Pht (cm)	SD (cm)	HD (cm)	S/H	ES (%)	SW (g)	Y/P (g)	Yield (t/ha)
SHA 1	67.0 <sup>abcde</sup>	85.3 <sup>abc</sup>	75.2 <sup>de</sup>	0.89 <sup>cd</sup>	10.0 <sup>bcd</sup>	436 <sup>a</sup>	11.1 <sup>abc</sup>	33.8 <sup>efg</sup>	15.2 <sup>def</sup>	1.08 <sup>ef</sup>
SHA5	66.0 <sup>e</sup>	84.0 <sup>c</sup>	76.3 <sup>cde</sup>	0.99 <sup>bc</sup>	12.3 <sup>bc</sup>	564 <sup>a</sup>	11.7 <sup>abc</sup>	34.3 <sup>defg</sup>	16.4 <sup>def</sup>	1.17 <sup>def</sup>
SHA 6	66.7 <sup>bcd</sup>	85.3 <sup>abc</sup>	70.2 <sup>e</sup>	0.70 <sup>d</sup>	8.8 <sup>d</sup>	402 <sup>a</sup>	9.7 <sup>bc</sup>	35.4 <sup>cdefg</sup>	20.9 <sup>cde</sup>	1.49 <sup>cde</sup>
SHA 7	67.2 <sup>abcde</sup>	85.7 <sup>abc</sup>	77.8 <sup>cde</sup>	0.91 <sup>cd</sup>	12.5 <sup>b</sup>	585 <sup>a</sup>	8.0 <sup>c</sup>	36.1 <sup>bcd</sup>	24.4 <sup>bcd</sup>	1.74 <sup>cd</sup>
SHA 11	66.2 <sup>de</sup>	84.0 <sup>c</sup>	86.8 <sup>bcd</sup>	1.30 <sup>ab</sup>	12.1 <sup>bc</sup>	455 <sup>a</sup>	12.3 <sup>abc</sup>	38.4 <sup>abc</sup>	18.1 <sup>def</sup>	1.21 <sup>def</sup>
SHA 13	68.0 <sup>ab</sup>	86.7 <sup>ab</sup>	89.4 <sup>bcd</sup>	1.19 <sup>b</sup>	12.5 <sup>b</sup>	523 <sup>a</sup>	14.5 <sup>ab</sup>	36.4 <sup>bcd</sup>	18.5 <sup>def</sup>	1.31 <sup>def</sup>
SHA15	68.3 <sup>a</sup>	86.7 <sup>ab</sup>	88.1 <sup>bcd</sup>	1.19 <sup>b</sup>	11.5 <sup>b</sup>	539 <sup>a</sup>	13.6 <sup>abc</sup>	36.4 <sup>bcd</sup>	22.6 <sup>cde</sup>	1.61 <sup>cde</sup>
SHA 17	67.3 <sup>abcde</sup>	85.7 <sup>abc</sup>	92.4 <sup>bcd</sup>	1.20 <sup>b</sup>	11.7 <sup>bcd</sup>	569 <sup>a</sup>	16.9 <sup>a</sup>	37.0 <sup>bcd</sup>	29.0 <sup>bc</sup>	2.07 <sup>bc</sup>
SHA 18	66.3 <sup>cde</sup>	84.7 <sup>bc</sup>	76.3 <sup>cde</sup>	1.01 <sup>bc</sup>	10.6 <sup>bcd</sup>	471 <sup>a</sup>	13.4 <sup>abc</sup>	38.6 <sup>ab</sup>	19.4 <sup>def</sup>	1.51 <sup>cde</sup>
SHA 22	67.2 <sup>abcde</sup>	85.3 <sup>abc</sup>	83.4 <sup>bcd</sup>	1.04 <sup>bc</sup>	12.6 <sup>b</sup>	527 <sup>a</sup>	14.3 <sup>ab</sup>	38.0 <sup>abc</sup>	23.4 <sup>bcd</sup>	1.65 <sup>cde</sup>
SHA 25-1	67.7 <sup>abcd</sup>	86.3 <sup>ab</sup>	84.7 <sup>bcd</sup>	1.00 <sup>bc</sup>	10.8 <sup>bcd</sup>	374 <sup>a</sup>	12.8 <sup>abc</sup>	30.9 <sup>h</sup>	13.9 <sup>ef</sup>	1.03 <sup>ef</sup>
SHA 25-2	67.8 <sup>abc</sup>	86.3 <sup>ab</sup>	96.0 <sup>bc</sup>	1.18 <sup>b</sup>	12.5 <sup>a</sup>	619 <sup>a</sup>	12.7 <sup>abc</sup>	40.4 <sup>a</sup>	13.4 <sup>b</sup>	2.43 <sup>b</sup>
SHA 29	67.0 <sup>abcde</sup>	85.3 <sup>abc</sup>	79.4 <sup>cde</sup>	1.01 <sup>bc</sup>	11.4 <sup>bcd</sup>	492 <sup>a</sup>	3.4 <sup>d</sup>	33.4 <sup>fgh</sup>	21.6 <sup>cde</sup>	1.58 <sup>cde</sup>
SHA 30	67.2 <sup>abcde</sup>	86.0 <sup>abc</sup>	100.5 <sup>b</sup>	1.24 <sup>b</sup>	12.2 <sup>bc</sup>	486 <sup>a</sup>	10.3 <sup>bc</sup>	36.9 <sup>bcd</sup>	11.3 <sup>f</sup>	0.79 <sup>f</sup>
SHA 32	66.7 <sup>bcd</sup>	85.3 <sup>abc</sup>	90.4 <sup>bcd</sup>	1.23 <sup>b</sup>	11.9 <sup>bc</sup>	545 <sup>a</sup>	9.7 <sup>bc</sup>	32.7 <sup>gh</sup>	14.8 <sup>def</sup>	1.06 <sup>ef</sup>
SHA 35	66.5 <sup>bcd</sup>	85.0 <sup>bc</sup>	79.8 <sup>cde</sup>	1.13 <sup>bc</sup>	11.5 <sup>bcd</sup>	521 <sup>a</sup>	16.4 <sup>a</sup>	36.2 <sup>bcd</sup>	17.7 <sup>def</sup>	1.23 <sup>def</sup>
SHA 37	67.5 <sup>abcde</sup>	86.0 <sup>abc</sup>	90.5 <sup>bcd</sup>	1.30 <sup>ab</sup>	12.6 <sup>b</sup>	595 <sup>a</sup>	13.3 <sup>abc</sup>	38.8 <sup>bcd</sup>	19.9 <sup>def</sup>	1.32 <sup>def</sup>
SHA 41	66.5 <sup>bcd</sup>	85.0 <sup>bc</sup>	78.7 <sup>cde</sup>	0.88 <sup>cd</sup>	9.3 <sup>cd</sup>	388 <sup>a</sup>	15.4 <sup>ab</sup>	36.8 <sup>bcd</sup>	15.0 <sup>def</sup>	1.08 <sup>ef</sup>
SHA42-m	66.8 <sup>abcde</sup>	85.7 <sup>abc</sup>	81.4 <sup>bcd</sup>	1.08 <sup>bc</sup>	9.7 <sup>bcd</sup>	479 <sup>a</sup>	14.3 <sup>ab</sup>	37.2 <sup>bcd</sup>	16.7 <sup>def</sup>	1.19 <sup>def</sup>
Hysun 33	68.3 <sup>a</sup>	87.3 <sup>a</sup>	12.47 <sup>a</sup>	1.49 <sup>a</sup>	16.6 <sup>a</sup>	683 <sup>a</sup>	10.4 <sup>bc</sup>	38.3 <sup>abc</sup>	53.7 <sup>a</sup>	3.62 <sup>a</sup>
Mean	67.1	85.6	86.1	1.10	11.7	513.0	12.2	36.1	21.2	1.51
CV (%)	1.8	1.8	16.3	23.5	19.5	38.9	34.7	6.1	32.6	35.0

\* DF, DM, Pht., SD, HD, S/H, ES, SW and Y/P are days to flowering, days to maturity, plant height, stem diameter, head diameter, no. of seeds/head, empty seed, 1000-seed weight and seed yield/plant, respectively. \* Any means having the same letter(s) are non-significantly different according to Duncan multiple range test at 5% level of significance.

### Phenotypic, genotypic, and environmental variances

Estimation of phenotypic ( $\sigma_{ph}^2$ ), genotypic ( $\sigma_g^2$ ) and environmental variances ( $\sigma_e^2$ ) indicate the genetic components background that reflects the divergent differences among the materials. In this study, phenotypic variances were greater than genotypic ones all characters in both seasons. The values of all variances for all characters in season 2013 were greater than their respective ones in season 2012. On the other hand, in season 2013, the phenotypic ( $\sigma_{ph}^2$ ), genotypic ( $\sigma_g^2$ ) and environmental ( $\sigma_e^2$ ), variances were greater in characters, plant height, head diameter, number of seeds/head, empty seeds% and seed yield/plant, except in genotypic variance in characters empty seeds% and 1000-seeds weight (Table 5). Similar results were reported by Mahmood and Mehdi (2003), Arshad et al. (2007), Zannou et al. (2008), Izquierdo and Aguirrezabal (2008) and Fadlalla (2010) who reported that genotypic variances were smaller than their corresponding phenotypic one for all characters studied in sunflower. In contrast, Sajid (2004) showed that genotypic and phenotypic coefficient of variation was high for all seedling traits.

### Phenotypic and genotypic coefficient of variations, heritability, and genetic advance

Estimates of phenotypic (PCV%) and genotypic (GCV%) coefficient of variations, heritability in broad sense ( $h^2$ ), genetic advance (GA) and genetic advance as percentage of the grand mean (GA%) for the first and second seasons are displayed in Table 6. In this study all the undertaken characters showed greater phenotypic coefficient of variations than their respective genotypic ones. These estimates were greater in season 2013 than those in 2012 for all characters, except for stem diameter, number of seeds/head, seed yield/plant and seed yield/ha. The highest PCV estimate was 52.72%. It was scored for seed yield/plant, while the lowest PCV% was 1.88%. It was scored by days to maturity in season 2012. However, in 2013 the highest PCV was 39.38 recorded for empty seeds, whereas the lowest was 3.33%, recorded for days to maturity. Regarding the heritability ( $h^2$ ) estimates, most of the characters had lower values ( $h^2 < 0.60$ ) in both seasons, except seed yield/plant (g) in season 2012 (0.62). The highest  $h^2$  estimate was 0.62 given by seed yield/plant (g), while the lowest  $h^2$  estimate was 0.01, given by number of seeds/head in season



**Table 4.** Means of 10 characters of 20 sunflower hybrids evaluated at Khordunia in season 2013.

Hybrid	DF	DM	Pht (cm)	SD (cm)	HD (cm)	S/H	ES (%)	SW (g)	Y/P (g)	Yield (t/ha)
SHA 1	59.5 <sup>a</sup>	93.8 <sup>a</sup>	86.4 <sup>a</sup>	1.12 <sup>a</sup>	12.3 <sup>a</sup>	450 <sup>e</sup>	20.6 <sup>a</sup>	59.4 <sup>ab</sup>	22.5 <sup>a</sup>	1.61 <sup>a</sup>
SHA5	57.3 <sup>a</sup>	92.7 <sup>a</sup>	121.6 <sup>a</sup>	1.40 <sup>a</sup>	16.8 <sup>a</sup>	790 <sup>abc</sup>	13.7 <sup>abcd</sup>	53.7 <sup>ab</sup>	33.4 <sup>a</sup>	2.38 <sup>a</sup>
SHA 6	57.7 <sup>a</sup>	92.7 <sup>a</sup>	106.9 <sup>a</sup>	1.05 <sup>a</sup>	11.9 <sup>a</sup>	454 <sup>de</sup>	18.4 <sup>abcd</sup>	52.8 <sup>ab</sup>	21.8 <sup>a</sup>	1.56 <sup>a</sup>
SHA 7	58.0 <sup>a</sup>	91.8 <sup>a</sup>	107.7 <sup>a</sup>	1.03 <sup>a</sup>	12.0 <sup>a</sup>	697 <sup>abcde</sup>	20.4 <sup>ab</sup>	58.0 <sup>ab</sup>	27.6 <sup>a</sup>	1.98 <sup>a</sup>
SHA 11	57.3 <sup>a</sup>	93.7 <sup>a</sup>	130.3 <sup>a</sup>	1.58 <sup>a</sup>	15.0 <sup>a</sup>	831 <sup>abc</sup>	14.7 <sup>abcd</sup>	62.4 <sup>ab</sup>	37.7 <sup>a</sup>	2.70 <sup>a</sup>
SHA 13	60.3 <sup>a</sup>	93.5 <sup>a</sup>	131.0 <sup>a</sup>	1.64 <sup>a</sup>	17.5 <sup>a</sup>	807 <sup>abc</sup>	16.6 <sup>abcd</sup>	58.3 <sup>ab</sup>	38.1 <sup>a</sup>	2.72 <sup>a</sup>
SHA15	59.7 <sup>a</sup>	91.2 <sup>a</sup>	97.0 <sup>a</sup>	1.26 <sup>a</sup>	12.3 <sup>a</sup>	728 <sup>abcde</sup>	12.9 <sup>abcd</sup>	52.5 <sup>b</sup>	33.7 <sup>a</sup>	2.41 <sup>a</sup>
SHA 17	57.0 <sup>a</sup>	92.5 <sup>a</sup>	109.5 <sup>a</sup>	1.26 <sup>a</sup>	14.2 <sup>a</sup>	613 <sup>bcde</sup>	18.9 <sup>abc</sup>	62.8 <sup>ab</sup>	33.2 <sup>a</sup>	2.38 <sup>a</sup>
SHA 18	57.7 <sup>a</sup>	92.2 <sup>a</sup>	110.3 <sup>a</sup>	1.48 <sup>a</sup>	14.0 <sup>a</sup>	681 <sup>abcde</sup>	17.3 <sup>abcd</sup>	58.6 <sup>ab</sup>	33.0 <sup>a</sup>	2.36 <sup>a</sup>
SHA 22	58.8 <sup>a</sup>	95.2 <sup>a</sup>	119.7 <sup>a</sup>	1.42 <sup>a</sup>	15.2 <sup>a</sup>	622 <sup>bcde</sup>	15.8 <sup>abcd</sup>	56.3 <sup>ab</sup>	34.3 <sup>a</sup>	2.45 <sup>a</sup>
SHA 25-1	57.8 <sup>a</sup>	94.0 <sup>a</sup>	95.9 <sup>a</sup>	1.38 <sup>a</sup>	14.3 <sup>a</sup>	433 <sup>e</sup>	20.9 <sup>a</sup>	56.1 <sup>ab</sup>	20.1 <sup>a</sup>	1.44 <sup>a</sup>
SHA 25-2	60.5 <sup>a</sup>	93.5 <sup>a</sup>	102.1 <sup>a</sup>	1.35 <sup>a</sup>	14.8 <sup>a</sup>	870 <sup>ab</sup>	12.1 <sup>bcd</sup>	63.3 <sup>a</sup>	36.0 <sup>a</sup>	2.57 <sup>a</sup>
SHA 29	57.5 <sup>a</sup>	91.3 <sup>a</sup>	122.4 <sup>a</sup>	1.49 <sup>a</sup>	15.4 <sup>a</sup>	954 <sup>a</sup>	13.5 <sup>abcd</sup>	60.4 <sup>ab</sup>	40.5 <sup>a</sup>	2.89 <sup>a</sup>
SHA 30	58.8 <sup>a</sup>	92.8 <sup>a</sup>	138.3 <sup>a</sup>	1.39 <sup>a</sup>	14.0 <sup>a</sup>	708 <sup>abcde</sup>	10.5 <sup>d</sup>	55.8 <sup>ab</sup>	39.1 <sup>a</sup>	2.80 <sup>a</sup>
SHA 32	56.7 <sup>a</sup>	96.0 <sup>a</sup>	113.9 <sup>a</sup>	1.28 <sup>a</sup>	13.6 <sup>a</sup>	775 <sup>abc</sup>	14.0 <sup>abcd</sup>	59.4 <sup>ab</sup>	35.0 <sup>a</sup>	2.50 <sup>a</sup>
SHA 35	58.2 <sup>a</sup>	90.7 <sup>a</sup>	119.8 <sup>a</sup>	1.32 <sup>a</sup>	14.3 <sup>a</sup>	655 <sup>abcde</sup>	18.7 <sup>abcd</sup>	60.4 <sup>ab</sup>	31.0 <sup>a</sup>	2.21 <sup>a</sup>
SHA 37	57.7 <sup>a</sup>	91.8 <sup>a</sup>	120.8 <sup>a</sup>	1.42 <sup>a</sup>	13.8 <sup>a</sup>	640 <sup>abcde</sup>	14.2 <sup>abcd</sup>	52.2 <sup>b</sup>	32.3 <sup>a</sup>	2.31 <sup>a</sup>
SHA 41	57.2 <sup>a</sup>	92.5 <sup>a</sup>	111.0 <sup>a</sup>	1.57 <sup>a</sup>	14.9 <sup>a</sup>	523 <sup>cde</sup>	17.6 <sup>abcd</sup>	61.9 <sup>ab</sup>	25.2 <sup>a</sup>	1.80 <sup>a</sup>
SHA42-m	57.5 <sup>a</sup>	93.3 <sup>a</sup>	88.5 <sup>a</sup>	1.21 <sup>a</sup>	11.5 <sup>a</sup>	765 <sup>abcd</sup>	11.2 <sup>cd</sup>	62.4 <sup>ab</sup>	34.5 <sup>a</sup>	2.47 <sup>a</sup>
Hysun 33	60.3 <sup>a</sup>	94.0 <sup>a</sup>	107.3 <sup>a</sup>	1.40 <sup>a</sup>	14.6 <sup>a</sup>	589 <sup>bcde</sup>	14.5 <sup>abcd</sup>	57.0 <sup>ab</sup>	28.9 <sup>a</sup>	2.07 <sup>a</sup>
Mean	58.3	93.0	112.0	1.35	14.1	679.0	15.80	58.2	31.9	2.28
CV (%)	4.1	3.3	23.9	26.0	22.6	33.2	37.1	13.0	37.1	37.2

\* DF, DM, Pht., SD, HD, S/H, ES, SW and Y/P are days to flowering, days to maturity, plant height, stem diameter, head diameter, no. of seeds/head, empty seed, 1000-seed weight and seed yield/plant, respectively. \* Any means have the same letter(s) are non-significantly different according to Duncan multiple range test at 5% level of significance.

**Table 5.** Phenotypic ( $\delta^2_{ph}$ ), genotypic ( $\delta^2_g$ ), and environmental ( $\delta^2_e$ ) variances for 10 characters of 20 sunflower hybrids evaluated at Khordunia for two seasons 2012 and 2013.

Character	Season 2012			Season 2013		
	( $\delta^2_{Ph}$ )	( $\delta^2_g$ )	( $\delta^2_e$ )	( $\delta^2_{Ph}$ )	( $\delta^2_g$ )	( $\delta^2_e$ )
Days to 50% flow	1.73	0.21	1.51	6.13	0.50	5.63
Days to maturity	2.60	0.35	2.25	9.57	0.18	9.39
Plant height (cm)	305.60	108.89	196.67	787.10	73.58	713.52
Stem diameter	0.09	0.02	0.07	0.13	0.01	0.12
Head diameter (cm)	7.08	1.91	5.17	11.00	0.82	10.18
No. of seed / head	40316	566	39751	62864	12070	50793
Empty seed (%)	24.68	6.75	17.92	38.82	4.35	34.47
1000-seed weight (g)	9.18	4.28	4.90	66.79	9.55	57.24
Seed yield/plant (g)	124.80	77.14	47.65	151.18	10.70	140.48
Seed yield (t/ha)	0.62	0.35	0.28	0.77	0.05	0.72

2012. However, in season 2013, the highest was 0.19, given by number of seeds/head, while the lowest one was 0.02 given by days to maturity (Table 8). Like the trend of the heritability estimate, the values of the expected genetic advance under selection (GA%) changed over seasons. GA% value scored for seed

yield/plant (g) was 52.78% as highest score in 2012, but it scored 1.49% in 2013, respectively. The highest estimate of GA% was 6.40%. It was recorded for number of seeds/head, whereas the lowest one 0.02% and scored by days to maturity in 2013 (Table 8). The rest of the characters showed low and staple values in the

**Table 6.** The phenotypic (PCV %), genotypic (GCV %) coefficient of variations, heritability (h<sup>2</sup>) estimates, genetic advance (GA) and genetic advance as percentage of the mean (GA%) in 10 characters of 20 sun flower hybrids evaluated at Khordunia for two seasons 2012 and 2013.

Characters	Season 2012					Season 2013				
	PCV (%)	GCV (%)	h <sup>2</sup>	GA	GA (%)	PCV (%)	GCV (%)	h <sup>2</sup>	GA	GA (%)
Days to 50% flowering	1.96	0.68	0.12	0.12	0.17	4.25	1.21	0.08	0.12	0.20
Days to maturity	1.88	0.69	0.13	0.16	0.19	3.33	0.46	0.02	0.02	0.02
Plant height (cm)	20.31	12.12	0.38	7.66	8.90	25.15	7.66	0.09	1.65	1.47
Stem diameter	27.18	13.58	0.25	0.08	6.98	26.73	6.15	0.05	0.01	0.67
Head diameter (cm)	22.85	11.88	0.27	0.77	6.62	23.50	6.40	0.07	0.14	0.98
No. of seed / head	39.17	4.64	0.01	0.69	0.13	36.91	16.18	0.19	43.46	6.40
Empty seed (%)	4.66	21.27	0.27	1.47	11.99	39.38	13.18	0.11	0.48	3.04
1000-seed weight (g)	8.38	5.72	0.47	1.98	5.49	14.05	5.31	0.14	0.91	1.56
Seed yield/plant (g)	52.72	41.45	0.62	11.18	52.78	38.53	10.25	0.07	0.48	1.49
Seed yield (t/ha)	52.40	39.03	0.55	0.67	44.62	38.53	10.23	0.07	0.03	1.48

different seasons. In agreement, Mamta et al. (2017b) stated that PVC was slightly high than GCV in sunflower hybrids. They also reported that heritability was high for seed yield/plant. Similar results were reported by Farooq et al. (2006) and Fadlalla (2010). On the other hand, Monica and Lauren (2003) expressed that heritability was lower in inbreeding species. Similar results for genetic advance were reported by Mamta et al. (2017a) who stated that genetic advance as percent from mean was high for seed yield/plant followed by seed weight.

