African Journal of Agricultural Research

Volume 12 Number 7 16 February 2017
ISSN 1991-637X

Academic Journals

ABOUT AJAR

The African Journal of Agricultural Research (AJAR) is published weekly (one volume per year) by Academic Journals.

African Journal of Agricultural Research (AJAR) is an open access journal that publishes high-quality solicited and unsolicited articles, in English, in all areas of agriculture including arid soil research and rehabilitation, agricultural genomics, stored products research, tree fruit production, pesticide science, postharvest biology and technology, seed science research, irrigation, agricultural engineering, water resources management, marine sciences, agronomy, animal science, physiology and morphology, aquaculture, crop science, dairy science, entomology, fish and fisheries, forestry, freshwater science, horticulture, poultry science, soil science, systematic biology, veterinary, virology, viticulture, weed biology, agricultural economics and agribusiness. All articles published in AJAR are peer- reviewed.

Contact Us

Editorial Office: ajar@academicjournals.org

Help Desk: helpdesk@academicjournals.org

Website: http://www.academicjournals.org/journal/AJAR

Submit manuscript online http://ms.academicjournals.me/

Editors

Editorial Board

African Journal of Agricultural Research

Table of Contents: Volume 12 Number 7, 16 February, 2017

ARTICLES

African Journal of Agricultural Research

Table of Contents: Volume 12 Number 7, 16 February, 2017

ARTICLES

academicJournals

Vol. 12(7), pp. 459-466, 16 February, 2017
DOI: 10.5897/AJAR2016.11817
Article Number: 3AC100162757
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Leaf area of sugarcane varieties and their correlation with biomass productivity in three cycles

Vinicius Santos Gomes da Silva[1]*, Mauro Wagner de Oliveira[2], Terezinha Bezerra Albino Oliveira[2], Bruno Campos Mantovanelli[1], Abraão Cícero da Silva[1], Adrielle Naiana Ribeiro Soares[3] and Paulo Ricardo Aprígio Clemente[2]

[1]Departamento de Agronomia, Universidade Federal Rural de Pernambuco, Av. Dom Manoel de Medeiros, s/n, 52171-900, Recife, Pernambuco, Brazil.
[2]Centro de Ciências Agrárias, Universidade Federal de Alagoas, BR 104, Km 85, s/n, 57100-000. Rio Largo, Alagoas, Brazil.
[3]Departamento de Agricultura e Biodiversidade, Universidade Federal de Sergipe, - UFS, São Cristóvão, Brazil.

Received 11 October, 2016; Accepted 19 January, 2017

The objective of this work is to study the leaf area and biomass production of 4 sugarcane varieties in cane plant cycles, first and second regrowth. The experiment was conducted at Fazenda Jequiá in the state of Alagoas, Brazil. A randomized block design with 5 replications was used. The treatments were 4 cultivars: RB92579, SP813250, RB867515 and VAT90212. The length and width of the sheet + 3 were determined as well as the following parameters; the number of tillers per square meter, the number of green leaves, leaf area, and leaf area index in most growing sugarcane in the three crop cycles. The productivity of shoot dry biomass was determined at the time of cane maturity in 3 cycles. Univariate and multivariate analyses were performed. The larger leaf area index of 4.46 m^2 m^{-2} was observed for RB92579 in plant cane cycle. The dry biomass yield was not influenced by varieties having average values of 47, 41 and 31 t ha^{-1} in the sugarcane plant cycles in the first and second regrowth, respectively. The principal component analysis enabled us to identify from Current Population Survey (CPs) variance which component can contribute to the explanation of the data.

Key words: Dry matter accumulation, leaf area index (LAI), principal component analysis.

INTRODUCTION

Sugarcane is among the largest crops in Brazil; the lower acreage was used for only soybeans and corn. The 2015 harvest of planted area of cane was 8.6 million hectares, while soybean and corn area was 33 million

*Corresponding author. E-mail: vinicius.agro2008.1@gmail.com. Tel: 55-81-33206237. Fax: 55-81-33206200.

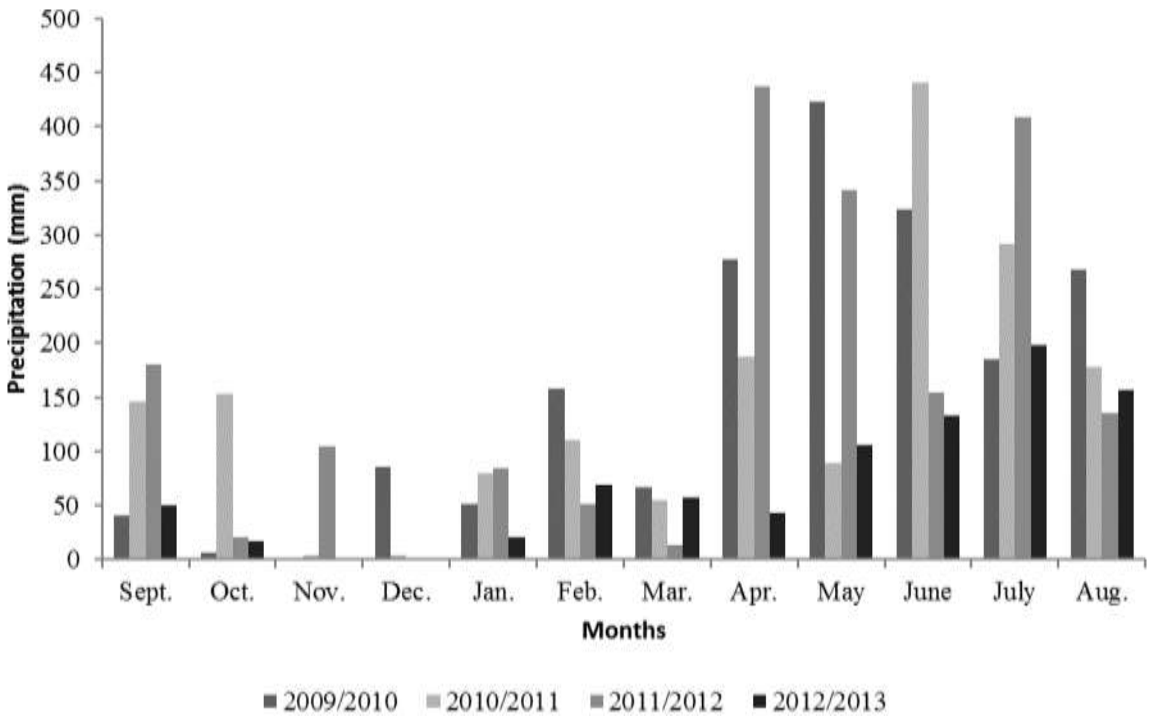


Figure 1. Monthly precipitation during the period studied.

and 15 million hectares, respectively (Ibge, 2016). This average productivity is 76 t ha^{-1}, which corresponds to less than 25% of the biological potential of the crop (Landell et al., 2008; Conab, 2016). Thus, agricultural practices such as liming, gypsum, chemical fertilizer, irrigation, inoculating with endophytic bacteria, use of compost and green manures have been taken by the sugar-energy sector to increase the productivity of sugarcane, making it a more competitive culture (Oliveira et al., 2007b; Bastos et al., 2016; Cunha et al., 2016; Pedula et al., 2016; Silva et al., 2016). Another practice that deserves attention is the choice of most productive sugarcane varieties that better adapt to specific soil and climatic conditions. This technology when properly employed helps to increase the productivity of labor, work, fertilizer efficiency and other inputs, with consequent reduction in costs and increase in the production system efficiency.

There are several varieties of sugarcane with good agronomic and industrial features such as: High response to improving soil fertility, upright growth and resistance to tipping, which facilitates harvest; high productivity stems; force of sprouts, resistance to pests and diseases, and broth with high sucrose content and easy industrialization. Regarding the characteristics of varieties, one of which most relate to agricultural income is the leaf area, due to its relationship with photosynthetic capacity. Increased leaf area provides an increase in the plant's ability to harness solar energy in order to perform photosynthesis and thus can be used to evaluate the productivity (Reis et al., 2013).

Leme et al. (1984) report that the leaf area index (LAI) is effective to evaluate the end yield, and the highest values during the development cycle would be related to the higher end production of culms. In this sense, the knowledge of the dynamics of leaf area and leaf of the system architecture in different cultivars of sugarcane may allow a better understanding of the relationship of these characteristics with the final yield.

The objective of this study is to evaluate the leaf area in the phase of maximum growth of four varieties of sugarcane, in the cycles of plant cane, first and second regrowth in Alagoas wild, correlating its effect on the production of dry biomass of the aerial part.

MATERIALS AND METHODS

The research was conducted in the municipality of Anadia, wild Alagoas, with Latitude 09° 41'04 "S and longitude 36° 18'15" W. The climate of the study area is tropical rainy, with dry summers, according to Koppen classification; its average annual rainfall is 1500 mm (Figure 1) and has average annual temperature of 29°. The terrain varies from flat to gently rolling.

The soil of the experimental area is classified as Yellow Latosol (Embrapa, 2013). It has medium texture, whose chemical characterization was carried out on soil samples collected in layers from 0.0 to 0.2 and 0.2 to 0.4 m (Table 1). The correction of soil

Table 1. Chemical analyses of soil samples at the layers 0-20 and 20-40 cm.

Layers	pH	P	K	Ca	Mg	Al	H+Al	SB	T
cm		**mg dm^{-3}**				**cmolc dm^{-3}**			
00-20	5.9	103.0	40	1.8	0.8	0.0	3.80	2.70	6.5
20-40	5.0	21.6	20	0.6	0.3	0.6	4.62	0.95	5.57
Prof	**t**	**V**	**m**	**MO**	**Zn**	**Fe**	**Mn**	**Cu**	**B**
cm	**cmolc dm^{-3}**	**%**		**dag kg^{-1}**			**mg dm-^{3}**		
00-20	2.70	42	0	1.8	2.5	75.6	9.7	1.1	0.4
20-40	1.55	17	39	0.8	0.4	53.4	0.3	0.2	0.3

pH in H_2O (Ratio 1:2.5). P K, Fe, Zn, Mn, and Cu: Mehlich extractor. Ca, Mg and Al: KCl extractor. H+Al: Calcium acetate extractor. B: Hot water extractor; S: Monocalcium phosphate in acetic acid extractor.

acidity was performed using dolomitic limestone at a dose of 150 kg ha^{-1}, calculated by the method which seeks to increase the base saturation to 60%, according to Oliveira et al. (2007a). After application of the calcareous, the soil was plowed and meshed; then the grooves were opened. Planting density fluctuated around 15 to 18 buds per meter of furrow.

The experimental design was randomized blocks, with 5 replications, consisting of 4 varieties of sugarcane: SP813250, RB867515, RB92579 and VAT90212 grown in 6 furrows of 10.0 m long portions, with space of 1 m, totaling 60 m^2 of total area. It was considered useful area of each plot, the 4 central lines with 6 meters long, totaling 24 m^2.

The fertilization of the soil was based on the recommendation of the Triunfo plant, according to the results of soil analysis (Table 1); 60, 100 and 150 kg ha^{-1} of N, P_2O_5 and K_2O were applied at the bottom of the groove. The plant cane cycle was harvested at 14 months, after the sugarcane harvest the search in the first and second regrowth continued, with each having a duration of 12 months. These cycles treatments received 500 kg ha^{-1} of formula 20-05-25.

The evaluation of leaf area was done during the maximum growth stage of the plants, in the plant cane cycle 8 months after planting and regrowth, 6 months after cutting of plant cane and first regrowth, respectively. For this evaluation, the number of green leaves was counted (fully expanded sheet with a minimum of 20% of green area, from the +1 sheet) and the leaves +3 were measured. The length and width of the third middle part of the leaf blade were obtained (Hermann and Câmara, 1999): AF = L × W × 0.75 × (N + 20), where C is the length of the sheet +3, W is the width of the sheet +3, 0.75 is the correction factor for the crop leaf area, and N is the number of expanded leaves at least 20% green area. For tiller number and counting, the number of green sheets of each tiller was also sampled at 1 m groove.

The production of dry biomass of shoot was done when the sugarcane had matured at 14 months after planting the sugarcane plant, and during the first and second regrowth at 12 months. The determination was performed by harvesting 2 m^2 of each plot and their extrapolation to 1 ha; after the determination of the fresh matter, subsamples of all shoot in each treatment was passed into chopper forage and dried in forced ventilation oven at 65ºC to constant weight.

The univariate and multivariate analyses were performed. Univariate statistical analyses were performed with the computer program Sisvar (Ferreira, 2008). The variables were subjected to analysis of variance by F test and those where the F was significant were compared to the averages using Scott Knott test at 5% probability. In order to group the variables into a more meaningful set (represented by the components) and identify which variables belong to which components and how each variable explains each component, the study of Principal Component Analysis (PCA) was done. Thus, the initial set of 8 variables came to be characterized by 2 new orthogonal latent variables, which allowed its location in two-dimensional figures (sort of access for main components), which are linear combinations of the original variables created with the 2 largest eigenvalues of the data covariance matrix (Hair et al., 2005). The appropriateness of this analysis is verified by the full information of the original variables held in the main components showing eigenvalues greater than the unit, or lower eigenvalues of which do not have the relevant information. The linear correlation between the production of dry biomass of shoots and length, the size of leaves, number of tillers, number of green leaves and leaf area were determined. All multivariate statistical analyses were processed in software STATISTICA® versão 7.0 .

RESULTS AND DISCUSSION

Table 2 shows the mean values of length and width of the sheet +3 and the number of plants per m^2 of four varieties of sugar cane in cycles cane plant - first and second regrowth. There was variety effect of the 3 variables. The smaller +3 sheet lengths were observed in RB92579 during the 3 cycles of culture, approximately 10% lower than the other varieties. Regarding the width of the third middle part of the leaf +3, there was a significant difference only in the second regrowth; for RB92579, the greatest width was approximately 9% higher than the average of other cultivars.

The number of plants was influenced by the variety of cane plant cycles and first regrowth. In cane plant, RB867515, SP813250 and VAT 90212 did not differ, but obtained a number of tillers of 20% less than RB92579. In the first regrowth, RB92579 and SP813250 were similar and about 18% superior to the other 2 genotypes (Table 3). In the coefficients of variation for length of the sheets to the width of the sheets, the number exhibits a range from 2.37 to 14.86%. In the works consulted, the coefficients of variation for the biometric parameters of sugarcane in the maximum growth phase were less than 15% (Almeida et al., 2008; Oliveira et al., 2011; Silva et al., 2012) which confirm this study.

Table 2. Valores average length and width of +3 leaf of four varieties of sugarcane in the maximum growth stage, in the plant cane cycles (PC), first regrowth (PR) and second regrowth (SR).

Varieties	Length of leaves (cm)				Width of leaves (cm)			
	CP	PR	SR	Average	CP	PR	SR	Average
RB92579	149b	138b	120b	135	4.78[a]	4.17[a]	3.89[a]	4.28
RB867515	164[a]	153[a]	134[a]	150	4.72[a]	4.28[a]	3.59[b]	4.19
SP813250	159[a]	150[a]	133[a]	147	4.50[a]	4.23[a]	3.49[b]	4.07
VAT90212	167[a]	155[a]	135[a]	152	4.82[a]	4.37[a]	3.55[b]	4.24
Average	160	149	132	147	4,.70	4.26	3.63	4.19
C.V(%)	3.78	2.37	4.52		4.83	3.86	4.34	

Means followed by the same letter do not differ significantly by the Scott-Knott test at 5% probability.

Table 3. Valores average of number of plants m^{-2} and green leaves $tiller^{-1}$ of four varieties of sugarcane in the maximum growth stage, in the plant cane cycles (PC), first regrowth (PR) and second regrowth (SR).

Varieties	Number of plants m^{-2}				Number of green leaves $tiller^{-1}$			
	CP	PR	SR	Average	CP	PR	SR	Average
RB92579	10.40[a]	8.70[a]	8.40[a]	9.16	7.85[a]	7.98[a]	6.73[a]	7.52
RB867515	7.60[b]	6.90[b]	6.90[a]	7.13	7.14[a]	7.07[a]	7.02[a]	7.07
SP813250	9.10[b]	8.70[a]	8.90[a]	8.9	6.38[a]	5.88[a]	5.84[a]	6.03
VAT90212	8.40[b]	7.50[b]	7.10[a]	7.66	7.28[a]	7.12[a]	6.62[a]	7.00
Average	8.87	7.95	7.82	8.21	7.16	7.01	6.56	6.91
C.V(%)	8.51	12.83	14.86		21.52	21.39	17.31	

Means followed by the same letter do not differ significantly by the Scott-Knott test at 5% probability.

Table 4. Valores average of leaf area (LA cm^2 $tiller^{-1}$). leaf area index LAI (m^2 m^{-2}) of four varieties of sugarcane in the maximum growth phase, the plant cane cycles (CP), the first regrowth (FR) and second regrowth (SR).

Varieties	LA (cm^2 $perfilho^{-1}$)				LAI (m^2 m^{-2})			
	CP	FR	SR	Average	CP	FR	SR	Average
RB92579	5.296[a]	4.326[a]	3.071[a]	4.231	4.46[a]	3.05[a]	2.04[a]	3.18
RB867515	5.126[a]	4.476[a]	3.304[a]	4.302	3.19[b]	2.49[a]	1.82[a]	2.50
SP813250	4.552[a]	3.789[a]	2.753[a]	3.698	3.22[b]	2.54[a]	1.87[a]	1.27
VAT90212	5.606[a]	4.650[a]	3.096[a]	4.45	3.76[b]	2.78[a]	1.70[a]	2.74
Average	5.145	4.31	3.056	4.17	3.66	2.72	1.86	2.74
C.V(%)	16.42	16.29	13.71		16.36	18.91	11.09	

Means followed by the same letter do not differ significantly by the Scott-Knott test at 5% probability.

In studies conducted in state of Alagoas by Silva (2007), it was found that in the plant cane cycle, the number of plant of RB92579 was statistically always above that of RB867515. Also according to Silva (2007), the maximum growth phase of the average population density of RB867515 and RB92579 was respectively 10 and 14 plants per square meter similar to values obtained in this work. Similar results for the first regrowth were observed by Almeida et al. (2008)'s study with 4 varieties of sugarcane conducted in Rio Largo. The population density fluctuated around 11 plants per square meter. Megda et al. (2012) had a number of 12 plants per square meter tillers in most growing variety of SP891115, therefore 30% higher than that observed in this study.

Tables 3 and 4 presents the means values of number of leaves per tillers, leaf area, leaf area index of 4 varieties of sugar cane in cycles cane plant - first and second regrowth. Only the variable leaf area index in

Table 5. Values of dry biomass productivity average of shoots of four varieties of sugarcane in the maximum growth phase, the cycles of plant cane, and first and second regrowth.

Varieties	Dry biomass productivity t ha^{-1}			
	Plant cane	First Regrowth	Second Regrowth	Average
RB92579	46.81[a]	44.51[a]	31.76[a]	41.02
RB867515	49.91[a]	38.95[a]	31.00[a]	39.95
SP813250	47.75[a]	40.56[a]	33.25[a]	40.52
VAT90212	46.05[a]	40.23[a]	31.60[a]	39.29
Average	47.63	41.06	31.90	40.19
C.V(%)	11.32	12.06	9.30	

Means followed by the same letter do not differ significantly by the Scott-Knott test at 5% probability.

plant cane cycle presented varietal effect; the highest LAI was observed in the variety RB92579. This variety has shorter leaves, but then the number of plants per square meter exceeds the others, reflecting in leaf area approximately 24% higher than the average of the others. The average number of green leaves per tiller was 7.16, 7.01 and 6.56, for the sugarcane plant cycles, the first and second regrowth, respectively (Table 3). Machado et al. (2009) found similar results in a study conducted with 2 genotypes of sugarcane in plant cane cycle in São Paulo; they obtained 7 leaves per plant at the stage of maximum crop growth. For the leaf area by tiller, it is observed that for decreased cycles, the average of the 4 varieties on the cane was about 40% higher than that found in the second regrowth. The mean values obtained in plant cane, first and second regrowth were 5,145, 4,310 and 3,056 cm^2, respectively. Oliveira et al. (2007a) in a study conducted with 3 varieties of sugarcane (RB72454, RB855113 and RB855536) in plant cane cycle in red Latosol in Northwestern Paraná observed that the maximum growth phase leaf area per tiller was close to 5,000 cm^2, corroborating with the results of this study.

The leaf area index of RB92579 observed by Almeida et al. (2008) in studies conducted in the CECA/UFAL, in 180 after planting was 4.5, one of the largest in the studies reviewed. In a research also conducted in CECA/UFAL, with assessments conducted at intervals of 30 days, Silva (2007) found that the average rate of leaf area of the variety RB92579 was statistically always greater than the RB867515. In 130 to 370 days after planting the cane, the average value of leaf area index of RB867515 and RB92579 was respectively 2.3 and 3.0. In the maximal growth phase, leaf area index of RB92579 was close to 5.0, whereas for RB867515 it was approximately 4.0 . Silva et al. (2012), in a study conducted in sub-middle São Francisco Valley, evaluated the leaf area of the first regrowth of RB92579 irrigated; the LAI ranged from 1.07 to 180 days after cutting (DAC), and from 5.55 to 332 DAC.

Table 5 presents the average values of dry biomass productivity of the aerial part of the 4 varieties in cycles cane - plant first and second regrowth. There was no effect on the range of 3 cycles of cultivation. The dry biomass productivity of the shoot was 47, 41 and 31 tons per hectare in plant cane cycles, first and second regrowths, respectively. Mendes (2006), in a research conducted with 8 varieties of sugarcane in a Ultisol in Minas Gerais, obtained for RB867515 variety in plant cane cycle and first regrowth an accumulation of dry matter was 43 and 33 t ha^{-1}, respectively; the results corroborate that of the present study. Calheiros et al. (2012) in a study conducted in the area of Alagoas in Oxisoil obtained 35 t ha^{-1} of dry matter accumulation for RB92579 and 30 t ha^{-1} for RB867515.

The aboveground biomass productivity observed in the present work could be medium- high, since Alagoas phase maximum growth of sugarcane occurs in short days and, therefore, under low light, unlike Center -South of Brazil, where the increased brightness coincides with the greater water availability. The non-coincidence of the maximum water availability with light influences negatively the photosynthetic rates, resulting in lower productivity of sugarcane in Alagoas, compared to South-Central (Oliveira et al., 2011).