## Conclusion

From the results of this study it concluded that there were significant differences among the undertaken hybrids. The phenotypic coefficients of variation values were greater than their corresponding genotypic ones. Heritability values were low for all characters, except for seed yield/plant. Genetic advance as percentage from the overall mean values were greater in 2012 than their corresponding ones in 2013. More investigation should be done for some promising hybrids. They were SHA258-2, SHA29 and SHA30.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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## Full Length Research Paper

## Prospection and production of Solanaceae species resistant to the root knot nematode

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Plant species of Solanaceae family are affected by numerous pathogens worldwide. Among them is the root-knot nematode which hinders the establishment of crops in the field, reducing their production capacity. This work aims to select Solanaceae species tolerant or resistant to root-knot nematode under the climatic conditions of Fortaleza, Ceará. Two experiments were performed. For both, the followings were evaluated: 'Santa Clara' tomato, hybrid 'T92', cherry 'Carolina' and 'Laranja'; gilo 'Comprido Grande Rio'; eggplant 'Comprida Roxa'; Pepper 'Cayenne'; and chili 'All Big'. Each treatment had six replicates. In the first trial, the number of twigs and egg masses per root, the aboveground height and fresh root weight at 60 days after inoculation (DAI) were evaluated. In the second trial, the reproduction factor of the 130 DAI, total number and weight of fruits, as well as productivity were evaluated. The tomato 'Santa Clara', 'Carolina', 'Laranja', the eggplant and gilo had the major infestations of *Meloidogyne incognita*; however, only tomato 'Santa Clara' showed decreased productivity among all the cultivars. On the other hand, in the hybrid 'T92' and chili, there was no nematode reproduction, no egg mass and reproduction factor was zero. Thus, it is concluded that Pepper 'Cayenne', 'All big' Chili 'All Big' and 'T92' hybrid tomato are immune to *Meloidogyne incognita*, so they can be tested in resistant rootstock trials for susceptible commercial tomato plants.

**Key words:** Root-knot nematode, *Solanum lycopersicum*, *Solanum melongena*, *Capsicum annuum*, *Solanum gilo*, *Meloidogyne incognita*.

### INTRODUCTION

Vegetable production in Brazil has great economic, social and nutritional importance. Due to new technologies and developed cultivation techniques the production of vegetables increased by 33% and their productivity by

38% (Carvalho, 2013). According to the Brazilian Association of Seeds and Seedlings Commerce (Associação Brasileira do Comércio de Sementes e Mudas - ABCSEM, 2014), more than 94 billion reais were

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moved in 2012 in the agricultural chain that involves the vegetable production segment. In total, approximately 20 million tons of -vegetable crops were produced. Socially, the production of this group of plants favored employability in Brazil, since only in 2012, about 2 million people were directly employed in this type of activity (ABCSEM, 2014).

In addition to the above, the production of vegetables promotes human health, by providing a nutritionally diet rich in fiber, vitamins and minerals (Machado, 2008), which are key components to a healthy and balanced diet.

Despite the wide applicability, consumption and productivity of the species of the Solanaceae family, edaphoclimatic factors such as temperature, light, wind, nutrient concentration and phytosanitary problems may adversely affect the establishment of these crops in the field, thus reducing their production capacity and making them more expensive to purchase by consumers. Some of these factors are often addressed through the planning and selection of areas for cultivation. However, issues relating to phytosanitary problems such as the presence of unwanted organisms cannot always be predicted, given the impossibility of having full control of the entrance of a given pathogen in the production area. Among the many organizations that affect the production of tomato, great emphasis can be given to the root-knot nematode (*Meloidogyne* spp). This pathogen belongs to the genus *Meloidogyne* (Goeldi - 1887), which has four main species: *Meloidogyne incognita*, *Meloidogyne javanica*, *Meloidogyne arenaria* and *Meloidogyne hapla* with widespread distribution throughout the world. It composes the nematode group of greater importance for vegetables (Pimenta and Carneiro, 2005), affecting the vast majority of cultivated solanaceae (Pernezny et al., 2003).

Cultural control methods including crop rotation, fallowing, the use of pesticides, and solarization are often employed in the management of root-knot nematodes. Nevertheless, although such techniques present some efficiency, some have operational and economic drawbacks that make it difficult and often hamper their implementation by producers. For example, the use of solarization limits the production area for crops, which is of particular importance for small farmers that cannot grow in the area being solarized, or cannot solarize its infested area because of the rainy season (Baptista et al., 2007).

Furthermore, the use of chemical pesticides is often associated with negative environmental impacts and increased production costs. The development and deployment of nematode-resistant cultivars is an efficient, cost-effective and environmentally-friendly option for nematode management. In previous reports, the use of host plant resistance decreased nematode reproduction with effectivity comparable to chemical control (Cook and Evans, 1987; Starr and Roberts, 2004).

Given the above, the objective of this study is to identify species and cultivars of the Solanaceae family that have tolerance or resistance characteristics to most frequent worldwide species of root-knot nematode, *M. incognita*.

## MATERIALS AND METHODS

For the development of this work two trials were conducted. The work completion period was from September 2014 to February 2015. For both tests, the design was completely randomized in a factorial 2 x 8 (two ground conditions, infested and uninfested with root-knot nematodes, and eight species / cultivars: 1) Tomato 'Santa Clara' from Santa Cruz group (susceptible to root-knot nematode); 2) Hybrid tomato 'T92'; 3) Cherry tomato (*S. lycopersicum* var *cerasiforme*) 'Carolina'; 4) Cherry tomato 'Laranja'; 5) Gilo (*Solanum gilo* Raddi) 'Comprido Grande Rio'; 6) Eggplant (*Solanum melongena* L.) 'Comprida Roxa'; 7) Chili (*Capsicum annuum*) 'Cayenne'; and 8) Chili 'All Big' (*C. annuum* var *annuum* L.). One plant was cultivated per pot and each species was replicated six times under each of the two treatments.

The seedlings were grown in polyethylene trays with 162 cells filled with substrate produced from organic compound and vermiculite in the proportion of 9:1. The physical and chemical composition of the soil used for filling the trays is shown in Table 1. After sowing, the trays were allocated in a covered structure with 30% shading, where they remained for about 30 days until they were transplanted to the final location for production. The physical and chemical characteristics of the compound used for filling the trays and pots cultivation of Solanaceae are as follows (Table 1).

When the seedlings had three to four true leaves, they were transplanted to the pots with 8 L capacity, and cultivated until harvest. Fifteen days after transplanting the plant growth was supported by vertical staking according to the protocol by Guimarães et al. (2007, 2008), using polyethylene strings. Irrigation was carried out twice a day with a system characterized as micro-sprinkler type. Ridge planting was performed 30 days after transplanting. Other cultural practices such as thinning and hoeing were performed as needed following the recommendations of Filgueira (2008).

To obtain the inoculum, the nematode was cultured on the roots of cherry tomato 'Carolina', (*S. lycopersicum* var *cerasiforme*) grown on site. 60 days after inoculation the infested roots were removed from the soil, carefully washed, wrapped in plastic bags and taken to the Phytopathology Laboratory in Universidade Federal do Ceará. Eggs of the root-knot nematode population were extracted using Coolen and D'Herde's (1972) technique and approximately 5,000 eggs were injected into the soil of each plant at a depth of 3 cm two days after transplanting.

For evaluation of the results, the following parameters were considered: number of galls (NG); galls index (GI); mass number of external eggs (ME); egg mass index (EMI). The GI and EMG in the roots were represented by a scale of 1 to 5, according to Taylor and Sasser (1978), modified by Hadisoeganda and Sasser (1982). After 130 days of inoculation, total fresh roots were harvested to determine the reproduction factor (RF) and reproduction index (RI) as an estimate of the reproductive capacity of the nematodes:  $RF = Fp/Ip$ , where  $Fp$  = final nematode population and  $Ip$  = initial nematode population. In this parameter, plant species are classified as immune (RF = 0), resistant (RF < 1.0) and susceptible (RF > 1.0) (Oostenbrink, 1966).

The rendering index (RI) was obtained by the ratio between the average number of eggs per gram of root plants of each treatment and the average number of eggs per gram of root tomato 'Santa Clara' (pattern) multiplied by 100 (Taylor, 1967). The classification corresponds to susceptible (S) when RI > 50%, slightly resistant (SR) if RI 26 to 50%, moderately resistant (MR) if RI from 11 to

**Table 1.** Physical and chemical composition of the soil used for the production of Solanaceae seedlings.

Chemical characteristics	Value	Unit	Puller
pH	6.80	mg dm <sup>-3</sup>	In H <sub>2</sub> O, KCl and CaCl <sub>2</sub> – Ratio 1:2:5
P	50.40	mg dm <sup>-3</sup>	Puller Mehlich 1
K	57.00	cmol <sub>c</sub> dm <sup>-3</sup>	Puller Mehlich 1
Ca <sup>2+</sup>	20.30	cmol <sub>c</sub> dm <sup>-3</sup>	KCl – 1 mol L <sup>-1</sup>
Mg <sup>2+</sup>	6.90	cmol <sub>c</sub> dm <sup>-3</sup>	KCl – 1 mol L <sup>-1</sup>
Al <sup>3+</sup>	0.00	cmol <sub>c</sub> dm <sup>-3</sup>	KCl – 1 mol L <sup>-1</sup>
H+AL	1.49	cmol <sub>c</sub> dm <sup>-3</sup>	Acetate calcium 0.5 mol L <sup>-1</sup> - pH 7.0
SB	27.35	cmol <sub>c</sub> dm <sup>-3</sup>	Exchangeable basic sum
CTC (t)	27.35	cmol <sub>c</sub> dm <sup>-3</sup>	Effective capacity of cation exchange
CTC (T)	28.84	%	Capacity of cation exchange to the pH 7.0
V	95.00	%	Base saturation index
M	0.00	%	Aluminum saturation index

**Table 2.** Susceptibility classification of the plants according to the number of root-knot and egg mass (Taylor and Sasser, 1978) modified by Hadisoeganda and Sasser (1982).

Number of root-knot and egg masses	Index number	GI/EMI	Plants classification
0	0	0.0-1.0	Highly resistant
01-02	1	1.1-3.0	Very resistant
03-10	2	3.1-3.5	Moderately resistant
11-30	3	3.6-4.0	Lightly resistant
31-100	4	4.1-5.0	Susceptible
>100	5	-	-

25%, very resistant (VR) if RI from 1 to 10%, highly resistant (HR) if RI < 1.0% and immune (I) when there is no reproduction (Table 2).

To characterize the productivity of Solanaceae species, periodic harvests of fruits were performed, to assess the number of fruits produced and as well as total fruit fresh weights.

Statistical analysis was performed on collected data using the Scott - knott. The obtained data were submitted to analysis of variance by the software Sisvar (Ferreira, 2010) and, once difference between the treatments was observed, they were compared by the Scott-Knott group test's at 5% level of significance.

## RESULTS AND DISCUSSION

After the evaluation of the plants used in the determination of susceptibility (Table 2) it can be seen that tomatoes 'Laranja' and 'Carolina'; eggplant 'Comprida Roxa' an gilo 'Comprido Grande Rio' - showed the highest average number of galls and number of egg masses 60 days after inoculation (DAI) as well as the reproduction factor (RF) in the 130 DAI of the plants.

Such results indicate that the susceptibility of the species to *M. incognita*, gilo (*S. gilo*) IR showed less than 50% indicating a slight resistance (SR) to the nematode (Table 3). The susceptibility of other tomato cultivars to more

widespread nematodes in the middle of agricultural production (*M. javanica*, *M. incognita*, *M. arenaria* and *M. enterolobii*) has been reported by other researchers in other studies (Talavera et al., 2009; Bitencourt and Silva, 2010).

In comparison with other plant species, hybrid tomato 'T92', the pepper 'All Big and chili 'Cayenne' are considered immune to nematode infection (Table 3) and immune (I) as FR and RI were zero, according to the criteria of Oostenbrink (1966) and Taylor (1967), respectively. This result was expected for the long life hybrid tomato 'T-92' that commercially has, as one of its main features, resistance to nematode galls *M. incognita*, *M. arenaria* and *M. javanica*. The results obtained with the chili 'Cayenne' were similar to those found by other authors for other species within the genus *Capsicum*. Carneiro et al. (2000) observe two cultivars of sweet chili (*C. annuum*) resistant to *M. javanica* and *M. arenaria*. Recently, Rosa (2013) observed immunity to *M. javanica* in different cultivars of chili.

Pepper cultivar "All Big" showed immunity to root-knot nematode infection which was comparable to previous studies involving a variety of *Capsicum* spp. (Carnerio et al., 2000). In general, what could be seen from the

**Table 3.** Evaluation of parameters: number of galls, egg mass, number of eggs and reproduction factor of *Meloidogyne incognita*.

Species/Cultivars	Number of root-knot <sup>1,2</sup>	GI	Number of egg mass	EMI	Number of eggs <sup>1</sup>	Reproduction factor <sup>1</sup>	RI
Tomato 'Laranja'	495	5	77.67	4	250,667	50.13	89
Tomato 'Carolina'	500	5	75.83	4	444,000	88.80	96
Tomato 'T92'	0	0	0.00	0	0	0	0
Chilli 'All Big'	0	0	0.00	0	0	0	0
Pepper 'Cayenne'	1	1	0.00	0	0	0	0
Eggplant 'Comprida Roxa '	500	5	78.50	4	1,538,667	307	145
Gilo 'Comprido Grande Rio'	500	5	72.00	4	464,000	93	47
Tomato 'Santa Clara'	500	5	72.83	4	535,000	107	100

<sup>1</sup>Average of six replicates; accounted for up to a maximum of 500 root-knot. GI = Galls index of root-knot nematode; EMI = Egg mass index; RI = reproduction index.

various research results already published and the information generated from this study is that the genetic diversity among different cultivars of pepper and chili available can be considered the main factor responsible for resistance or susceptibility of plants to the species of the root-knot nematodes, since there are wide varieties that exhibit resistance to more than one kind of nematode; simple resistance, being resistant to one kind of nematode; and complete absence of resistance (susceptibility).

For the observed characteristic of the shoot height, no difference was observed between the plants grown under different conditions of soil infestation for cultivars of tomato cherry group, hybrid tomato, pepper, chili and eggplant. However, for tomato 'Santa Clara' and gilo, differences were found between the plants grown in soil infested with the pathogen or not. For the tomato 'Santa Clara' there was greater shoot height for plants grown in soil without infestation, while gilo (*S. gilo*) showed higher shoot values for plants grown in an environment infested by the pathogen (Table 4).

With the exception of plant cultivars "T92", "Comprido Grande Rio" and "Comprida Roxa", all species demonstrated increases in root mass during cultivation in nematode-infested soil. For the characteristic evaluated root mass, except for the tomato hybrid 'T92' and eggplant's cultivar that had lower average values for plants grown in an environment infested with nematodes, and the gilo's cultivar (*S. gilo*), which showed no significant difference in mass between plants grown in soil infested by nematode or not, all other cultivars of each evaluated species produced higher average of root mass when grown in infested soil with the study pathogen (Table 5).

In contrast to Sharma et al. (2005), increased shoot height and root mass was observed in pepper variety "Cayenne". The highest fresh weight of roots observed, generally, in infested treatments is related mainly with the thickening of galls on nematodes infestation, which

causes certain increase in mass at the site of infestation. The galls on each root system showed that this accumulation contributes to a significant increase in fresh root mass compared to uninfested plants.

For production characteristics, for all other species and cultivars no differences in yield were observed between the two soils used for cultivation, both infested and non-infested. On the other hand, in the tomato 'Santa Clara' it was observed fewer fruit number, total fresh mass weight and mass average of fruit per plant grown in the soil infested with nematodes (Table 5).

Root-knot nematodes are often associated with high reductions in the productivity of tomato in Brazil. For the cherry tomato plants 'Carolina' and 'Laranja', eggplant, gilo pepper and chili, there were no reductions in the production of plants grown in soil infested when compared with the plants grown in uninfested soil. An exception was observed for tomato 'Santa Clara', which showed high susceptibility to the pathogen.

As for productivity, it is observed in Figure 1 that, with the exception of tomato 'Santa Clara', in which it was evident the negative influence of infestation by root-knot nematode, the other species and cultivars there was no difference between plants propagated in nematode-infested or non-infested soil types. This is suggestive of a measure of tolerance.

According to Sikota and Fernandez (2005), the nematodes of the genus *Meloidogyne* have been one of the main soil pathogens that commonly affect vegetable crops. In general, the major symptoms that appear in the shoot of plants are wilts caused by the reduction of the capacity of water absorption by the roots. In more severe infestation conditions, it was observed that nutritional deficiency was caused mainly by reducing the transport of essential nutrients in plants, regulated by the root system damaged. The galls are important because they directly compromise the physiology of the plant, causing, in general, reduction in crop production and reduction in quality of the final product (Abad et al., 2009).

**Table 4.** Productive characters of species and cultivars of Solanaceae grown in soil infested or not with root-knot nematode.

Species/cultivars	Not infested soil	Infested soil	C.V. (%)
<b>Number of total fruits</b>			
Cherry tomato 'Laranja'	0.80 <sup>ns</sup>	0.79 <sup>ns</sup>	19.59
Cherry tomato 'Carolina'	1.08 <sup>ns</sup>	1.09 <sup>ns</sup>	6.17
Hybrid tomato 'T92'	1.36 <sup>ns</sup>	1.28 <sup>ns</sup>	20.50
Chili 'All Big'	0.48 <sup>ns</sup>	0.50 <sup>ns</sup>	23.40
Pepper 'Cayenne'	0.42 <sup>ns</sup>	0.53 <sup>ns</sup>	27.62
Eggplant 'Comprida Roxa '	1.16 <sup>ns</sup>	1.10 <sup>ns</sup>	19.53
Gilo 'Comprido Grande Rio'	0.95 <sup>b</sup>	1.21 <sup>a</sup>	9.91
Tomato 'Santa Clara'	1.22 <sup>a</sup>	1.10 <sup>b</sup>	7.72
<b>Root mass (g. plant<sup>-1</sup>)</b>			
Cherry tomato 'Laranja'	3.58 <sup>b</sup>	14.63 <sup>a</sup>	24.46
Cherry tomato 'Carolina'	26.08 <sup>b</sup>	37.47 <sup>a</sup>	21.03
Hybrid tomato 'T92'	24.15 <sup>a</sup>	14.02 <sup>b</sup>	33.91
Chili 'All Big'	17.90 <sup>b</sup>	32.77 <sup>a</sup>	19.64
Pepper 'Cayenne'	2.58 <sup>b</sup>	7.30 <sup>a</sup>	25.02
Eggplant 'Comprida Roxa '	48.08 <sup>a</sup>	31.02 <sup>b</sup>	24.24
Gilo 'Comprido Grande Rio'	85.10 <sup>ns</sup>	68.92 <sup>ns</sup>	29.04
Tomato 'Santa Clara'	14.28 <sup>b</sup>	38.95 <sup>a</sup>	34.08

\*Means followed by the same lowercase letters do not differ at the level of 5% probability by Scott-Knott test's. ns- not significant.

**Table 5.** Productive characters of species and cultivars of Solanaceae grown in soil infested or not with root-knot nematode.

Species/Cultivars	Not infested soil	Infested soil	C.V. (%)
Cherry tomato 'Laranja'	61 <sup>ns</sup>	51 <sup>ns</sup>	25.21
Cherry tomato 'Carolina'	38 <sup>ns</sup>	36 <sup>ns</sup>	17.26
Hybrid tomato 'T92'	25 <sup>ns</sup>	27 <sup>ns</sup>	10.19
Chili 'All Big'	21 <sup>ns</sup>	26 <sup>ns</sup>	18.45
Pepper 'Cayenne'	28 <sup>ns</sup>	27 <sup>ns</sup>	16.72
Eggplant 'Comprida Roxa '	24.00 <sup>ns</sup>	25.50 <sup>ns</sup>	9.35
Gilo 'Comprido Grande Rio'	7 <sup>ns</sup>	5 <sup>ns</sup>	36.90
Tomato 'Santa Clara'	24 <sup>a</sup>	8 <sup>b</sup>	16.58
<b>Pasta fresh total fruits (kg.plant<sup>-1</sup>)</b>			
Cherry tomato 'Laranja'	0.65 <sup>ns</sup>	0.58 <sup>ns</sup>	28.87
Cherry tomato 'Carolina'	0.50 <sup>ns</sup>	0.42 <sup>ns</sup>	25.96
Hybrid tomato 'T92'	2.14 <sup>ns</sup>	1.80 <sup>ns</sup>	20.12
Chili 'All Big'	0.86 <sup>ns</sup>	0.93 <sup>ns</sup>	13.94
Pepper 'Cayenne'	0.09 <sup>ns</sup>	0.10 <sup>ns</sup>	30.61
Eggplant 'Comprida Roxa '	2.92 <sup>ns</sup>	3.28 <sup>ns</sup>	30.42
Gilo 'Comprido Grande Rio'	0.23 <sup>ns</sup>	0.16 <sup>ns</sup>	32.53
Tomato 'Santa Clara'	2.75 <sup>a</sup>	0.64 <sup>b</sup>	26.64

\*Means followed by the same lowercase letters do not differ at the level of 5% probability by Scott-Knott test's. ns- not significant.

For the other species and cultivars studied, both production and productivity parameters showed a good

production similarly in both soils infested and uninfested. This suggests that there is a tolerance of those to



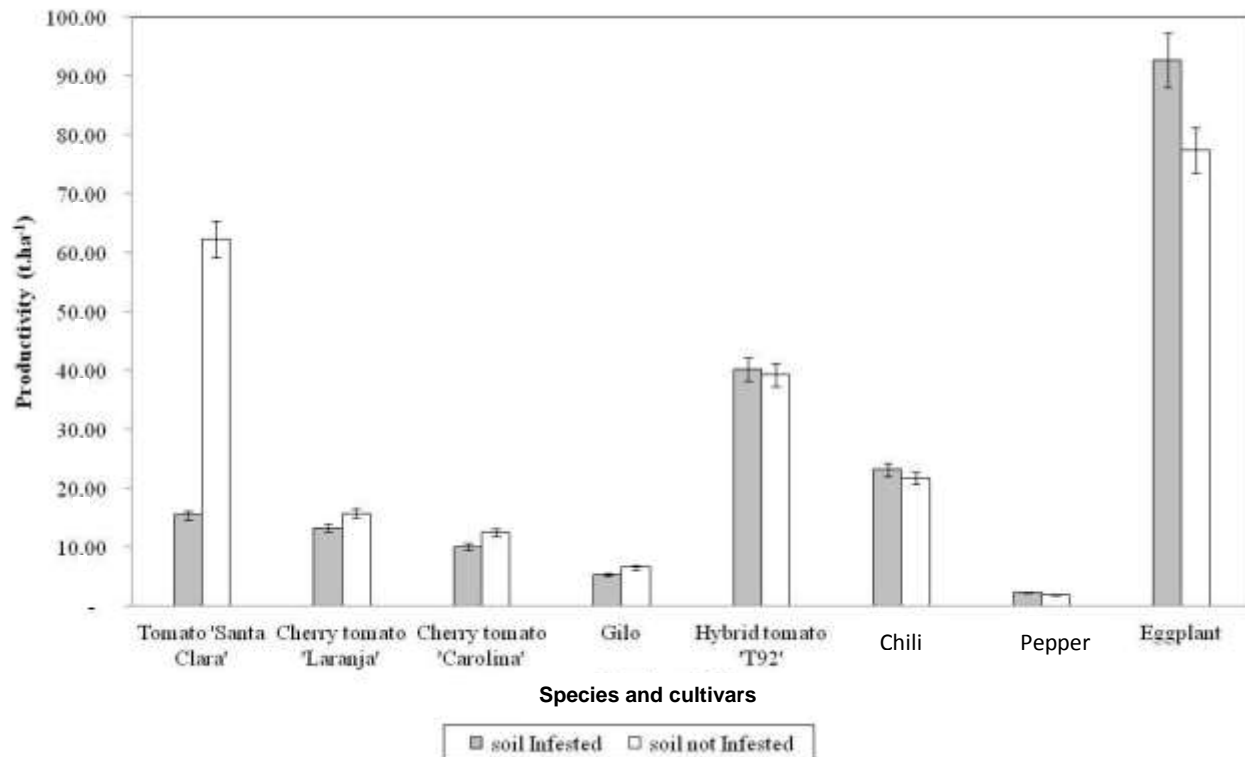


Figure 1. Productivity of species and cultivars of Solanaceae grown in nematode infested or non-infested soil.

infestation by nematode; this is consistent with the non-observation of difference between those physiological factors studied between treatments.

## Conclusion

The Pepper 'Cayenne', the Chili 'All Big' and hybrid tomato 'T92' are immune to *M. incognita*. Therefore, they can be used in subsequent studies of resistant – control cultivars using other plant varieties.

The cherry tomato "Laranja" and "Carolina", eggplant 'Comprida Roxa' and Gilo 'Comprido Grande Rio' are tolerant to *M. incognita*, since they were able to produce despite the high infestation. Physiological analysis of the plants must be performed during the production stage, preferably after the start of the fruit harvest, to further evaluate the relationship between the susceptibility to the pathogen and production of the plant.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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*Full Length Research Paper*

## Effects of market price, cultivating area and price regulation on cotton production in China

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**This study examines the quantitative effects of market price in cotton producing areas of China. It also analyzes the qualitative effects of price regulation on cotton production. Secondary time series data were collected from National Bureau of Statistics in China, between 1990 and 2013. Calculation of the growth rate of cotton in production was done using linear trend model and multi-regression model to analyze the correlations between production, area and market price. The results of regression between the dependent variable (cotton production) and independent variables such as the previous year's area, current year's area and the previous year's market price showed that the R-Square and adjusted R-Square values are 0.89 and 0.87, respectively, and the t-statistics of all independent variables rejected null hypothesis of no correlation at the 1% significant level. This infers that the market price and cultivating area of cotton crop has a highly significant relationship with the production. What's more, the coefficients of current year's area and previous year's market price are higher than 0, denoting a positive impact on production. However, coefficient of previous year's area is smaller than 0, implicating a negative influence on cotton production. In addition, the value of dry weight calculated is 1.94, which means no series auto-correlation exists. Despite this, it can be concluded from the regression results is that cultivating area and market price have time lag impacts on cotton production. Furthermore, price regulation has indirect positive impacts on cotton production.**

**Key words:** Cotton productivity, growth rate model, multi-regression.

### INTRODUCTION

Cotton is one of the most important non-food cash crops and it is an important source of foreign currencies earning

in China. Four major countries like China, India, USA and Pakistan produce cotton; however, China ranks 1st

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#Equal Contribution, Order of Authorship was selected by coin through toss

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among these four countries (Wang et al., 2009). Twenty four provinces out of thirty one provinces produce cotton in China; about 300 million Chinese are involved directly or indirectly in cotton production activities (National Bureau of Statistics in China, 2008). From 1978 to 1984, cotton production of China increased steadily and reached a significant level of 6.26 million metric tons in 1984 (UNEP, 2002). Later the production increased from 4.1 to 5.7 million tons. However, it decreased to 3.8 million tons in 1999. Cotton production value accounted for 13% in the added value of the economy and contributed to the agricultural gross domestic product by 0.6% (Stat, 2015). In China, the area used for cotton plantation is about 4,219 thousand hectares, with a production of about 616.0 million tons of cotton in 2014. Cotton consumption in China was expected to remain stable at a level of around 8 million tons, which accounts for one third of the global consumption. In 2015, the cotton consumption in China surpassed that of neighboring countries like India and Pakistan (ICAC, 2015). In addition, cotton provides raw material for textile industries, which is the biggest sector in China and which also offers employment for about 10 million people (China National Garment Association, 2014; Fashion United, 2013). Therefore, China has positively followed the trend of technology advancement and upgraded its textile industries (Studwell, 2013). The rise in China's exports over the past 10 years has contributed to some of the complexities in its import (Wang and Wei, 2008; Amity and Freund, 2010). China has three major cotton growing areas (Yang and Cui, 2010), which are Xinjiang, Yangtze River Basin (Which includes Jiangsu and Hubei province) and Huang-He districts (which includes Hubei, Henan, and Shandong). Cotton is, also a labor intensive crop which provides work opportunities for rural people, especially for women (picking) (Lokhande, 1995).

Cotton is cultivated in almost every country of the world. The major producers, consumers and exporters are China, USA, the previous Soviet Union, India, and Pakistan. It accounts for more than 20% of world production and nearly 20% of world consumption. The development of China has had a significant impact on cotton market of the world. In 1997, China imported 783 thousand metric tons of cotton which was 13.6% of the total world import, but since 1999, China has become a net-exporter at a trading level of 330 thousand tons (UNEP, 2002). Before 1993, cotton domestic price in China was very low, around 32.91 US cents/pound. Thus, the Chinese government increased the cotton purchasing price to 47% in 1994. Again in 1995, Chinese government increased the purchasing price of cotton by 29%, which instigated a massive increase in domestic cotton price, then the purchasing cotton price in domestic market went up to 76.79 US cents/pound. In 1999, Chinese government decided to make cotton price free. Subsequently, the domestic cotton price dropped to 42.02 US cents/pound, while in 2000 the price went from 54.72 to 63.47 US

cents/pound (UNEP, 2002).

### The overview of Chinese cotton policies

As cotton is an important economic crop in China, the production and price of cotton have significant effects on the agricultural development of the country. This has made the Chinese government implement many policies to keep the cotton industry stable. From 1949 to 1954, the Chinese government has implemented free marketing policy and the official has also used many methods such as cotton cooperation and order advancement to make the cotton industry stable. From 1954 to 1985, the Chinese government implemented the cotton reserving plan bill. During this period only the government can purchase or sell cotton, and private cotton business was illegal. Later from 1985 to 1999, in order to keep up with the change of agriculture industry structure and develop the commodity economy in agriculture, Chinese government changed unified purchase and sale policy into contracts transaction. After 1999, China has started the market reform with the development. China's cotton policy transited in market-oriented systems. However, in 2010, the price of cotton rose sharply in the world and domestic market. In order to stabilize the price in domestic market, in March 2011, the Chinese government enacted price regulation policy of cotton to protect the farmers' cotton cultivation profits and consequently achieve the target of cotton production. The target price as the form of price regulation lasted for 3 years, from 2011 to 2013. In 2011, target price was 19,800 yuan/ton and in 2012 and 2013, it was 20,400 yuan/ton. China also adopted price distinguished policies for staple food grains, such as rice, wheat and corn. The policy aimed at rice and wheat was described as a minimum procurement price policy, and a policy of complete or virtual self-sufficiency in which these grains can be predictable for the foreseeable future (Zhu, 2011; Gale, 2013). When the price of cotton floated dramatically, the government would carry out price regulation policy. For example, when the market price declined to a large extent, the government will purchase the cotton from farmers at the target price. Currently, the target price policy is merely implemented in Xinjiang, which serves as a policy testing area.

In 2014, China's policy makers gradually started to diminish the market value of cotton, which was vended from intervention stockpiles. Because of the high degree of price uncertainty shaped through the scope of China's stock. The gradually reducing prices due to the standard of China's stock made itself the main significant factor for policy changes including going back to the price support and offering a lower level of support policy for cotton. However the sales from stock was diverse in different ways. During 2009-2010, stock auctions were assumed as target price increases, nonetheless during 2012-2013

the planning of stock sales was combined with efforts to support price. Stock product was offered for sale with the value of 5-10 percent lower than the price of the product purchased, although the edge price was 60% lower than the purchased price. By the acknowledged price floor, the auctions aimed at cotton reserve were open to public mechanisms of current discussions between textile industry and government. The government restrictions on cotton imports, which increased the price of stock cotton and permitted the government to connect the delivery of import quota to purchases at the stock sales, is an essential module of the planning of reserve auctions prices. During 2012-2013 the sum of stock auctions was greater than 6.5 million tons, nevertheless not sufficient to stop the reserve from increasing to an unparalleled level.

During the period of 2011-2013, China's interference with purchasing cotton at the support price level was extended from previous practices, but such interventions was not fully preoccupied beforehand. The disruption from 2012 to 13 was that the stockpile of China's cotton planning was converted to piece support organization, with reserve consciously increasing to unparalleled scope and flattering the possible source of world market unpredictability rather than gadget of price predictability. In 2014, Chinese government lowered the auction price for interference reserve, gesturing to decline the price support (Mcdoland 2015). Following are the researchers working on cotton price related topics in China. Du and Min (2010) carried out research on Investigation and Analysis on Production and Market Behavior of Cotton Growers in China; Gale & Fared(2013) performed a topic on Growth and Evolution in China's Agricultural Support Policies; FAO(2013) implemented a research about The United Nations and International Cotton Advisory Committee; Hua Liu(2013) expressed his insight on Thoughts of the Implementation of Macro Regulatory Policy for Cotton and CNCRC's Development Strategy; Ge et al(2010) did some research on Cotton market integration and the impact of China's new exchange rate regime; Huang et al (2013) shared his investigation on The subsidization of farming households in China's agriculture; Cater et al(2012) made explorations on Advances in Chinese Agriculture and its Global Implications; ITMF(2014) conducted Cotton Contamination Surveys in 2003, 2005, 2007, 2009, 2011, 2013 & 2014.

The allocation of the rest parts is as follows: Section 2 outlines the methodologies of the study; Section 3 displays the empirical results and discussion; Section 4 analyses the effects of price regulation on cotton production; Section 5 lists out the conclusion.

## METHODOLOGY

The study covers a period of 24 years from 1990 to 2013. Data were collected from various government publications from the National Bureau of Statistics in China. All variables for multi-

regression analysis are in natural logarithms.

## Growth rate model

Data of time series of cotton production, area and yield were analyzed to estimate their growth rate by subsequence of growth model (Koondhar et al., 2016; Magsi 2012), which is as follows:

$$g_x = \left( \frac{X_T}{X_0} \right)^{1/T} - 1$$

where  $g_x$  = geometric average growth rate,  $X_0$  = initial value of variable X,  $X_T$  = final value of variable X, 0 = base year, and T = final year.

## Multi-regression model

Test of assumptions is an important task which a researcher utilizes in a multi-regression model. Serious assumption violations can result from biased estimates of relationships, over or under-confident estimates of the precision of regression coefficients (that is, biased standard errors), and untrustworthy confidence intervals and significance tests (Chatterjee and Hadi, 2012; Cohen et al., 2003). To figure out the relations between cotton production, market price and cultivating area, multi-regression model was used as the method of analyses. The following are the different equation design hypothesis based on general economic regularities:

$$\text{Equation hypothesis 1: } \ln Y_t = \alpha_0 + \beta_1 \ln A_t + \beta_2 \ln P_t + \varepsilon \quad (1)$$

where  $Y_t$  = current year's cotton production,  $\alpha_0$  = intercept,  $A_t$  = current year's cultivating area,  $\beta_1$  = coefficient of current year's cultivating area, which is expected to be above 0,  $P_t$  = current year's market price,  $\beta_2$  = coefficient of current year's market price, which is expected to be above 0, and  $\varepsilon$  = error term.

$$\text{Equation hypothesis 2: } \ln P_t = C_1 + \beta_1 \ln P_{t-1} + \varepsilon \quad (2)$$

where  $P_t$  = current year's market price,  $C_1$  = intercept,  $\beta_1$  = coefficient of previous year's market price, which is expected to be below 0,  $P_{t-1}$  = previous year's market price, and  $\varepsilon$  = error term.

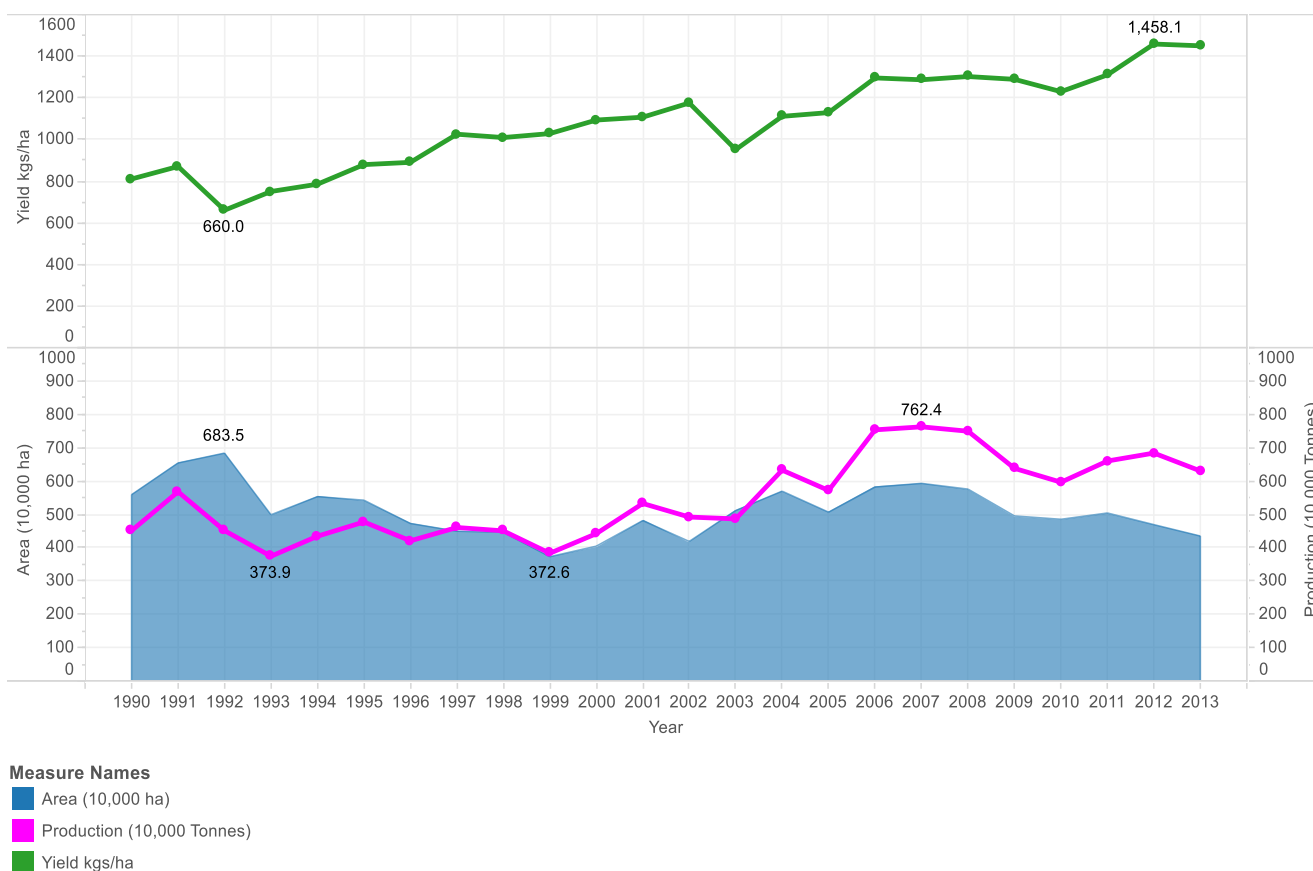
$$\text{Equation hypothesis 3: } \ln Y_t = C_3 + \beta_1 \ln A_t + \beta_2 \ln A_{t-1} + \beta_3 \ln P_{t-1} + \varepsilon \quad (3)$$

where  $Y_t$  = current year's production,  $C_3$  = intercept,  $\beta_1$  = coefficient of current year's area which is expected to be above 0,  $A_t$  = current year's cultivating area,  $\beta_2$  = coefficient of previous year's cultivating area which is expected to be below 0,  $A_{t-1}$  = previous year's cultivating area,  $\beta_3$  = coefficient of previous year's market price which is expected to be above 0,  $P_{t-1}$  = previous year's market price, and  $\varepsilon$  = error term.

These equations were estimated using ordinary least square method.

## EMPIRICAL RESULTS AND DISCUSSION

Figure1 represents the trend of cotton cultivation in China, which is fluctuate upward and downward from the period of 2006-2013 may due to water shortage, climate changes, pest and insect attack, rainfall and may also fluctuation of the cotton market price. Subsequently the interception data of cotton production in 2007 was



**Figure 1.** Cotton production, area and yield in China during the financial years 1990-2013

**Table 1.** Average growth rate of cotton cultivating area, cotton production and yield of cotton in china from 1990 to 2013.

Year	Area	Production	Yield
1990-1995	9.4	0.4	2.5
1996-2000	1.1	5.7	4.6
2001-2007	3.3	6.1	3.0
2008-2013	-4.9	-2.8	2.7
Average growth rate	2.2	2.3	3.2

Data Source: Calculated by author through the use of Excel 2016.

recorded 762.4 million tons, but the largest cotton cultivated area was recorded 6,835, thousand hectares in 1992 and the highest yield was estimated 1458 kg/ha in 2012. When it comes to 2010, the relationship of cotton area and production is more worrying due to the sharp price change in domestic cotton market and some technical reasons. From 2007 to 2013, the yield of cotton was stable, but the cotton production decreased due to cotton cultivated area.

Table 1 indicates the average growth rate of area, production and yield of cotton in China. From 1990 to 1995, the average growth rate was 9.4% in area, 0.4% in

production and 2.5% in yield respectively. Nevertheless, from 1996 to 2000, it was recorded 1.1% growth in area, 5.7% in production and 4.6% in yield. The average growth rate of production increased 5.3%, yield 2.1% but growth of area was much lower in 1996-2000 compared with period of 1990-1995. Again, the growth rate of area rose to 3.3% from 2001 to 2007. In this period, production increased 0.4% and the yield decreased 1.6% compared with the period of 1996-2000. And from the period 2008-2013, the average growth rate of cotton cultivated area was -4.9%, production -2.8% and yield 2.7%. In the period 2008-2013, the average growth rate of cotton

**Table 2.** The relationship between current year's market price and previous year's price.

Variable	Coefficient	Standard Error	t-test	Sig. level
Intercept (C <sub>1</sub> )	0.491	0.453	1.082	0.291
Previous Year's market price	0.926	0.076	12.115	0.000

R<sup>2</sup>= 0.87; Adj.R<sup>2</sup>= 0.86; F-calculate= 146.79; Durbin-Watson value= 2.07.

**Table 3.** The relations between production, area and market price.

Variable	Coefficient	Standard Error	t-test	Sig. level
Intercept (C <sub>2</sub> )	1.873	1.021	1.833	0.081
Current year's area	0.344	0.121	2.851	0.010
Current year's market price	0.249	0.024	10.547	0.000

R<sup>2</sup>= 0.86, Adj.R<sup>2</sup>= 0.85, F-calculate= 64.68, Durbin-Watson value= 1.81.

**Table 4.** The relationships between production and current year's area, previous year's area and market price.

Variable	Coefficient	Standard error	t-test	Sig. level
Intercept (C <sub>3</sub> )	3.065	1.079	2.841	0.010
Current year's area	0.624	0.145	4.317	0.000
Previous year's area	-0.417	0.147	-2.834	0.010
Previous year's market price	0.248	0.022	11.038	0.000

R<sup>2</sup>= 0.89, Adj.R<sup>2</sup>= 0.87, F-calculate= 49.68, Durbin-Watson value= 1.94.

cultivated area and production is much lower than the period 1990-1995, which is due to some technical issues during cultivation and the price decrease of cotton in domestic market. According to the history data of cotton in China during 1990-2013, the total average growth rate of area is 2.2%, production w 2.3% and yield 3.2%.

Table 2 shows the significant relationship between current year's market price and previous year's market price. The value of R<sup>2</sup> and adjust R<sup>2</sup> are 0.87 and 0.86 respectively, which means previous year's market price has highly linear tendency with current year's market price of cotton. Therefore, the result of D.W value has positive relationship which is 2.07 means independent variables does not have auto-correlation. Furthermore the variable of previous year's market price reaches 1% significant level, and it's coefficient is 0.926, which represents it has strongly positive linear relationship with current year's market price. Besides it means if the previous year's market price increase 1%, the current year market price will proportionately increase 0.926%. The positive coefficient of previous year's market price obviously goes against the equation hypothesis 2. According to hypothesis of rational economic body, if the previous year's price is relatively high, then the farmers will be apt to cultivate more cotton, ultimately resulting in higher supply than demand, which will subsequently leads to price decline. As there is time lag in the reaction of market mechanism, therefore, rise in the previous

year's price will decrease the price in the current year.

Table 3 shows that the R-square and adjust R-square value are 0.86 and 0.85, which indicate that the equation has a good correlation, furthermore the value of D.W is 1.81 which indicate that there is no serious autocorrelation between current year cultivated area under cotton crop and market price. Therefore, the intercept passed T-test at the significant level is 10%, and the current year area and market price reached 1% significant level. The above results identify that the independent variables (current year's market price and current year's cultivated area) has correlative with dependent variable (current year's production). Apart from what's explained above, the coefficients in the table connotes that current year's area and market prices have positive impacts on the production. It means when current year's area increases 1%, the production will rise by 0.344%, and when current year's market price increases 1%. The production will rise by 0.249%, which conforms to the equation hypothesis 1.