The graphical representation of the main components (Figure 2) allowed the characterization of the variables most discriminated in the formation and differentiation through the variables evaluated. As the percentage of variance explained by the PLCs, it appears that for sugarcane plant (Figure 2A), the first and second components account for 89.63% of the total variance, being 58.75% in the PC1 and 30.88% CP2 for the first regrowth (Figure 2B); it represents the PLCs 94.34% of the total variance while for the second regrowth (Figure 2C), it corresponds to 98.20% of the total variance. Mardia et al. (1979) confirmed that in a principal component analysis the first two or three components accumulate relatively high percentage of the total variation, generally over 70%; they explain the variability

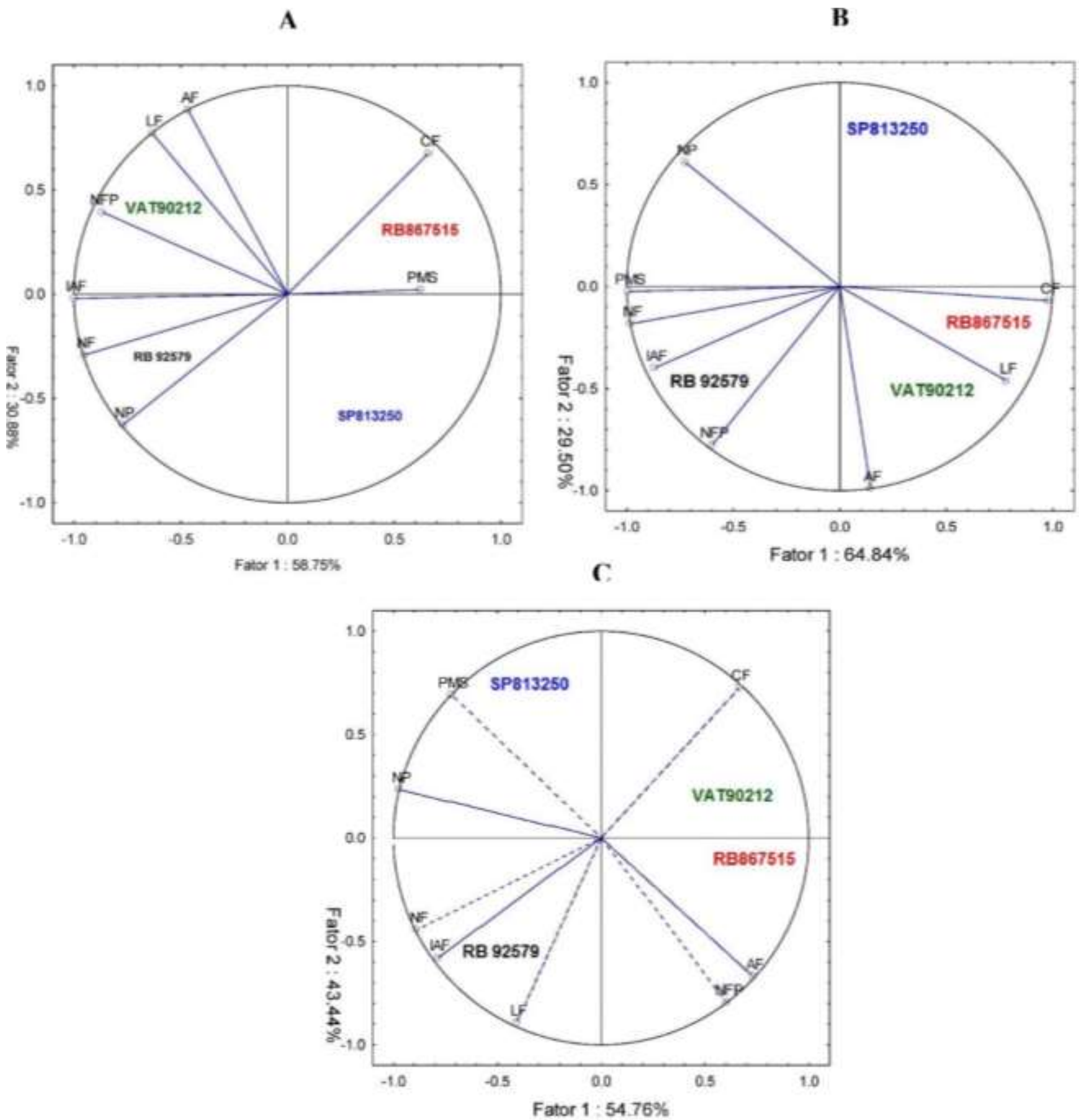


Figure 2. Components chart top of groups of varieties and variables of sugarcane in three different cycles. A = Plant cane; B = first regrowth; C = second regrowth.

manifested among cultivars and variables, which occurred in this study, since the two first principal components accounted for more than 70% of the total variance available.

The analysis of components made it possible to distinguish different patterns of distribution of variables when correlated with the stock in 3 different cycles. In this pattern of distribution, it is observed that to cultivate SP813250, isolating it in the second quadrant showed no degree of relationship with the variables studied (Figure 1A). This indicates that this cultivar is not suited for these variables, and from agronomic point of view, it was not be influenced, so that there was greater production. Pincelli and Silva (2012) studied morphological changes in sugarcane cultivar; they also find no relation variables such as LAI and leaf area per tiller (LA) for the production of the variety SP813250. In contrast to the other cultivars, there was an association between dry matter production (DMP) and length sheet (LS) to cultivate RB867515; to cultivate RB92579, there was association between number of plants (NP), number of green leaves (GL) and LAI; while to cultivate VAT90212, there was was no relation in the number of leaves per plant, leaf width and leaf area per tiller.

From the graph component of the first regrowth (Figure 2B), a similar pattern of distribution was observed between VAT90212 and RB867515 cultivars in the variables leaf area, leaf width and length. This similarity in the distribution obtained for these 2 varieties, shows the similarity in their genetic characteristics, reflected in low variability of the evaluated parameters; such justification is related to the second production cycle. As observed in plant cane, to cultivate SP813250, there was no association with any group of variables for the first production cycle. For the variable number of plants (Figure 2B), it is observed that this is not related to any cultivar. The cultivar RB92579 was associated with NFP, LAI, NF and PMS, presenting for the second cycle the same distribution pattern as observed for the plant cane cycle, but in this situation the PMS and NFP variables have greater relationship with this cultivar in the period assessed.

The second regrowth (Figure 2C) shows a pattern of high differentiation when compared to the others, for all cultivars showed relation with the other variables. According to Hair et al. (2005), this ratio can firstly be explained by the greater variance of 98.20%, which favors a pattern of relatively homogeneous distribution between the variables and cultivars. In the cultivate SP813250, only the second regrowth showed similarity with the variables studied; PMS and NP are those mostly identified with this cultivar. As is shown in Figure 2, there is no overlap between the variables; this indicates the non-occurrence of the graphic representation, thereby showing the ability to represent the largest accumulation of explained variance (Hair et al., 2005). Another important fact is that some variables are close to the unit circle, showing that they have a greater contribution in relation to variables that are farther apart.

Conclusion

The larger leaf area index was seen in RB92579. The dry biomass yield was not influenced by varieties. The principal component analysis enabled us to identify from CPs variance what each component can contribute to the data explanation, and prove effective in differentiating between the studied variables and cultivars groups for different cycles, showing the importance of each cultivation in different production cycles of sugar cane.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Almeida ACS, Souza JL, Teodoro I, Barbosa GVS, Moura Filho, G, Ferreira Junior RA (2008). Vegetative development and production of sugarcane varieties as a function of water availability and thermic units. Ciênc. Agrotec. 32:1441-1448.

Bastos AVS, Silva MV, Silva EC, Teixeira MB, Muraoka T, Soares FAL, Coelho RD (2016). Agroindustrial yield of sugarcane grown under different levels of water replacement and nitrogen fertilization. Afr. J. Agric. Res. 11:2623-2629.

Calheiros AS, Oliveira MW, Ferreira VM, Barbosa GVB, Santiago AD, Aristides EVS (2012). Production of biomass, from sugar and protein in function of sugarcane varieties and phosphorous fertilization. Semin. Cien. Agrar. 3:809-818.

Conab - Companhia Nacional de Abastecimento (2016). Safra brasileira da cana-de-açúcar, 2016/17. Levantamento de agosto de 2016. < http://www.conab.gov.br >.

Cunha FN, Silva NIF, Teixeira MB, Morais WA, Vidal VM, Cunha ES, Gomes FH, Araujo AO (2016). Agronomic performance and industrial yield of sugarcane under water-saving irrigation in cerrado soil. Afr. J. Agric. Res. 11(32):3056-3064.

Embrapa (2013). Sistema brasileiro de classificação de solos. (3th ed). Brasília.

Ferreira DF (2008). SISVAR: um programa para análises e ensino de estatística. Rev. Symp. 6:36-41.

Hair JF, Anderson RE, Tatham RL, Black WC (2005). Análise multivariada de dados. Bookman, Porto Alegre.

Hermann ER, Câmara GMS (1999). Um método simples para estimar a área foliar de cana-de-açúcar. Stab. 17:32-34.

Ibge – Intistuto Brasileiro de Geografia e Estatística (2016). Levantamento sistemático da produção agrícola. Disponível em: http://www.ibge.gov.br/home/estatistica/indicadores/agropecuaria/lspa/lspa_201105_4.shtm>.

Landell MGA, Bressiani JA (2008). Melhoramento genético e manejo varietal. In: Dinardo. Cana-de-Açúcar. 1 ed. Instituto Agronômico de Campinas, Campinas.

Leme EJA, Maniero MA, Guidolin JC (1984). Estimativa da área foliar da cana-de-açúcar e sua relação com a produtividade. Cad. Planal. 2:3-22.

Machado RS, Ribeiro RV, Marchiori PER, Machado DFSP, Machado EC, Landell MGDA (2009). A Biometric and physiological responses to water deficit in sugarcane at different phenological stages. Pesqui. Agropecu. Bras. 44:1575-1582.

Mardia KV, Kent JT, Bibby JM (1979). Multivariete Analysis. Academic Press, London.

Megda MXV, Trivelin PCO, Franco HCJ, Otto R, Vitti AC (2013). Agronomic efficiency of nitrogen fertilizers on green harvesting sugarcane ratoon. . Pesqui. Agropecu. Bras. 47:1681-1690.

Mendes LC (2006). Eficiência nutricional de cultivares de cana-de-açúcar. Dissertação Mestrado. Universidade Federal de Viçosa, Viçosa. 46p.

Oliveira ECA, Oliveira RI, Andrade BMT, Freire FJ, Lira Jr MA, Machado PR (2007a). Crescimento e acúmulo de matéria seca em variedades de cana-de-açúcar cultivadas sob irrigação plena. Rev. Bras. Eng. Agríc. Ambient. 14:951-960.

Oliveira MW, Freire FM, Macêdo GAR, Ferreira JJ (2007b). Nutrição mineral e adubação da cana-de-açúcar. Inf. Agropecu. 28:30-43.

Oliveira MW, Magrini J, Lyra FE, Pereira MG, Aristides EVS (2011). Produção da RB867515 influenciada pela aplicação de substâncias húmicas, aminoácidos e extrato de algas marinhas. Stab. 30:30-33.

Oliveira RA, Daros E, Zambon JLC, Weber H, Ido OT, Bespalhok-Filho JC, Silva DKT (2007). Leaf area in three sugarcane cultivars and its relationship with biomass production. Pesqui. Agropecu. Trop. 37:71-76.

Pedula RO, Schultz N, Monteiro RC, Pereira W, Araujo AP, Urquiaga S, Reis VM (2016). Growth analysis of sugarcane inoculated with diazotrophic bacteria and nitrogen fertilization. Afr. J. Agric. Res. 11:2786-2795.

Pincelli RP, Silva MA (2012). Leaf morphological changes in sugarcane cultivars in response to water deficit. Biosci. J. 28:546-556.

Reis LS, Azevedo CA, Albuquerque AW, Junior JF (2013). Índice de área foliar e produtividade do tomate sob condições de ambiente protegido. Rev. Bras. Eng. Agríc. Ambient. 17:386-391.

Silva ET (2007). Área foliar, crescimento e produção de sacarose por duas cultivares de cana-de-açúcar (*Saccharum spp*) influenciada por doses de fósforo. Dissertação (Mestrado em Agronomia) – Universidade Federal de Alagoas, Maceió.

Silva MOS, Freire FJ, Sobral, JK, Oliveira ECA, Freire MBGS, Apolinário VXO (2016). Bacteria associated with sugarcane in Northeastern Brazil. Afr. J. Microbiol. Res. 10:1587-1594.

Silva TGF, Moura, MSB, Zolnier S, Carmo JFA, Souza LSB (2012). Biometrics of the sugarcane shoot during irrigated ratoon cycle in the Submedio of the Vale do São Francisco. Rev. Cien. Agron. 48:500-509.

academicJournals

Vol. 12(7), pp. 467-476, 16 February, 2017
DOI: 10.5897/AJAR2016.11430
Article Number: 5943BBB62760
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Response of phosphorous fertilizer and its recommendation for food barley (*Hordium vulgare* L.) production on Nitisols of central Ethiopian highlands

Legesse Admassu

Ethiopian Institute of Agricultural Research, Holeta Agricultural Research Center, P. O. Box 2003,Addis Ababa, Ethiopia.

Received 13 July, 2016; Accepted 28 September, 2016

Nowadays, making available proper and balanced fertilizer recommendations is of paramount importance in order to confirm security and increase crop productivity in sustainable way for farmers and other stakeholders. Soil test based phosphorous calibration study was conducted for barley (*Hordium vulgare* L.) on Nitosols of farmers' fields in West Shewa, in the central highlands of Ethiopia. The experiment was arranged in a randomized complete block design with six levels of phosphorous fertilizer (0, 10, 20, 30, 40 and 50 kg ha^{-1}) with three replications. Results revealed that yield and yield components of food barley were significantly affected by P fertilizer application. Phosphorous fertilizer application at different rates increased grain yield of food barley by 23 to 46% compared to the control. Available soil test P concentrations analyzed three weeks after planting were affected significantly by P fertilizer application rate. Relative yield and Bray-2 soil test phosphorous value correlation indicated that soil test phosphorous values greater than 13 mg kg^{-1} was found to be sufficient for food barley production. The average phosphorous requirement factor (P_f) calculated from soil test phosphorous values of all treatments for study area was 10.2. Most sites tested had Bray 2 P values <10 mg kg^{-1}. In the absence of a soil test, a recommendation of 40 kg P ha^{-1}, resulting in the best response overall, could be made for the first year of application. It was also recommend that to prevent a potential loss of barley yield, a maintenance application of at least 5 to 10 kg P ha^{-1} be applied every year, irrespective of the calculated recommended rate, in order to replace P exported from the field in the form of grain and straw yield. Further field trials are required to determine interactions between P response and the effects of climate, soil properties, and other management practices.

Key words: Food barley, phosphorous calibration, nitisols, phosphorous requirement factor, critical concentration.

INTRODUCTION

Soil fertility decline due to continued degradation of agricultural soils is a major concern in African agriculture in general and in Ethiopia in particular. Among the bio-physical factors, it remains to be single most important

E-mail: legesse.admassu@gmail.com.

constraint to food security in sub-Saharan Africa (SSA) (Bekele and Drake, 2003). The International Centre for Soil Fertility and Agricultural Development estimates that Africa loses 8 million metric tons of soil nutrients per year and over 95 million hectare of land have been degraded to the point of greatly reduced productivity (Henao and Baanante, 2006). According to Stoorvogel et al. (1993, the annual average nutrient loss for SSA was 26 kg N, 3 kg P, and 19 kg K ha^{-1} $year^{-1}$ with that for Ethiopia being 47 kg N, 7 kg P and 19 kg K ha^{-1} $year^{-1}$ resulting in a negative nutrient balance (Omotayo and Chukwuka, 2009). In contrast, farms in North America and Europe have averaged net positive nutrient balances (Sanchez et al., 1994). Farmers in SSA seldom apply fertilizers in the recommended fertilizer rates at appropriate time that does not consider the crop nutrient requirement because of many socioeconomic constraints such as lack of supplies, cost of fertilizers, lack of access to financial credit, delivery delays, low and variable returns (Partey and Thevathasan, 2013). With rapid population growth, continuous and intensive cropping without restoration of the soil fertility, has depleted the nutrient base of most soils (Alice et al., 2012) resulting in poor crop yields. Nutrient depletion could be even worse in highly populated countries such as Ethiopia (Haileslassie et al., 2006).

Barley one of the most important food crops predominantly grown from 1500 to 3500 m above sea level in Ethiopia (Lakew et al., 1996). It covers an area of about 1.13 million ha, but its national average yield is low at 1.7 t ha^{-1} (CSA, 2014). Phosphorous (P) is usually the most yield limiting of soil-supplied elements, and soil P tends to decline when soils are used for agriculture (David and David, 2012). A high proportion of the grain becomes human food, and a consequent residue is not returned to the field as often as plant or animal wastes (Buol, 1995). The low solubility of phosphates and their rapid transformation to insoluble forms makes P less available or unavailable to crops (Smil, 2000). P is deficient in about 70% of the soils in Ethiopia (Mamo and Haque, 1991). Barley production in Nitisol areas of Ethiopia are marginally to severely deficient in P and constrained by soil acidity and low nutrient availability (Agegnehu et al., 2011; Regassa and Agegnehu, 2011) making it the main growth limiting factor (David and David, 2012). These highlands constitute 43% of the country but account for 95% of the cultivated area and support 88 and 75% of the human and livestock population, respectively (Yirga and Hassan, 2013)

Sound soil test based and site specific nutrient management is essential in reversing this trend and increase crop yield in agricultural land. It is essential for successful fertilization program and crop production. It is a reliable and accurate method to identify the nutrient rates required to attain a desired level of plant growth and yield. It is important that results of soil tests be calibrated against crop response from applications of the plant nutrients in question (Wortmann et al., 2013). A reliable soil test correlates soil nutrients to plant use, and fertilizer recommendations calibrate tests to field conditions for individual crops once relationship between soil test values, fertilizer rates and crop yield is known, it is possible to determine the most economical fertilizer rate for a given crop which can make fertilizer recommendations refined according to the requirements of each field in a given farm (Seif, 2013). Once critical nutrient level and crop requirement are worked out, farmers and producers could use this relatively simple tool to increase fertilizer profitability.

Calibration is a means of establishing a relationship between a given soil test value and the yield response from adding nutrient to the soil as fertilizer. It provides information how much nutrient should be applied at a particular soil test value to optimize crop growth without excessive waste. Calibration research predicts the probability of response from applying a given nutrient which must be determined experimentally in the field (Dahnke and Olsen, 1990). Calibrations are specific for each crop type, soil type, soil pH, climate plant species, and crop variety (Seif, 2013; Agegnehu and Lakew, 2013; Sonon and Zhang, 2014). Soil testing particularly soil P, tests can be used for evaluating soil P availability and fertilizer recommendations. The reliable nutrient status of farmers' fields' true will only be revealed through chemical soil testing during a particular growing season. The most widely used available soil P test is Bray II (Bray and Kurtz, 1945) on acidic soils (Bado et al., 2008). Instead of simple individual soil tests, soil calibrations of the relationship between soil test and yields of a specific plant are needed for fertilizer recommendation. A critical limit of available P and P requirement factors for a specific soil and crop have been conducted for some crops recently, but the critical limits of available P and P requirement factor are not established for many crops including food barley. Different methods can be used to examine such a relationship. One example of simple graphical method is the Cate Nelson graphical method (Nelson and Anderson, 1977).

The blanket recommendation of 46% P and 64% N for food barley in the central highlands of Ethiopia (Bekele et al., 1993) does not consider the differences in agro-ecological environments (Agegnehu and Lakew, 2013) which may not be applicable under the current production system and for the foreseeable future. Since the spatial and temporal fertility variations in soils were not considered, farmers have been applying same P fertilizer rate to their fields regardless of soil fertility differences. Almost all soil properties exhibit variability as a result of dynamic interactions between natural environmental factors, that is, climate, parent material, vegetation and topography (Jenny, 1941). Soil properties and in turn plant growth are significantly controlled by variation in landscape attributes including slope, aspect and elevation which influence plant nutrient distribution

(Rezaei and Gilkes, 2005) specially in Ethiopian highlands where the steep and dissected terrain topography make soils susceptible to soil erosion and degradation (Hurni, 1988). For this reasons, the blanket recommendation will make inefficient use of these expensive nutrients which contribute to the depletion of scarce financial resources, increased production costs and potential environmental risks (Tarekegne and Tanner, 2001).

Currently, soil fertility research improvement is geared towards site specific fertilizer recommendation. The establishment of a reliable soil test is able to assist in the determination of P requirements. It involves a correlation to find an extractant for soil nutrients for a laboratory test that will best mine an amount of a nutrient proportional to what a plant extracts (Seif, 2013). This will be followed by a calibration to relate soil test numerical value with field nutrient response in the form of crop yield from the addition of the fertilizer nutrient to the soil (Shaver, 2014). Therefore, the objectives of this study were to correlate the Bray-2 soil test P with relative grain yield response of food barley across selected Nitisol areas of West Shewa to establish preliminary agronomic interpretations, and determine the critical P concentration and P requirement factor.

MATERIALS AND METHODS

Experimental site

Phosphorous response trials with food barley were conducted on farmers' fields from 2012 to 2015 during the main cropping seasons in West Shewa in the central highlands of Ethiopia. Food barley is grown mainly by subsistence farmers in the highlands of the country. The rainfall is bimodal with long-term average annual rainfall 1100mm, about 25% of which falls from June to September and the rest from January to May and average minimum and maximum air temperature of 6.2 and 22.1°C, respectively. The environment is seasonally humid and major soil type of the trial sites is Eutric Nitisol (FAO classification).

For the selection of representative trial sites across the area over 600 soil samples (0 to 20 cm depth) were collected in three years from farmers' fields before the onset of the trial. Soil samples were analyzed for pH using in a ratio of 2.5 ml of water to 1 g soil available P using Bray II method (Bray and Kurz, 1945) organic C content using Walkley and Black method (1954), total N using Jehldahl method (Bremner and Mulvaney, 1982), exchangeable cations and cation exchange capacity (CEC) using ammonium acetate method (Chapman, 1965). The available soil P (using Bray-2 method) ranges prior to planting considered for classification were <10 mg kg^{-1} for low, 10 to 25 mg P kg^{-1} for medium and >25 for high (Table 1). Based on this categorization 9 farmers with low and medium fields available P were selected for the first year, 5 farmers for the second year and 4 farmers with the same categories for the last two years, respectively.

Experimental setup

The experiment was arranged in a randomized complete block design with six levels of phosphorous (0, 10, 20, 30, 40 and 50 kg P ha^{-1}) with three replications. The plot size was 4 m by 5 m (20 m^2) and the spacing between plots and blocks were 0.5 m and 1 m, respectively. The harvested plot size was 16 m^2. Barley (*var. HB-1307)* was seeded at the recommended rate of 125 kg ha^{-1}. The experiment was planted in June. The sources of N and P were urea and triple super phosphate (TSP), respectively. The P fertilizer was applied at planting. While the recommended N fertilizer (60 kg ha^{-1}) was applied two doses; half at planting and half at tillering stage. Other agronomic practices were applied based on local research recommendations.

The first weeding was done 30 to 35 days after planting and the second weeding was carried out a month after the first weeding. Agronomic parameters collected were grain yield, aboveground total biomass, thousand seed weight, test weight, seed weight (g/100 spikes), spike size and plant height (average of 10 plants). One site in the third year was dropped due to poor crop performance and 18 sites were considered for harvesting, data analysis and interpretation in three years. To estimate total biomass and grain yields the entire plot was harvested at maturity in November. After threshing seeds were cleaned weighed and adjusted at 12% moisture level. Total biomass and grain yields recorded on plot basis were converted to kg ha^{-1} for statistical analysis.

Determination of critical P concentration (Pc)

To correlate relative yield vs. soil test P values and determine critical P concentration, the available P was extracted from the soil samples taken three weeks after planting from each plot of all experimental fields using Bray 2 method and three replications for each treatment.

The Cate-Nelson graphical method (Nelson and Anderson, 1977) was used to determine the critical P value using relative yields and soil test P values obtained from 18 P fertilizer trials conducted at different sites, to assess the relationship between grain yield response to nutrient rates and soil test P values, relative grain yields in percent were calculated as follows:

$$Relative\ yield\ (\%) = \frac{Yield}{maximum\ yield}\ X\ 100 \quad (1)$$

The scatter diagram of relative yield (Y-axis) versus soil test value (X-axis) was plotted. The range in values on the Y-axis was 0 to 100%. A pair of intersecting perpendicular lines was drawn to divide the data into four quadrants. The vertical line defines the responsive and non-responsive ranges. The observations in the upper left quadrants overestimate the P fertilizer P requirement while the observations in the lower right quadrant underestimate the fertilizer requirement. The intersecting lines were moved about horizontally and vertically on the graph, always with the two lines parallel to the two axes on the graph, until the number of points in the two positive quadrants was at a maximum (or conversely, the number of points in the two negative quadrants was at a minimum). The point where the vertical line crosses the X-axis was defined as optimum critical soil test level (Nelson and Anderson, 1977).

Determination of P requirement factor (Pf)

Phosphorous requirement factor (Pf) is the amount of P in kg needed to raise the soil P by 1 mg kg^{-1}. It enables to determine the quantity of P required per hectare to raise the soil test by 1 mg kg^{-1}, and to determine the amount of fertilizer required per hectare to bring the level of available P above the critical level (Nelson and Anderson, 1977). It was calculated using available P values in samples collected from unfertilized and fertilized plots.

Table 1. Soil nutrient contents of the trial sites before planting food barley in 2012.

Farmers names	pH (1:2.5 H_2O)	Total N (%)	P (mg kg-1)	K ($cmol_c\ kg^{-1}$)	Na ($Cmol_c\ kg^{-1}$)	CEC ($Cmol_c\ kg^{-1}$)
Lelissa	4.62	0.19	5.0	0.82	0.15	16.8
Gadissie	5.63	0.18	5.2	1.94	0.11	17.1
Teshome	5.41	0.16	6.6	1.39	0.11	19.2
Getu	5.23	0.14	9.2	4.52	0.15	19.4
Beyene	5.01	0.15	6.8	0.56	0.11	17.2
Aselefech	5.11	0.15	6.2	0.65	0.16	16.0
Abera	6.24	0.19	7.4	1.69	0.12	31.5
Mekonnen	5.38	0.16	14.4	0.78	0.12	26.4
Taffa	5.32	0.20	6.2	0.61	0.13	18.4
Gudisa	5.00	0.19	8.2	0.72	0.19	21.2
Bekele	5.51	0.14	4.6	0.68	0.15	17.1
Bizuayehu	4.98	0.20	8.8	0.52	0.14	22.4
Dereje	6.54	0.18	9.4	0.86	0.21	26.5

CEC: Cation exchange capacity.

Phosphorous requirement factor was expressed as:

$$Pf = \frac{kg\ P\ applied}{\Delta\ soil\ P} \quad (2)$$

Therefore, the rate of P fertilizer to be applied (Pa) was expressed in terms of critical P concentration (Pc), initial soil P value (Pi) and P requirement factor (Pf).

$$Pa = (Pc - Pi)\ X\ Pf \quad (3)$$

Statistical analysis

The data were subjected to analysis of variance using the procedure of the of SAS statistical package version 9.0 (SAS Institute, 2001). The total variability for each trait was quantified using the following model.

$$Tijk = \mu + Yi + Rj(i) + Pk + PY(ik) + eijk \quad (4)$$

where T_{ijk} is the total observation, μ = grand mean, Yi = effect of the i^{th} year, Rj(i) is the effect of the j^{th} replication (within the i^{th} year), P_k is the of the k^{th} treatment, PY $_{(ik)}$ is the interaction of the k^{th} treatment with i^{th} year and e_{ijk} is the random error. Means for the main effects were compared using the means statement with least significant difference (LSD) test at the 5% level.

RESULTS

Weather

The total rainfall amount and precipitation pattern for 2012 was significantly higher compared with long-term average, 2013 and 2014 (Figure 1). The rainfall amounts recorded for July and September were considerably higher in 2012 than in 2013 and 2014. When compared with a 30 year average, rainfall in July was higher by 41 mm in 2012, but lower by 122 and 126 mm in 2013 and 2014, respectively, which entails average moisture received in 2012 was conducive for barley growth and development. Moisture deficiency in July and September critically affects tillering and grain filling, respectively.

Yield and yield components

The responses of grain yield and yield components of food barley to phosphorus fertilization, year and interaction of year by phosphorous of the combined data of over three years are presented in Table 2. The three cropping year data analysis of variance indicated that grain yield and yield components of food barley were significantly affected by year and P fertilizer. Analysis of variance over three cropping seasons revealed that the year effect was highly significant ($p<0.001$) for grain and yield components of barley (Table 2). The year by P fertilizer rate interaction was not significant for grain yield and yield components of barley. The highest mean grain yield (5050 kg ha^{-1}) was obtained in the year 2012 compared to the lowest (2313 kg ha^{-1}) recorded in 2014. The maximum total crop biomass, harvest index, thousand seed weight, seed weight, test weight, seed weight, spike length and plant height also recorded in the same cropping season (Tables 3 and 4).

Grain yield, total above ground biomass, harvest index, thousand grain weight, test weight, seed weight per spike and plant height of food barley significantly responded ($p<0.01$ and $p<0.001$) to P fertilizer application rate (Table 2). Spike size and moisture content were significantly affected by year only but not by P ($p<0.05$) for grain yield and yield components (Table 2). Grain yield significantly ($p<0.001$) affected by P rate.

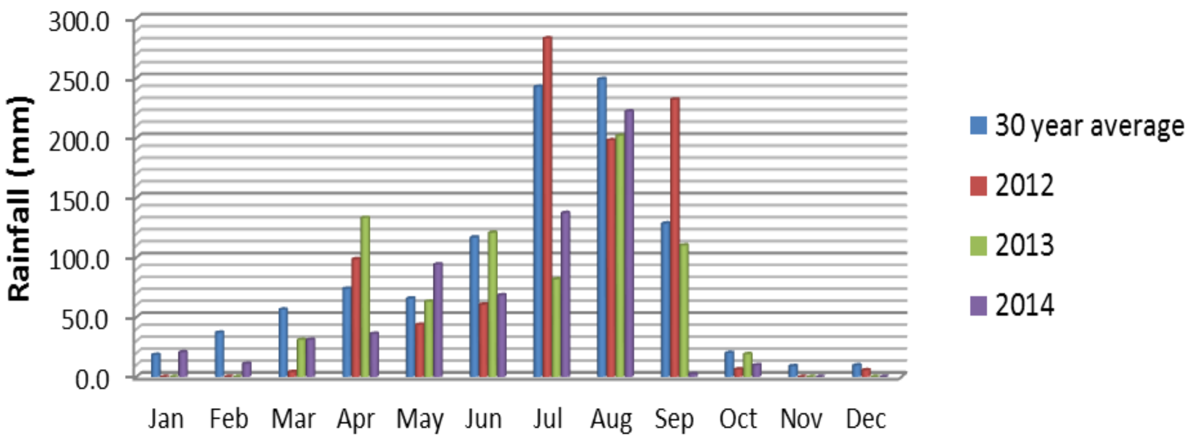


Figure 1. Monthly total rainfall for 30 year average, 2012, 2013 and 2014 cropping season's rainfall at Holeta and around the trial sites.

Table 2. Effects of year, P fertilizer rate and their interaction on yield and yield components of food barley across sites in 2012, 2013, and 2014.

Parameter	Year (Y)	Phosphorous (P)	Y×P
Grain yield	***	***	ns
Total biomass	***	***	ns
Harvest index	***	***	ns
Thousand seed weight	***	***	ns
Seed weight	***	**	ns
Test weight	***	ns	ns
Spike size	***	***	ns
Plant height	***	***	ns
Moisture content	***	ns	ns

Significant at **P ≤ 0.01, ***P ≤ 0.001; ns, not significant.

Table 3. Table of means for main effects of P application year and fertilizer rate on food barley crop parameters in 2012, 2013 and 2014.

Factor / Year	Grain yield (kg/ha)	Biomass yield (kg/ha)	Harvest index (%)	Thousand seed weight (g)	Test weight (kg L^{-1})
2012	5050[a]	10536[a]	48.1[a]	45.3[a]	65.2[a]
2013	2505[b]	6004[b]	42.0[b]	42.6[b]	62.6[b]
2014	2313[b]	6293[b]	36.5[c]	40.6[c]	63.0[b]
Phosphorous					
0	2923[c]	7149[c]	40.2[c]	40.8[c]	63.6
10	3551[b]	8113[b]	43.1[b]	41.8[c]	64.1
20	3636[b]	8158[b]	43.6[ab]	41.5[c]	64.0
30	3788[b]	8408[b]	44.1[ab]	43.6[b]	64.3
40	4268[a]	9120[a]	45.6[a]	46.4[a]	64.0
50	4168[a]	9166[a]	44.4[ab]	46.2[a]	64.2
CV	18.3	18.6	10.6	6.2	2.8
P_{linear}	***	***	**	***	ns

Within each column, means with different letters are significantly different at p < 0.05; CV, coefficient of variation

Table 4. Table of means for main effects of P application year and fertilizer rate on food barley crop parameters in 2012, 2013 and 2014.

Factor / Year	Seed weight (g/100 spikes)	Spike length (cm)	Plant height (cm)	Moisture content (%)
2012	156.0[a]	6.9[a]	102.9[a]	10.9
2013	141.4[b]	6.4[b]	95.4[c]	9.8
2014	126.3[c]	6.3[b]	100.4[b]	10.3
Phosphorous				
0	138.5[c]	6.3[c]	96.7[d]	10.5
10	141.3[bc]	6.5[bc]	99.8[c]	10.5
20	142.4[bc]	6.4[bc]	100.2[c]	10.5
30	144.5[abc]	6.7[ab]	100.5[bc]	10.4
40	152.1[a]	6.8[a]	102.7[ab]	10.4
50	147.5[ab]	6.9[a]	103.5[a]	10.4
CV	13.1	7.3	5.2	3.5
P_{linear}	**	***	***	ns

Within each column, means with different letters are significantly different at $p < 0.05$; CV, coefficient of variation.

Significantly a higher grain yield was obtained from the application of 40 kg P ha^{-1}. The application of P fertilizer rate of 10, 20, 30, 40, and 50 kg ha^{-1} increased grain yields of food barley by 21, 24, 30, 46 and 43%, respectively, compared to the control (without P fertilizer). Application of P fertilizer consistently increased total biomass (linear, r^2 = 0.9), grain yield, harvest index, Plant height, thousand seed weight consistently increased as P rate increased, but showed slight decrease beyond 40 kg ha^{-1}. However, statistically significant differences were not obtained among P levels for hectoliter weight and moisture content (Table 3). The combined analysis of variance across all experimental locations signify that barley yield and yield components differed significantly (P<0.001) among trial locations (data not shown). Physical observations revealed that heading and flowering stages were earlier and higher plant height was recorded in plots that received P fertilizer compared with untreated plots.

Critical P concentration (Pc) and P requirement factor (Pf)

Soil P values determined three weeks after planting differed significantly (P<0.01) among P levels. The main effect of P fertilizer resulted in mean soil test P values of 8.5 to 17.4 mg kg^{-1}. Bray-2 soil test P values below 10 mg kg^{-1} are considered low. The increase in soil P response to P fertilizer application was linear up to 50 kg P ha^{-1}. The highest mean soil P concentration (17.4) was recorded from 50 kg P ha^{-1} (Figure 2).

The correlation between relative food barley grain yield response and soil P measured with Bray-2 method is indicated in Figure 3. The critical P concentration (P_c) was determined from the scatter diagram drawn using relative grain yields of food barley and the subsequent soil test P values for all P levels (0 to 50 kg P ha^{-1}). The Pc defined by the Cate Nelson method in this study was about 13 mg P kg^{-1}, with mean relative yield response of about 80% (Figure 3). When the soil test value is below the critical value additional information is needed on the quantity of P required to elevate the soil P to the required level. This is the P requirement factor (P_f), the amount of P required to raise the soil test P level by 1mg kg^{-1}, computed from the difference between available soil test P values from plots that received 0 to 50 kg P ha^{-1} using the second formula mentioned above. Accordingly the calculated Pf were 8.5 to 13 and the overall average P_f of all treatments for the study area was 10.2 (Table 5). Thus the rate of P fertilizer required per hectare can be calculated using the soil critical P concentration, initial soil P determined for each site before planting (Table 1) and the P requirement factor as indicated above in the third formula.

DISCUSSION

Cropping season disparity has brought about significant differences in yield and yield components. Results have indicated that the amount of seasonal rainfall received and in the growing season greatly impacts the response to P fertilizer application in increasing productivity of food barley. In 2013 and 2014, lower yield and yield components were recorded due to early insufficient amount of rainfall in all trial sites during the tillering period in the month of July. The yield obtained was lower in

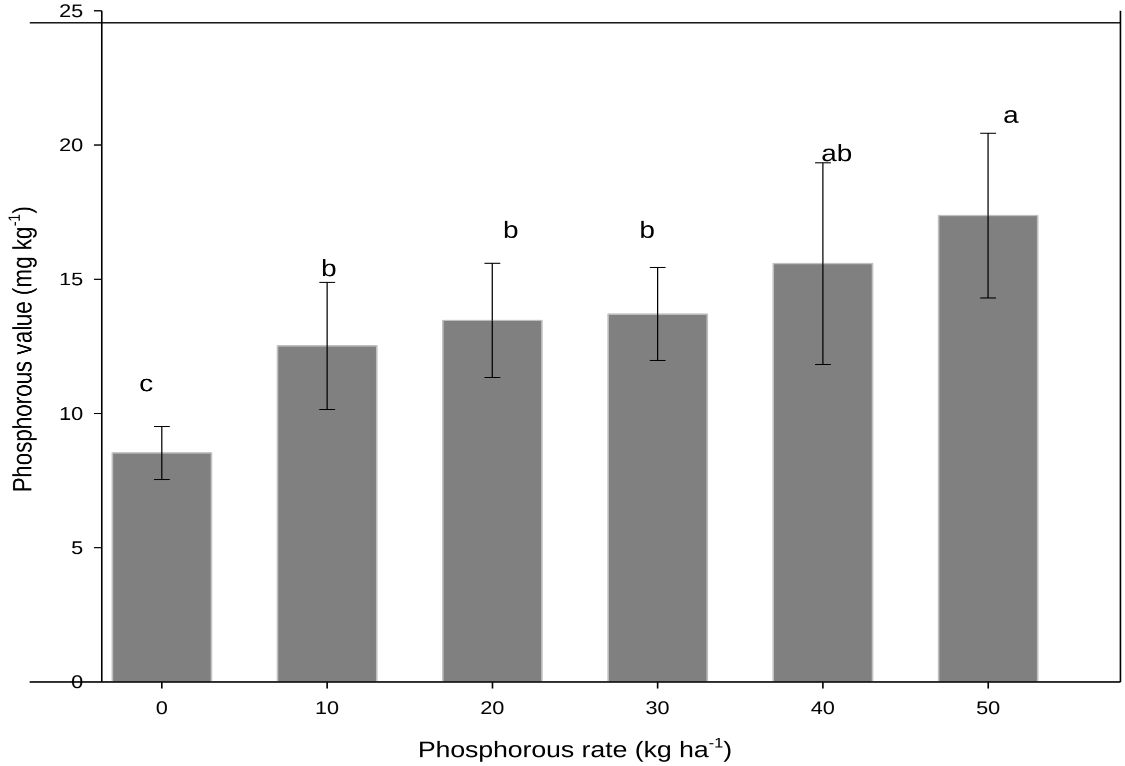


Figure 2. Effect of available soil test phosphorous value analyzed three weeks after planting to P fertilizer rate in 2012, 2013 and 2014. Error bars with standard error.

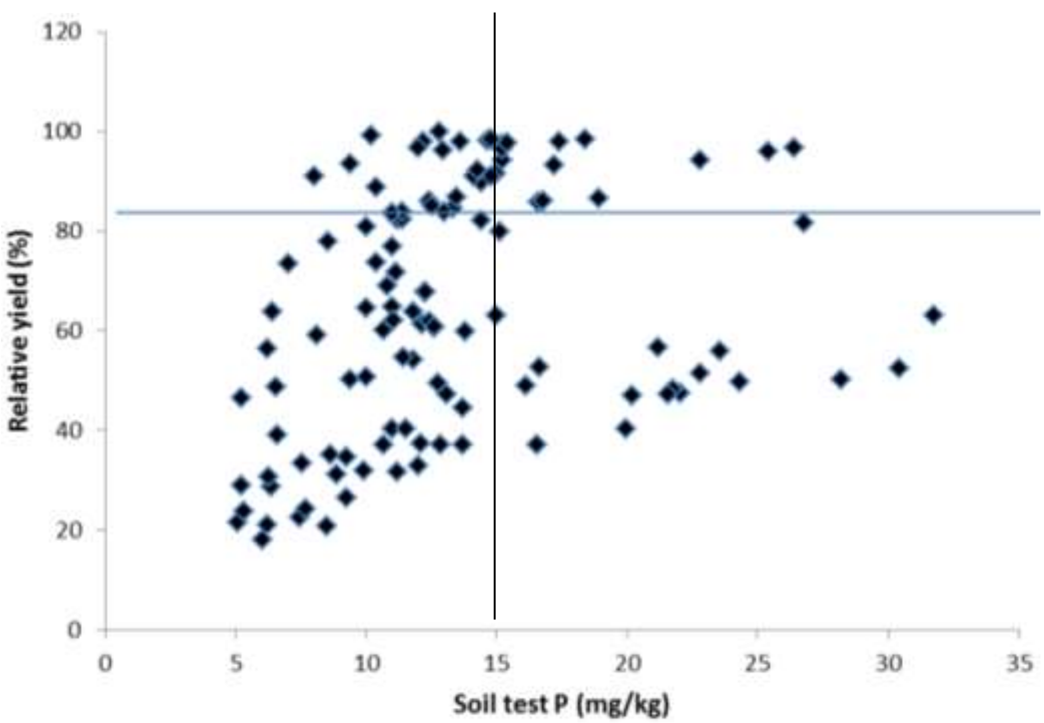


Figure 3. Relationships between soil extractable P measured using Bray-2 method and food barley relative yield (percentage of the maximum yield). Using the graphical method of Nelson and Anderson (1977), a critical limit of 13 mg P kg^{-1} of soil extractable P was identified.

Table 5. Determination of P requirement factor for food barley on Nitisols in 2012, 2013 and 2014.

Phosphorous rate (kg/ha)	Soil test P (Bray-II)		P increase over control	P requirement factor (Pf)
	Range	Average		
0	6.4-29.9	11.3		
10	7.0-28.2	12.5	1.2	8.5
20	6.6-27.9	13.3	2.0	9.8
30	7.2-24.3	13.6	2.3	13.0
40	6.6-30.4	15.3	4.0	9.9
50	7.1-31.8	16.4	5.1	9.8
Average	-	-	-	10.2

2014 compared to 2013 because the amount of moisture received in September 2014 was lower during the critical period of grain filling stage. The amount of precipitation received in July 2013 and 2014 was half and one third of the precipitation received in 2012, respectively.

Studies have indicated that grain yield and nutrient uptake of barley were greater in a relatively wetter season than the drier ones (Agegnehu et al., 2006). According to Jones et al. (2011) low nutrient uptake early in a plant's growth lowers nutrient quantity for the seed affecting yield. Crop uptake of nutrients is affected by soil and climatic conditions. One of the constraints is low soil moisture that restrict uptake of plant nutrients. This indicates that a successful soil test fertilizer program is reliant on rainfall and soil moisture status which influences the response of crops and yield to a greater extent than fertilizer applications.

Many different factors combine to limit the success of any soil test and the suitability of the recommendation for a given situation. Food security in Ethiopia is strongly dependent on rainfall variability and soil management practices. Below average seasonal rainfall, little or no rainfall (dry spells), persistent moisture deficit l, severe soil erosion and runoff loss of water and the resultant low soil fertility are the prominent causes for the low agricultural productivity in the Ethiopian highlands. Moreover, the continuous removal of crop residues coupled with minimal use of farmyard manure results in the mining of nutrients, organic matter depletion and weakening of soil structure (Tulema et al., 2007). These processes lead to increased runoff and erosion losses that are strongly linked to topsoil. Therefore, the practice of judicial water conservation undoubtedly plays a significant role in increasing agricultural production in the sub-humid areas where agriculture is hampered by periodic droughts and low soil fertility (Oicha et al., 2010).

Analysis of variance revealed that phosphorous had a highly significant effect on yield and yield component of food barley. Grain yield consistently increased as the rate of P increased up to 40 kg P ha^{-1} then a slight decrease in yield was observed at the highest rate 50 kg P ha^{-1} (Table 3). This could be due to low pH or the lower amount of nitrogen (N) applied at a rate of 60 kg N ha^{-1} alike to all plots. Agegnehu and Lakew (2013) revealed that application of P significantly increased the grain yield of malting barley.

According to the Nelson and Anderson method, the critical level of Bray-2 P in the top 15 cm of soil was about 13 mg kg^{-1}. At values of greater than or equal to 13 mg kg^{-1}, the crop achieved about 80% of its maximal yield in the absence of P fertilizer application (Figure 3). This implies that P fertilizer application could be recommended for a buildup of the soil P to this critical value, or maintaining the soil P at this level. Increasing P beyond this level, the cost of additional P fertilizer to produce extra yield would likely be greater than the value of the additional yield. Thus the soils with available P status below 13 mg kg^{-1}, yield of food barley could show a significant response to applications of P fertilizers. Whereas in areas with available P status greater than 13 mg kg^{-1}, the P concentration in the soil exceeds crop needs so that further addition of P fertilizer may not result in a profitable yield increase. Agegnehu and Lakew (2013) reported that Critical concentration of 12 mg P ha^{-1} for malting barley using Bray II test.