Table 4 indicates that the comparing results of Tables 3 and 4 show that the R-square and adjust R-square values are 0.89 and 0.87 respectively, which concludes that the equation has a highly significant relationship. The value of D.W has calculated 1.94, which means no series auto-correlation exists. Furthermore, t-test results also have significant levels which are 1% it is more than Table 2. With respect to the variables' coefficients, current

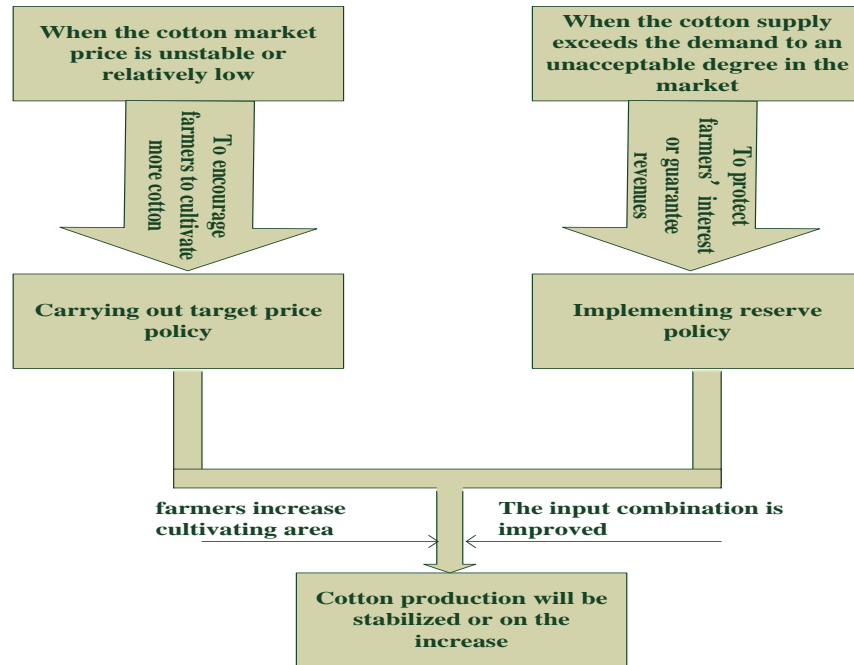


Figure 2. The process of how price regulation influence cotton production

year's area and previous year's market prices have positive correlations with production; however, previous year's area is negatively correlative with production, which means if previous year's area rises, the production will decrease. Furthermore, current year's market price or previous year's market price is positively proportional to the production. Results above are consistent with equation hypothesis 3. Nevertheless, the reason why previous year's area has negative influence on production remains to be considered in a deeper sense. If the previous year's cultivated area is large under the unchanged input ratio and technology, which means the production will correspondingly increase, the cotton supply in the market will rise. Under the circumstance that market demand is stable, the increasing supply will tend to exceed the demand, and consequently the market price is lower, which obeys the regularities of supply and demand. When the market price goes down, the farmers' cultivating interest will also decrease, subsequently reducing cultivated area or input, which will eventually bring down the production.

**Effects of price regulation on cotton production**

Since 2011, China has carried out cotton price regulation policies, one of which is cotton reserve policy, and the other is target price policy. Target price is the price which is announced by the government for willingness to buy farm products at a certain price, which is always announced before sowing. It is announced for encouraging farmers to steadily cultivate cotton (Motie et

al 1998). The reserve policy works when the cotton supply is on great excess over demand in the market, functioning as a market regulation method, which plays a vital role in protecting farmers' cotton planting revenue. What's different between target price policy and reserve policy is that the former is carried out beforehand, but in contrast, the latter is performed afterwards. It implies that the target price policy is encouragement and incentive, however the reserve policy serves as the compensation in case that farmers will suffer from loss. The oscillation arose in cotton production, and yielded in the period 1990-2012 due to climate change, attack of pests, rainfall. Furthermore, the key factor which affected the yield and production more is the reduction of cotton market price, on account of which the farmers were reluctant to cultivate more cotton by implementing more application of inputs so area and yield of cotton was decrease, which is the effect on the production of cotton. As cultivating cotton can only bring a relatively lower interest, the farmers will be more apt to reduce the cultivated area of cotton. To stabilize the cotton production, taking effective measures to guarantee the cotton planting farmer's revenues is of great importance. According to rules of supply and demand, if the target price goes up, the corresponding supply will also increase consequently, in which sense target price has close relations with cotton production. When the government carries out higher target price, then the farmer will have keen interest in increasing the area of cotton cultivation for achieving the government targets (Figure 2).The cotton price in domestic and international market changed greatly in 2010, making the production unstable. Consequently, the



Chinese government increased the target price for maintaining the production. In order to maintain the cotton production, protect the farmers' benefit and boost their willingness to cultivate cotton, Chinese government implemented the reserve policy during the year 2011-2013 and target price policy in the year 2014. According to these policies, the government purchased cotton at 19,800yuan /ton in 2011 and also increased to 20,400 yuan/ton in 2012 and 2013. In 2014 the government adjusted the price policy: set a target price in 2014 and 2015, which are 19,100yuan /ton to 19,800yuan /ton respectively. From 2011 to 2015 price regulation effected positively on cotton production in domestic China.

The consequences discoursed by important consultations clearly indicate that variation in production depends greatly on market price and price regulation of cotton. As market price changes more dramatically, price regulation has significant influence on stabilizing production and price in domestic market. Generally the rise of target price will reassure farmers so that they will cultivate cotton in a larger area or increase the usage of modern technologies and better inputs for achieving desirable production. Nevertheless, target price policy was only implemented in several places without being enlarged nationwide. Therefore, extension of target price policy remains to be done.

## Conclusions

Cotton is one of the most important non-food cash crops and great contributions to the foreign currency earning in China. This study analyzed the effects of market price, price regulation and cultivating area on cotton production. Multi-regression model was used to examine the quantitative impacts of market price and cultivating area on cotton production from 1990 to 2013. Moreover, qualitative effects of price regulation on cotton production were also discussed.

The regression results reveal that current year's market price and cultivating area have positive influence on cotton production, showing 1% rise in current year's market price. Increasing production by 0.249 and 1% led to current year's cultivating area increase. Increase in production by 0.344%, conforms to equation hypothesis 1. In addition, the empirical results display that current year's cultivating area and previous year's market price exerts positive effects on cotton production, nevertheless, negative impact of previous year's area on production was found, suggesting 1% increase in the current year's cultivating area, thus, increasing production by 0.624%; 1% rise in previous year's market price increases production by 0.248; 1% increase in previous year's cultivating area decreases production by 0.417%, denoting market price and cultivating area have time lag effects on production and the coefficients are consistent with equation hypothesis 3. Moreover, analyzing results connotes that previous year's market price has positive

influence on current year's market price, showing that 1% rise in previous market price increases current year's price, which disobeys equation hypothesis 2. Apart from the regressive analysis, qualitative analysis was made to evaluate the effects of price regulation on cotton production, which showed price regulation has indirectly positive effects on cotton production.

To stabilize the cotton production in China, on one hand, the government should actively adopt price regulation policies such as target price policy and reserve policy to cope with the unexpected change of the market price, which is sustainable to protect farmers' interests; On the other hand, the government ought to take effective measures to protect the farmland and guarantee a reasonable land scale of cotton cultivation, which means the government should take responsibility for maintaining the area of cotton cultivation within a proper range.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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## Full Length Research Paper

# Effect of calcium chloride dipping treatment on quality of *Ziziphus spina-christi* L. fruits during cold storage

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*Ziziphus spina-christi* L. is one of the wide varieties of plant grown in Al-Ahsa Oasis in Saudi Arabia and known locally as Alnabaq (buckthorn). The objective of this investigation was to study the effects of postharvest calcium chloride applications on fruit quality of buckthorn under cold storage condition at 2°C and 90% RH for 5 weeks. Fruits were dipped in calcium chloride at different concentrations (0, 2 and 4% W/V) for 10 min. Weight loss, total soluble solids (TSS), ascorbic acid content, fruit firmness and peroxidase activity were determined after Ca<sup>++</sup> treatment and during cold storage every one week. The results revealed that, both the storage periods and treatments significantly affected the postharvest quality of Alnabaq fruits during cold storage condition. A reduction in fruit firmness and ascorbic acid content was observed during storage period. In addition, the results showed that, fruit treated with calcium chloride recorded higher firmness than the control, while lower peroxidase activity was recorded during cold storage at 2°C for 5 weeks. Generally, these results indicated that post-harvest Ca treatments delayed fruit softening and decreased weight loss.

**Key words:** Storage, *Ziziphus spina-christi*, weight loss, firmness, CaCl<sub>2</sub>, peroxidase activity, ascorbic acid.

## INTRODUCTION

*Ziziphus spina-christi* L. (family Rhamnaceae) is a subtropical plant known as Alnabaq or Sedr, which is reported to be used as alternative medicine for human (Shahat et al., 2001). The fruits are rich in carbohydrates and Mg, Ca, Fe and Zn, whereas, the seeds are rich in crude fiber (Osman and Ahmed, 2009).

Al-Ahsa Oasis in Saudi Arabia is known for its wide varieties of biodiversity including *Z. spina-christi* L. trees, which grows wildly and is widely known locally as knar or

Alnabaq, and it tolerates salinity and high heat, but the fruits vary in size and taste, making it undesirable for consumption. Therefore, several types of Alnabaq were introduced from China and India, in the form of cuttings and grafted on the local variety (species), including the Chinese Nabq (*Ziziphus maruritiana* L.), the widespread "Beyuan" cv. in the Kingdom. These fruits are considered as an economic crop in the arid and semiarid regions.

However, many factors have influences on the quality

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of buckthorn fruit, such as agricultural practices and storage conditions (Soliva-Fortuny et al., 2002). The shelf life of buckthorn fruits is short, especially, at ambient temperature (Pareek et al. 2009). Deterioration of fruit quality during storage is mainly due to relatively high metabolic activity under unsuitable storage condition (Fattahi et al., 2010). Postharvest application of calcium has a potential role in keeping quality and prolonging storage life of fruits by delaying senescence and reducing respiration rate (White and Broadley, 2003; Lester and Grusak, 2004; Bhattara and Gautam, 2006). It is well known that the role of calcium is to delay senescence in horticultural crops (Misra and Gupta, 2006; Shirzadeh et al., 2011; Sohail et al., 2015). Liu et al. (2017) reported that treatment with 1% Ca followed by cold storage at 5°C significantly improved apricot fruit quality and shelf life. In addition, the application of CaCl<sub>2</sub> significantly decreased weight loss, TSS, total sugar and TSS/acid ratio, while ascorbic acid and fruit firmness increased with CaCl<sub>2</sub> treatments (Jan et al., 2015, 2016). In addition, CaCl<sub>2</sub> at 2% and chitosan at 1% concentrations proved to be effective in reducing weight loss, decay percentage and maintaining maximum firmness and prolong shelf life of peach fruits during cold storage (Abdel Gayed et al., 2017). There is little information on the storage of fruits. Therefore, this study aimed to investigate the impact of postharvest application of calcium chloride on Nabaq fruits, in extending the self-life of buckthorn (Nabaq) fruits and on the fruit quality during cold storage.

## MATERIALS AND METHODS

This study was conducted in 2016/2017 at College of Agricultural and Food Sciences, King Faisal University, KSA. Fruits were collected in February, from a private orchard at Alqatif, Saudi Arabia. Fruits were then transferred to laboratory. Fruits were sorted and fruits free of any damage, unripe or any defects were used. Fruits were divided into 3 groups, each group contained 300 fruits for each treatment in three replicates and dipped in two CaCl<sub>2</sub> (Sigma-Aldrich, Germany) solutions (2 and 4%) and in distilled water as a control treatment for 10 min. After that, they were dried for 24 h at ambient temperature. Subsequently, they were packed with ventilated polyethylene bags and then stored at 2°C and 90% RH (Shirzadeh et al., 2011) for 5 weeks. After 7, 14, 21, 28, and 35 days, 20 fruits per treatment were used for fruit quality evaluation. The physicochemical analysis for example weight loss, TSS, ascorbic acid content, fruit firmness and peroxidase activity were determined.

### Physicochemical analysis

The weight of fruits was recorded after treatments (initial weight) and after that, it was weekly recorded and the variation in weight loss was expressed as a percentage of accumulative weight loss from the initial weight of the fruits.

Total soluble solids content (Brix) in the fruit juice was determined using of a hand Refractometer (Atago Co., Tokyo, Japan) and the value reported as degree Brix. Fruit firmness was determined using Digital Fruit Firmness Tester, Penetrometer (FHP-803, Agriculture Solutions LLC, USA) fitted with an 8 mm diameter

flat tip and expressed as kg/cm<sup>2</sup>. Measurements were done with three fruits.

Ascorbic acid content (mg 100 g<sup>-1</sup> fresh weight) was measured by titration with 2,6- dichloroindophenol dye that turns to pink color according to AOAC (2006).

Peroxidase activity was determined according to Chance and Maehly (1955). 1 g of the fruit tissue after peeling was homogenized in a mortar with ice 200 mM potassium phosphate buffer (pH 7.0) containing 5 mM Na<sub>2</sub>EDTA, 10 mM Na<sub>2</sub>S<sub>2</sub>O<sub>5</sub> and 1% polyvinylpyrrolidone. Solution was centrifuged (15,000 xg, 15 min) then supernatant was utilized for POD activity determination.

### Statistical analysis

Data were subjected to analysis of variance (ANOVA) using the statistical software SPSS (SPSS Inc., Chicago, USA). Least significant difference test (LSD) at P<0.05 was used for the comparisons among treatment means.

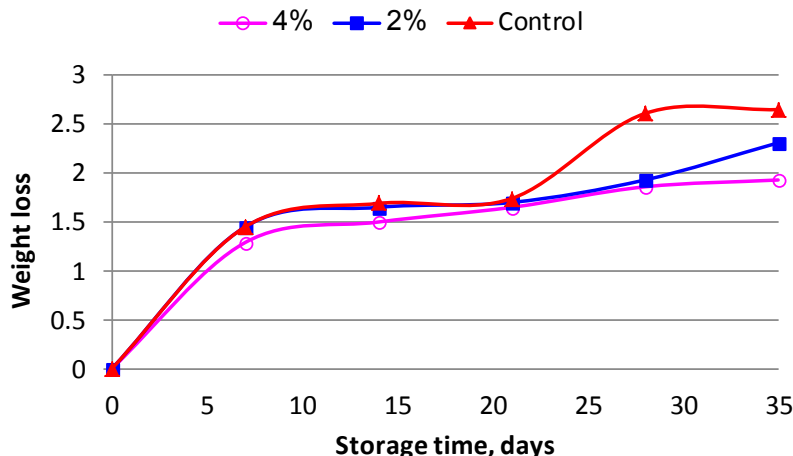
## RESULTS AND DISCUSSION

### Weight loss

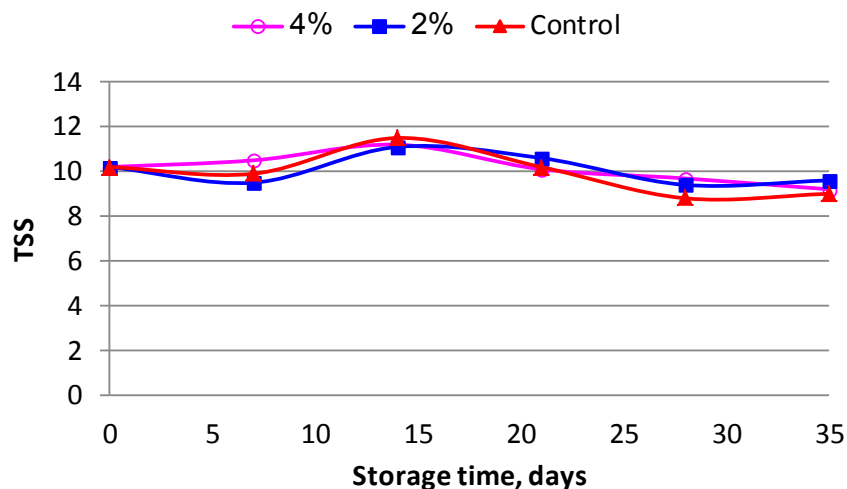
The results in the Figure 1 shows weight loss during cold storage of untreated fruits as compared to fruits treated with 2 and 4% CaCl<sub>2</sub>. But, weight loss of control fruits at the end of experiment (35 day) was higher (2.65%) than fruits treated with CaCl<sub>2</sub> (2.31 and 1.93%). CaCl<sub>2</sub> solution at both concentrations reduced weight loss in comparison with that of the control. All fruit samples recorded a rapid loss of weight at the first week then a gradual reduction was observed during storage (Figure 1). This might be due to a decline in respiration rate and less moisture loss from the fruits during storage. Fruit treated with 4% CaCl<sub>2</sub> recorded the lowest weight loss as compared to the control. During last two weeks of the experiment, untreated fruits recorded higher weight loss than the treated ones (Figure 1). The low weight loss recorded by CaCl<sub>2</sub> treated fruits could be related to the network formation by Ca and pectin in the fruit cell wall to restrict moisture loss (Genanew, 2013). Previous studies indicated that fruits dipped in calcium chloride solutions were the most effective in decreasing weight loss in comparison with the control (Mahajan and Dhatt, 2004; Sohail et al., 2015).

### Total soluble solids (TSS, Brix)

Initially, the total TSS content of fruits was 10.2%. Results showed an increasing trend irrespective of treatments until second week and then decreased during storage period in all treatments from 10.2 to 9 for the control and from 10.2 to 9.6 and 9.2 for 2 and 4% CaCl<sub>2</sub> treatment, respectively. The increasing TSS content until second week was likely due to concentrated juice content due to dehydration during storage (Akhtar et al., 2010). Figure 2 shows that application of calcium chloride had a



**Figure 1.** Effect of calcium chloride and storage period on weight loss in Nabaq fruits after 7, 14, 21, 28 and 35 days of storage at 2°C (LSD:0.66).



**Figure 2.** Effect of calcium chloride on total soluble of solids content of Nabaq fruits after 7, 14, 21, 28 and 35 days of storage at 2°C (LSD:0.75).

slight effect on TSS content during storage and the differences were not significant.

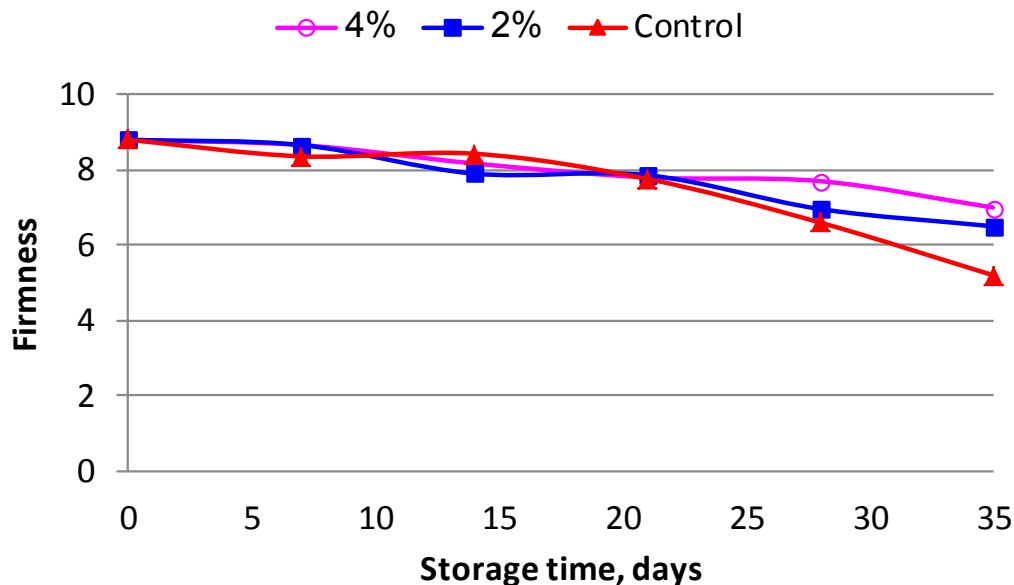
### Fruit firmness

It is clear from Figure 3 that there was a general increase in fruit softening in all the treatments. However, fruit dipped in CaCl<sub>2</sub> solutions had the highest firmness at the end of storage period as compared to the control treatment. At the end of the experiment, fruits treated with 2 and 4% CaCl<sub>2</sub> recorded a decrease of 26.1 and 20.4% in firmness, respectively, against 40.9% for untreated fruits. However, the highest fruit softening detected in the control may be due to the quick metabolic processes (breakdown of starch and proto-pectin to sugars and

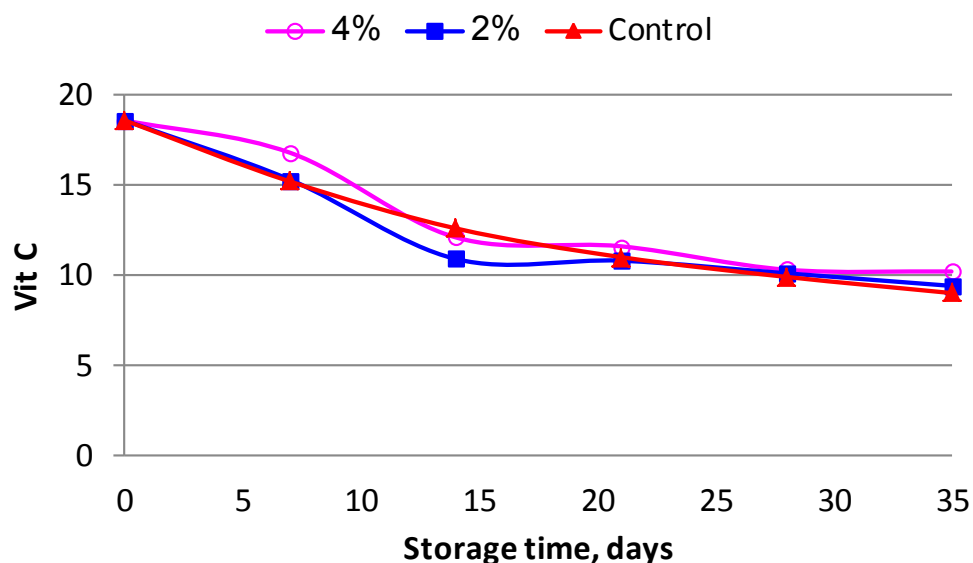
pectic acid, respectively) in comparison with the treated fruits. In addition, Anthon et al. (2005) reported that the interaction of calcium with pectin is known to be the mechanism for the calcium-firming role. Previous investigations with different crops indicated that the softening of fruits treated with different concentrations of CaCl<sub>2</sub> decreased but firmness was kept during storage (White and Broadley, 2003; Shirzadeh et al., 2011).

### Ascorbic acid content

Veltman et al. (2000) reported that ascorbic acid is very sensitive to decomposition as a result of its oxidation during food processing and storage. All the treatments recorded continuous rapid reduction in the content of



**Figure 3.** Effect of calcium chloride on firmness (Kg/cm<sup>2</sup>) of Nabaq fruits after 7, 14, 21, 28 and 35 days of storage at 2°C (LSD:0.18).

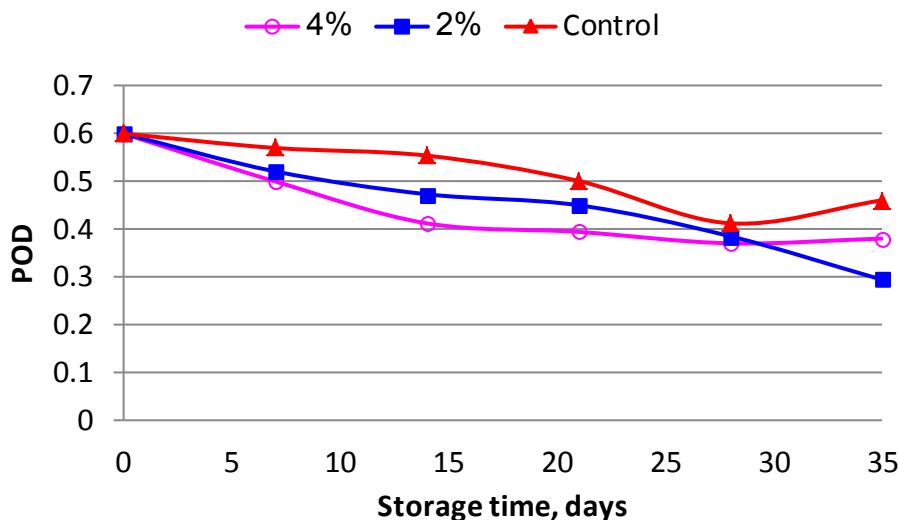


**Figure 4.** Effect of calcium chloride on ascorbic acid content of Nabaq fruits after 7, 14, 21, 28 and 35 days of storage at 2°C (LSD:0.21).

ascorbic acid during the first two weeks of storage (Figure 4). In contrast, Jan et al. (2016) found that ascorbic acid content increased when apples fruits were dipped in 9% of CaCl<sub>2</sub> solution in comparison with the control. After two weeks from the beginning of the experiment, there were no significant differences among treatments in ascorbic acid content (Figure 4). During storage, ascorbic acid content reduction could be due to its antioxidant activity (Davey et al., 2000).