According to the result of our study, some yield responsive sites to P fertilizer applications had soil test levels above the critical level. Hence, to protect potential loss of food barley, at least a maintenance application of 10 kg P ha^{-1} may be required depending on the grain yield goal and profitability.

Following the pre-planting of soil analysis results all of the trial sites had lower soil P values than the critical P concentration. This had a direct relationship with the crop growth and yields. In most cases, soil pH less than 5.5 is deficient in available P and exchangeable cations (Brady and Weil, 2010). In such soils the proportion of P fertilizer that could be available to a crop becomes inadequate (Brady and Weil, 2010), unless amended through organic matter maintenance or liming to increase soil pH between 6.5 and 7 (Wortmann, 2015) for acid neutralization and applied through proper placement to increase the efficiency of utilization of the applied fertilizer. Higher coefficient variability in grain yield of food barley on Nitisols may have been related to greater variability within and among less fertile sites.

Conclusion

Soil-test P fertilizer calibration for food barley on Nitisols was proposed based on the Bray 2 extraction. This calibration is based on the analysis of six different P-rate test sites in which crop response to P fertilizer was determined in three cropping seasons (years). The results of this field work clearly indicated the importance of soil test based P fertilizer application on achieving maximum yield and yield components of food barley under field conditions of West Shewa on Nitisols soil type. In this part of the country, soil fertility depletion is severe and use of external input is very low. The critical available soil P concentration (13 mg kg^{-1}) in Bray -2 method and the average P requirement factor (10.2) on Nitisols have been established for the study sites and similar areas. The results seem promising and could be used as a basis for soil test P fertilizer recommendations for the production of food barley on Nitisols areas of central Ethiopian highlands. They can also be used for future intensification in the other areas for developing a system for soil test based fertilizer recommendation. Nevertheless, to develop an effective guideline for wider applicability of soil test based fertilizer recommendations, additional research assisted by appropriate soil P extraction method is required to generate sufficient information for the most important crop-soil systems.

CONFLICT OF INTERESTS

The author has not declared any conflict of interests.

ACKNOWLEDGEMENTS

The authors are grateful to Chanyalew Mandefro, Beyene Ofa, Haile Biza, Kasech Berhanu, the Barley Improvement Research Team and Holeta Research Center Soil and Plant Analysis Laboratory for their technical assistance. The Ethiopian Institute of Agricultural Research is acknowledged for funding the experiment.

REFERENCES

Agegnehu G, Lakew B (2013). Soil test phosphorous calibration for malting barley (*Hordium vulgare* L.) production on Nitisols of Ethiopian highlands. Trop. Agric. 90:177-187.

Agegnehu G, Liben M, Molla A, Feyisa A, Bekele A, Getaneh F (2011). "Research achievements in soil fertility management in relation to barely in Ethiopia." In Barley research and development in Ethiopia edited by B. Mulatu and S. Grando, ICARDA, Aleppo, Syria. pp. 137-152.

Agegnehu G, Ghizaw A, Sinebo W (2006). Yield performance and land use efficiency of barley and faba bean mixed cropping in Ethiopian highlands. Eur. J. Agron. 25:202-207.

Alice AS, Masateru M, Kengo I (2012). Effects of Soil Fertility Management on Growth, Yield, and Water-Use Efficiency of Maize (*Zea mays* L.) and Selected Soil Properties. Commun. Soil Sci. Plant Anal. 43(6):924-935.

Bado BV, DeVries ME, Haefele SM, Marco MC, Ndiaye MK (2008). Critical limit of extractable phosphorous in a Gleysol for rice production in the Senegal River valley of West Africa. Commun. Soil Sci. Plant Anal. 39:202-206.

Bekele W, Drake L (2003). Soil and water conservation behavior of subsistence farmers in the Eastern highlands of Ethiopia: A case study of the Hunde lafto area. Ecol. Econ. 46:437-451.

Bekele T, Ashagre A, Tulema B, GebreKidan G (1993). Soil fertility management in Barley. In: Barley research in Ethiopia: past work and future prospects. Proceedings of the first barley research review workshop.16-19 October 1993, Addis Ababa: IAR/ICARDA. Addis Ababa, Ethiopia.

Brady NC, Weil RR (2010). Elements of the nature and properties of soils. Pearson Education International. New Jersey.

Bray RH, Kurz LT (1945). Determination of total, organic, and available forms of phosphate in soil. Soil Sci. 59:39-45.

Bremner JM, Mulvaney CS (1982). Nitrogen total. In Methods of soil analysis, ed. A. L. Madison, WI: American Society of Agronomy, SSSA. pp. 595-624.

Buol SW (1995). Sustainability of soil use. Ann. Rev. Ecol. Syst. 26(1):5-44.

Chapman DD (1965). Determination of exchangeable Ca, Mg, K, Na, Mn, and effective cation exchange capacity in soil. In Methods of soil analysis, ed. C. A. Black, Madison, WI: ASA, SSSA. pp. 902-904.

Central Statistical Agency (CSA) (2014). Area and production of major crops for 2013/2014 Meher (main rainy) season for private peasant holdings in Ethiopia. Statistical bulletin. Addis Ababa.

David ME, David JT (2012). Modeling an Improvement in Phosphorus Utilization in Tropical Agriculture. J. Sust. Agric. 36(1):18-35.

Dahnke WC, Olsen RA (1990). Soil test correlation, calibration, and recommendation. P 45-71. In: R.L. Westerman (ed.) soil testing and plant analysis, 3rd ed., Soil science society of America, Madison, WI. SSSA Book Series: 3.

Haileslassie A, Priess JA, Veldkamp E, Lesschen JP (2006). Smallholders' soil fertility management in the central highlands of Ethiopia: Implications for nutrient stocks, balances and sustainability of agroecosystems. Nutr. Cycl. Agroecosyst. 75:135-146.

Henao J, Baanante C (2006). Agricultural production and soil nutrient mining in Africa: Implication for resource conservation and policy development. IFDC Tech. Bull. International Fertilizer Development Center. Muscle Shoals, Al. USA.

Hurni H (1988). Degradation and conservation of the resources in the Ethiopian highlands. Mt. Res. Dev. 8:123-130.

Jenny H (1941). Factors of soil formation: a system of quantitative pedology by Hans Hans Jenny. McGraw-Hill, New York.

Jones C, Olson-Rutz K, Dinkins CP (2011). Nutrient Uptake Timing by Crops: to assist with fertilizing decisions. Montana State University. USA.

Lakew B, Gebre H, Alemayehu F (1996). Barley production and research in Ethiopia. In: Gebre G, van Leur J, editors. Barley research in Ethiopia: past work and future prospects. Addis Ababa: IAR/ICARDA. pp. 1-8.

Mamo T, Haque I (1991). Phosphorous status of some Ethiopian soils.II. Forms of and distribution of inorganic phosphates and their relation to available phosphate. Trop. Agric. 68:2-8.

Nelson LA, Anderson RL (1977). Partitioning soil test–crop response probability. In Soil testing: Correlating and interpreting the analytical results, ed. T. R. Peck. Madison, WI: American Society of Agronomy pp. 19-39.

Oicha T, Cornelis WM, Verplancke H, Nyssen H, Govaerts B, Behailu M, Haile M, Deckers J (2010). Short term effects of conservation agriculture on Vertisols under tef (Eragrostis tef (Zucc.) Trotter) in the Northern Ethiopian highlands. Soil Till. Res. 106:294-302.

Omotayo OE, Chukwuka KS (2009). Soil fertility restoration technique in sub-Saharan Africa using organic resources. Afr. J. Agric. Res. 4:144-150.

Partey ST, Thevathasan NV (2013). Agronomic Potentials of Rarely Used Agro-forestry Species for Smallholder Agriculture in Sub-Saharan Africa: An Exploratory Study. Commun. Soil Sci. Plant Anal. 44(11):1733-1748.

Regassa H, Agegnehu G (2011). "potentials and limitations of acid soils in the highlands of Ethiopia: A review." In Barley research and development in Ethiopia edited by B. Mulatu and S. Grando. ICARDA, Aleppo, Syria pp. 103-112.

Rezaei SA, Gilkes RJ (2005). The effects of landscape attributes and community on soil chemical properties in the rangelands. Geoderma 125:167-176.

Sanchez PA, Woomer PL, Palm CA (1994). Agro-forestry approaches for rehabilitating degraded lands after tropical deforestation. In: Rehabilitation of degraded lands in the tropics: technical approach. JIRCAS International Symposium Series; 1992 Sep. 16-17; Tsukuba Japan. Tsukuba (Japan): Japan Int. Res. Center Agric. Sci. pp. 108-119.

SAS Institute (2001). SAS/STAT User's Guide, Version 8.2. SAS Institute, Cary, NC.

Seif J (2013). Soil testing. Fact sheet No. 0.501. Crop/soil series. Colorado state university. USA.

Shaver TM (2014). Soil testing and Nutrient recommendations. University of Nebrasaka.USA.

Smil V (2000). Phosphorus in the environment: natural flows and human interferences. Ann. Rev. Energy Environ. 25(1):53-88.

Sonon LS, Zhang H (2014). Soil test calibration work in southern USA. University of Texas A and M university. USA.

Tarekegne A, Tanner D (2001). Effects of fertilizer application on N and P uptake, recovery and use efficiency of bread wheat grown on two soil types in central Ethiopia. Ethiop. J. Nat. Res. 3:219-244.

Tulema B, Jens BA, Tor AB (2007). Availability of organic nutrient sources and their effects on yield and nutrient recovery of tef [*Eragrostis tef* (Zucc.) Trotter] and on soil properties. J. Plant Nutr. Soil Sci. 170:543-550.

Walkley A, Black CA (1954). An examination of the Degtjareff methods for determining soil Organic matter and proposed modification of the chromic acid titration methods. Madison, WI: ASA and SSSA.

Wortmann CS (2015). Principles of Fertility. nutrient management for agronomic crops in Nebraska. University of Nebraska–Lincoln Extension, Lincoln, NE. P 17.

Wortmann CS, Helmers M, Malarino A, Barden C, Devlin D, Pierensky G, Lory J, Massey R, Holz J, Shapiro C, Kovar J (2013). Agricultural phosphorous management and water quality protection in the mid west. Research publication Research publication RP187, Revised. University of Nebraska-Lincoln Extension, Lincoln, NE.

Yirga C, Hassan RM (2013). Determinants of inorganic fertilizer use in the mixed crop livestock farming systems of central highlands of Ethiopia. Afr. Crop Sci. J. 21:669-668.

academicJournals

Vol. 12(7), pp. 477-486, 16 February, 2017
DOI: 10.5897/AJAR2016.11580
Article Number: FBB2D0C62762
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Automation in the control of a water mixer used for sanitation of bovine milking unit

Carlos Eduardo Camargo Nogueira, Gilberto Carlos Arnauts, Enerdan Fernando Dal Ponte*, Rosemar Cristiane Dal Ponte, Jair Antonio Cruz Siqueira, Samuel Nelson Melegari de Souza and Eder Soares Machado

Postgraduation Program in Agriculture Energy Engineering, Western Paraná State University, UNIOESTE, Cascavel, PR, Brazil.

Received 19 August, 2016; Accepted 21 November, 2016

The milking unit sanitization is necessary after use. The processes are usually divided in steps and each one needs water at different temperatures for proper cleaning. However, to get the right temperature for each step cannot be easily achieved manually in rural properties. A hot water and cold water flow are needed. The farmer will need to properly mix the two water flows in order to reach a desired temperature for the cleaning step, and due to temperature fluctuations and flow, a constant intervention is necessary to maintain the same optimal temperature of the mixture. In this work, a mixer with automatic control of cold water flow, coupled to an electric heating system, was developed with the aim of reaching an ideal final temperature required for the sanitation of milking units, regardless of temperature variations and flow at the entrance. Two control boards were compared, the Arduino and the IOIO during the first seconds of operation of the system, the opening angle of the cold water registered is calculated from the hot and cold temperatures. After this gradual adjust in the angle was performed. The temperature was considered to be attained when it reached a maximum deviation of 1°C relative to the desired temperature. The system proved to be slightly more satisfactory with the IOIO, since the desired temperature was stabilized at lower average time, 24 s after system initialization.

Key words: Automation, sanitation, milking unit.

INTRODUCTION

The Brazilian herd for the production of milk is estimated to be 40 million animals, 27% of the total herd. Of these, 14 million are for cow milking (LEDIC, 2000). Such production is necessary since the average Brazilian consumption is 130 L per year, still below the recommended by the WHO, which is 175 L per year (SANTO, 2001).

The sanitization of milking units is with hot water,

*Corresponding author. E-mail: enerdan.ponte@pr.senai.br.

because the high temperature facilitates the removal of dirt in places difficult to reach. The optimum water temperature depends on the type of equipment to be cleaned, as well as of the brand, in addition to the phase of activity, following the manufacturer's specifications. This temperature, in case of milking units, is around 30 to 80°C. If the water does not reach the ideal temperature, cleaning cannot be effective, and if the temperature exceeds the upper limit, there may be damage to the equipment, causing losses.

For a proper cleaning of milking equipment, a few steps are necessary (MILKPARTS, 2002):

1. Pre-rinse: After milking, the equipment must be placed in the milk jar, and dipped in 5 to 10 L of water at 45°C. Thus, the milk that was left in the equipment will be drained into the milk jar.
2. Chlorinated alkaline cleaning: With 10 L of water at 80°C, the chlorine must be diluted in water. This step provides the removal of fats and proteins. During the passage of water through the equipment, it must be dipped and removed so it enters some air in the hoses, generating turbulence and increasing cleaning efficiency. This action must be made until the water cools to 50°C.
3. Rinse: This step must be repeated with the water at 30°C, so that the resulting detergent in the hoses is eliminated.

What many farmers use for water heating is a system with electric resistance or a Liquefied Petroleum Gas heater (SANTOS, 2002). These two systems, in most models, allow a good control of the temperature, from manually or automatically adjusted thermostats. In contrast, the costs of these systems are very high. Other farmers use water heating systems with solar energy. Although this system is cheaper, the method does not allow a precise control of water temperature, as this depends on factors such as room temperature and solar radiation.

The sun is considered an inexhaustible source of energy, greatly exploited by man. However, the use of solar energy is not yet made in significant scale in relation to other energy sources (Roger, 2004). The total amount of solar energy that reaches the atmosphere outside Earth is about 35,000 times greater than that consumed in the planet, according to STOUT data (1980).

A complete system of solar water heating is basically composed by solar collectors, heat reservoirs of accumulation and components that include an auxiliary energy source and a hot water distribution network, and are sized to provide enough hot water for the consumption points, at desired temperature and flow rate, as specified in project (Fossa et al., 2008; Chaguri, 2009; Sousa, 2009). The solar collectors can be flat or concentrators, in accordance with the purpose (Baptista, 2006; Costa, 2007; Lafay, 2005). Currently, the most used collectors are flat closed, most used worldwide, and the vacuum tubes flat, with growing market due to the excellent performance in cold climate regions (Rosa, 2012).

In general, the use of solar energy for water heating requires a low initial investment and very little spent on maintenance, being an optimal solution in the medium and long term.

To control the temperature in a manner that it would not be more expensive, for the heating project solar energy, the combination of two volumes of water at different temperatures, is used as an alternative in the correct amount to achieve the desired temperature. This is a complex process to be performed by the farmer, since some calculations are required to reach the correct mixture.

The development of a system that controls the mixing of water at different temperatures in order to obtain a desired final temperature can be financially feasible. A controller available for sale, and used for initial projects, is the *Arduino*. The Arduino system is a microcontroller capable of interacting with the outside through input and output devices, such as sensors and motors, respectively. It can be programmed for tasks such as measuring water temperature and control motors responsible for opening and closing registers, the program being written in Arduino Programming Language, with the use of IDE Arduino Development Environment (Arduino, 2014).

Another similar system is the IOIO, which is a board of input and output of data with several digital and analog connections. The ones with input can be connected to temperature sensors, water flow, luminosity and many others that are needed. This board is only an interface, so do not have the capability to run a program to interact with peripherals. The program has to run on a server with the operational system Android, as a mobile phone, which can run Java language with high complexity API. The communication of the board with the phone is via USB or Bluetooth (Ytai, 2011).

In this context, the present work proposes the development and evaluation of an automatic system for mixing hot and cold water, using the Arduino and IOIO platforms, in order to get water at the desired temperature for the sanitation process of a mechanical milking unit.

MATERIALS AND METHODS

The work was developed at the State University of Western Paraná, Campus Cascavel (PR) with Latitude 24°59' South, Longitude 58°23' West and average altitude of 785 m above sea level. The average annual temperature is 23°C (Brasil Channel, 2001).

The following items are described as the main equipment that were used or developed.

Boiler

The boiler equipment is responsible for storing hot water without large losses of temperature. This is basically formed by the reservoir, which stores the water efficiently to minimize heat losses,

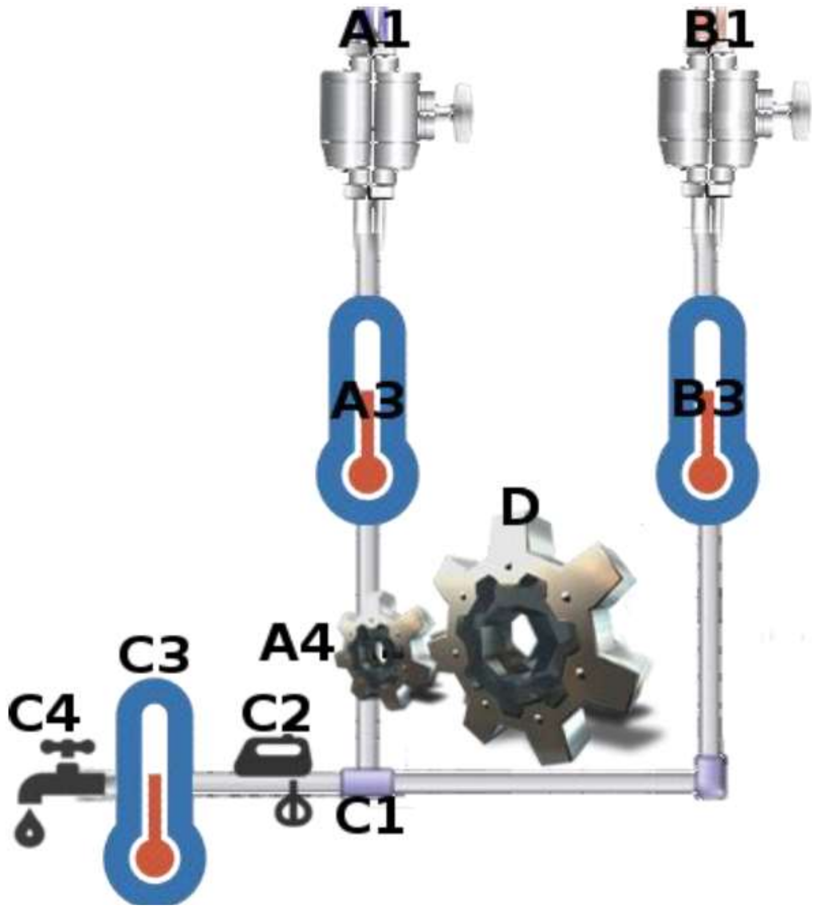


Figure 1. Scheme of the mixer developed.

and an electrical resistance, responsible for simulating a solar water heating system. A Solar Life brand boiler with the following features was used: Stainless steel 304, 200 L, low pressure, 3000 W auxiliary heating resistance.

Mixer

The mixer is the equipment developed to receive two water flows at different temperatures, having as output a single flow of water at the desired temperature. To be able to perform this function, the mixer is formed by several components, some specific of *IOIO* board, others of the *Arduino* board and other common to both. The mixer scheme details are shown in Figure 1.
Each mixer item is detailed:

A1: Cold water inlet;
B1: Hot water inlet;
A3: Cold water temperature sensor;
B3: Hot water temperature sensor;
A4: Registry for cold water flow control;
D: Controller engine for the A4 registry;
C1: Junction of cold and hot water flows;
C2: Section to cause turbulence;
C3: Final temperature sensor;
C4: Water outlet for use in the sanitation of the milking units.

Besides the common hydraulic devices, such as pipes and diverse connections, some other mixer components deserve attention and are described subsequently.

Two types of temperature sensors have been used in each one of the three points shown in the previous Figures A3, B3 and C3 one compatible with *Arduino* and the other with the *IOIO*. The first is the DS18B20 waterproof model, and the second is the NTC 10KTermopar model.

To control the flow of water, a sphere registry for cold water was used. The model used is metal with ¾ inch diameter.

To trigger the sphere registry, the servo motor brand Blue Bird, BMS-I530MG model was used. The main features of this servo motor are: Operating voltage between 4.8 and 7.4V dimensions 55 x 30 x 60 mm, 200 Ncm torque and opening of 141°. These features allow the servo motor to be used for opening and closing the water registry, from 0 to 90°.

The electrical power supply of the entire mixer was performed through a *Bestapress* source, model BP-503. It receives 127 or 220 V input and provides an output of 5 and 12 V.