#### Peroxidase activity

Results in Figure 5 show that the storage time has a significant effect on peroxidase activity (POD). The highest POD activity was found in the control fruits as compared to fruits treated with CaCl<sub>2</sub>, while lowest POD activity was recorded in treated fruits. In this context, this result is in line with the results reported by Shirzadeh et al. (2011) in apple. In addition, Lamikanra and Watson



**Figure 5.** Effect of calcium chloride on peroxidase activity in Nabaq fruits after 7, 14, 21, 28 and 35 days of storage at 2°C (LSD:0.16).

(2001) indicated that the level of oxidative stress in cut fruits was related to ascorbate dependency of peroxidase enzymes. However, calcium appears to be necessary for post-harvest treatment of some fruits, since isoperoxidase could cross-link the chains of polygalacturonan (Penel et al., 1999).

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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*Full Length Research Paper*

## Saflufenacil and indaziflam herbicide effects on agricultural crops and microorganisms

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The herbicides saflufenacil and indaziflam have recently been registered in Brazil for weed control in sugarcane crops; however, little information exists regarding their residual effects or influences on soil microorganisms. Therefore, the present study aimed: (a) to determine the effects of saflufenacil and indaziflam on soil microorganisms and (b) to evaluate the residual and dose effects of these herbicides on soybean, sunflower, sunn hemp and peanut crops. The herbicides indaziflam (100 g a.i. ha<sup>-1</sup>) and saflufenacil (120 g a.i. ha<sup>-1</sup>) were applied to dark red latosol samples, and the CO<sub>2</sub>-C released by soil basal respiration was measured at 7, 14, 21, 28, 35, 42, 49 and 56 days after treatment (DAT), in an experiment with a completely randomized design (CRD) and five replicates. The microorganisms were quantified via the use of different culture media, each replicated three times at 0, 15, 30 and 60 DAT. No significant difference occurred among the treatments for the carbon content of the microbial biomass. Regarding the basal respiration, the soils treated with saflufenacil showed a decrease in the carbon released by the soil at 49 DAT, whereas the carbon released by the soils treated with indaziflam increased until the last day of evaluation. The responses of the fungal and bacterial populations and the amyolytic and cellulolytic microorganisms differed among the treatments. The residual effect of the herbicides on the crops was evaluated via a CRD, in a 6 (doses) × 5 (sowing times) factorial arrangement with four replicates. The different indaziflam and saflufenacil doses were sprayed separately at pre-emergence. At 0, 10, 20, 40 and 60 days after the herbicide applications, soybean, sunn hemp, sunflower and peanut were sown. The phytotoxicity of saflufenacil to the crops declined throughout the evaluations for all the doses and species. Indaziflam was highly phytotoxic to all the crop species until 60 days after application, preventing the field sowing of the crops during that period.

**Key words:** Phytotoxicity, sugarcane, carryover, microbial degradation.

### INTRODUCTION

The herbicides saflufenacil and indaziflam have been recently registered in Brazil for weed control in sugarcane crops; nevertheless, little information exists regarding their effects on other crops, especially those used

rotationally in sugarcane fallow areas.

Saflufenacil can be pre-plant incorporated and applied pre- and post-emergence in crops such as sugarcane, maize, soybean, wheat and cotton. This herbicide is used

for the control of eudicots and belongs to the pyrimidinedione chemical class, inhibiting the enzyme protoporphyrinogen oxidase (PROTOX). The main physicochemical characteristics of saflufenacil include a vapor pressure (VP) of  $2 \times 10^{-14}$  mm Hg at 25°C (a nonvolatile herbicide), a half-life ( $t_{1/2}$ ) of one to five weeks, a pKa of 4.3 (a weak acid) and a water solubility of 30 mg L<sup>-1</sup> at pH 5.0 and 2100 mg L<sup>-1</sup> at pH 7.0 (BASF, 2008). This herbicide is absorbed by both roots and leaves, with its translocation occurring mainly in the xylem and its mobility limited in the phloem. Susceptible plants show symptoms of injury within a few hours and die within one to three days (Soltani, 2010).

Soil organic matter has a high affinity for the saflufenacil molecule; therefore, soils with high organic matter content have a relatively small amount of the molecule available for plant absorption (Monquero et al., 2012). Gannon et al. (2014) observed that the saflufenacil phytotoxicity to canola was dependent on the soil properties and that those soils with high contents of organic matter and clay showed relatively low toxicity. Soltani (2010) observed that saflufenacil at 100 and 200 g a.i. ha<sup>-1</sup> caused 51 to 99% injury and reduced height by 25 to 93%, shoot dry weight by 92 to 99% and seed yield by 56 to 99% in cranberry and in adzuki, Lima, snap and white beans. Soybean and pea were the crops most tolerant to saflufenacil.

Indaziflam belongs to the alkylazine chemical class and acts on cell wall biosynthesis without affecting the synthesis of polysaccharide polymers. The action is inhibitory and most likely occurs at some point during the cross-linking stage of cellulose microfibrils. Inhibition of cell division in meristematic tissues has also been proposed as a secondary mode of action. Indaziflam is used as a pre-emergent herbicide for the control of monocotyledonous weeds and some eudicots in perennial crops, such as citrus, coffee and sugarcane. Its physicochemical characteristics are as follows: a VP of  $1.875 \times 10^{-10}$  mm Hg at 20°C (a nonvolatile herbicide), a half-life of approximately 150 days, a pKa of 3.5 and a water solubility of 0.044 to 0.0017 g L<sup>-1</sup> at pH 4.0 and 0.0028 to 0.0012 g L<sup>-1</sup> at pH 9.0 (U.S. EPA, 2010; Alonso et al., 2011). In the US, the labeled rate for indaziflam in Florida citrus ranges from 73 to 95 g a.i. ha<sup>-1</sup>. Indaziflam provides three to four months of residual weed control in citrus, depending on humidity and temperature (Jhala and Singh, 2012). For the control of annual grasses sensitive to this herbicide, the doses range from 25 to 100 g ha<sup>-1</sup>, reaching up to 150 g ha<sup>-1</sup> for more tolerant species (Kaapro and Hall, 2012).

According to Guerra et al. (2014), studies conducted

during the initial development of maize, millet, sorghum, soybean, sunflower, cotton, beet and cucumber crops indicated that all the species were sensitive to soil-applied indaziflam in the field. The only symptom observed in the different species after planting in soil containing this herbicide was the nonemergence of seedlings, except for sunflower. Cotton and maize did not emerge only when sown in the soil with the highest indaziflam dose (100 g ha<sup>-1</sup>). Soybean plants emerged in the soil treated with the two lowest indaziflam doses (20 and 40 g ha<sup>-1</sup>) but died after a few days. In contrast, sorghum, millet, cucumber and beet did not emerge even in soil treated with the lowest indaziflam dose (20 g ha<sup>-1</sup>) (Guerra et al., 2014).

Herbicides may directly or indirectly affect microbial activity in the soil. The direct effects include toxicity to the soil microbiota, whereas the indirect effects include damage to the crops that affects their physiology, reducing plant-microorganism interactions. For example, Arruda et al. (2001) reported that sulfentrazone application reduced root nodulation and the exudation of amino acids by soybean xylem. Note that agrochemicals may positively or negatively affect soil microorganisms. Positive effects occur when the product is metabolized by the soil microorganisms, and negative effects occur when the chemicals poison them (Santos et al., 2005; Vivian et al., 2006).

Therefore, the present study aimed to determine the residual effect of saflufenacil and indaziflam and the dose-response relationship of these herbicides regarding the growth and development of *Glycine max* (soybean), *Crotalaria juncea* (sunn hemp), *Helianthus annuus* (sunflower) and *Arachis hypogaea* (peanut) together with the effects of these herbicides on amyolytic and cellulolytic microorganisms, fungi and total bacteria.

## MATERIALS AND METHODS

### Effects of the herbicides saflufenacil and indaziflam on soil microorganisms

The herbicides saflufenacil and indaziflam were applied on August 09, 2016. The experimental units consisted of trays (28 × 43 × 4.5 cm) containing 2 kg of dark red latosol, composed of four single samples. The soil used in the experiment was collected in a native forest with no history of herbicide use, at a depth of 10 cm. The chemical analysis of the soil samples indicated the following: P = 15 mm dm<sup>-3</sup>; organic matter = 24%; pH CaCl<sub>2</sub> = 5.1; K = 2.5 mmol<sub>c</sub> dm<sup>-3</sup>; Ca = 28%; Mg = 12%; H + Al = 0.4%; sum of bases (SB) = 42.5%; cation exchange capacity (CEC) = 82.5%; percentage of base saturation (V%) = 52; and clay, sand and silt = 600, 150 and 190 g kg<sup>-1</sup>, respectively. Herbicides were applied at doses of 120 g

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a.i. ha<sup>-1</sup> of saflufenacil and 100 g a.i. ha<sup>-1</sup> of indaziflam; the control group received no herbicide treatment. The herbicides were applied via a CO<sub>2</sub> backpack sprayer equipped with three TeeJet DG 110.03 VS nozzles (Drift Guard), with a 0.50-m spacing and a flow rate of 200 L ha<sup>-1</sup>. At the time of application, the wind speed was 2.1 m s<sup>-1</sup>, the humidity was 56.9%, and the temperature was 30°C. The soil samples were subsequently crushed and sieved through a 2-mm mesh and homogenized, with the moisture adjusted to 60% of field capacity.

#### Microbial biomass carbon content

The microbial biomass was determined using the fumigation-extraction method described by Vance et al. (1987). The samples were analyzed in triplicate; that is, each soil sample was divided into six subsamples of 20 g each and placed in 100-ml glass bottles. Three subsamples were subjected to the fumigation-extraction process, and the other three were subjected to immediate extraction after weighing (non-fumigated samples). The carbon content in the soil extracts was calculated as follows:

$$\text{MBC } (\mu\text{g C g}^{-1} \text{ soil}) = (\text{F-NF}) / \text{Kc},$$

Where MBC = microbial biomass carbon; F = fumigated samples; NF = nonfumigated samples; Kc = 0.33, a correction coefficient.

#### Microbial activity assessed by basal respiration (respirometry)

Glass jars with lids were used as respirometers. The microbial activity was evaluated by the amount of CO<sub>2</sub> released at 7, 14, 21, 28, 35, 42, 49 and 56 days from non-fumigated soil samples in a static system (GRISI, 1995). A completely randomized design (CRD) was used, with five replicates for each of the following treatments: blank, control (no herbicide application), indaziflam and saflufenacil. Fifty-gram samples of sieved soil within snap cap glass flasks were placed inside a jar along with another flask containing 10 ml of 1 N NaOH to capture the CO<sub>2</sub> released by the soil. The flasks were hermetically sealed and incubated at 25 ± 2°C in the dark. Jars containing only 10 ml of 1 N NaOH were incubated as well (blanks). Every seven days of incubation, the NaOH solution was titrated with a standard solution of 0.5 N HCl, by adding 2 ml of saturated 10% BaCl<sub>2</sub> solution to precipitate the Na<sub>2</sub>CO<sub>3</sub> and two drops of 1% phenolphthalein solution as an indicator. Soil basal respiration was quantified using the following equation:

$$\text{SBR (mg of CO}_2\text{-C kg}^{-1} \text{ soil hour}^{-1}) = ((\text{Vb-Va}) * 0.5 * 6 * 1000) / (\text{Wd} / \text{T})$$

where SBR = carbon from soil basal respiration, Vb = volume (mL) of hydrochloric acid used to titrate the control solution (blank), Va = volume (mL) of hydrochloric acid used for sample titration, Wd = dry weight (g) and T = time (hours).

#### Quantification of total bacteria, fungi and amylolytic and cellulolytic microorganisms

The bacteria and fungi present in the soil samples were quantified by counting the colony-forming units (CFU) via the serial dilution technique and plating in the following culture media: nutrient agar (NA) for bacteria, Martin's medium for fungi, cellulose for cellulolytic microorganisms and starch agar for amylolytic microorganisms. The NA medium was prepared by adding 28 g of NA per liter of distilled water. Subsequently, the NA medium was autoclaved for 20 min at

120°C and 1 atm, and 2 ml of nystatin L<sup>-1</sup> was added to inhibit fungal growth before pouring the medium into incubation plates. Martin's medium consisted of 1 g of KH<sub>2</sub>PO<sub>4</sub>, 0.5 g of MgSO<sub>4</sub>·7H<sub>2</sub>O, 5 g of peptone, 10 g of dextrose, 0.03 g of rose Bengal, 20 g of agar and 1 L of distilled water. Martin's medium was autoclaved for 20 min at 120°C and 1 atm, and 0.2% streptomycin was added at the rate of 2 mL L<sup>-1</sup> before the medium was poured into plates. The cellulolytic microorganisms were quantified using potato dextrose agar (PDA) medium containing 20% potato, 2% dextrose, 2% agar and tetracycline (100 mg L<sup>-1</sup>). After incubation, the plates were flooded with 10 mL of concentrated Congo red solution (2.5 g L<sup>-1</sup>) for colony quantification. The starch agar medium used to quantify amylolytic microorganisms consisted of 20 g of agar, 20 g of soluble starch and 1.0 L of distilled water.

Ten-gram samples of soil were added to 90 ml of saline (0.85% NaCl) and homogenized for 15 min with the aid of a shaker. Next, 1 ml aliquots of this suspension were transferred to test tubes containing 9 ml of saline solution. The dilution process was continued until obtaining the 10<sup>-1</sup>, 10<sup>-2</sup> and 10<sup>-3</sup> serial dilutions, which were used for amylolytic microorganisms, for fungi and cellulolytic microorganisms and for bacteria, respectively. Aliquots containing 100 µl of the dilutions were inoculated in triplicate onto plates containing the respective media, and the plates were incubated for 24 h at 35°C for bacteria and for 48 h at 30°C for fungi. Afterwards, the dilutions having between 30 and 300 colonies were counted, and the results are expressed as CFU g<sup>-1</sup> of soil.

The data from all the experiments with microorganisms were subjected to analysis of variance by the F test, and the means of the treatments were compared by the Tukey test at the 5% probability level.

#### Effects of the herbicides saflufenacil and indaziflam on agricultural crops

The experiment was conducted in a greenhouse and used a CRD in a 6 (doses) × 5 (sowing times) factorial arrangement for each species and herbicide, with four replicates. For the experiment, 5-L pots were filled with dark red latosol samples (chemical analysis as described above) and sprayed with either saflufenacil (0, 7.5, 15, 30, 60 and 120 g a.i. ha<sup>-1</sup>) or indaziflam (0, 6.5, 12.5, 25, 50 and 100 g a.i. ha<sup>-1</sup>). The herbicides were applied on February 14, 2017, via a CO<sub>2</sub> backpack sprayer, which was equipped with a spray boom with 110.03 fan nozzles, at a constant pressure of 245.16 kPa and a flow rate of 200 L ha<sup>-1</sup>. The relative air humidity and temperature during herbicide application were monitored with a weather station, reaching 58% and 21°C, respectively. The pots were housed in a greenhouse with automatic irrigation (5 mm H<sub>2</sub>O per day). At 0, 10, 20, 40 and 60 days after herbicide application, the following species were sown: *Glycine max* (soybean variety 95Y52), *Crotalaria juncea* (sunn hemp), *Helianthus annuus* (sunflower variety Rajado) and *Arachis hypogaea* (peanut variety Runner 886). Each experimental unit consisted of six plants.

The herbicide effects were evaluated at 32 days after sowing (DAS) by a percentage scale of scores, where zero (0) represents the absence of symptoms and 100 represents the death of all plants (ALAM, 1974). The following were also determined at 32 DAS: the chlorophyll concentration with a chloroLOG chlorophyll meter (FALKER), the leaf area (by a nondestructive method based on the use of a Li-COR 3000 leaf area meter) and the dry biomass of the shoots after cutting the plants close to the ground. The dry mass was obtained by placing the plants in a forced-air oven at 65°C, until reaching a constant weight. The results were subjected to analysis of variance and regression. The regression curves were adjusted with the SigmaPlot software.

## RESULTS AND DISCUSSION

### Effects of the herbicides saflufenacil and indaziflam on soil microorganisms

As seen in Table 1, no significant difference occurred among the treatments regarding the total carbon content of the microbial biomass; that is, the herbicides did not affect the microbial biomass, which is responsible for the transformation of organic matter, for nutrient cycling and for energy flow (Wardle, 1992). According to Moorman (1989), herbicides have little effect on the soil microbial biomass, but the populations and activities of certain functional groups are affected.

Voos and Groffman (1997) evaluated the relationship between the microbial biomass and the dissipation of 2,4-D and dicamba by soil microorganisms and found a positive relationship between the size of the microbial biomass and the degradation of these herbicides in the soil. Thus, these results may be useful in predicting the behavior of herbicides in different ecosystems. Additionally, Moreno et al. (2007) attributed an increase in the carbon content of the microbial biomass after atrazine application to adaptation by the microorganisms, which used the atrazine as a source of carbon and energy. Hart and Brooks (1996) reported that effects of 19 years of cumulative annual field application of benomyl, chlorfenvinphos, aldicarb, triadimefon and glyphosate, either singly or in combination, therefore had no measurable long-term harmful effects on the soil microbial biomass.

Table 2 shows that at 0 and 15 days after application (DAA), significantly more CFU occurred in the saflufenacil-treated soil samples than in the control samples. Starting at 30 DAA, a decrease occurred in the microbial population compared with the populations observed during the first evaluations. This result can be explained by the product's half-life in the soil, since herbicides can serve as nitrogen, carbon and energy sources or as a cometabolism substrate, in which microorganisms can transform a herbicide without depleting the energy needed for their development (Fournier et al., 1997). Thus, the bacteria most likely first used the carbon present in the herbicide to grow; after product degradation, the number of colonies decreased but did so without differing statistically from that of the control group.

For indaziflam, the colony number increased throughout the evaluations. At 0, 15 and 30 DAA, no significant difference from the control group existed, which most likely transpired because the microorganisms were undergoing an acclimation phase. At 60 DAA, a greater number of CFU occurred in the indaziflam-treated soil samples than in the control group, perhaps due to the high persistence of indaziflam in the soil and its subsequent role as a microbial energy source, leading to

**Table 1.** Total carbon content of the microbial biomass at 17 days after treatment (DAT).

Treatments	Total carbon biomass ( $\mu\text{g}\cdot\text{g}^{-1}$ soil)
Control	320.28
Saflufenacil	356.79
Indaziflam	250.15

**Table 2.** Number of bacterial colony-forming units (CFU) detected with nutrient agar (NA) for saflufenacil- and indaziflam-treated soil samples at different days after application (DAA). Log-transformed data are shown.

Parameter	UFC $\text{g}^{-1}$ soil			
	Days after herbicide application (DAA)			
	0	15	30	60
Control	1.33 <sup>bA</sup>	2.00 <sup>bA</sup>	2.46 <sup>aA</sup>	1.93 <sup>bA</sup>
Saflufenacil	6.33 <sup>aA</sup>	7.73 <sup>aA</sup>	3.16 <sup>aB</sup>	3.10 <sup>abB</sup>
Indaziflam	1.23 <sup>bB</sup>	2.30 <sup>bAB</sup>	3.9 <sup>aA</sup>	4.40 <sup>aA</sup>
VC (%)	33.85			
SD columns	2.28			
SD lines	2.52			

Equal lowercase letters between columns and equal capital letters between lines do not differ statistically at 5% significance.

an increased microbial population (Table 2).

Tu et al. (1992) applied eight herbicides, atrazine, butylate, ethalfluralin, imazethapyr, linuron, metolachlor, metribuzin and trifluralin to loamy sand to determine if these materials caused any serious effects on microbial and enzymatic activities related to soil fertility. Some herbicides showed an effect on bacteria and fungi for the first week of incubation, but, subsequently, the populations returned to levels similar to those obtained in the controls. Results indicated that the herbicidal treatments at the level tested were not drastic enough to be considered deleterious to soil microbial and enzymatic activities which are important to soil fertility.

Dzantor and Felsot (1991) reported the effects of simulated spills of alachlor alone or as a mixture with atrazine, metolachlor, and trifluralin on microbial activity. Simulated spills initially inhibited bacteria, but after 7 days, bacterial numbers had recovered to levels similar to those in untreated controls. Fungal populations were drastically reduced after 1 day and became undetectable after 7 and 21 days of incubation in the mixed herbicide and alachlor-only treatments, respectively (Dzantor and Felsot, 1991)

The number of fungal colonies in the saflufenacil-herbicide-treated soil decreased throughout the study, and a statistically significant difference occurred between the treated and control groups at 30 and 60 DAA (Table

**Table 3.** Number of fungal colony-forming units (CFU) detected with Martin's culture medium for saflufenacil- and indaziflam-treated soil samples at different days after application (DAA). Log-transformed data are shown.

Parameter	UFC g <sup>-1</sup> soil			
	Days after herbicide application (DAA)			
	0	15	30	60
Control	1.27 <sup>aA</sup>	1.50 <sup>aA</sup>	1.23 <sup>aA</sup>	1.33 <sup>aA</sup>
Saflufenacil	1.63 <sup>aA</sup>	1.03 <sup>abAB</sup>	0.53 <sup>bB</sup>	0.36 <sup>bB</sup>
Indaziflam	0.96 <sup>aB</sup>	0.83 <sup>bB</sup>	1.75 <sup>aA</sup>	1.53 <sup>aAB</sup>
VC (%)	25.25			

Equal lowercase letters between columns and equal capital letters between lines do not differ statistically at 5% significance.

**Table 4.** Number of colony-forming units (CFU) for amylolytic microorganisms in saflufenacil- and indaziflam-treated soil samples detected with starch agar at different days after application (DAA). Log-transformed data are shown.

Parameter	UFC g <sup>-1</sup> soil			
	Days after herbicide application (DAA)			
	0	15	30	60
Control	2.00 <sup>bA</sup>	3.20 <sup>bA</sup>	2.47 <sup>aA</sup>	2.93 <sup>aA</sup>
Saflufenacil	5.27 <sup>aA</sup>	4.63 <sup>aA</sup>	1.83 <sup>aB</sup>	0.96 <sup>bB</sup>
Indaziflam	1.87 <sup>bA</sup>	1.83 <sup>cA</sup>	1.56 <sup>aA</sup>	1.10 <sup>bA</sup>
VC (%)	24.52			

Equal lowercase letters between columns and equal capital letters between lines do not differ statistically at 5% significance.