The IOIO board used has, in addition to the input and output connections, also an input for power supply, as well as a USB connection, which in this work was used to attach the Bluetooth device, responsible for communication with the *Android* server. The language used for development of the software was Java. The *Android* server used was of a cell phone model *Philco phone 530*. This phone has support for human interface devices and platform for development of new software, besides a mini SD card attached, which was used to store the values of temperature and water flow

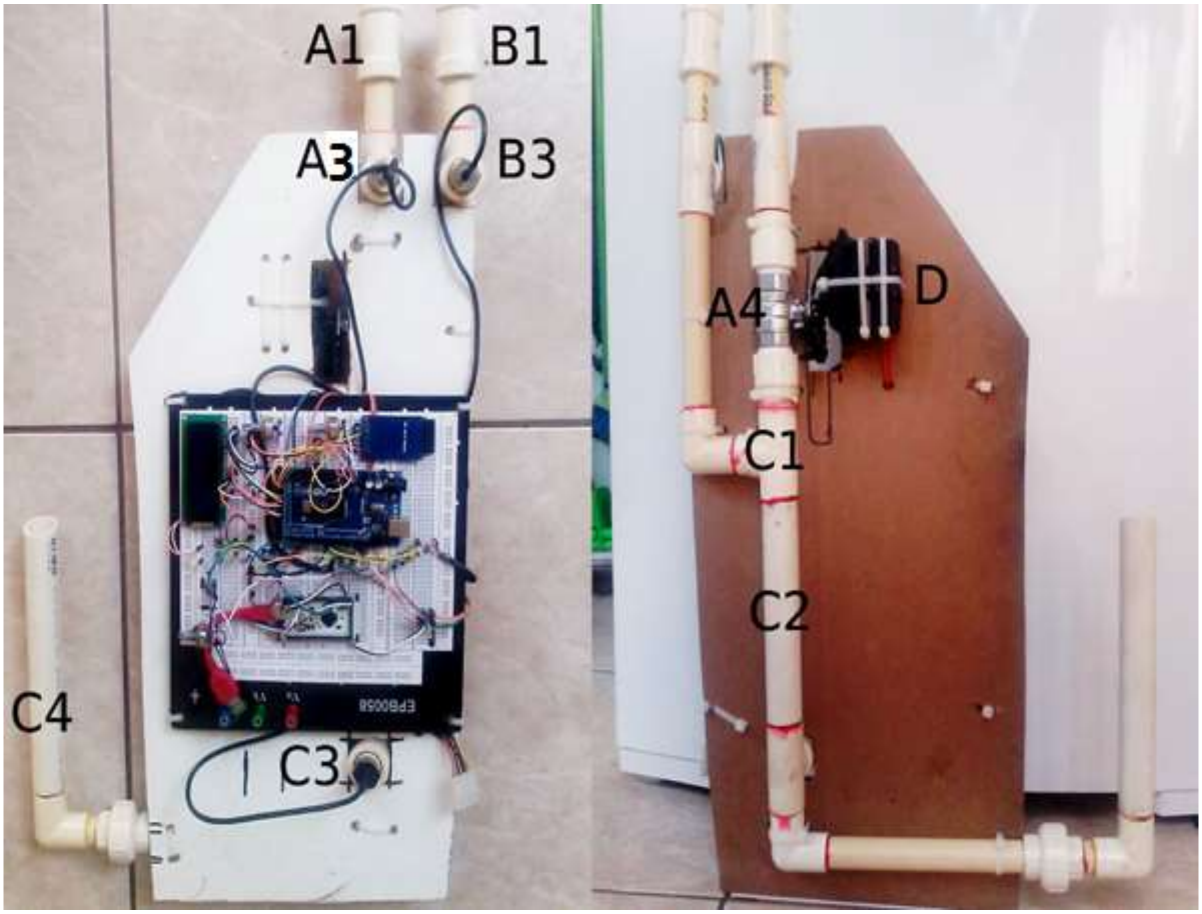


Figure 2. Mixer system details.

at every second.

For the solution with Arduino, the model Mega 2560 was used. This model was chosen due to the amount of entries and sufficient communication outputs for connecting all sensors and motors.

The information collected for temperature and water flow is stored in a mini SD card. This card has been attached in a *shield* in order to be recorded and read.

The information of water flow and temperature of each sensor, as well as temperature selected by the user, were displayed on a LCD display 16×2 model.

Mixer system details are presented in Figure 2.

Methodology

The equipment was installed at the place of the experiment. Cold water was obtained through a water tank without any temperature control, stored in the external environment, with a capacity of 100 L and refilled by public water network. The hot water was obtained through an electrical resistance installed in the boiler, controlled by a thermostat, aiming to simulate a solar water heating system. The maximum temperature verified in the boiler was 95°C.

The two water inlet flows, hot and cold, are directed to the mixer that, to the user command, will provide water at the desired temperature. This mixer has a temperature sensor for hot water, other for cold water, and another for mixed water. A water registry is responsible for controlling the flow of cold water, and the hot water flow has free passage. Finally a servo motor is responsible for opening or closing the water registry, in order to control the temperature of the mixed water. To increase the efficiency of the mixing, an uneven part of the pipe causes turbulence inside the pipeline (C2). Soon after the turbulence, the water passes through another temperature sensor, which should indicate the final temperature selected by the user. The control of the final temperature is made from the activation of the servo motor connected to the registry, being executed by the *Arduino* and *IOIO* controller boards. These two types of controllers were compared about the efficiency in controlling the final temperature of the water, as follows: At the beginning of the process, from the temperature data collected from the sensors, the registry opening angle is calculated and positioned at a point considered optimal to achieve the desired temperature. Thereafter, the water temperature is constantly adjusted through the automatic opening and closing of the cold water registry until it is stabilize at the desired temperature. Thus, the most efficient controller is considered the one that presented less time for stabilization of this temperature. A flowchart of the control system is presented in Figure 3 and the experiment structure is presented in Figure 4.

Turnaround time

The time between change in the registry angle and change in temperature has direct effects on the system performance, and this

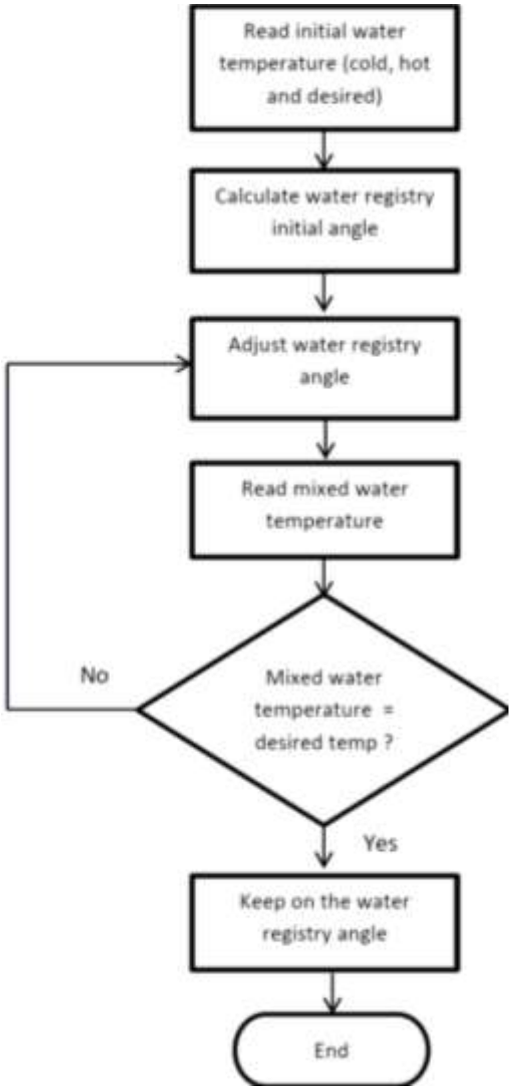


Figure 3. Flowchart of the control system.

depends on several factors, such as sensor response time, distance between the registry and the temperature sensor, heat loss, among others. In this work, final response time was investigated, which represents the time interval that occurs for stabilization of the desired temperature, after the beginning of adjustments in the registry angle.

Algorithm

The algorithm used to adjust the registry opening angle and, consequently, the desired temperature setting, is the same for *Arduino* and *IOIO*. This runs at every second, as well as all other functions of the mixer, such as temperature reading of the sensors, and SD card record. Also, at every second, the change is made in the registry angle, based on a function that determines the value of change.

The definition of the initial angle of opening the registry was made considering three methods: for the first, the angle is established at 45°; the second, the angle is calculated using an equation set from the ratio of hot and cold water flows; and the third, the angle is calculated using an equation set from the temperature data of the hot, cold and desired water, collected in previous experiments. These equation adjustments were performed with the software Matlab R2012a. The methodology that presented lower error in the initial angle calculation was adopted in the final algorithm to test the *Arduino* and *IOIO* platforms.

RESULTS AND DISCUSSION

From the temperature data of hot and cold water collected from the system, the opening angles of the cold water registry were calculated, so that the desired water temperature is reached. The system performance was evaluated for the *Arduino* and *IOIO*, using each one of the three methods applied in the stabilization of the temperature, which establishes the initial opening angle of the registry.

Starting angle at 45°

The first algorithm was developed with initial fixed angle at 45°, because, disregarding the temperatures of hot and cold water, this is the intermediate position between 0 and 90º.

Starting angle based on the flow ratio

The second algorithm was developed by varying the opening angle of the registry in 10 by 10°, from 0° (fully open registry) until 90° (fully closed registry), obtaining multiple flow data. It was verified that the maximum flow of hot water was 18 L min^{-1} (obtained with the registry angle at 90°, where there is no cold water flow), the maximum flow of cold water was 29, 30 L min^{-1} (obtained with the interruption of the hot water supply), and the maximum flow of hot and cold water together was 33, 16 L min^{-1} (obtained with the registry angle at 0°). From all the flow data collected, the relations between the flow of hot and cold water were calculated. Using the software Matlab R2012a, the best equation that relates the opening angle of the registry, with the relation of flow rates presented below was adjusted:

$$f(x) = -42.23 * x^{-1.015} + 81.79 \quad (1)$$
$$R^2 = 0.96$$

Where: *f(x)* = Angle, degrees; *x* = relation between the flow of hot and cold water, dimensionless;
R^2 = Determination coefficient.

Starting angle based on the temperatures collected

The third algorithm was developed from the water temperature data (cold, hot and desired), collected after

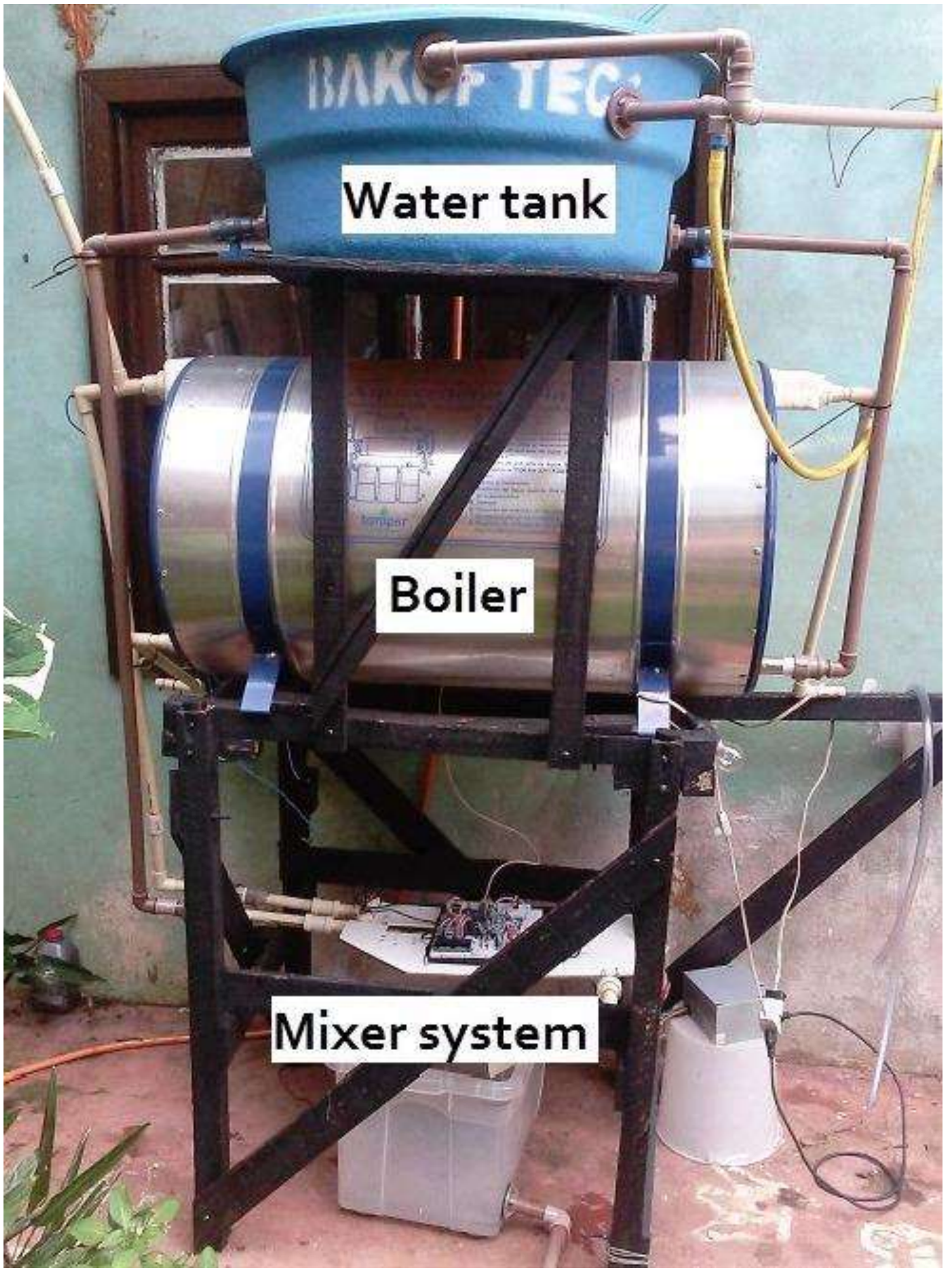


Figure 4. Experiment structure.

the stabilization of the desired temperature, and the registry opening angle data. In a similar manner, the previous method was adjusted, using the software Matlab R2012a. The best equation that relates the opening angle of the registry with the relation between the flow of hot and cold water is presented as follows:

$$f(x) = -27.19 * x^{-1.844} + 74.83 \quad (2)$$
$$R^2 = 0.964$$

Where: *f(x)* = Angle, degrees; *x* = relation between the flow of hot and cold water, dimensionless; R^2 = Determination coefficient.

In this case, the flow relation (value of x) was calculated from the temperature data collected, using the 1st Law of Thermodynamics, considering the occurrence of a mixture of fluids with different temperatures. The equation is presented as follows (Moran, 2011):

$$\frac{Q_{AQ}}{Q_{AF}} = \frac{T_{AD} - T_{AF}}{T_{AQ} - T_{AD}} \quad (3)$$

Where: Q_{AQ} = Hot water flow, L s^{-1}; Q_{AF} = Cold water flow, L s^{-1}; T_{AD} = Desired water temperature, °C; T_{AF} =

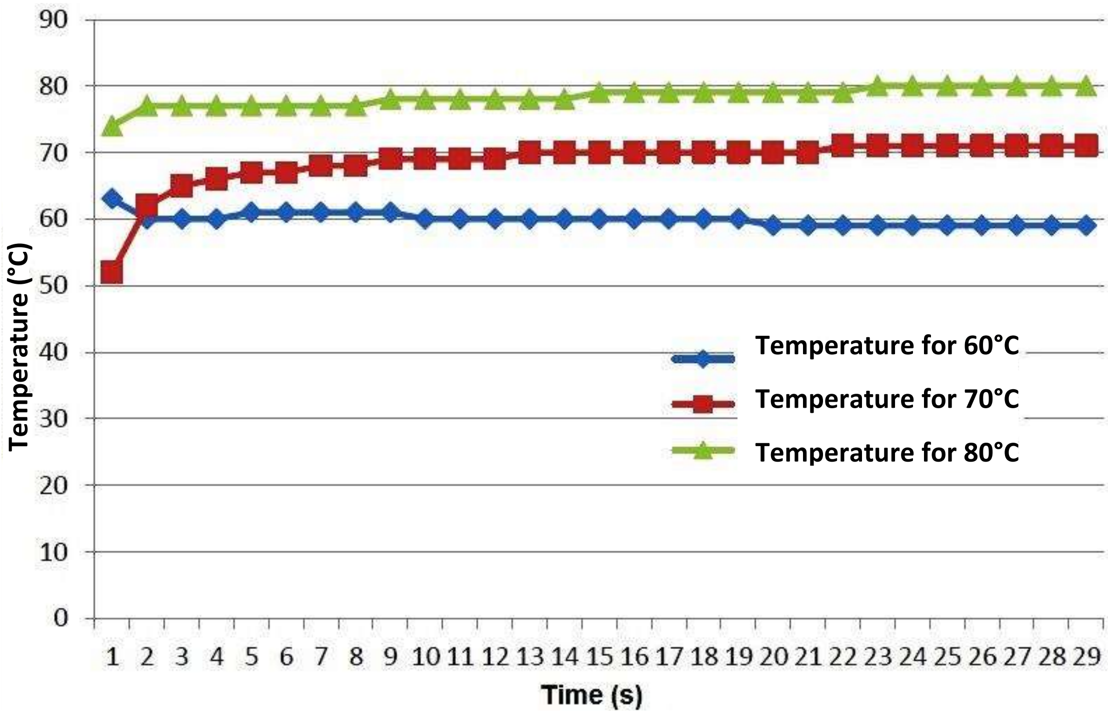


Figure 5. Temperatures for 3 experiments with Arduino.

Cold water temperature, °C; T_{AQ} = Hot water temperature, °C.

Comparison between the real and calculated opening angles

From the two equations adjusted to calculate the initial opening angle of the registries and the collected temperatures of hot, cold and desired water, the opening angles were calculated and compared with the real angles obtained in the experiment. Calculating the standard deviation of the errors of the real angles obtained in the experiment and calculated by the equations, the values of 4.5 and 6.2 respectively for the Equations 1 and 2 were obtained. It was therefore verified that Equation 1 had the lowest standard deviation, and therefore, was applied to the final algorithm to define the initial angle of registry openings. From this parameter, the comparison tests between the *Arduino* and *IOIO* response times were performed*.*

Comparison between the temperature stabilization times

The response time for the stabilization of the desired temperature were obtained considering two different situations of interest: initial angles fixed at 45º, or initial angles calculated by Equation 1, considered as the equation that presented the best angle adjustments. The calculations were made for both the Arduino and IOIO. It was verified that the use of Equation 1 greatly decreases the system response time, given that it provides the algorithm with an initial opening angle of the registry next to the real value, minimizing the number of iterations needed for temperature stabilization. The average values obtained for the stabilization times, using the angle at 45º and the angle obtained by Equation 1 were, 64.7 and 31.6s for Arduino, and 46.0 and 24.1 s, for IOIO respectively.

Taking into account the better performance of Equation 1, the data collected with Arduino and IOIO, for the desired temperatures of 60, 70 and 80ºC, are presented in Figures 5 to 8. These three temperatures were chosen to make the simulation and comparison of the systems, in order to verify which one presents better performance. However, the developed water temperature control system could be used for any other temperatures, within the range set for cleaning (from 30 to 80°C) (Milk Parts, 2002). For all tests performed, the average boiler temperature was around the maximum temperature controlled by the thermostat (95°C). Figures 5 and 7 shows respectively that for Arduino and IOIO, the outlet temperatures, over time, will converge to the desired temperatures. Figures 6 and 8 displays respectively for Arduino and IOIO that the registry opening angles, over time, will converge to the correct opening angles, which cause the outlet temperatures to be equal to the temperatures desired. It is important to emphasize that

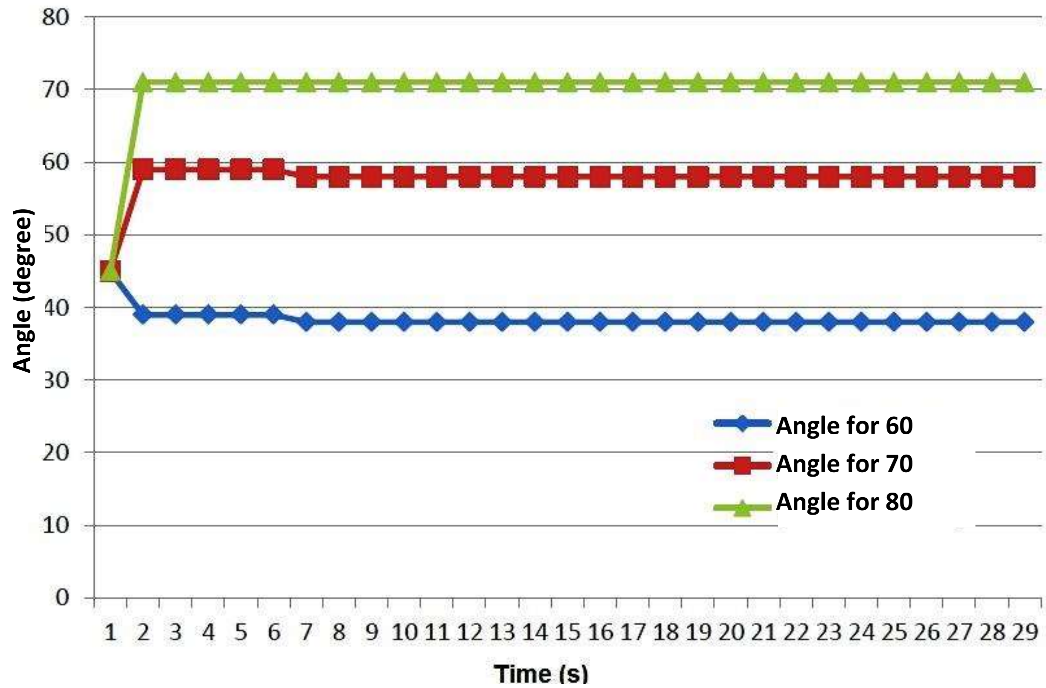


Figure 6. Angles for 3 experiments with Arduino.

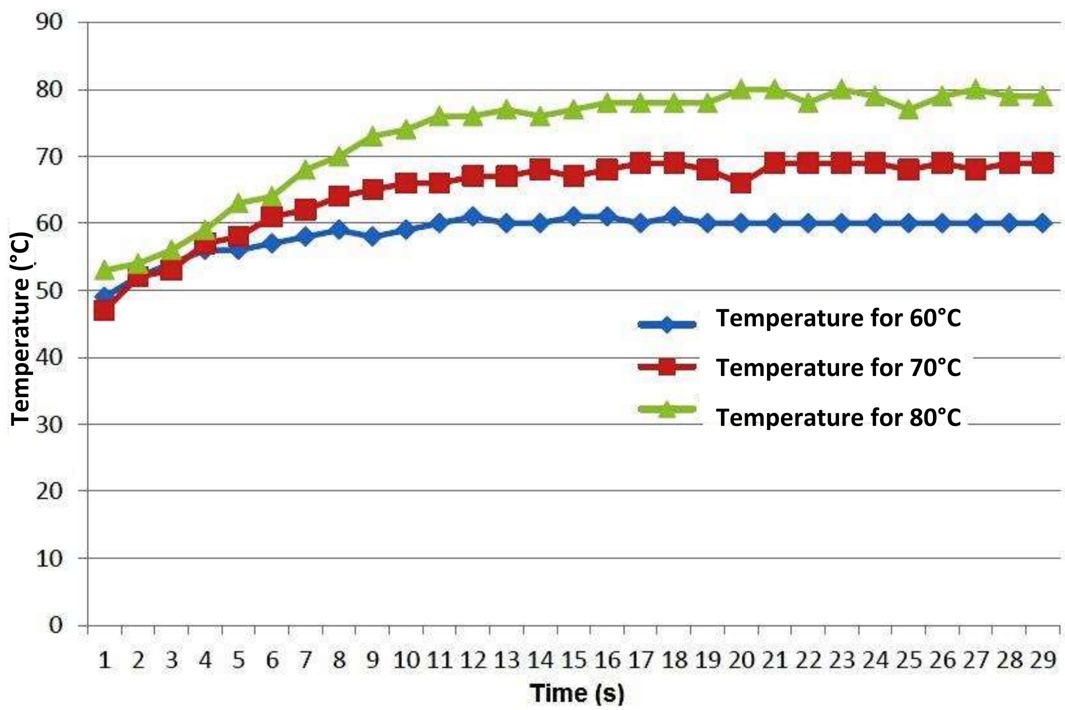


Figure 7. Temperatures for 3 experiments with IOIO.

the initial angle, calculated by Equation 1, is presented for t = 2s.

It is evident that the final result of the two systems is very similar; what differs is the oscillation that occurs in the adjustment of the outlet temperature and the opening angle of the registry; however it did not significantly

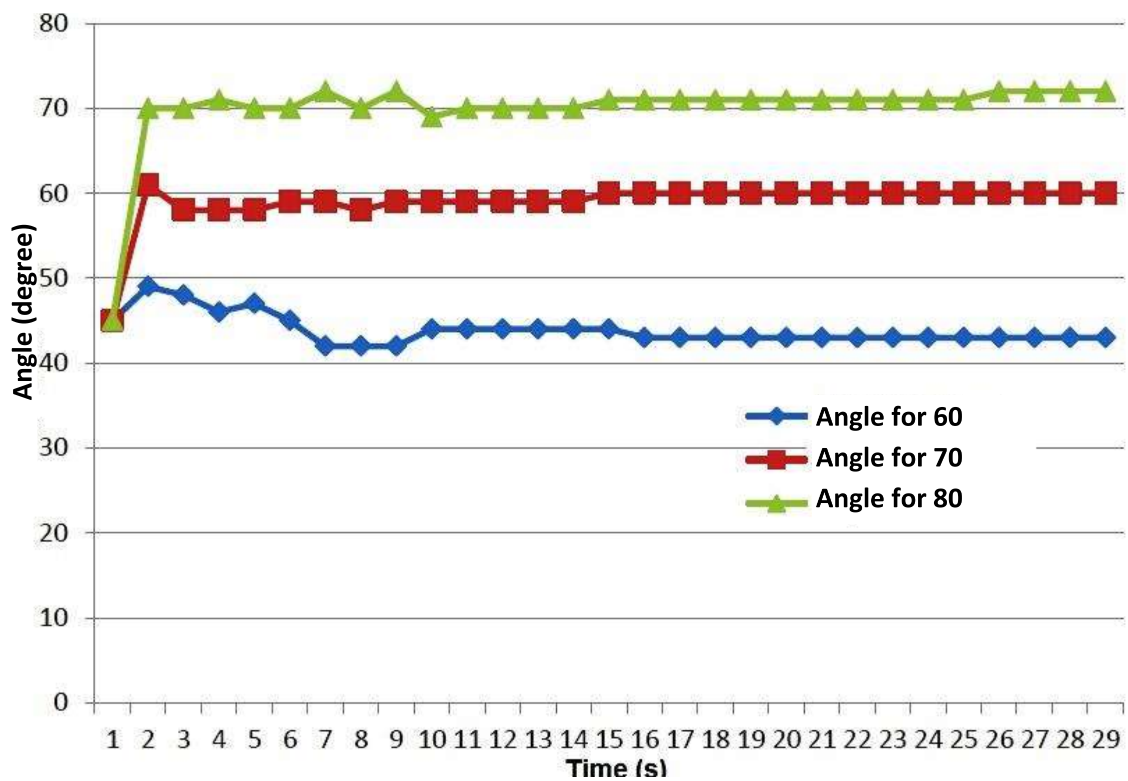


Figure 8. Angles for 3 experiments with IOIO.

influence the final result.

Conclusions

The results demonstrate that the required temperatures are reached within a few seconds after system startup and kept stable in the course of operation. This was possible by the success achieved in the stages of construction for the simulation of a solar heating system, the hydraulic unit, and its efficient control through IOIO and Arduino. The stabilization of the outlet temperature is linear in order to achieve the desired temperature without large oscillations (less than 1°C). The average time of stabilization of the outlet temperature was 31.6 s for Arduino, and 24.1 s for IOIO. The calculation of the initial angle of registry opening, based on the temperatures of hot, cold and desired water, quite sped up the arrival process at the desired temperature. Thus, the use of these systems (Arduino and IOIO) to control water temperature used for cattle milking units was shown to be very promising.

Suggestions for future researches

It is suggested to verify the influences of the temperature sensors response time, and the distance between register and sensors, in the results obtained in the experiment.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interest.

REFERENCES

ARDUINO (2014). Arduino. Accessed in May, 18, 2014. http://www.arduino.cc.

Baptista ASC (2006). Análise da Viabilidade Econômica da Utilização de Aquecedores Solares de Água em Resorts no Nordeste do Brasil. COPPE/UFRJ.

BRASILCHANNEL (2001). São Miguel do Iguaçu - PR. Accessed in July, 15, 2014. http://brasilchannel.com.br/municipios/mostrar_municipio.asp?nome=S% E3o+Mi guel+do+Igua%E7u&uf=PR.

Chaguri JJJ (2009). Sistemas prediais de aquecimento de água: parâmetros de dimensionamento e gerenciamento. Universidade de São Paulo.

Costa RNA. (2007). Viabilidade térmica, econômica e de materiais de um sistema solar de aquecimento de água a baixo custo para fins residenciais. Universidade Federal do Rio Grande do Norte.

Fossa JA, Santos EM, Junior JC, Fagá MTW (2008). Perspectivas tecnológicas para instalações internas de gases combustíveis em clientes residenciais. Cátedra Do Gás.

Lafay JMS (2005). Análise energética de sistemas de aquecimento de água com energia solar e gás. UFRGS.

Ledic ILG (2000). O grande trunfo da nossa pecuária leiteira. Editora Peirópolis.

MILKPARTS (2002). Manual de instruções ordenhadeira. http://milkparts.com.br/

Moran MJ, Shapiro HN (2011). Princípios de termodinâmica para Engenharia. Centro Federal de Educação Tecnológica Celso Suckow da Fonseca - CEFET/RJ. 6 ed.
Roger H (2004). Energia e Meio Ambiente. Editora Cengage Learning Editores.
Rosa FN (2012). Aplicabilidade de coletores solares com tubo evacuado no Brasil. UFRG.
Santo BRE (2001). Os caminhos da agricultura brasileira (2nd ed.). Orton IICA / CATIE.
Santos EM (2002). Gás natural: Estratégias para uma energia nova no Brasil. Editora Annablume.
Sousa AGL (2009). Desenvolvimento de software para projeto de sistemas centralizados de aquecimento solar de agua. Rio de Janeiro.
Stout BA (1980). Energia para a agricultura mundial. Itália: FAO.
YTAI (2011). IOIO Documentation Accessed in May, 15, 2014. https://github.com/ytai/ioio/wikiraphy.

academicJournals

Vol. 12(7), pp. 487-493, 16 February, 2017
DOI: 10.5897/AJAR2016.11726
Article Number: 4C4B54D62765
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Long-term application of swine manure on soybean grown in no-till system in Savannah soils

June Faria Scherrer Menezes[2], Mariana Pina da Silva[1*], Veridiana Cardozo Gonçalves Cantão[2], Jeander Oliveira Caetano[2], Vinícius de Melo Benites[3], Gabriela Wilk Baião Campos[4] and Bruno Lobato Rodrigues dos Santos[5]

[1]Universidade Estadual de Goiás, Campus de Ipameri Rodovia Go 330 Km 241 Anel Viário S/N Bairro: Setor Universitário CEP: 75780-000 Ipameri, Goiás, Brazil.
[2]Universidade de Rio Verde, Fazenda Fontes do Saber, CEP: 75901-970 Rio Verde, Goiás, Brazil.
[3]Pesquisador, Embrapa Solos. Rua Jardim Botânico 1024, CEP 22460-000 Rio de Janeiro, Rio de Janeiro, Brazil.
[4]Mestranda em Produção Vegetal Pela Universidade de Rio Verde, Fazenda Fontes do Saber, CEP: 75901-970 Rio Verde, Goiás, Brazil.
[5]Graduado em Agronomia pela Universidade de Rio Verde, Fazenda Fontes do Saber, CEP: 75901-970 Rio Verde, Goiás, Brazil.

Received 19 September, 2016; Accepted 27 January, 2017

The application of liquid swine manure (LSM) in the soil as biofertilizer is one of the most recycling alternatives adopted, because the nutrients contained in this residue can override, in part or totally, commercial fertilizers, crop productivity with a reduction in production costs. The goal with this work was to evaluate the use of LSM as a substitute of mineral fertilization on soybean culture in conditions of Savannah in 2014/2015. The experimental design was of randomized blocks with three replications. The treatments consisted of the use of mineral fertilizer recommended for soybean culture; without fertilization; and doses of LSM (25; 50; 75 and 100 m³ ha^{-1}). The levels of N, P, K, Ca, Mg, Cu, Zn, Mn and Fe in leaves, seeds and dry matter from the shoot, plant height and productivity of grain was assessed. There was no statistical difference between treatments that received mineral fertilization (P and K) and the treatments that received pig manure, indicating that the use of pig waste as bio fertilizer on soybean fertilization may be a promising alternative. The mineral fertilizer in soybean culture can be replaced by shots of LSM from 80 m³ ha^{-1} with no losses in grain productivity in a clayey dystroferric Red Latosol. The applications of doses of pig manure generally did not promote increase in the content of nutrients in grains and in the extraction of nutrients by the aerial part of the soybeans plants.

Key words: Bio fertilizer, successive applications, waste, organic fertilization, animal production, organic matter.

INTRODUCTION

Soybean has great economic and social importance in the world, because it is the main oilseed crop produced and consumed. Its production is destined for animal consumption, through the soybean meal, as well as for human consumption through oil and other products (Hirakuri and Lazzarotto, 2014). In ideal conditions of environment, the soybean responds with high productivity. One of the most important management techniques to get them is the fertilization. Aiming to improve utilization and reduce dependence on fertilizers, the Brazil alternative is search of soil fertilization. In many regions, there is use of local waste connections, being viable option, when used properly (Hoffmann et al., 2001).

The use of animal waste as fertilizer is a rational alternative and of great interest in terms of environmental, economic, social and agronomic traits. Apply organic waste in the soil system promotes infiltration and water absorption, improving the ability to exchange cations, resulting in better yields (Higashikawa et al., 2010). According to Lopes and Caixeta (2000), pig farming is an activity of economic importance mainly in the agricultural sector in the region of southwest Goiano. The Rio Verde region stands out for its dynamic economy and high grain production prompted the incentive of public policies for the State of Goiás, drawing huge agribusiness of pig meat to the region.

Currently, in this region, there are a large number of pig farms, where animals are kept in confinement system which is concentrated in small areas, large volumes of waste in liquid form. The proper management of these wastes is essential to the sustainability of the Brazilian swine demonstrating the need to know the environmental aspects of the scrap (Menezes et al., 2003). The pig manure, consisting of a mixture of feces, urine, food scraps, and a variable amount of water are typically handled in liquid form (Aita and Giacomini, 2008).

Various alternatives have been proposed for utilization of such waste in agriculture, and use as fertilizer in the soil, one of the most promising, since applied with discretion (Amorim et al., 2005; Sediyama et al., 2005; Corrêa et al., 2001; Santos et al., 2012; Lourenzi et al., 2014; Sediyama et al., 2014; Sousa et al., 2014). These technical criteria take into account the extraction of nutrients, nutritional requirement of the crop, soil fertility and waste analysis (Correa et al., 2011). Successive applications of liquid swine manure in the same area may lead to environmental problems such as the possibility of contamination of the water table (Zaman and Blennerhassett, 2010; Gonzatto et al., 2013; Aita et al., 2014; Giacomini et al., 2014).

The problem of pig manure management is complex and there is no *a priori* solution, but several possibilities have both positive and negative points. The researches developed so far offer motivating results that guide the optimization, in the field, of these procedures, and of new alternatives that integrate the productivity of pigs with the environmental preservation. Rising costs of commercial fertilizers, and increased environmental pollution make the use of organic waste in agriculture an attractive option, from the point of view due to nutrient cycling. These facts generate an increase in demand for the purpose of evaluating the technical and economic feasibility for the provision of some of these residues in agricultural soils (Santos et al., 2011).

However, due to the high cost of agricultural production, mainly by the use of mineral fertilizers and for being a residue that contains high levels of organic matter and of other nutrients, primarily nitrogen and phosphorus, the use of liquid pig manure becomes viable, because currently, agriculture research aimed to increase productivity and cost reduction. As a result of this, the work aimed to evaluate the effectiveness of the use of pig manure on productivity and the supply of nutrients for the crop of soybeans grown in no-tillage system in areas of Savannah.

MATERIALS AND METHODS

The experiment was conducted in the field in an area of Savannah named “Cerrado” in Brazil, at an experimental farm in the region of Rio Verde Goiás State (17° 48 'South 50° 55' West, elevation 760 m). The soil of the region is characterized as clayey dystroferric Red Latosol with texture showing 420, 470 and 110 g kg^{-1} of sand, silt and clay, respectively (Embrapa, 2006). The main characteristics of the soil (0 to 0.20 m depth, in natural conditions) with: pH: 4.5; Ca: 1.6 cmolc dm^{-3}; Mg: 0.6 cmolc dm^{-3}; K: 0.14 cmolc dm^{-3}; Al: 0.13 cmolc dm^{-3}; P (Mehlich-1): 3 mg dm^{-3}; saturation bases: 21% and organic matter content: 23 g kg^{-1}.

The region has a typical tropical climate, alternately wet and dry with cold average temperature exceeding 18°C, the rainfall is less than 2000 mm per year with rains in the summer and fall, according to the Köppen classification (Alvares et al., 2013). The first experiment was installed in 1999/2000. The soil was plowed, securely and acidity of the soil was corrected with lime (2.2 Mg ha^{-1}), as is usually done in the Cerrado, in order to raise the soil pH to 5.5 - 6.0 and bases saturation 60%, which is recommended for growing soybeans. The no-tillage system in the straw was adopted in all years subsequent farming.

Soy and corn crops were interspersed in succession and waste was applied annually in the following rates: 0 (control), 25, 50, 75 and 100 m^3 ha^{-1}, when soy was grown and 0, 25, 50, 100 and 200 m^3 ha^{-1} for the cultivation of corn. For comparison, the additional treatments with application of mineral fertilizers (NPK) we used.

For the 2014/2015 cropping, the soil received the 15th waste application. The experimental design was of randomized blocks with three replications. The treatments consisted of four doses of LSM (no LSM, 25 m^3 ha^{-1}, 50 m^3 ha^{-1}, 75 m^3 ha^{-1} and 100 m^3 ha^{-1}) and mineral fertilization with PK suggested by Shankar and Lobato (2004) at a ratio of 188 kg ha^{-1} of P_2O_5 in the form of monoammonium phosphate (MAP)+ 80 kg ha^{-1} of K_2O in the form of KCl applied before seeding culture of soybean with a total of 6 treatments and 18 installments. The area of each installment is 150 m^2 (15 m x 10 m).

Waste products used were from a farm of vertical system terminator with a capacity of 4000 piglets having remained 120 days in advance of anaerobic stabilization pond and was spread the day, 10/28/2014, ten days before seeding of soybean, by measuring

*Corresponding author. E-mail: mari_agro@hotmail.com.

Table 1. Chemical composition of the liquid waste of pigs used in soil in 2014.

N[1]	P[2]	K[3]	Ca	Mg
---- kg m^{-3} ----				
1.3	0.83	0.60	1.51	0.74
pH	Density	MO*	MS*	$S\text{-}SO_4$
	-- g cm^{-3} --	----- % ----		-- kg m^{-3} --
7.53	1.009	1.96	2.94	0.29

Percentage of conversion of nutrients applied: N = 50%[1], P=60%[2] e K = 100%[3] (CFSEMG, 1999)*.

the quantity of wastes by the flow of the distributor in a given period.

Manure samples were taken for determination of dry matter and chemical profiling (Pavan et al., 1992). The results of the chemical analysis of the used waste are presented in Table 1.

Organic and dry matters

Sowing was done in 11/6/2014 day, through mechanized planting; variety seeds were NS 7300 IPRO2 with 0.5 m spacing and planting density corresponding to 19 seeds per meter. During the cycle, all cultural practices needed for the culture were conducted (Embrapa, 2008).

In the flowering stage, R1/R2, foliar tissue samples were collected for determination of the levels of N, P, K, Ca, Mg, Cu, Zn, Mn and Fe as the methodology described by Sousa and Lobato (2004). After sampling, the leaves were washed, dried and then taken to an oven, at 65°C. After this process, the leaves were ground into Wiley type mill, stainless steel, passed sieves of mesh sizes of 0.84 mm^2 and wrapped in paper packaging properly identified for chemical analyses. In the dry material and ground, sulphuric and digestion was determined with the content of N by Kjeldahl distillation method. For the determination of the levels of P, Ca, Mg, Cu, Fe, Mn, Zn and K, the plant material underwent nitric-perchloric acid digestion method (AOAC, 1990), and then quantified by UV-vis spectrophotometry (UV-vis), and flame Atomic absorption spectrophotometry (Silva et al., 2009).

In determining the mass production of the shoot, three plants were collected per plot. The plant material collected was taken to the laboratory oven for drying, at 65°C. In the dry material, the levels of N, P, K, Ca, Mg, S, Cu, Fe, Mn and Zn was determined in accordance with the methodology described by Silva et al. (2009). At the time of harvest, the average height of five plants (cm) from ground level to the upper terminal part of the plant was determined.

For the harvest, soybean plants were collected from an area of 7.5 m^2 (6 rows with spacing of 0.5 and 2.5 m in length each). The grains of the plants of each plot were collected and weighed in the balance and given the moisture of grains of each parcel. The productivity of grain was adjusted to 13% moisture. The mass of 100 grains for each treatment was determined based on recommendation of the rules for seed analysis (Brasil, 1992). The beans were sent for chemical analysis and determination of the levels of macro and micro-nutrients according to Silva et al. (2009).

Foliar levels of macronutrients and micronutrients in soybean culture were compared with the critical levels of their nutrients suggested by Shah and Lobato (2004). The Sisvar software (Ferreira, 2011) was used for statistical analysis of the variables. The averages of the treatments, when significant, were compared, by the Tukey test, to 5% of probability. When significant difference in the interaction among the factors was found, regression analysis was carried out.

RESULTS AND DISCUSSION

For the chemical composition of the liquid pig manure (LSM) as shown in Table 1 and the mineral fertilizers applied on the plots, the amounts of nutrients applied in function of the treatments were calculated (Table 2).

It was observed that the fertilization with liquid pig manure (LSM) overcame the mineral nitrogen fertilization, regardless of the dose applied. The dose of 75 m^3 ha^{-1} of LSM was similar to phosphorus mineral fertilizer and the dose of 100 m^3 ha^{-1} of LSM was similar to the mineral potassium fertilizers.

The mineral fertilization did not provide Ca, Mg, and S, whereas with the LSM, the higher the dose, the greater the supplies of these nutrients (Table 2). Unlike mineral fertilizers, which have minimum composition set for each condition of soil and culture, the pig waste composition is extremely unbalanced, varying according to the power supply, water management, storage conditions and the age of the animals (Scherer et al., 1995; Konzen and Barros, 1997) making hard to recommend. Therefore, continuous applications can cause imbalance of nutrients in the soil and the gravity of the problem will depend on the composition and amount of waste, time of application, the ability to extract by crops and the soil type.

According to Konzen and Barros, (1997), it is fundamental to develop a technical management and fertilization plan so that it becomes a viable alternative to increase water availability, which can reduce treatment costs and also serve as a source of nutrients for the plants, and the costs of acquiring commercial chemical fertilizers. The foliar contents of N, P, K, Ca, Mg, S, Fe, Mn, Cu and Zn in soybeans were not affected ($p > 0.05$) by treatments (Table 3). Similar results were obtained by Santos et al. (2012), Sediyama et al. (2014), Seidel et al. (2010) and Bócoli et al. (2016), which detected no changes in foliar concentrations of N, P and K in squash, peppers and corn, respectively, with the application of pig manure. However, Corrêa et al. (2008) noted that, with increasing doses of organic waste, N content increased in soybean leaves. Araújo et al. (2013) verified that the application through mineral fertilizer reduced foliar N content in soybeans.

Comparing the levels of macronutrients in soybean

Table 2. Amount of nutrients applied in soil in 2014 on the basis of the treatments.

Treatments	N	P	K	Ca	Mg	S (SO_4)
	------------------------ kg ha^{-1} --------------------					
Control (0)	0	0	0	0	0	0
25 m^3 ha^{-1} de LSM	32.5	20.7	15.0	37.7	18.5	7.2
50 m^3 ha^{-1} de LSM	65.0	41.5	30.0	75.5	37.0	14.4
75 m^3 ha^{-1} de LSM	97.5	62.3	45.0	113.5	55.5	21.6
100 m^3 ha^{-1} de LSM	130	83.0	60.0	151.0	74.0	28.8
Mineral fertilizer	16.9	64.8	66.7	0	0	0

Table 3. Nutrient content in foliar tissue of soybeans due to the application of liquid swine manure.

Variable	M.F.	Doses of liquid waste of swine (m^3 ha^{-1})					Average	F value	CV (%)
		0	25	50	75	100			
N (g kg^{-1})	36.7	37.0	42.3	31.1	41.7	34.7	37.3	1.30^{NS}	17.32
P (g kg^{-1})	3.3	3.0	3.1	3.7	3.4	4.1	3.44	2.84^{NS}	12.59
K (g kg^{-1})	17.3	18.5	17.0	19.1	18.4	19.5	18.32	0.78^{NS}	10.81
Ca (g kg^{-1})	8.5	10.6	10.8	10.1	10.4	9.6	10.01	2.02^{NS}	10.27
Mg (g kg^{-1})	3.2	4.1	4.0	4.0	4.2	4.0	3.91	2.12^{NS}	11.13
S (g kg^{-1})	1.8	1.5	1.5	1.7	1.8	2.0	1.73	1.24^{NS}	16.07
Fe (mg kg^{-1})	87.0	78.3	81.3	74.2	82.8	87.2	81.82	1.06^{NS}	10.30
Mn (mg kg^{-1})	55.2	61.4	61.4	56.5	55.7	51.7	57.01	2.24^{NS}	7.68
Cu (mg kg^{-1})	7.0	5.8	6.2	6.3	66.0	6.0	6.32	1.16^{NS}	10.53
Zn (mg kg^{-1})	32.9	28.2	28.3	32.8	33.4	41.2	32.8	2.20^{NS}	16.84

^{NS}- Not significant by the F test ($p > 0.05$); M.F.- mineral fertilization; CV- coefficient of variation.

plant tissue with their respective critical levels (reference) according to Sousa and Lobato (2004), it was found that the levels of P, K, Ca and Mg were suitable, demonstrating nutritional balance for these elements. Detected deficiency of nitrogen (N) and sulphur (S) is independent of treatments (Table 3). Although results of other authors indicate that fertilization with pig manure is efficient for nutrition and productivity of crops, a dose suited to the requirement of culture is applied (Konzen and Alvarenga, 2005). However, in this experiment, the fertilization failed to meet the nutritional demands of the plant in N and S (Table 3).