3). In turn, the number of fungal colonies in the indaziflam-treated soils differed significantly from that in the control group only at 15 DAA, and an increase in the number of colonies was observed at 30 and 60 DAA. These results are consistent with those reported by Reis et al. (2008), who showed that ametryn and trifloxysulfuron-sodium, alone or in combination, and 2,4-D caused a reduction in the soil fungal population density only after 15 DAA. The density was restored in the subsequent evaluations because of either a metabolic adjustment in the subpopulations affected by the herbicide or the lower residual herbicide concentrations in the soil (Reis et al., 2008).

The number of amylolytic microbial colonies observed for the saflufenacil-treated soils differed from that observed for the control group at 0, 15 and 60 DAA; the number of these colonies gradually decreased during the evaluations (Table 4). These results might be explained by an initial use of the energy contained in the herbicides to increase the microbial population, followed by subsequent decreases in population size with the dissipation of these products in the soil due to microbial

**Table 5.** Number of colony-forming units (CFU) for cellulolytic microorganisms detected with BDA medium in saflufenacil- and indaziflam-treated soil samples at different days after application (DAA). Log-transformed data are shown.

Parameter	UFC g <sup>-1</sup> soil			
	Days after herbicide application (DAA)			
	0	15	30	60
Control	2.47 <sup>bA</sup>	2.90 <sup>cA</sup>	2.40 <sup>bA</sup>	2.20 <sup>aB</sup>
Saflufenacil	6.67 <sup>aB</sup>	10.43 <sup>aA</sup>	5.80 <sup>aB</sup>	4.66 <sup>aB</sup>
Indaziflam	4.37 <sup>abB</sup>	5.70 <sup>bA</sup>	3.67 <sup>abAB</sup>	2.77 <sup>aB</sup>
VC (%)	28.72			

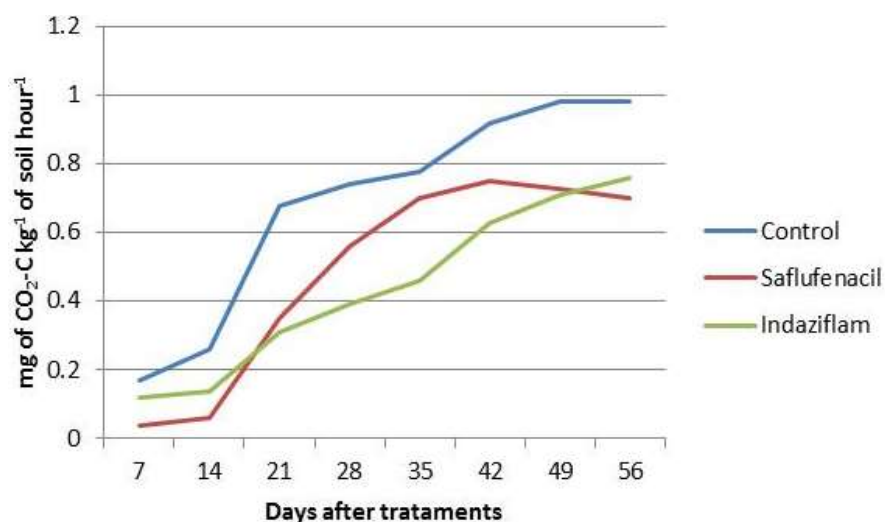
Equal lowercase letters between columns and equal capital letters between lines do not differ statistically at 5% significance.

action. At 15 and 60 DAA, fewer amylolytic microbial colonies occurred for the indaziflam-treated soils than for the control group soils, but no statistically significant differences existed at any of the evaluation times (Table 5). An important point to emphasize is that the starch agar medium is used to select the amylase-producing microorganisms involved in the transformation of carbon and nitrogen compounds in the soil.

For cellulolytic microorganisms, the number of CFU in the saflufenacil-treated soil samples at 0, 15 and 30 DAA was statistically significantly higher compared with that in the control samples. The indaziflam-treated soils produced more CFU for cellulolytic microorganisms compared with the control soils at all the evaluation times; however, the only statistically significant difference was observed at 15 DAA (Table 5).

Cellulose is a polysaccharide composed of approximately 40 glycosidic chains cross-linked into compact bundles. Each chain has a degree of polymerization approximating 10,000 glucose units linked by  $\beta$ 1-4 bonds and cannot be metabolized by most animals because most lack an enzyme that hydrolyzes these bonds. However, this material does not accumulate in the environment due to the activity of fungi and bacteria, which produce cellulolytic enzymes (Lehninger et al., 2006). Thus, the presence of microorganisms that break the cellulose chain is of extreme importance.

As shown in Figure 1 for the saflufenacil-treated soils, the amount of CO<sub>2</sub> declined at 56 DAT, in agreement with the manufacturer's information indicating that saflufenacil is a nonvolatile herbicide (VP of  $2 \times 10^{-4}$  mm Hg) and has a half-life of one to five weeks (BASF, 2008). By contrast, a growing curve was observed for indaziflam. Similarly, Kaapro and Hall (2012) reported that indaziflam has a high residual period in the soil, greater than 150 days, persisting longer than other pre-emergent herbicides. Between 7 and 14 days, a phase of microbial acclimation to the herbicide is believed to have occurred, with the herbicide then released as CO<sub>2</sub> over time, according to its



**Figure 1.** Amount of CO<sub>2</sub> released at 7, 14, 21, 28, 35, 42, 49 and 56 days after treatment (DAT) in mg of CO<sub>2</sub>-C kg<sup>-1</sup> of soil hour<sup>-1</sup>.

persistence.

### Effects of the herbicides saflufenacil and indaziflam on agricultural crops

The phytotoxicity of saflufenacil to soybean plants was above 40% when sowing was performed at 0 and 10 DAA, and a relationship existed between the plant response and the saflufenacil dose. The phytotoxicity of a commercial dose of saflufenacil was close to 20% at 20 and 40 DAA and was 15% at 60 DAA (Figure 2A). Monquero et al. (2012) studied the residual effect of saflufenacil after drought periods (0, 15, 30, 45, 60 and 90 days) in a dystrophic red latosol (clay texture) and reported that the phytotoxicity of a bioindicator (cucumber) was greater than or equal to 80% until up to 28 days of drought.

The phytotoxicity of saflufenacil to crotalaria plants was less than 30% in all the doses used, except when sowing was performed on the day of herbicide application, when phytotoxicity reached 40% (Figure 2B). For sunflower plants, saflufenacil phytotoxicity exceeded or equaled 80% when sowing was performed at 0 or 20 DAA, respectively (Figure 2C). Saflufenacil phytotoxicity decreased at the other sowing times; however, even at 60 DAA, phytotoxicity close to 40% was observed for the commercial dose, which could lead to yield losses. These results corroborate those of Brighenti (2015), who reported that saflufenacil decreased the plant stand and sunflower yield (kg ha<sup>-1</sup>), with the highest dose also reducing the weight of 1000 achenes. Peanut plants presented a phytotoxicity below 20% only at 60 DAA, with their highest phytotoxicity (40%) observed at 0 DAA

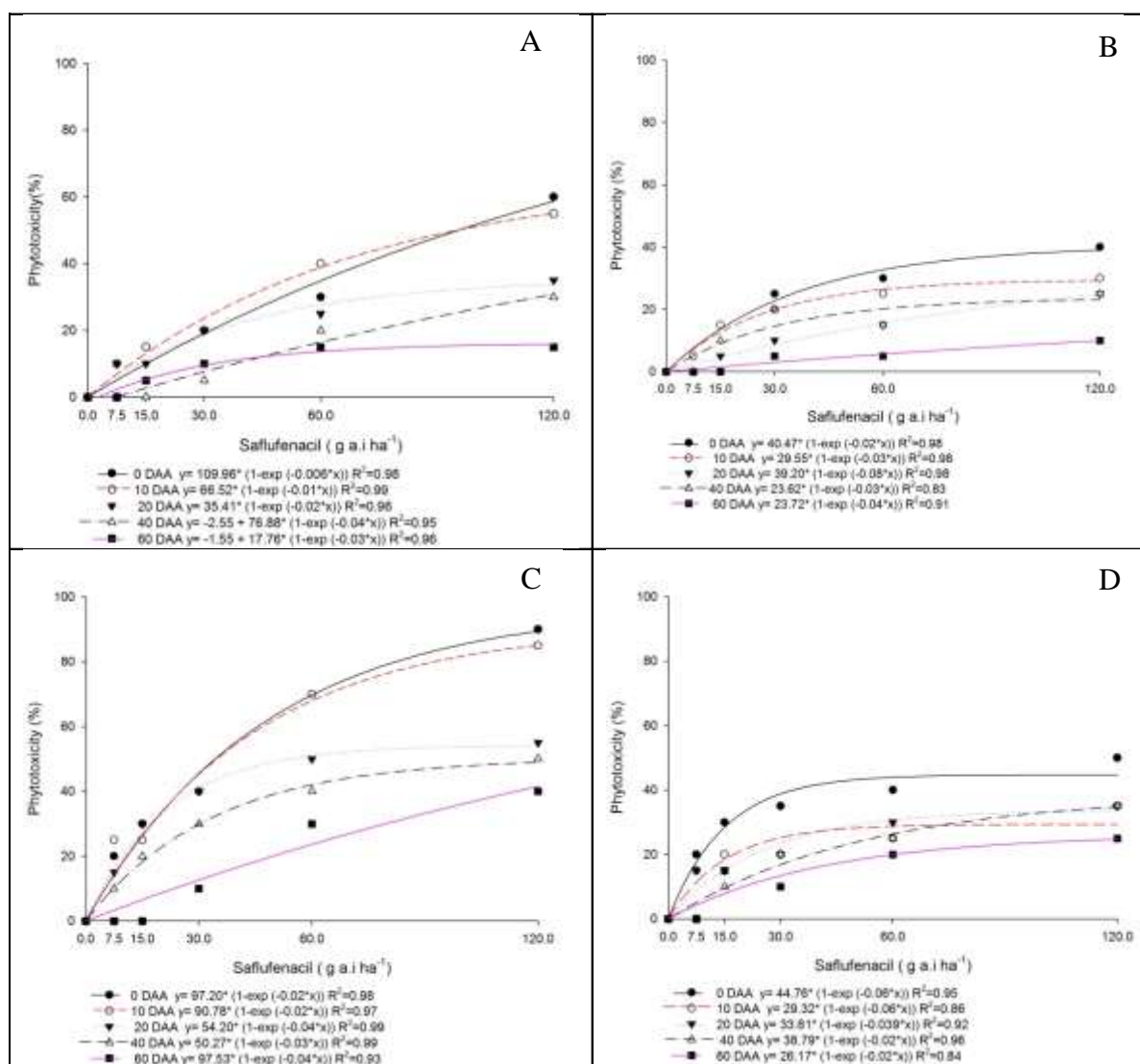
(Figure 2D).

Soltani (2010) observed that saflufenacil at 100 and 200 g a.i ha<sup>-1</sup> caused 51 to 99% injury and reduced height by 25 to 93%, shoot dry weight by 92 to 99% and seed yield by 56 to 99% in cranberry and in adzuki, Lima, snap and white beans. The most saflufenacil-tolerant crops were soybean and pea. In other research, Soltani et al. (2009) reported that addition of an adjuvant to saflufenacil applied POST caused 99% injury to corn at three-leaf stage and reduced yield up to 59% compared to saflufenacil applied without adjuvant.

According to Papiernik et al. (2012), the half-life of saflufenacil ranges from 13 (in the arable layer) to 32 days (in the subsurface layer), with low soil sorption and rapid dissipation. However, injuries were observed in rotational crops (pumpkin, cucumber, carrots, garlic, pepper and beet) up to one year after high herbicide doses (100 to 200 g a.i. ha<sup>-1</sup>) were applied, the same doses recommended in Brazil (Robinson and Mcnaughton, 2012).

Indaziflam was highly phytotoxic to soybean plants regardless of the sowing period. For example, the phytotoxicity was 70% at 60 DAA, and a direct positive relationship existed between increased dose and plant phytotoxicity (Figure 3A). These results corroborate those of Guerra et al. (2014), who reported that not only soybean but also sorghum, millet, cucumber and beet showed an indaziflam dose below 5 g ha<sup>-1</sup> that caused 50% injury to the plants (I<sub>50</sub> dose), with the dose commonly used in countries where this herbicide has already been registered 20 times higher than 5 g ha<sup>-1</sup>. The authors concluded that most of the species tested were highly sensitive to indaziflam (Guerra et al., 2014).

In a study on the effect of simulated indaziflam drift



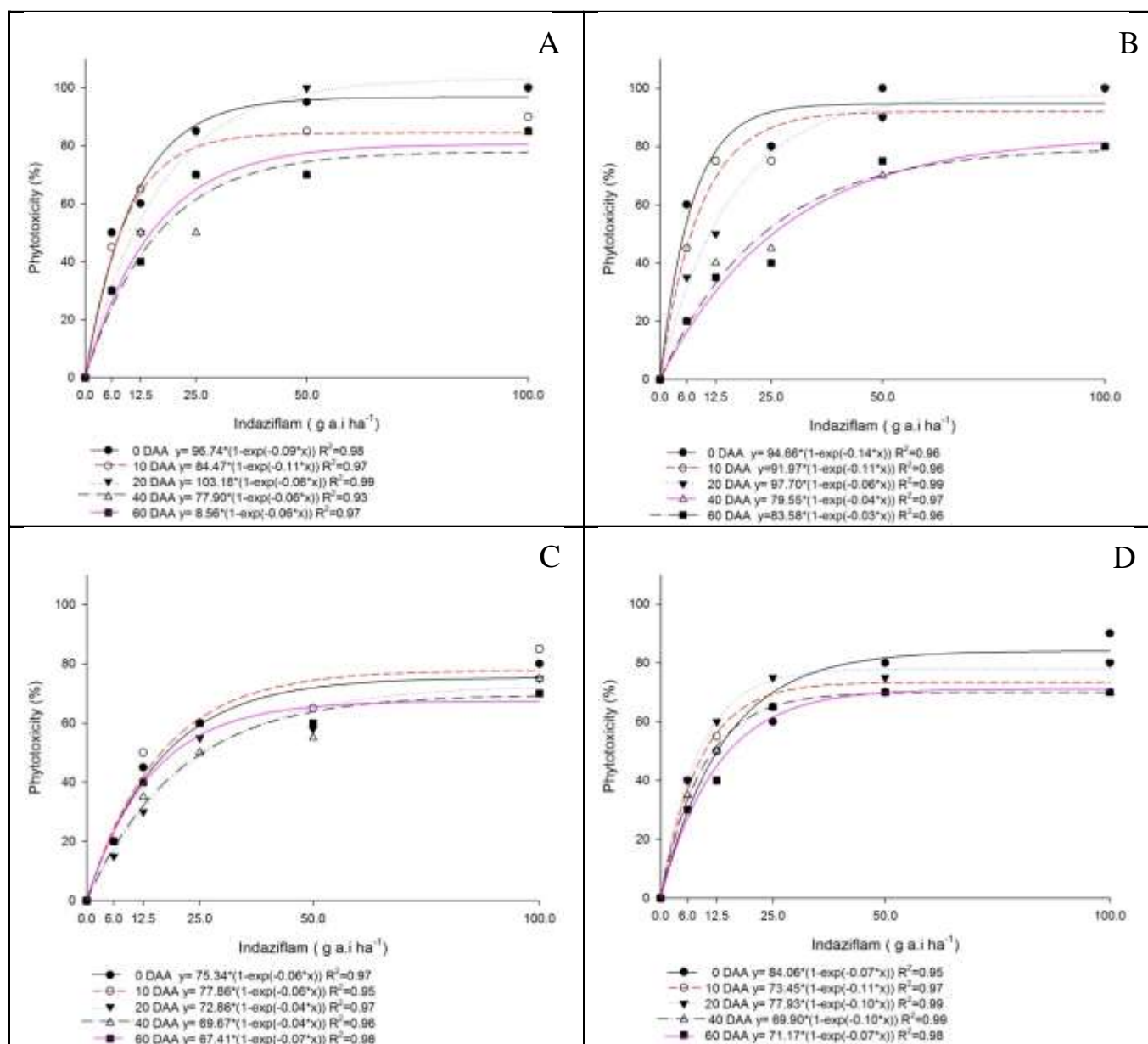
**Figure 2.** Phytotoxicity (%) of different saflufenacil doses to *Glycine max* (A), *Crotalaria juncea* (B), *Helianthus annuus* (C) and *Arachis hypogaea* (D) at 32 DAS and 0, 10, 20 and 60 DAA.

(doses of 100, 20, 10, 5 and 2.5% of the commercial dose of  $73 \text{ g a.i ha}^{-1}$ ) on the growth of selected crops, the crops were ranked as follows according to the observed indaziflam level at which susceptibility occurred: cotton < tobacco < tomato < pumpkin < pepper < soybean. For cotton, the most sensitive crop, 2.5% of the commercial dose caused a 20% reduction in the root mass (Jeffries et al., 2014).

For sunn hemp, indaziflam was also highly phytotoxic. Except for sowing at 40 and 60 DAA, the phytotoxicity at all the other sowing times was equal to or greater than 80%, with a direct response to increased dose (Figure 3B). The sunflower plants presented high phytotoxicity values that ranged from 60% (sowing between 40 and 60

DAA) to 80% (sowing at the other times) (Figure 3C). These results are similar to those found by Guerra et al. (2014), who showed that sunflower was the most indaziflam-tolerant species of those evaluated, presenting only a slight decrease in the fresh weight of roots. Furthermore, even at the highest dose tested ( $100 \text{ g ha}^{-1}$ ), insufficient injury occurred to reach the  $I_{50}$  value (the dose of herbicide required to cause a 50% reduction in the plant fresh weight, relative to the weight obtained without herbicide treatment). Guerra et al. (2014) also found that maize and cotton had an intermediate tolerance to indaziflam, with an  $I_{50}$  of 86 and  $63.5 \text{ g ha}^{-1}$ , respectively.

Jhala and Hanson (2011), when studying the effect of



**Figure 3.** Phytotoxicity (%) of different indaziflam doses to *Glycine max* (A), *Crotalaria juncea* (B), *Helianthus annuus* (C) and *Arachis hypogaea* (D) at 32 DAS and 0, 10, 20 and 60 DAA.

indaziflam on sunflower, cotton, maize, soybean, millet, cucumber, beet and sorghum, observed that all the species studied were sensitive to indaziflam. The only symptom observed in the different species after planting in soil containing indaziflam was the nonemergence of seedlings, except for sunflower. Cotton and maize did not emerge only when sown in the soil treated with the highest dose of indaziflam (100 g ha<sup>-1</sup>). For soybean, emergence occurred in the soil treated with the two lowest doses (20 and 40 g ha<sup>-1</sup>); however, the plants died

after a few days. Additionally, sorghum, millet, cucumber and beet did not emerge, even in the soil treated with the lowest dose of this herbicide (20 g ha<sup>-1</sup>). The exact mechanisms of action of this herbicide are not yet fully understood but are believed to involve the prevention of cell wall formation in new cells, stopping plant growth. Indaziflam cannot be considered selective for peanuts at any of the tested doses or sowing times, since peanuts presented phytotoxicity between 60 and 85% (Figure 3D).

One of the options to reduce expenses during



sugarcane fallow that is important to remember is the adoption of MEIOSI (Inter-Rotational Methods of Simultaneous Occurrence - Método Inter Rotacional Ocorrendo Simultaneamente). According to Rocha Neto (2013), the MEIOSI system has been gaining prominence because the simultaneous rotation of sugarcane with a legume, such as soybean or peanut, or with green manure reduces the need for nitrogen fertilization and provides better planting logistics. The MEIOSI system consists of planting the sugarcane (September/October) in a 2:8 ratio, that is, two sugarcane rows: eight crop rows first occupied by the chosen legume within a sugarcane-free area. Subsequently, at the end of the rainy period (February/March), the first-planted sugarcane is used as a seedling and is planted in the area previously occupied by the legume. If green manure is used, the manure must be incorporated into the soil to provide nutrients to the sugarcane crop. However, for the MEIOSI system to work, the herbicide must be chosen with care to avoid carryover, which can occur with indaziflam.

When the soybean plants were sown in soils treated with saflufenacil, the biomass differed significantly between 0 and 10 DAA, with a lower accumulation at the highest dose used. Within each dose used, a statistically significant difference was observed among the sowing times at doses of 120, 60 and 30 g a.i. ha<sup>-1</sup>, with a higher biomass accumulation as the period between the herbicide application and the sowing time increased. For indaziflam, statistically significant differences were observed at all the doses tested, and the biomass produced declined as the herbicide dose increased (Table 6).

For the soils treated with saflufenacil, the leaf area of the soybean plants differed significantly compared with that of the control plants at 10 and 20 DAA, and the lowest leaf area was observed when saflufenacil was used at the commercial dose. For the soils treated with indaziflam, a statistically significant difference existed at all the sowing times, with a lower leaf area at the highest doses and reductions of up to 100% (Table 6). These results explain why Kuva and Salgado (2016), when evaluating the effect of indaziflam on weeds in a sugarcane crop, emphasized the need to consider the residual effect on rotational crops, avoiding intervals shorter than one year between indaziflam applications and the sowing of any crop.

No significant decrease in the chlorophyll content occurred with any of the saflufenacil treatments. By contrast, indaziflam reduced the chlorophyll content at all the sowing times, with differences detected among the commercial dose, one-half of that dose and one-quarter of that dose.

In the case of soybean, the effects of saflufenacil were only observed for the sowing times closest to the herbicide application. Soltani (2010) also reported that soybean and pea were the most tolerant crops

among the several crops tested. Regarding the development of crotalaria plants, the commercial dose of saflufenacil negatively affected the biomass production of plants sown at 0, 10 and 20 DAA. At 40 DAA, the lowest biomasses were observed at the lowest doses; however, these biomasses were the same as those of the control group. Thus, the herbicide was not responsible for the reduction (Table 7). By contrast, the use of indaziflam reduced the biomass of crotalaria plants at all the sowing times, especially at the commercial dose.

For the leaf area of crotalaria plants, saflufenacil negatively affected leaf expansion at 0 and 20 DAA with the use of the commercial dose and at 10 DAA with the commercial and the lowest doses. In turn, indaziflam affected the leaf area of crotalaria plants at all the sowing times (Table 7). Regarding the chlorophyll content, no statistically significant difference occurred between the groups treated with saflufenacil and the control group at any of the sowing times. With indaziflam, changes in the chlorophyll content were observed at 20, 40 and 60 DAA (Table 7).

For sunflower, saflufenacil affected shoot biomass accumulation not only at 10 DAA with the one-half and the full commercial dose but also at 40 DAA with the highest dose, in which case the plants died. Indaziflam negatively affected shoot biomass accumulation at 0, 10, 20 and 60 DAA at the highest doses used (Table 8).

Saflufenacil affected the leaf area of the sunflower plants at 0 and 10 DAA with the one-half and full commercial doses and at 40 DAA with the full commercial dose. Indaziflam, however, affected the leaf area of the sunflower plants at all the sowing times evaluated (Table 8). The chlorophyll content was lowest in sunflower plants with the use of one full commercial dose of saflufenacil at 40 and 60 DAA and at the commercial indaziflam dose when sowing was performed at 20 and 60 DAA (Table 8).

The biomass accumulation in the peanut plants that developed in the presence of a commercial saflufenacil dose presented statistically significant differences at 10 and 40 DAA. For the peanut plants sown in the soils treated with the commercial indaziflam dose, the biomass accumulation showed significant differences at all the sowing times (Table 9).

The leaf area of peanut plants was negatively affected by the highest dose of saflufenacil when sowing was performed at 10 and 20 DAA. For the commercial dose of indaziflam, a reduction in the leaf area of peanut plants was observed at all the sowing times (Table 9). For the chlorophyll content, significant differences occurred at 0 and 40 DAA in plants exposed to saflufenacil and at 20 and 60 DAA in the indaziflam-exposed plants.

In general, the phytotoxicity of saflufenacil decreased starting at 40 DAA in all crop species evaluated, which was expected because this herbicide has a half-life of one to five weeks. The same was not observed for indaziflam, whose half-life exceeds 150 days. Therefore,

**Table 6.** Dry biomass of aerial part (g), leaf area (cm<sup>2</sup>) and chlorophyll content of soybean plants sown at different times after application of saflufenacil and indaziflan (0, 10, 20, 40 and 60 DAA).

Dry biomass (g)											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	2.35 <sup>aA</sup>	2.87 <sup>aA</sup>	1.67 <sup>aA</sup>	1.58 <sup>aA</sup>	1.71 <sup>aA</sup>	0	2.10 <sup>aA</sup>	2.87 <sup>aA</sup>	0.66 <sup>abB</sup>	1.59 <sup>aB</sup>	1.71 <sup>aAB</sup>
7.5	1.91 <sup>aA</sup>	2.19 <sup>abA</sup>	1.53 <sup>aA</sup>	2.08 <sup>aA</sup>	2.17 <sup>aA</sup>	6.0	1.24 <sup>abAB</sup>	2.26 <sup>abA</sup>	0.67 <sup>abB</sup>	1.27 <sup>abAB</sup>	1.22 <sup>abAB</sup>
15	1.87 <sup>aA</sup>	2.39 <sup>abA</sup>	1.34 <sup>aA</sup>	1.80 <sup>aA</sup>	2.20 <sup>aA</sup>	12.5	0.71 <sup>bB</sup>	1.58 <sup>abcA</sup>	1.28 <sup>abA</sup>	1.29 <sup>abA</sup>	0.92 <sup>abA</sup>
30	1.58 <sup>aB</sup>	2.02 <sup>abAB</sup>	1.31 <sup>aB</sup>	1.28 <sup>aB</sup>	2.23 <sup>aA</sup>	25	0.19 <sup>bB</sup>	0.88 <sup>cAB</sup>	0.00 <sup>bB</sup>	1.59 <sup>aA</sup>	0.33 <sup>bB</sup>
60	1.61 <sup>aB</sup>	2.05 <sup>abA</sup>	1.27 <sup>aB</sup>	1.32 <sup>aB</sup>	1.79 <sup>aAB</sup>	50	0.24 <sup>bA</sup>	0.98 <sup>bcA</sup>	0.00 <sup>bA</sup>	0.92 <sup>abA</sup>	0.37 <sup>bA</sup>
120	1.28 <sup>bB</sup>	1.37 <sup>bB</sup>	0.85 <sup>aB</sup>	1.67 <sup>aA</sup>	2.02 <sup>aA</sup>	100	0.14 <sup>bA</sup>	0.66 <sup>cA</sup>	0.00 <sup>bA</sup>	0.07 <sup>bA</sup>	0.33 <sup>bA</sup>
VC %			33.69						55.20		
Leaf area (cm <sup>2</sup> )											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	78.98 <sup>aB</sup>	92.17 <sup>aA</sup>	136.10 <sup>aA</sup>	57.66 <sup>aB</sup>	63.17 <sup>aB</sup>	0	78.98 <sup>aB</sup>	102.17 <sup>aA</sup>	136.10 <sup>aA</sup>	51.23 <sup>abB</sup>	63.17 <sup>aB</sup>
7.5	66.42 <sup>aB</sup>	51.62 <sup>bB</sup>	123.01 <sup>aA</sup>	71.88 <sup>aB</sup>	88.52 <sup>aAB</sup>	6.0	66.29 <sup>abB</sup>	90.61 <sup>abA</sup>	48.06 <sup>bcB</sup>	29.58 <sup>abB</sup>	52.35 <sup>abB</sup>
15	81.49 <sup>aAB</sup>	56.15 <sup>bB</sup>	120.92 <sup>aA</sup>	65.90 <sup>aB</sup>	78.55 <sup>aAB</sup>	12.5	49.53 <sup>abcB</sup>	92.07 <sup>aA</sup>	65.02 <sup>bB</sup>	51.01 <sup>abB</sup>	31.06 <sup>abB</sup>
30	73.50 <sup>aAB</sup>	50.17 <sup>bB</sup>	101.12 <sup>abA</sup>	69.02 <sup>aAB</sup>	70.66 <sup>aAB</sup>	25	17.57 <sup>bcBC</sup>	68.89 <sup>bA</sup>	0.00 <sup>cC</sup>	57.42 <sup>aB</sup>	7.26 <sup>bC</sup>
60	51.66 <sup>aB</sup>	43.70 <sup>bB</sup>	104.68 <sup>abA</sup>	70.39 <sup>aAB</sup>	66.06 <sup>aAB</sup>	50	38.95 <sup>abB</sup>	69.97 <sup>bA</sup>	0.00 <sup>cB</sup>	36.94 <sup>abB</sup>	12.71 <sup>bB</sup>
120	69.63 <sup>aA</sup>	38.96 <sup>bA</sup>	64.94 <sup>bA</sup>	54.14 <sup>aA</sup>	63.82 <sup>aA</sup>	100	15.24 <sup>cB</sup>	47.78 <sup>cA</sup>	0.00 <sup>cB</sup>	7.65 <sup>bB</sup>	9.94 <sup>bB</sup>
VC%			28.50						37.33		
Chlorophyll content											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	34.55 <sup>aA</sup>	33.17 <sup>aA</sup>	38.90 <sup>aA</sup>	37.55 <sup>aA</sup>	40.10 <sup>aA</sup>	0	34.55 <sup>aA</sup>	33.17 <sup>aA</sup>	38.90 <sup>aA</sup>	37.55 <sup>aA</sup>	40.10 <sup>aA</sup>
7.5	35.67 <sup>aA</sup>	35.05 <sup>aA</sup>	38.02 <sup>aA</sup>	40.37 <sup>aA</sup>	41.35 <sup>aA</sup>	6.0	34.42 <sup>aA</sup>	34.12 <sup>aA</sup>	36.37 <sup>aA</sup>	26.52 <sup>aA</sup>	37.60 <sup>abA</sup>
15	37.02 <sup>aA</sup>	34.22 <sup>aA</sup>	37.82 <sup>aA</sup>	35.70 <sup>aA</sup>	36.35 <sup>aA</sup>	12.5	30.22 <sup>aA</sup>	35.17 <sup>aA</sup>	31.62 <sup>aA</sup>	35.80 <sup>aA</sup>	33.00 <sup>abA</sup>
30	33.82 <sup>aBC</sup>	32.20 <sup>aC</sup>	41.95 <sup>aA</sup>	40.27 <sup>aAB</sup>	37.07 <sup>aBC</sup>	25	27.90 <sup>aAB</sup>	35.42 <sup>aA</sup>	0.00 <sup>bC</sup>	31.72 <sup>aA</sup>	15.97 <sup>cdB</sup>
60	34.02 <sup>aA</sup>	34.67 <sup>aA</sup>	36.80 <sup>aA</sup>	37.87 <sup>aA</sup>	40.52 <sup>aA</sup>	50	25.62 <sup>aB</sup>	37.55 <sup>aA</sup>	0.00 <sup>bC</sup>	26.52 <sup>aAB</sup>	22.90 <sup>bcB</sup>
120	33.47 <sup>aAB</sup>	30.72 <sup>aB</sup>	35.57 <sup>aAB</sup>	35.85 <sup>aAB</sup>	39.17 <sup>aA</sup>	100	13.77 <sup>bA</sup>	20.45 <sup>bA</sup>	0.00 <sup>bB</sup>	7.57 <sup>bB</sup>	6.80 <sup>dB</sup>
VC %			9.72						25.26		

The averages followed by the same letter do not differ statistically from each other, lower case letters are compared vertically and upper case horizontal by the Tukey test 5%.

**Table 7.** Dry biomass of aerial part (g), leaf area (cm<sup>2</sup>) and chlorophyll content of crotalaria plants sown at different times after application of saflufenacil and indaziflan (0, 10, 20, 40 and 60 DAA).

Dry biomass (g)											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	0.57 <sup>aA</sup>	0.72 <sup>abA</sup>	0.46 <sup>aA</sup>	0.57 <sup>abA</sup>	0.48 <sup>aA</sup>	0	0.58 <sup>aA</sup>	0.72 <sup>abA</sup>	0.46 <sup>aA</sup>	0.57 <sup>abA</sup>	0.48 <sup>aA</sup>
7.5	0.34 <sup>aB</sup>	0.77 <sup>abA</sup>	0.41 <sup>aB</sup>	0.23 <sup>bB</sup>	0.52 <sup>aAB</sup>	6.0	0.60 <sup>aA</sup>	0.98 <sup>aA</sup>	0.52 <sup>aA</sup>	0.94 <sup>aA</sup>	0.49 <sup>aA</sup>
15	0.39 <sup>aA</sup>	0.47 <sup>bA</sup>	0.55 <sup>aA</sup>	0.24 <sup>bA</sup>	0.35 <sup>aA</sup>	12.5	0.14 <sup>bA</sup>	0.72 <sup>abA</sup>	0.11 <sup>aB</sup>	0.29 <sup>bAB</sup>	0.39 <sup>aAB</sup>
30	0.56 <sup>aB</sup>	0.92 <sup>aA</sup>	0.39 <sup>aB</sup>	0.38 <sup>bB</sup>	0.38 <sup>aB</sup>	25	0.15 <sup>bA</sup>	0.47 <sup>abA</sup>	0.33 <sup>aA</sup>	0.29 <sup>bA</sup>	0.17 <sup>aA</sup>
60	0.41 <sup>aB</sup>	1.04 <sup>aA</sup>	0.35 <sup>abB</sup>	0.32 <sup>bB</sup>	0.37 <sup>aB</sup>	50	0.15 <sup>bA</sup>	0.27 <sup>bA</sup>	0.38 <sup>aA</sup>	0.22 <sup>bA</sup>	0.45 <sup>aA</sup>
120	0.35 <sup>aB</sup>	0.53 <sup>bB</sup>	0.29 <sup>abB</sup>	0.95 <sup>aA</sup>	0.41 <sup>aB</sup>	100	0.20 <sup>abA</sup>	0.23 <sup>bA</sup>	0.01 <sup>bA</sup>	0.18 <sup>bA</sup>	0.09 <sup>abA</sup>
VC %	37.58					75.15					
Leaf area (cm <sup>2</sup> )											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	23.20 <sup>aA</sup>	40.06 <sup>aA</sup>	56.07 <sup>aB</sup>	22.28 <sup>aA</sup>	29.95 <sup>aA</sup>	0	23.20 <sup>aB</sup>	60.06 <sup>aA</sup>	56.07 <sup>aA</sup>	22.28 <sup>aB</sup>	29.95 <sup>aB</sup>
7.5	22.69 <sup>aB</sup>	42.67 <sup>aAB</sup>	40.21 <sup>abA</sup>	16.38 <sup>aB</sup>	30.13 <sup>aAB</sup>	6.0	28.85 <sup>aB</sup>	68.80 <sup>aA</sup>	17.90 <sup>bcB</sup>	18.61 <sup>aB</sup>	29.95 <sup>aB</sup>
15	19.07 <sup>aB</sup>	27.97 <sup>bAB</sup>	45.63 <sup>abA</sup>	12.29 <sup>aB</sup>	20.99 <sup>aB</sup>	12.5	24.95 <sup>aB</sup>	60.61 <sup>aA</sup>	11.80 <sup>bcB</sup>	18.77 <sup>aB</sup>	22.35 <sup>abB</sup>
30	20.39 <sup>aBC</sup>	52.42 <sup>aA</sup>	42.45 <sup>abA</sup>	16.94 <sup>aC</sup>	21.68 <sup>aBC</sup>	25	19.10 <sup>aB</sup>	62.07 <sup>aA</sup>	27.64 <sup>bB</sup>	16.99 <sup>aB</sup>	17.87 <sup>abB</sup>
60	21.92 <sup>aB</sup>	56.94 <sup>aA</sup>	32.02 <sup>abB</sup>	13.64 <sup>aB</sup>	35.30 <sup>aAB</sup>	50	18.60 <sup>aB</sup>	59.97 <sup>aA</sup>	11.64 <sup>bcB</sup>	13.01 <sup>aB</sup>	30.86 <sup>aB</sup>
120	18.76 <sup>abA</sup>	44.21 <sup>aA</sup>	32.06 <sup>abA</sup>	14.97 <sup>aA</sup>	29.14 <sup>aA</sup>	100	3.80 <sup>bB</sup>	45.54 <sup>abA</sup>	2.61 <sup>bcB</sup>	12.44 <sup>aB</sup>	13.73 <sup>abB</sup>
VC %	38.17					29.78					
Chlorophyll content											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	28.67 <sup>aA</sup>	30.17 <sup>aA</sup>	33.02 <sup>aA</sup>	35.50 <sup>aA</sup>	34.02 <sup>aA</sup>	0	28.67 <sup>aA</sup>	30.17 <sup>aA</sup>	33.02 <sup>aA</sup>	35.50 <sup>aA</sup>	34.02 <sup>aA</sup>
7.5	30.20 <sup>aA</sup>	30.05 <sup>aA</sup>	31.10 <sup>aA</sup>	34.80 <sup>aA</sup>	32.72 <sup>aA</sup>	6.0	35.40 <sup>aA</sup>	33.20 <sup>aAB</sup>	22.10 <sup>abB</sup>	33.60 <sup>abB</sup>	35.52 <sup>aA</sup>
15	28.02 <sup>aB</sup>	28.92 <sup>aB</sup>	32.92 <sup>aAB</sup>	31.95 <sup>aAB</sup>	37.40 <sup>aA</sup>	12.5	29.17 <sup>aAB</sup>	31.50 <sup>aAB</sup>	21.90 <sup>abB</sup>	30.12 <sup>abA</sup>	35.07 <sup>aA</sup>
30	32.32 <sup>aA</sup>	29.47 <sup>aA</sup>	28.22 <sup>aA</sup>	29.97 <sup>aA</sup>	31.05 <sup>aA</sup>	25	27.60 <sup>aAB</sup>	29.67 <sup>aAB</sup>	20.35 <sup>abB</sup>	33.27 <sup>abA</sup>	24.27 <sup>aAB</sup>
60	31.62 <sup>aA</sup>	32.27 <sup>aA</sup>	30.72 <sup>aA</sup>	33.00 <sup>aA</sup>	37.37 <sup>aA</sup>	50	26.97 <sup>aAB</sup>	29.75 <sup>aAB</sup>	17.47 <sup>bB</sup>	22.90 <sup>abB</sup>	34.82 <sup>aA</sup>
120	31.05 <sup>aAB</sup>	32.57 <sup>aAB</sup>	29.37 <sup>aAB</sup>	27.80 <sup>aB</sup>	37.20 <sup>aA</sup>	100	22.10 <sup>aBC</sup>	34.37 <sup>aA</sup>	0.00 <sup>cD</sup>	26.02 <sup>abA</sup>	8.87 <sup>bCD</sup>
VC%	13.37					24.77					

The averages followed by the same letter do not differ statistically from each other, lower case letters are compared vertically and upper case horizontal by the Tukey test 5%.

**Table 8.** Dry biomass of aerial part (g), leaf area (cm<sup>2</sup>) and chlorophyll content of sunflower plants sown at different times after application of saflufenacil and indaziflan (0, 10, 20, 40 and 60 DAA).

Dry biomass (g)											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	1.56 <sup>aA</sup>	1.83 <sup>aA</sup>	1.31 <sup>aA</sup>	1.29 <sup>bcA</sup>	1.31 <sup>aA</sup>	0	1.56 <sup>aA</sup>	1.84 <sup>aA</sup>	1.31 <sup>aA</sup>	1.29 <sup>aA</sup>	1.31 <sup>abA</sup>
7.5	0.98 <sup>abB</sup>	1.50 <sup>abB</sup>	1.57 <sup>abB</sup>	2.93 <sup>aA</sup>	1.29 <sup>abB</sup>	6.0	1.03 <sup>abA</sup>	1.46 <sup>abA</sup>	1.11 <sup>aA</sup>	1.29 <sup>aA</sup>	1.38 <sup>abA</sup>
15	1.68 <sup>aA</sup>	1.84 <sup>aA</sup>	1.04 <sup>aA</sup>	1.01 <sup>bcA</sup>	1.64 <sup>aA</sup>	12.5	0.15 <sup>cC</sup>	0.60 <sup>bcBC</sup>	1.34 <sup>aAB</sup>	1.41 <sup>aA</sup>	0.56 <sup>bcBC</sup>
30	1.19 <sup>aA</sup>	0.49 <sup>abcA</sup>	1.49 <sup>aA</sup>	1.60 <sup>abA</sup>	1.35 <sup>aA</sup>	25	0.90 <sup>abcA</sup>	0.77 <sup>bcA</sup>	0.65 <sup>aA</sup>	0.66 <sup>aA</sup>	0.63 <sup>bcA</sup>
60	0.59 <sup>abC</sup>	0.0 <sup>cC</sup>	1.02 <sup>abC</sup>	2.11 <sup>abA</sup>	1.64 <sup>aAB</sup>	50	0.52 <sup>bcA</sup>	1.12 <sup>abcA</sup>	0.80 <sup>aA</sup>	1.10 <sup>aA</sup>	0.67 <sup>bcA</sup>
120	1.67 <sup>aA</sup>	0.43 <sup>bcAB</sup>	0.72 <sup>aAB</sup>	0.00 <sup>cB</sup>	1.28 <sup>aA</sup>	100	0.55 <sup>bcA</sup>	0.43 <sup>cA</sup>	0.40 <sup>bA</sup>	0.99 <sup>aA</sup>	0.33 <sup>cA</sup>
VC %			49.05						39.02		
Leaf area (cm <sup>2</sup> )											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	94.01 <sup>ab</sup>	186.92 <sup>aA</sup>	140.88 <sup>ab</sup>	136.67 <sup>ab</sup>	81.41 <sup>ab</sup>	0	23.20 <sup>ab</sup>	60.06 <sup>aA</sup>	56.07 <sup>aA</sup>	22.28 <sup>ab</sup>	29.95 <sup>ab</sup>
7.5	101.90 <sup>ab</sup>	162.86 <sup>aA</sup>	101.80 <sup>ab</sup>	85.41 <sup>abB</sup>	96.92 <sup>ab</sup>	6.0	28.85 <sup>ab</sup>	68.80 <sup>aA</sup>	17.90 <sup>bcB</sup>	18.61 <sup>ab</sup>	29.95 <sup>ab</sup>
15	83.67 <sup>ab</sup>	199.77 <sup>bA</sup>	90.76 <sup>ab</sup>	75.19 <sup>abB</sup>	85.91 <sup>ab</sup>	12.5	24.95 <sup>ab</sup>	60.61 <sup>aA</sup>	11.80 <sup>bcB</sup>	18.77 <sup>ab</sup>	22.35 <sup>abB</sup>
30	110.60 <sup>aA</sup>	32.18 <sup>bB</sup>	80.10 <sup>aA</sup>	62.34 <sup>abA</sup>	35.30 <sup>ab</sup>	25	19.10 <sup>ab</sup>	62.07 <sup>aA</sup>	27.64 <sup>bB</sup>	16.99 <sup>ab</sup>	17.87 <sup>abB</sup>
60	38.21 <sup>abA</sup>	17.67 <sup>cA</sup>	47.79 <sup>aA</sup>	76.76 <sup>abA</sup>	29.15 <sup>aA</sup>	50	18.60 <sup>ab</sup>	59.97 <sup>aA</sup>	11.64 <sup>bcB</sup>	13.01 <sup>ab</sup>	30.86 <sup>ab</sup>
120	21.54 <sup>bB</sup>	25.09 <sup>bcB</sup>	66.01 <sup>aA</sup>	0.00 <sup>bB</sup>	81.93 <sup>aA</sup>	100	3.80 <sup>bB</sup>	45.54 <sup>abA</sup>	2.61 <sup>bcB</sup>	12.44 <sup>ab</sup>	13.73 <sup>abB</sup>
VC %			39.12						29.78		
Chlorophyll content											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	30.65 <sup>aA</sup>	32.17 <sup>aA</sup>	36.05 <sup>aA</sup>	35.70 <sup>aA</sup>	33.55 <sup>abA</sup>	0	30.65 <sup>aA</sup>	32.17 <sup>aA</sup>	36.05 <sup>aA</sup>	35.70 <sup>aA</sup>	33.55 <sup>aA</sup>
7.5	28.75 <sup>aA</sup>	30.65 <sup>aA</sup>	33.70 <sup>aA</sup>	36.15 <sup>aA</sup>	31.47 <sup>bcA</sup>	6.0	30.20 <sup>aA</sup>	31.95 <sup>aA</sup>	29.70 <sup>abA</sup>	31.05 <sup>aA</sup>	33.05 <sup>aA</sup>
15	31.30 <sup>aA</sup>	30.35 <sup>aA</sup>	30.75 <sup>aA</sup>	37.52 <sup>aA</sup>	35.92 <sup>aA</sup>	12.5	26.80 <sup>aA</sup>	30.65 <sup>aA</sup>	27.42 <sup>abA</sup>	30.32 <sup>aA</sup>	31.82 <sup>aA</sup>
30	28.70 <sup>ab</sup>	29.87 <sup>aAB</sup>	34.07 <sup>aAB</sup>	31.10 <sup>aAB</sup>	37.07 <sup>aA</sup>	25	29.00 <sup>aA</sup>	32.17 <sup>aA</sup>	29.17 <sup>abA</sup>	27.02 <sup>aA</sup>	23.00 <sup>abA</sup>
60	28.55 <sup>aA</sup>	29.55 <sup>aA</sup>	31.77 <sup>aA</sup>	13.42 <sup>bB</sup>	26.77 <sup>bcA</sup>	50	29.67 <sup>aA</sup>	32.52 <sup>aA</sup>	31.00 <sup>abA</sup>	26.30 <sup>aA</sup>	22.37 <sup>abA</sup>
120	23.45 <sup>ab</sup>	28.82 <sup>aAB</sup>	33.15 <sup>aA</sup>	0.00 <sup>cC</sup>	24.02 <sup>cB</sup>	100	28.70 <sup>aA</sup>	30.50 <sup>aA</sup>	18.57 <sup>bAB</sup>	24.45 <sup>aAB</sup>	14.90 <sup>bB</sup>
VC%			13.89						20.14		

The averages followed by the same letter do not differ statistically from each other, lower case letters are compared vertically and upper case horizontal by the Tukey test 5%.