In the culture of soybean, nitrogen fertilizer is not recommended due to biological nitrogen fixation (BNF) and doses greater than 30 kg ha^{-1} of N may interfere negatively with BNF (Hungria et al., 2001). Another factor that may have limited the use of N may have been N losses by volatilization of ammonia ($N\text{-}NH_3$) during the application of the waste. According to Sommer and Hutchings (2001), N losses by volatilization of ammonia can be up to 75% of the ammoniacal N of the waste.

The recommendation of sulfur for crops is of 20-30 kg ha^{-1} (Sousa and Lobato, 2004), and the doses of 75 and 100 m^3 ha^{-1} of LSM would supply this requirement, though inefficient in plant nutrition. Possibly, most of the S contained in the waste is in organic form for digestion. Although there has been a deficiency of N and S visual symptoms of these nutrients were not observed.

According to the data in Table 4, no significant difference among the treatments with regard to productivity and mass of 100 grains of soy was seen. The different doses of manure used on the base fertilizer, as compared to the chemical fertilizer (NPK), did not differ statistically, demonstrating the efficiency of waste as bio fertilizers on grain production this crop (Table 4). Some authors also performed work with pig manure on soybean culture (Konzen, 2003; Menezes et al., 2007; Blanco, 2015) and verified these beneficial effects on productivity of culture. This demonstrates that the fertilization of soybean culture in this soil was required, even if the nutrient supplies were carried out with application of mineral fertilizer or through pig manure. The pig manure provided production gains as compared to the mineral fertilization; therefore, total or partial form in fertilization programs can be replaced.

The average yield of grain with the experiment was above the national average (2999 kg ha-1) and the State of Goias (2595 kg ha^{-1}) in 2014/2015 (Conab, 2015). According to the data in Table 5, it can be seen that only significant differences occurred between treatments (p <

Table 4. Yield kg grain ha^{-1} (PG) and weight of 100 grains g (M100G) of soybeans due to the application of liquid swine manure.

Variable	M.F.	Doses of liquid waste of swine (m^3 ha^{-1})					Average	F value	CV (%)
		0	25	50	75	100			
PG	4002	3682	3785	4550	4314	4369	4117	0.96^{NS}	14.88
M100G	16.4	12.0	17.1	15.9	16.1	15.7	15.53	1.48^{NS}	16.57

^{NS}- Not significant by the F test (p > 0.05); ** significant at 1% probability; PG- grain productivity; M100G- mass of 100 grains; M.F.- mineral fertilization; CV- coefficient of variation.

Table 5. Levels of nutrients in soy beans related to application of liquid swine manure.

Variable	M.F.	Doses of liquid waste of swine (m^3 ha^{-1})					Average	F value	CV (%)
		0	25	50	75	100			
N (g kg^{-1})	44.1	47.8	47.8	54.0	44.1	53.7	46.54	2.72^{NS}	15.66
P (g kg^{-1})	4.8	4.3	4.3	5.0	4.6	5.2	4.67	1.29^{NS}	14.02
K (g kg^{-1})	19.5	18.0	18.0	19.2	18.0	18.6	18.57	1.11^{NS}	5.80
Ca (g kg^{-1})	2.8	2.5	2.5	2.8	2.6	3.0	2.81	0.46^{NS}	18.18
Mg (g kg^{-1})	3.0	3.0	3.0	3.1	3.0	3.0	3.02	0.61^{NS}	5.51
S (g kg^{-1})	2.9	3.0	3.0	2.8	2.7	3.2	2.89	0.93^{NS}	11.85
Fe (mg kg^{-1})	623.2^{ab}	461.9^{abc}	461.9^{abc}	559.2^{ab}	682.1^{a}	305.5^{c}	507.1	7.66**	17.34
Mn (mg kg^{-1})	39.2^{b}	60.8^{a}	60.8^{a}	43.7^{b}	37.8^{b}	34.8^{b}	44.28	8.64**	12.74
Cu(mg kg^{-1})	9.4	11.1	11.1	11.2	11.0	9.2	10.27	2.05^{NS}	10.90
Zn (mg kg^{-1})	35.2	35.4	35.4	37.0	36.6	37.0	35.93	0.35^{NS}	9.30

^{NS}- Not significant by the F test (p > 0.05); ** significant at 1% probability; M.F.- mineral fertilization; CV coefficient of variation.

0.05) for contents of Fe and Mn in soy beans. Despite the significance in the doses of liquid pig waste, the equations showed no adjustment, with low coefficient of determination. It is observed in Table 5 that while the application of pig waste at a dose of 100 m^3 ha^{-1} were observed with lower levels of iron and manganese in soy beans.

As compared with the work carried out by Embrapa (2008) in relation to quantity exported by the nutrient culture of soybean grain (Table 6), similar values observed in relation to levels of N, P, K, Ca, Mg, Mn, Cu and Zn and lower values relative to Fe (70 mg kg^{-1}) and above in relation to the S (5.4 g kg^{-1}). This occurs due to nutritional requirement of soybeans and export potential of culture is characteristics determined by genetic factors, but influenced by climatic factors, the fertility of the soil and the cultural management. Nutrient content in the dry matter of the aerial part of the culture of soybeans were not affected by treatments except for sulphur and zinc nutrients in which significant differences were observed (Table 6). Despite the significance in the doses of liquid pig waste, the equations showed no adjustment, with low coefficient of determination.

It can be concluded from Table 6 that the extraction of nutrients by the aerial part of the plant of soybean occurred in the following order: K>Ca>N>Mg>P>S> Fe> Mn>Zn>Cu. Contrary to the one obtained by Pedrinho Junior et al. (2004) working with soybeans in protected environment, it was found that the greatest accumulation of nutrients in soy was to P, Mg and S. Data was obtained by Caires and Fonseca (2000) working with soybeans in no-tillage system showing that nutrient extraction from the shoot by plant soybeans have the following sequence N>K>Ca>P>Mg>S. Certainly, these values vary according to the levels available in each soil, with the environment and with the ability to extract each cultivar. The average yield of soya beans increased depending on the dose of LSM, fitting by regression to the second degree polynomial function (Figure 1), and the dose that presented the greatest productivity was 80.3 m^3 ha^{-1} of liquid pig manure, 4412 kg ha^{-1} of grain. Similar results were found by Penha et al. (2015). The authors applying doses of LSM in the culture of soybean in 2008/2009 in the same experimental area of this study found that the maximum efficiency (4111 kg ha^{-1}) was obtained with the application of 88 m^3 ha^{-1} of liquid pig manure.

In accordance with results obtained by Borre (2008) in which the biggest returns in productivity from soybeans occurred with doses of 50 and 75 m^3 ha^{-1} of liquid manure from pigs with 3918 and 3870 kg ha^{-1}, respectively, Konzen (2003) also obtained with the dose of 50 m^3 ha^{-1} of liquid pig manure, the highest rates in productivity of soya beans, which is higher than the

Table 6. Nutrient content in the dry matter of the aerial part of the soy culture related to the application of liquid swine manure.

Variable	M.F.	Doses of liquid waste of swine (m^3 ha^{-1})					Average	F value	CV (%)
		0	25	50	75	100			
N (g kg^{-1})	5.4	5.4	5.1	6.1	6.0	6.2	5.70	0.38^{NS}	21.98
P (g kg^{-1})	0.7	0.8	0.7	0.7	1.1	0.9	0.80	2.46^{NS}	20.73
K (g kg^{-1})	13.4	12.7	13.4	11.1	15.6	16.9	13.85	1.58^{NS}	20.68
Ca (g kg^{-1})	7.8	7.1	7.8	8.3	8.3	8.0	7.87	0.41^{NS}	15.23
Mg (g kg^{-1})	5.2	4.2	4.8	5.2	5.5	5.5	5.07	0.99^{NS}	16.41
S (g kg^{-1})	0.8a	0.4cd	0.4c	0.3d	0.9a	0.6b	0.57	80.04**	8.83
Fe (mg kg^{-1})	447.4	433.4	551.3	563.6	473.2	458.2	487.9	1.89^{NS}	14.37
Mn (mg kg^{-1})	31.6	26.7	30.4	33.0	25.2	28.6	29.30	0.84^{NS}	19.30
Cu(mg kg^{-1})	4.0	4.3	4.4	4.1	4.0	4.2	4.20	0.56^{NS}	8.87
Zn (mg kg^{-1})	4.3 a	3.7ab	3.4ab	4.3a	2.4b	4.5a	3.76	5.67**	15.27

^{NS}- Not significant by the F test (p > 0.05); ** significant at 1% probability; M.F.- mineral fertilization; CV- coefficient of variation.

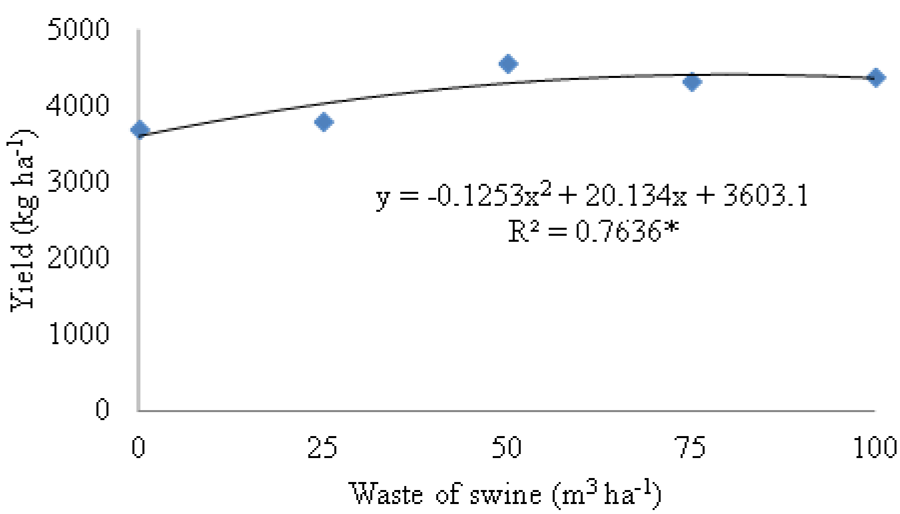


Figure 1. Productivity of soya beans in increasing doses function of pig manure, Cropping 2014/2015. Value of F-1.047 * – by the Tukey test at 5%.

maximum dose (100 m^3 ha^{-1}) used in the work.

Conclusions

There was no statistical difference between treatments that received mineral fertilizer (NPK) and treatments that received pig manure, indicating that pig waste utilization as bio fertilizer can be one of the alternatives to this culture of soybean.

The mineral fertilizer in soybean culture can be replaced by doses of liquid pig manure from 80 m^3 ha^{-1} in a clayey dystroferric Red Latosol without damage of the components of yield and not significant increase in grain productivity from this dose. Successive applications of doses of pig manure generally did not promote increase in the content of nutrients in grains and in the extraction of nutrients by the aerial part of the plant.

CONFLICTS OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

The authors thank the Cnpq/Fapeg for the financial support.

REFERENCES

Aita C, Giacomini SJ (2008). Nitrato no solo com a aplicação de dejetos líquidos de suínos no milho em plantio direto. Rev. Bras. Ciênc. Solo 32:2101-2111.

Aita C, Gonzatto R, Miola ECC, Santos DB, Rochette P, Angers DA, Chantigny MH, Pujol SB, Giacomini DA, Giacomini SJ (2014). Injection of dicyandiamide-trated pig slurry reduced ammonia volatilization without enhancing soil nitrous oxid e emissions from no-till corn in Southern Brazil. J. Environ. Qual. 43:789-800.

Alvares CA, Stape JL, Sentelhas PC, Gonçalves JLM, Sparovek G (2013). Köppen's climate classification map for Brazil. Meteorol. Zeitschrift 22(6):711-728.

Amorim AC, Lucas Jr J, Resende KT (2005). Compostagem e vermicompostagem de dejetos de caprinos: Efeito das estações do ano. Eng. Agríc. 25(1):57-66.

Araújo K, Mendes FL, Debiasi H, Balbinot Jr AA, Franchini JC (2013). Produtividade e teor foliar de nitrogênio na soja em função da pressão de pastejo, época de dessecação e aplicação de nitrogênio em sistema lavoura-pecuária. In: JORNADA ACADÊMICA DA EMBRAPA SOJA, 8., 2013, Londrina. Resumos Expandidos… Londrina: Embrapa Soja pp.123-126.

Association of official analytical chemists (AOAC) (1990). Official methods of analysis. 15.ed. Arlington, 1990. 1117p.

Blanco DB (2015). Adubação da cultura da soja com dejetos de suínos e cama de aviário. Dissertação de mestrado em engenharia de energia na agricultura- universidade estadual do oeste do paraná campus cascavel 36 f.

Bócoli ME, Mantovani JR, Miranda JM, Marques DJ, Silva AB (2016). Soil chemical properties and maize yield under application of pig slurry biofertilizer. Rev. Bras. Eng. Agríc. Amb. 20(1):42-48.

Borre IC (2008). Viabilidade agronômica da cultura da soja com a utilização de dejetos líquidos de suínos. 2008. 49f. (Monografia de graduação). Fesurv, Rio Verde, GO.

Brasil Ministério da Agricultura e Reforma Agrária (1992). Regras para análise de sementes. Brasília: SNDA/ DNDV/CLAV. 364 p.

Caires EF, Fonseca AF (2000). Absorção de nutrientes pela soja cultivada no sistema plantio direto em função da calagem na superfície. Bragantia, Campinas 59(2):213-220.

Conab – companhia nacional de abastecimento (2015). Indicadores da agropecuária. Ano XXII, nº 01, Jan 2015, Brasília.

Corrêa JB, Reis Jr RA, Carvalho JG, Guimarães PTG (2001). Avaliação da fertilidade do solo e do estado nutricional de cafeeiros do Sul de Minas Gerais. Ciênc. Agrotecnol. 25:1279-1286.

Corrêa JC, Büll LT, Crusciol CAC, Tecchio MA (2008). Aplicação superficial de escória, lama cal, lodos de esgoto e calcário na cultura da soja. Pesqui. Agropecu. Bras. 43:1209-1219.

Embrapa - Empresa Brasileira de Pesquisa Agropecuária (2006). Sistema brasileiro de classificação de solos. 2. ed. Rio de Janeiro: Embrapa Solos. 306 p.

Embrapa (2008). Tecnologias de Produção de Soja - Região Central do Brasil 2009 e 2010. Sistema de Produção, n. 13 - Londrina: Embrapa Soja 262 p.

Ferreira DF (2011). Sisvar: a computer statistical analysis system. Ciênc. Agrotecnol. 35(6):1039-1042.

Giacomini DA, Aita C, Pujol SB, GiacominI SJ, Doneda A, Cantú RR, Dessbesell A, Lüdtke RC, Silveira CAP (2014). Mitigação das emissões de amônia por zeólitas naturais durante a compostagem de dejetos de suínos. Pesqui. Agropecu. Bras. 49:521-530.

Gonzatto R, Miola ECC, Doneda A, Pujol SB, Aita C, Giacomini SJ (2013). Volatilização de amônia e emissão de óxido nitroso após aplicação de dejetos líquidos de suínos em solo cultivado com milho. Ciênc. Rural 43:1590-1596.

Higashikawa FS, Silva CA, Bettiol W (2010). Chemical and physical properties of organic residues. Rev. Bras. Ciênc. Solo Viçosa 34(5):1743-1752.

Hirakuri MH, Lazarotto JJ (2014). O agronegócio da soja nos contextos mundial e brasileiro [recurso eletrônico] - Londrina: Embrapa Soja 70:349.

Hoffmann I, Gerling D, Kyiogwom UB, Mané-Bielfeldt A (2001). Farmers management strategies to maintain soil fertility in a remote area in northwest Nigeria. Agric. Ecosyst. Environ. Amster. 86(3):263-275.

Hungria M, Campo RJ, Mendes IC (2001). Fixação biológica do nitrogênio na cultura da soja. Londrina: Embrapa Soja 48p. (Embrapa Soja. Circular Técnica, 35; Embrapa Cerrados. Circular Técnica, 13).

Konzen EA, Barros LC (1997). Lagoas de estabilização natural para armazenamento de dejetos líquidos de suínos. Sete Lagoas: EmbrapaMilho Sorgo 14 p. (Embrapa Milho e Sorgo).

Konzen EA, Alvarenga RC (2005). Manejo e utilização de dejetos de suínos: aspectos agronômicos e ambientais. Sete Lagoas: Embrapa Milho e Sorgo,. 16 p. (Embrapa Milho e Sorgo. Circular técnica, 63).

Konzen EA (2003). Fertilização de lavoura e pastagens com dejetos de suínos e cama de aves. Sete Lagoas: Embrapa Milho e Sorgo Bol. Técnico P 31.

Lopes RL, Caixeta Filho JV (2000). Suinocultura no Estado de Goiás: aplicação de um modelo de localização. Pesqui. Oper. 20(2):213-232.

Lourenzi CL, Ceretta CA, Brunetto G, Girotto E, Tiecher TL, Vieira RCB, Cancian A, Ferreira PAA (2014). Pig slurry and nutrient accumulation and dry matter and grain yield in various crops. Rev. Bras. Ciênc. Solo 38:949-958.

Menezes JFS, Alvarenga RC, Andrade CLT, Konzen EA, Pimenta FF (2003). Aproveitamento de resíduos orgânicos para a produção de grãos em sistema de plantio direto e avaliação do impacto ambiental. Rev. Plantio Dir. 12(73):30-35.

Menezes JFS, Konzen EA, Silva GP, Santos SCG, Piment AFF, Lopes JPC, Alvarenga RC, Andrade CLT (2007). Aproveitamento de dejetos de suínos na produção agrícola e monitoramento do impacto ambiental. Rio Verde: Universidade de Rio Verde 46 p. (Boletim Técnico, 6)

Pavan MA, Bloch MFM, Zempulski HC, Miyazawa M, Zocoler DC (1992). Manual de análise química de solo e controle de qualidade. Londrina, Iapar. (Circular, 76).

Pedrinho Júnior AFF, Bianco S, Pitelli RA (2004). Acúmulo de massa seca e macronutrientes por plantas de Glycine max e Richardia brasiliensis. Planta Daninha, Viçosa 22(1):53-61.

Penha HGV (2015). Impacto ambiental de elementos-traço em solos adubados com fertilizantes fosfatados e lodo de esgoto em longo prazo. Tese (Doutorado em Ciência do Solo) - Universidade Federal de Lavras, Lavras. 99 p.

Santos DH, Silva MA, Tiritan CS, Foloni JSS, Echer FR (2011). Qualidade tecnológica da cana-de-açúcar sob adubação com torta de filtro enriquecida com fosfato solúvel. Rev. Bras. Eng. Agríc. Amb. 15:443-449.

Santos MR, Sediyama MAN, Moreira MA, Megguer CA, Vidigal SM (2012). Rendimento, qualidade e absorção de nutrientes pelos frutos de abóbora em função de doses de biofertilizante. Hortic. Bras. 30:160-167.

Scherer EE, Baldissera IT, Dias LFX (1995). Método rápido para determinação da qualidade do esterco líquido de suínos a campo. Agropecuária Catarinense, Concórdia: empasc 8(2):40-43.

Sediyama MAN, Santos MR, Vidigal SM, Pinto CLO, Jacob LL (2014). Nutrição e produtividade de plantas de pimentão colorido, adubadas com biofertilizante de suíno. Rev. Bras. Eng. Agríc. Amb. 18:588-594.

Sediyama MA, Vidigal SM, Garcia NCP (2005). Utilização de resíduos da suinocultura na produção agrícola. Informe Agropecuário, Belo Horizonte 26(224):52-64.

Seidel EP, Gonçalves Junior AC, Vanin JP, Strey L, Schwantes D, Nacke H (2010). Aplicação de dejetos de suínos na cultura do milho cultivado em sistema plantio direto. Acta Sci. Tecnol. 32:113-117.

Silva FC (2009). Manual de análises químicas de solos, plantas e fertilizantes. 2. ed. Brasília, DF: Embrapa Informação Tecnológica; Rio de Janeiro: Embrapa Solos.

Sommer SG, Hutchings NJ (2001). Ammonia emission from field applied manure and its reduction: invited paper. European Journal of Agronomy, Amsterdam 15(1):1-15.

Sousa FA, Silva EB, Campos AT, Gandini AMM, Corrêa JM, GrazziottI PH (2014). Atividade microbiana e produção da lavoura cafeeira após adubação com dejetos líquidos de suínos. Biosci. J. 30:1041-1049.

Sousa DMG, Lobato E (2004). Cerrado: correção do solo e adubação. 2 ed. Brasília: Embrapa Cerrados 416 p.

Zaman M, Blennerhassett JD (2010). Effects of the different rates of urease and nitrification inhibitors on gaseous emissions of ammonia and nitrous oxide, nitrate leaching and pasture production from urine patches in an intensive grazed pasture system. Agric. Ecosyst. Environ. 136:236-246.

academicJournals

Vol. 12(7), pp. 494-501, 16 February, 2017
DOI: 10.5897/AJAR2015.10763
Article Number: 01E4D7A62764
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Impact of climate change on cotton production in Burkina Faso

Abdoulaye Diarra[1]*, Bruno Barbier[2], Bétéo Zongo[1] and Hamma Yacouba[1]

[1]Institut International d'Ingénierie de l'Eau et de l'Environnement (2iE), Rue de la Science - 01 BP 594, Ouagadougou 01, Burkina Faso.
[2]CIRAD, UMR G-eau, Montpellier 34398, France.

Received 24 December, 2015; Accepted 24 June, 2016

This paper evaluated the impact of climate change on cotton production in Burkina Faso. An econometric analysis resulted in identifying the major factors influencing cotton yields and evaluating the likely effects of future climate change. The results of our study regarding the potential impact of future climate change on cotton yield indicated that further increases in global temperature would significantly reduce the yield of cotton. Future changes in rainfall would also affect cotton production, but compared with the effects of temperature, the effects of rainfall are relatively lesser. Therefore, strategies for reducing the impacts of climate change on cotton production should emphasize the development of heat resistant cultivars rather than drought resistant ones in order to mitigate and adapt them to the effects of climate change.

Key words: Climate change, cotton, economic impact, adaptation, Sahel, Burkina Faso.

INTRODUCTION

Climate change represents one of the greatest environmental, social, and economic threats with which the planet is confronted today. Several climate models suggest that in West Africa, the average temperature is likely to increase while rainfall is likely to decrease. This change will have a significant impact on the livelihoods and living conditions of the population, particularly the poor. According to a report by the Intergovernmental Panel on Climate Change report (IPCC, 2007) developing countries are more vulnerable to climate change than developed countries because agriculture is mainly rainfed and is still the largest sector in their economies.