**Tabela 9.** Dry biomass of aerial part (g), leaf area (cm<sup>2</sup>) and chlorophyll content of peanuts plants sown at different times after application of saflufenacil and indaziflan (0, 10, 20, 40 and 60 DAA).

Dry biomass (g)											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	2.71 <sup>aA</sup>	4.51 <sup>abA</sup>	2.84 <sup>aA</sup>	4.05 <sup>abA</sup>	1.41 <sup>aA</sup>	0	2.71 <sup>aA</sup>	4.51 <sup>aA</sup>	2.84 <sup>aA</sup>	4.06 <sup>aA</sup>	2.87 <sup>aA</sup>
7.5	2.85 <sup>aA</sup>	3.51 <sup>abA</sup>	1.85 <sup>aA</sup>	3.50 <sup>abA</sup>	1.78 <sup>abB</sup>	6.0	2.84 <sup>aA</sup>	2.97 <sup>aA</sup>	0.34 <sup>abB</sup>	1.77 <sup>abAB</sup>	1.65 <sup>abAB</sup>
15	2.33 <sup>aAB</sup>	3.99 <sup>abA</sup>	1.44 <sup>abB</sup>	2.93 <sup>abAB</sup>	1.58 <sup>abB</sup>	12.5	2.77 <sup>aAB</sup>	4.09 <sup>aA</sup>	0.69 <sup>abB</sup>	1.67 <sup>abB</sup>	1.86 <sup>abAB</sup>
30	2.06 <sup>abB</sup>	5.16 <sup>aA</sup>	1.58 <sup>abB</sup>	2.72 <sup>abB</sup>	2.58 <sup>aA</sup>	25	2.22 <sup>aAB</sup>	2.89 <sup>aA</sup>	0.31 <sup>bB</sup>	1.21 <sup>bAB</sup>	0.00 <sup>bB</sup>
60	2.82 <sup>aAB</sup>	4.80 <sup>abA</sup>	1.08 <sup>abB</sup>	1.94 <sup>bB</sup>	2.77 <sup>aAB</sup>	50	2.97 <sup>aA</sup>	2.99 <sup>aA</sup>	0.40 <sup>abB</sup>	2.06 <sup>abAB</sup>	0.35 <sup>bB</sup>
120	3.04 <sup>aA</sup>	2.74 <sup>bA</sup>	1.51 <sup>aA</sup>	2.12 <sup>bA</sup>	2.88 <sup>aA</sup>	100	0.77 <sup>bAB</sup>	2.08 <sup>bA</sup>	0.30 <sup>bAB</sup>	0.88 <sup>bAB</sup>	0.12 <sup>bB</sup>
VC%	39.07					60.14					
Leaf area (cm <sup>2</sup> )											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	99.97 <sup>abB</sup>	212.92 <sup>aA</sup>	80.54 <sup>bB</sup>	104.30 <sup>abB</sup>	86.58 <sup>abB</sup>	0	99.98 <sup>aA</sup>	142.93 <sup>aA</sup>	40.54 <sup>abB</sup>	104.30 <sup>aA</sup>	86.59 <sup>abB</sup>
7.5	99.21 <sup>aAB</sup>	177.67 <sup>aA</sup>	109.33 <sup>aA</sup>	53.87 <sup>abB</sup>	83.53 <sup>aAB</sup>	6.0	96.78 <sup>aAB</sup>	124.33 <sup>aA</sup>	47.0 <sup>abB</sup>	48.29 <sup>aAB</sup>	80.23 <sup>abAB</sup>
15	77.05 <sup>aA</sup>	145.26 <sup>abA</sup>	86.53 <sup>abA</sup>	62.71 <sup>aA</sup>	62.33 <sup>aA</sup>	12.5	99.32 <sup>aAB</sup>	126.06 <sup>aA</sup>	50.38 <sup>aAB</sup>	43.25 <sup>abB</sup>	85.82 <sup>abAB</sup>
30	87.62 <sup>aAB</sup>	146.34 <sup>abA</sup>	111.42 <sup>abA</sup>	37.78 <sup>abB</sup>	67.69 <sup>aAB</sup>	25	79.01 <sup>aAB</sup>	114.34 <sup>aA</sup>	36.58 <sup>aAB</sup>	38.18 <sup>aAB</sup>	0.00 <sup>bB</sup>
60	90.17 <sup>aA</sup>	114.57 <sup>abA</sup>	81.28 <sup>abA</sup>	35.90 <sup>aA</sup>	54.22 <sup>aA</sup>	50	78.16 <sup>abB</sup>	107.91 <sup>aA</sup>	44.06 <sup>aAB</sup>	20.81 <sup>abB</sup>	36.31 <sup>abAB</sup>
120	85.75 <sup>aA</sup>	113.54 <sup>abA</sup>	113.40 <sup>aA</sup>	17.81 <sup>aA</sup>	71.66 <sup>aA</sup>	100	40.61 <sup>bAB</sup>	108.25 <sup>aA</sup>	19.76 <sup>bB</sup>	18.30 <sup>bB</sup>	11.79 <sup>abB</sup>
VC %	56.12					56.14					
Chlorophyll content											
Saflufenacil						Indaziflan					
Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA	Doses g a.i ha <sup>-1</sup>	0 DAA	10 DAA	20 DAA	40 DAA	60 DAA
0	39.07 <sup>abA</sup>	37.70 <sup>aA</sup>	46.35 <sup>aA</sup>	36.85 <sup>abA</sup>	43.85 <sup>aA</sup>	0	39.07 <sup>aA</sup>	37.70 <sup>aA</sup>	46.35 <sup>aA</sup>	36.85 <sup>aA</sup>	43.85 <sup>aA</sup>
7.5	42.00 <sup>abA</sup>	33.85 <sup>aA</sup>	43.05 <sup>aA</sup>	32.82 <sup>abA</sup>	41.10 <sup>aA</sup>	6.0	41.75 <sup>aA</sup>	37.80 <sup>aA</sup>	28.10 <sup>bcA</sup>	40.62 <sup>aA</sup>	37.50 <sup>aA</sup>
15	40.75 <sup>abA</sup>	37.60 <sup>aA</sup>	41.35 <sup>aA</sup>	41.75 <sup>aA</sup>	39.72 <sup>aA</sup>	12.5	42.60 <sup>aA</sup>	36.47 <sup>aAB</sup>	23.10 <sup>bcB</sup>	38.32 <sup>aAB</sup>	38.87 <sup>aAB</sup>
30	39.02 <sup>abA</sup>	43.40 <sup>aA</sup>	46.27 <sup>aA</sup>	40.92 <sup>aA</sup>	40.90 <sup>aA</sup>	25	40.60 <sup>aA</sup>	39.15 <sup>aA</sup>	18.05 <sup>bcB</sup>	38.77 <sup>aA</sup>	0.00 <sup>bB</sup>
60	33.60 <sup>bAB</sup>	36.22 <sup>aA</sup>	40.97 <sup>aA</sup>	35.32 <sup>abA</sup>	37.45 <sup>aA</sup>	50	37.97 <sup>aA</sup>	41.52 <sup>aA</sup>	13.42 <sup>cB</sup>	29.67 <sup>aAB</sup>	5.22 <sup>bC</sup>
120	36.62 <sup>abA</sup>	33.20 <sup>aAB</sup>	41.77 <sup>aA</sup>	26.35 <sup>bB</sup>	37.42 <sup>aAB</sup>	100	36.30 <sup>aA</sup>	42.00 <sup>aA</sup>	13.57 <sup>cBC</sup>	34.17 <sup>aA</sup>	7.30 <sup>bB</sup>
VC%	15.21					28.19					

The averages followed by the same letter do not differ statistically from each other, lower case letters are compared vertically and upper case horizontal by the Tukey test 5%.

the pre-emergent herbicide to be applied during the last sugarcane harvest must be chosen carefully when adopting crop rotation during sugarcane fallow periods.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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*Full Length Research Paper*

# Integrated crop management (ICM) for increasing rice production in Barind area

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An experiment was conducted at the Agronomy Field Laboratory, University of Rajshahi during the period from June, 2015 to December 2015 to study the effect of Integrated Crop Management (ICM) practice for increasing rice production in Barind area, Bangladesh. The experiment consisted of two factors that is, two variety which is BRRI dhan56 and BRRI dhan57, and five management practices like control, only weed management, only pest management, farmers practices and ICM practice. The experiment was laid out in Randomized Completely Block Design (RCBD) with three replications. Among the management practices, ICM gave the highest number of tillers plant<sup>-1</sup>, effective tillers plant<sup>-1</sup>, panicle length, number of grains panicle<sup>-1</sup> and 1000-grain weight, and the lowest results were found in control. Between two varieties, BRRI dhan 56 produced the highest yield components like effective tillers plant<sup>-1</sup>, number of grains panicle<sup>-1</sup> and 1000-grain weight than BRRI dhan 57. BRRI dhan 56 produced the highest grain yield than BRRI dhan 57 when the field was treated with ICM. So it can be concluded that the farmers are advised to cultivate BRRI dhan 56 and adopt ICM for maximizing rice production in Barind area in Bangladesh.

**Key words:** Rice, variety, Integrated Crop Management (ICM), yield, Barind area.

## INTRODUCTION

Geographically, Bangladesh is highly vulnerable to climate change. In particular, impact of climate variability on the agriculture and consequence on different other sectors are already evident in the drought prone High Barind tract regions. The agriculture sectors in the High Barind Tract regions are very likely to face significant yield reduction due to climate change in future. Moisture capacity of High Barind Tract soil is poor due to critical

organic matter contents and low infiltration of water. These situation make the area drought prone along with poor crop productivity.

Moreover, recent report indicates that ground water level of High Barind Tract is rapidly falling due to over exploitation of deep tube well. Transplant aman rice is the major crop which suffered regularly due to early and late drought, and planting of post rainy crop. During that time,

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the drought resistant varieties can be cultivated under rainfall condition. Drought tolerant rice varieties provide food security for farmers in Bangladesh (Habiba et al., 2013). The yield of rice depends on many factors such as varieties and suitable agronomic practices. Varieties play an important role in producing high yield of rice. The growth process of rice plant under a given agro climatic condition differs with variety (Sarker et al., 2014).

Generally, the farmers apply different agro-chemicals such as fungicide, insecticide, herbicide etc in the field indiscriminately. They also used chemical fertilizer in higher dose with a view to increase the yield of rice. Continuous application of all these agro-chemicals results in the reduction of soil fertility and productivity, soil erosion, death of beneficial insect, reduction of biodiversity, occurrence of environmental pollution and disturbance of ecological balance which is very much dangerous and a great threat for mankind in the question of their survival on the earth (Hossain and Siddique, 2015).

Integrated Crop Management (ICM) is the best way for solving all of the aforementioned problems. It combines the best of traditional methods with appropriate modern technology, balancing the economic production of crops with positive environmental management. ICM practice plays a significant role in producing higher yield of rice among the different practices. It also helps in the maintenance of soil structure and fertility, improvement of soil fertility, prevent buildup of pests, diseases and weeds, prevent damage to soil, water, avoid loss of biodiversity and reduce environmental damage and production cost, while majority of the farmers are not generally not aware and not following ICM practices. Considering the aforementioned, the present study was undertaken with the following objectives:

1. To determine the suitable management practices for rice.
2. To produce the rice in an environmental friendly way.

## MATERIALS AND METHODS

The research was conducted at the Agronomy Field laboratory, University of Rajshahi, Bangladesh during the period from June to December, 2015. The experimental field was a medium high land with silty loam textured soil having pH value of 7.8. Status of nitrogen, phosphorus and cation exchange capacity was medium. The soil properties of the field were organic matter 1.8%, total available nitrogen 0.04%, available phosphorus 11.25 ppm, available potassium 58ppm and available sulphur 25.65 ppm. The treatment of the experiment include two drought tolerant varieties namely BRR1 dhan 56 and BRR1 dhan 57, and five management practices; control, only weed management, only pest management, farmer's practice and integrated crop management ICM. Components of ICM are seed management, soil test and fertility maintenance, modern cultivation practices, and integrated pest management. The experiment was laid out in randomized complete block design (RCBD) with three replications. The unit plot size was 10m<sup>2</sup>. (4 m x 2.5 m). The seed were sown in nursery bed on 18 June, 2015. The experimental plots were uniformly fertilized with

nitrogen, phosphorus, potash, sulphur and zinc fertilizers as recommended dose. One third nitrogenous fertilizer (urea) and all other fertilizers were applied during final land preparation as basal dose. The 2/3 urea were applied on top dressing in two equal splits, first at 30 DAT and 2nd at flowering stage. Finally, 30 days old seeding of two varieties were transplanted in the well puddled plots with three seedlings hill<sup>-1</sup> on 17 July, 2015. Data were recorded on yield and yield components like total tillers plant<sup>-1</sup>, effective tillers plant<sup>-1</sup>, non-effective tillers plant<sup>-1</sup>, panicle length, spikelets panicle<sup>-1</sup>, grains panicle<sup>-1</sup>, 1000-grain weight, grain yield, straw yield, biological yield and harvest index. Data were analysed following the analysis of variance (ANOVA) technique, and mean differences were adjudged by Duncan's New multiple range test (DMRT) (Gomez and Gomez, 1984), with the help of computer Package MSTAT-C.

## RESULTS AND DISCUSSION

The results showed that effective tillers plant<sup>-1</sup>, number of grains panicle<sup>-1</sup>, 1000-grain weight, grain yield and biological yield had significant effect on variety. From Tables 1 to 3, the result revealed that variety had no significant effect on total tillers plant<sup>-1</sup>, non-effective tillers plant<sup>-1</sup>, panicle length, spikelets panicle<sup>-1</sup>, straw yield and harvest index. The maximum number of effective tillers plant<sup>-1</sup>, number of grains panicle<sup>-1</sup> and 1000-grain weight were observed from BRR1 dhan 56. Among two varieties, BRR1 dhan 56 produced the higher grain yield and biological yield (BRR1, 2013). This result was obtained for genetic makeup of the variety.

Different crop management practices also had a significant effect on most of the yield and yield contributing character like total tillers plant<sup>-1</sup>, panicle length, number of spikelets panicle<sup>-1</sup>, 1000-grain weight, number of grains panicle<sup>-1</sup>, grain yield, straw yield, biological yield and harvest index. The result showed that the field which received integrated crop management gave the highest total tillers plant<sup>-1</sup>, effective tillers plant<sup>-1</sup>, panicle length, number of spikelets panicle<sup>-1</sup>, 1000-grain weight, number of grains panicle and harvest index.

The maximum grain yield, straw yield and biological yield were obtained from the field that practiced ICM. This results were found because all favourable conditions received the field ICM practice. This result was supported by Wang et al. (2017). The interaction of variety and different management practices had no significant influence on yield and yield components. The effective tillers plant<sup>-1</sup>, panicle length, spikelets panicle<sup>-1</sup>, grains panicle<sup>-1</sup>, and 1000-grain weight were positively correlated with grain yield.

From the aforementioned discussion, it could be concluded that farmers are suggested to cultivate transplant aman rice BRR1 dhan 56 in the field which is managed by ICM practices for maximizing grain yield.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.



**Table 1.** Effect of variety and management practices on yield and yield components of transplant aman.

Variety	No. of total tillers plant <sup>-1</sup>	No. of effective tillers plant <sup>-1</sup>	No. of non-effective tillers plant <sup>-1</sup>	Panicle length (cm)	No. of spikelets panicle <sup>-1</sup>	No. of grains panicle <sup>-1</sup>	1000 grain weight (g)	Grain yield (t ha <sup>-1</sup> )	Straw yield (t ha <sup>-1</sup> )	Biological yield (t ha <sup>-1</sup> )	Harvest index (%)
BRR1 dhan 56	10.24	8.40 <sup>a</sup>	1.84	19.87	10.12	61.65 <sup>a</sup>	29.07 <sup>a</sup>	3.98 <sup>a</sup>	4.93	8.39 <sup>a</sup>	46.25
BRR1 dhan 57	9.86	7.59 <sup>b</sup>	1.27	19.78	10.05	58.06 <sup>b</sup>	24.17 <sup>b</sup>	3.79 <sup>b</sup>	4.40	8.19 <sup>b</sup>	45.96
LS	NS	0.01	NS	NS	NS	0.01	0.01	0.05	NS	0.01	NS
<b>Management practice</b>											
M <sub>0</sub>	8.76 <sup>c</sup>	6.66 <sup>c</sup>	2.52	18.80 <sup>b</sup>	9.78	56.91 <sup>c</sup>	25.82 <sup>c</sup>	2.42 <sup>d</sup>	3.28 <sup>d</sup>	5.7 <sup>d</sup>	42.50 <sup>b</sup>
M <sub>1</sub>	9.76 <sup>abc</sup>	7.88 <sup>b</sup>	1.16	19.77 <sup>b</sup>	9.88	58.56 <sup>bc</sup>	26.51 <sup>abc</sup>	3.61 <sup>c</sup>	3.95 <sup>c</sup>	7.66 <sup>c</sup>	46.41 <sup>a</sup>
M <sub>2</sub>	9.49 <sup>bc</sup>	7.65 <sup>bc</sup>	2.58	18.91 <sup>b</sup>	9.98	57.76 <sup>c</sup>	26.30 <sup>bc</sup>	3.53 <sup>c</sup>	4.06 <sup>c</sup>	7.47 <sup>c</sup>	47.00 <sup>a</sup>
M <sub>3</sub>	10.72 <sup>ab</sup>	8.77 <sup>ab</sup>	2.02	19.63 <sup>b</sup>	10.14	60.84 <sup>b</sup>	27.03 <sup>ab</sup>	4.633 <sup>b</sup>	5.35 <sup>b</sup>	9.98 <sup>b</sup>	47.14 <sup>a</sup>
M <sub>4</sub>	11.33 <sup>a</sup>	9.77 <sup>a</sup>	2.00	22.01 <sup>a</sup>	10.66	65.20 <sup>a</sup>	27.47 <sup>a</sup>	5.04 <sup>a</sup>	5.58 <sup>a</sup>	10.62 <sup>a</sup>	47.46 <sup>a</sup>
LS	0.01	0.01	NS	0.01	NS	0.01	0.01	0.01	0.01	0.01	0.01

In a column, figures having similar letters (s) or without letters (s) do not differ significantly, whereas figures that are having dissimilar letters (s) differ significantly having 1% level of probability (as per DMRT). NS = Non significant; M<sub>0</sub> = No Management; M<sub>1</sub> = Only weed management; M<sub>2</sub> = Only pest management; M<sub>3</sub> = Farmers practices; M<sub>4</sub> = Integrated crop management; LS = Level of significance.

**Table 2.** Interaction effect of variety and management practices on yield and yield components of transplant aman rice.

Treatment	No. of total tillers plant <sup>-1</sup>	No. of effective tillers plant <sup>-1</sup>	No. of non-effective tillers plant <sup>-1</sup>	Panicle length (cm)	No. of spikelets panicle <sup>-1</sup>	No. of grains panicle <sup>-1</sup>	1000 grain weight (g)	Grain yield (t ha <sup>-1</sup> )	Straw yield (t ha <sup>-1</sup> )	Biological yield (t ha <sup>-1</sup> )	Harvest index (%)
V <sub>1</sub> M <sub>0</sub>	8.98	6.66	3.22	18.84	9.78	57.50	28.40	2.46	3.26	5.72	43.00
V <sub>1</sub> M <sub>1</sub>	9.66	7.65	1.22	19.61	10.22	59.10	28.81	3.66	4.10	7.76	47.16
V <sub>1</sub> M <sub>2</sub>	9.55	7.87	2.66	18.66	9.72	58.76	29.00	3.58	4.00	7.58	47.22
V <sub>1</sub> M <sub>3</sub>	10.44	8.89	2.22	20.06	10.27	63.18	29.47	4.68	5.43	10.11	46.29
V <sub>1</sub> M <sub>4</sub>	10.66	9.55	2.05	22.20	10.61	69.61	29.70	5.11	5.66	10.77	47.44
V <sub>2</sub> M <sub>0</sub>	8.55	6.66	1.83	18.76	9.78	56.32	23.23	2.38	3.30	5.68	41.90
V <sub>2</sub> M <sub>1</sub>	10.22	8.11	1.11	19.93	9.55	57.92	24.20	3.55	4.03	7.58	46.83
V <sub>2</sub> M <sub>2</sub>	9.44	7.42	2.50	19.16	10.23	56.76	23.60	3.46	3.90	7.36	47.01
V <sub>2</sub> M <sub>3</sub>	11.00	8.66	1.83	19.20	10.01	58.50	25.47	4.58	5.26	9.84	46.54
V <sub>2</sub> M <sub>4</sub>	12.00	9.99	1.94	21.83	10.70	60.78	24.37	4.96	5.50	10.46	47.42
LS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

NS = Non significant; V<sub>1</sub>= BRR1 dhan56; V<sub>2</sub> = BRR1 dhan57; M<sub>0</sub> = No Management; M<sub>1</sub> = Only weed management; M<sub>2</sub> = Only pest management; M<sub>3</sub> = Farmers practices; M<sub>4</sub> = Integrated crop management'; LS = Level of significance.

**Table 3.** Simple correlation coefficient between yield and yield components of transplant aman rice.

Treatment	No. of total tillers plant <sup>-1</sup>	No. of effective tillers plant <sup>-1</sup>	No. of non-effective tillers plant <sup>-1</sup>	Panicle length (cm)	No. of spikelets panicle <sup>-1</sup>	No. of grains spike <sup>-1</sup>	1000grain weight (g)	Grain yield (t ha <sup>-1</sup> )
No. of total tillers plant <sup>-1</sup>	1	0.782**	0.041	0.443*	0.283	0.253	-0.040	0.657**
No. of effective tillers plant <sup>-1</sup>	-	1	-0.130	0.639**	0.339	0.526*	0.162	0.842**
No. of non-effective tillers plant <sup>-1</sup>	-	-	1	-0.226	0.288	0.053	0.119	-0.150
Panicle length (cm)	-	-	-	1	0.207	0.425*	0.189	0.518**
No. of spikelets panicle <sup>-1</sup>	-	-	-	-	1	0.317	0.131	0.318
No. of grains panicle <sup>-1</sup>	-	-	-	-	-	1	0.557**	0.689**
1000grain weight (g)	-	-	-	-	-	-	1	0.268
Grain yield (t ha <sup>-1</sup> )	-	-	-	-	-	-	-	1

\*\*Correlation is significant at the 0.01 level (2-tailed).

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