Despite the urgency of the issue, little research has been carried out regarding these effects in West Africa at the country level. Little is known about the extent of the potential damage in each country. Most studies in sub-Saharan African (SSA) have been conducted at the regional or global level (Rosenzweig and Parry, 1994; Darwin et al., 1995; IFPRI, 2009). However, since agricultural technologies and, environmental and socioeconomic conditions vary from one country to another, climate change is expected to have different effects; therefore, it is important to address the issue of climate change impacts on agricultural productivity at the national level.

This paper focuses on cotton production, a major

*Corresponding author. E-mail: abdoulaye.diarra@2ie-edu.org.

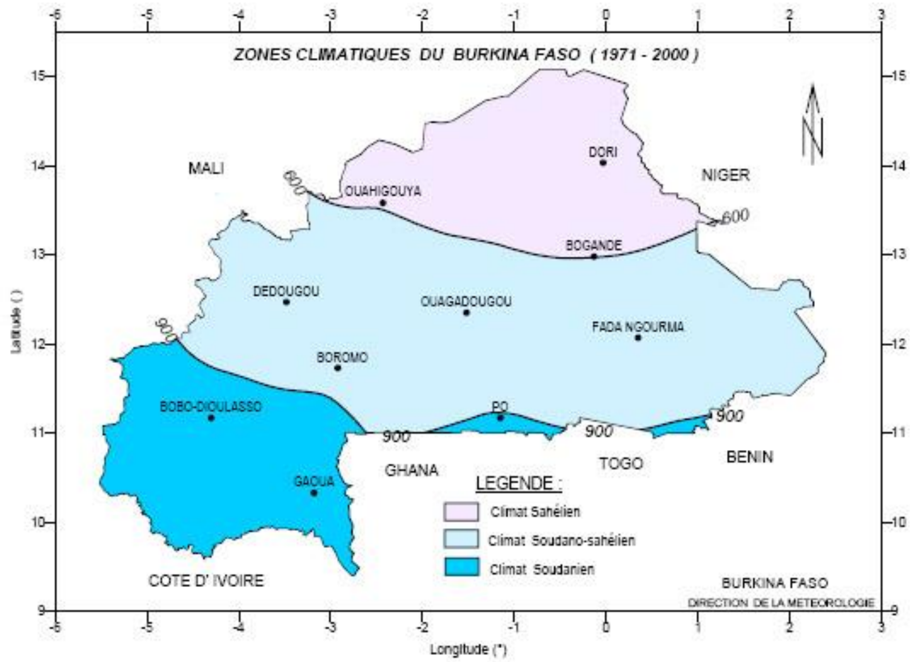


Figure 1. Agro-climatic zones in Burkina Faso.

sector of the economy and agriculture of Burkina Faso. In this country, agriculture represents a large share of the gross domestic product (22%). Cotton is the main export commodity in terms of value, and generates income for approximately 3 million people in the country. Most cotton-farms are family-owned and are small scale (an average of about 1.0 hectare of cotton per farm). Therefore, climate change could be potentially disastrous for the economy of Burkina Faso.

Two major methodologies have been used so far to study the relationship between climate and crop yields: a computer based simulation approach, the so-called crop growth models and a statistical model based on regression analysis. Both methodologies have their advantages and disadvantages (Cai, 2011). Crop growth modeling tools such as Decision Support System for Agrotechnology Transfer (DSSAT), Erosion Product Impact Calculator (EPIC) and Terrestrial Ecosystem Model (TEM) are mathematical representations of phenomena linked to different disciplines such as biology, physics and chemistry (Hoogenboom, 2000; Jones et al., 2003). For instance, EPIC has been used in numerous studies for a variety of purposes and has gained popularity throughout disciplines in agriculture.

However, crop growth modeling is highly complex and requires extensive information such as climate data, soil and management options in order to simulate the crop growing process. Such information is usually incomplete and sometimes unavailable. Because of this disadvantage of crop simulation modeling, regression analysis is an alternative method used for predicting yields in many yield forecasting studies (Schlenker et al., 2009; Yingjie, 2008; Chang, 2002; Lobell et al., 2008; Paeth et al., 2008). Compared to the crop growth modeling approach, data limitations are less of a concern in the regression model (Cai, 2011).

In this study, a non-linear regression model is used to investigate the impact of climate change on cotton yield in Burkina Faso. Coefficient estimates are used to predict the change in yield under alternative climate change scenarios.

MATERIALS AND METHODS

Climate in Burkina Faso

Burkina Faso has a semi-arid tropical climate. The dry season is characterized by the harsh harmattan wind which blows from the north-east to the south-west from October to March. April is the month of humid winds or trade winds bearing monsoons. The rainy season, from May or June to September is characterized by humid winds.

A mass of humid air from the Atlantic proceeds north from the Gulf of Guinea and reaches Burkina from the south-west where the rains start falling in April. At first sporadic, rains gradually cover the whole country from June onwards. August is the wettest month. The rains cease at the end of September. October is when the dry harmattan wind starts blowing throughout the country. The duration of the rainy season decreases progressively from the south-west to the north. The rainfall is very erratic and its volume also decreases from the south-west to the north. There are large seasonal shifts in temperature and their heat rises at night, particularly in the north of the country (Some and Sivakumar, 1994).

The country is divided into three climate zones (Figure 1). These agro-climatic zones also constitute the phyto-geographical regions

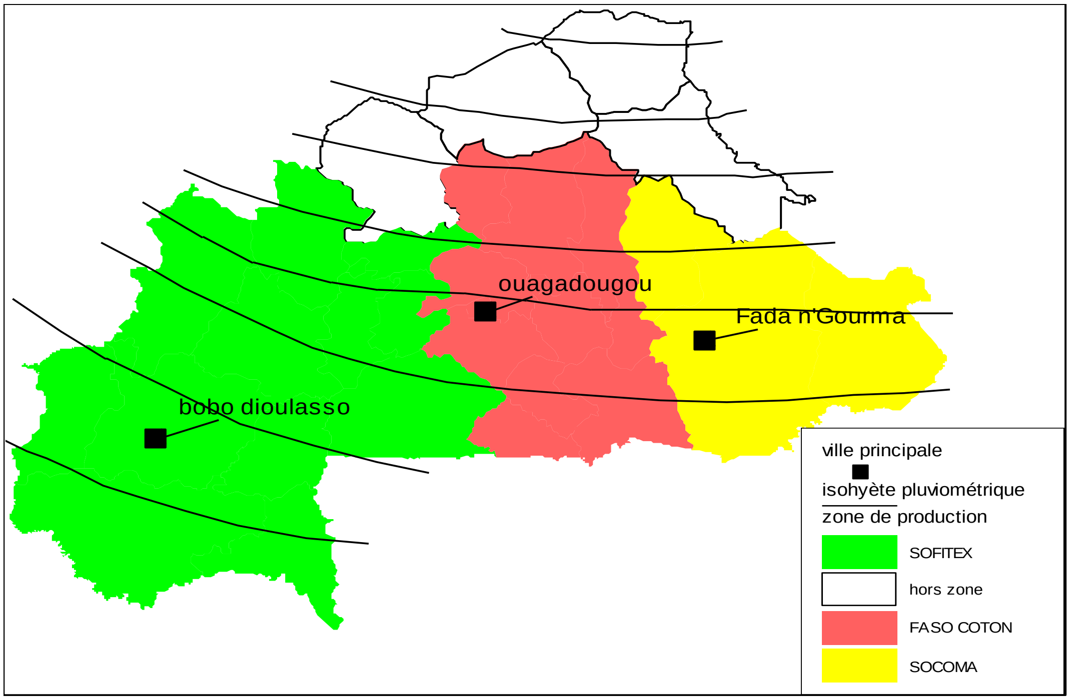


Figure 2. The three cotton zones of Burkina Faso.

of the country (Ouedraogo, 2006):

(1) The south Sudanese zone, located at the south of the 11°30' parallel with an average annual rainfall between 900 and 1200 mm. The rainy season here lasts six months. This is the domain of gallery forests along the rivers.
(2) The Sudano-sahelian region is situated within the 11°30' and 14°00'N parallels. This zone has an average annual rainfall of between 800 and 900 mm during four to five months. Here, there are more dense forest formations, and the herbaceous cover is more continuous. This is the largest zone and the one which is the most affected by human activity.
(3) The Sahelian zone is situated in north of 14°00'N. This zone has an average annual rainfall of between 300 and 600 mm and lasts for only three months. The vegetation there consists of steppes with trees, shrubs and thick bushes.

Study region: West of Burkina Faso

Zones of cotton production

The western region of Burkina Faso is the principal region of cotton production. In 2006, the production of cotton in the country was estimated at 676,065 tons, and the total amount of cotton farms was estimated at 325,000 ha. These farms are divided into three cotton producing zones in the country (Figure 2): the cotton zone of SOFITEX (Burkina Faso Textile Fibre Company) located in the west is the largest with 87% of national cotton production, the cotton zone of FASO COTON (Cotton company of Burkina Faso) located in the center with 8% of the production and the cotton zone of SOCOMA (Cotton Company of Gourma) located in the east with 5% of the production.

The cotton zone area of SOFITEX was chosen as the region of study for several reasons. One reason is that the region is the principal and historical area of cotton production. Furthermore, there are long series of reliable data for analysis in this region. The time series data for the other two sites is relatively short for the purpose of statistical analysis.

Temperature and rainfall in the western of Burkina Faso

Over the past 45 years, the shift in temperature is obviously less dramatic than the shift in rainfall. In the Figure 3, total rainfall and monthly average temperature of the cotton growing season (May to October) in the western of Burkina Faso are presented. There is an increasing trend of temperature for the last 45 years, which is not seen in rainfall.

Functional form of the regression model

The choice of variables to include in the regression analysis is crucial for the validity of the model. In the literature, temperature and rainfall have been demonstrated to have significant impacts on crop yields. Agronomically, high temperature affects soil moisture levels which could decrease crop yields if the supply of irrigation water supply is not sufficient. On the other hand, precipitation maintains necessary soil moisture for crop growth. Shifts in temperatures could change growing season lengths, thus inducing variations in crop yields. For example high temperature tends to shorten many crop growing seasons. A short growing season exposes crops to less solar radiation needed for photosynthesis (Caï, 2011).

The effects of weather conditions on crop yields are not simple linear relationships (Deschenes and Greenstone, 2007; Schlenker and Roberts, 2009). Most recent studies have adopted a non-linear specification for each climate variable where linear and quadratic terms are used as regressors, reflecting the effect of a physiological optimum on yield (Yingjie, 2008; Chang, 2002; Schlenker et al., 2009). This approach also allows a non-monotonic relationship

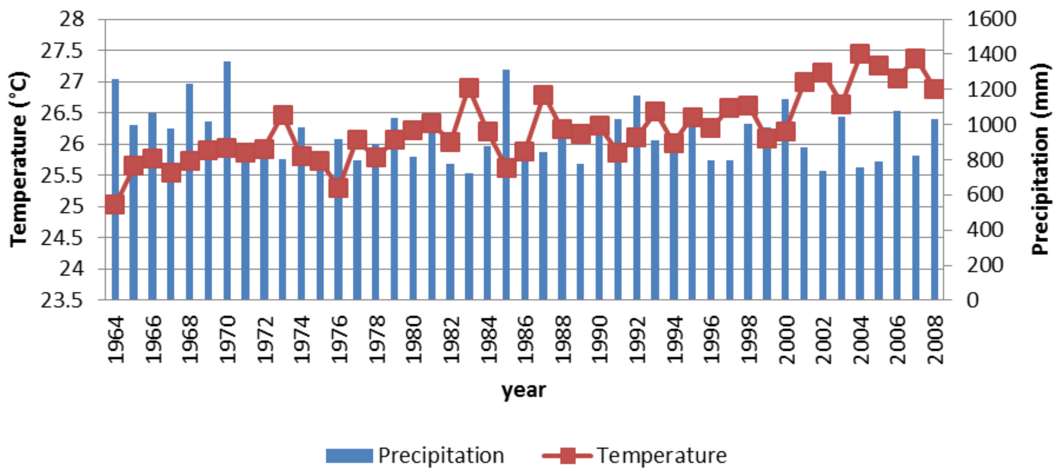


Figure 3. Total rainfall and monthly average temperature of the growing season of cotton (May to October) in western of Burkina Faso.

between climate and yield; warming might increase crop yields in cooler areas but decrease yields in warmer regions (Segerson and Dixon, 1999).

Besides the mean value of weather indicators, climate change has effects crop yields. Previous studies (Shaw et al., 1994; Mendelsohn et al., 1999) have shown that omitting the variation terms biased the effects of global warming; Chang (2002) included these two climate variation terms to estimate the potential impact of climate change on Taiwan's agriculture.

Crop yields do not depend only on weather conditions. Other factors can also affect crop yields such as crop prices. Several econometric studies provide evidence of the responsiveness of crop yields to crop price (Choi and Helmberger, 1993; Yingjie, 2008). The type of agricultural equipment and the level of intensification can also explain variability in crop yield (Huang et al., 2010).

Crop yields are expected to increase over time because of technological innovations such as the adoption of new varieties, improved application of fertilizers and irrigation, and expansion or contraction of crop acreage. Technological innovation is usually represented by a linear or quadratic time trend in empirical studies (Choi and Helmberger, 1993; Kaufmann and Schnell, 1997; Mc Carl et al., 2008).

Determinant of cotton yield in Burkina

Cotton yield response is typically estimated from field data using measurement of climate and non-climate-related variables to identify the physical effect of climate change on yield. In this study, the growing season (May to October) was divided into growing stages (S). The first stage is the planting time and germination (1st May to 30th June), the second is the vegetative growth (1st July to 1st September), the third is the development of fruiting and vegetative branches (1st September to 30th September) and the forth is maturity (1st October to 30th October).

The explanatory variables include: T_s, decadal average temperature of growing stage (°C); R_s, decadal average rainfall of growing stage (mm); VT_s, standard deviation of decadal temperature of the growing stage; VR_s, standard deviation of decadal precipitation of the growing stage; P_c, current price of cotton with regard to fertilizer price and D, time trend to consider technical progress. The average annual yield Y is the dependent variable (Equation 1):

$$Y = f(T_s, T_s^2, R_s, R_s^2, VT_s, VR_s, P_c, D) \quad (1)$$

This model adopts a non-linear specification for each climate variable where linear and quadratic terms are used as regressors, reflecting the effect of a physiological optimum on yield. Changes in temperature and rainfall from historical values are also included to capture the effect of an extreme event on yield. The model is estimated with time-series data over the period 1964-2008.

The samples consisted of secondary data for the western of Burkina Faso corresponding to SOFITEX company area. The data for average cotton yield (kg/ha), annual price of cotton (FCFA/kg) and annual price of fertilizer were provided by the SOFITEX Company. Decadal weather data on temperature and rainfall were obtained from the Burkina Central Weather Bureau. Temperatures are in degrees Celsius and rainfall measured in millimeters.

The summary statistics are presented in Table 1. Over the past 45 years, the temperature shift is obviously less dramatic than the variation in rainfall.

RESULTS AND DISCUSSION

Estimation of the cotton yield regression model

The model was estimated using SAS statistical analysis. The results show that the explanatory variables have significant effects on cotton production and the model tends to have a good explanatory power as measured by the coefficient of determination (R^2 = 0.94). The Durbin Waston coefficient (d=2.3) is close to 2, therefore errors are not correlated.

The results of coefficients of the explanatory variables are given in Appendix 1. Table 2 shows the variability of cotton yield in response to climate and non-climate variables.

For the non-climate variables, the coefficients of the explanatory variables have the expected sign. The relative price of cotton has a positive and very significant impact

Table 1. Summary statistics of data used in cotton yield response regression.

Parameter	Mean	Standard error
Non-climate		
P_c	0.93	0.21
Climate		
T_1	28.0	0.72
T_2	25.0	0.55
T_3	25.17	0.64
T_4	27.0	0.75
R_1	157	109.2
R_2	321.6	207
R_3	131.6	72.3
R_4	44.2	33.0
Climate shifts		
VT_1	2.3	1.20
VT_2	0.4	0.27
VT_3	0.6	0.4
VT_4	0.55	0.46
VR_1	743.2	469.43
VR_2	2043.2	1662.36
VR_3	1204.3	1146
VR_4	442.6	567

Table 2. Variability of cotton yield in response to climate and non-climate variables.

Variable	Growth stages	Variability
Temperature	First	-1.32
	Second	-4.14
	Third	5.30
	Fourth	1.60
Rainfall	First	0.18
	Second	-0.04
	Third	0.03
	Fourth	0.04
Relative price		0.93
Time trend		0.44

on yield. More specifically, a 1% increases in the relative price of cotton increases the average yield of cotton by about 0.93%. Fertilizer use is very important for cotton. It can represent up to 60% of production cost. Technology, represented by a time trend has as expected a positive and significant effect on cotton yield.

For the climate variables, increasing temperature in the two first growing stages is mostly unfavorable for cotton yield, while it is favorable in the two last ones, particularly the third stage. Too much rain in the second stage is unfavorable for cotton yield but favorable in the three other stages. A number of studies have determined the impact of climate change on cotton yield using the data from field and/or laboratory controlled-experiments and various crop simulation models. The result of the relationship between rainfall and cotton yield is quite consistent with the results in Some et al. (2006). That study used a crop simulation model and concluded that at the end of the rainy season cotton needs irrigation to reach its potential yield.

Climate changes have an impact on cotton yield. Our results show that rainfall variations of the last growing stages have a significant and negative impact on yield. Temperature variation of the first growing stage has significant and unfavorable impact on cotton yield. The review by Adams et al. (1998) compared the climate effects on grain crops in Latin and North America stemming from various crop simulation models. Their comparison indicated that crop productivity in large areas of Latin America is negatively affected by the inter-annual variability and the occurrence of extreme events.

Specific climate change cases for Burkina Faso

Strzepek and Mc Cluskey (2006) have developed climate evolution models for many African countries that cover the period between 2050 and 2100 within the framework of the hydrology component for the Global Environment Facility of the World Bank. These models are based on the A2 and B2 scenarios of the Special Report on Emissions Scenarios (SRES) and provide specific forecasts by country. All models forecast a rise in temperatures for Burkina Faso. This increase will vary from 2.4 to 3.9°C in 2050 and from 5.7 to 9.7°C in 2100 for the A2 scenario. The B2 scenario projects an increase of between 2.4°C and 3.8°C in 2050 and between 4°C and 7.1°C in 2100. While both scenarios indicate a potentially serious climate change impact in Burkina Faso, the latter scenario looks less alarming.

The models provide the same forecasts for rainfall regardless the scenario (A2 or B2). Four of the five models predicted an increase in rainfall from 1% to 12% by 2050 and from 3% to 30% by 2100. Only one model foresees a decrease in rainfall of about 4% by 2050 and 9% by 2100.

In our analysis, we assume that future temperature will increase by 0.5 to 4°C while future rainfall will change by -20 to 20%. The effect of precipitation variability is also estimated.

Impact of climate change on cotton production

Because the data used in this study do not result in an estimation of the effects of changes in CO_2, all scenarios describe no change in the CO_2 level and thereby include no CO_2 fertilization effects. Therefore, it was also assumed that the temperature or rainfall change occurs in all the four growing stages. Thus, in each scenario all seasonal variables are adjusted simultaneously by the same amounts.

The observed effect of the likely future change in climate (temperature and precipitation) on crop yields is shown in Figure 3. The results show that when temperature increases by 1°C cotton yield increases to 3% relative to the mean yield but decreases when temperature is higher. It can be concluded that higher temperature is harmful for cotton production. Similar results have also been reported by Schlenker and Roberts (2009) for U.S cotton production. Using an extensive 1950-2005 county level panel regression for the Eastern U.S. and fixed-effects models, they find that yields increase with temperature up to 32°C for cotton, but that temperature above these thresholds are harmful. They predict that a 4°C increase in temperature can lead to cotton yield decrease by 25%. The result shows a decrease of 13% in cotton yield for 4°C increase.

Using the growing season temperature and employing the Just and Pope (1978) stochastic production function, Schimmelpfennig et al. (2004) examined state-level panel data and found that cotton yields is adversely affected by growing season temperature. Reddy et al. (2002) used crop simulation models to estimate the impact of climate change on cotton production in the State of Mississippi, United States, and concluded that if global warming occurs as projected, fiber production in the future environment will be reduced. In Zimbabwe, Gwimbi (2009) examined climate change and its impact on cotton production using a time series analysis of temperature and rainfall for a period of 30 years. That study found that cotton production levels declined as rainfall decreased and temperatures increased throughout the district.

The effects of future changes in rainfall show that a higher rainfall could lead to an increase of cotton yield (Figure 4), while a 20% reduction in precipitation will decrease cotton yield by 4.4 and a 20% increase in precipitation will increase yield by 3%. Compared with the effects of temperature, the effects of rainfall are relatively small.

Table 3 lists 5 combinations of alternative climate change cases, which include 0, +4°C for temperature and -20, 0 and +20% for precipitation. The scenarios of decreasing rainfall (-20%) and increasing temperature (+4°C) will be highly deleterious to cotton production, leading to a cotton yield decrease of about 17.7%. The impact will be severe and harmful because Burkina Faso's climate is already relatively hot and dry.

Economic impact of change in cotton yield

The chain value approach was used to evaluate the impact of changes in crop yield on farmers' gross income, foreign exchange earning and tax revenues. The data used is from a report on the cotton sector made by the Ministry of Agriculture of Burkina Faso (MAHRH, 2007) for the 2005 seasonal year (Table 4).

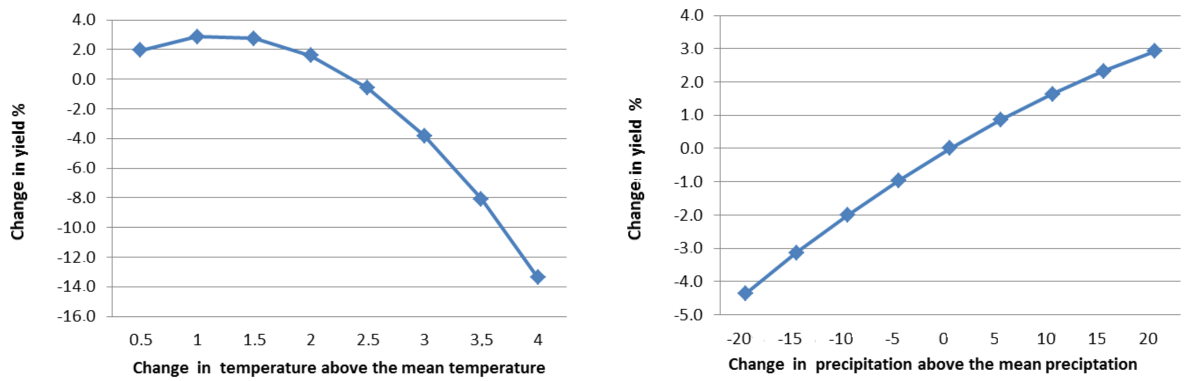


Figure 4. The impact of projected climate change on Burkina Faso cotton yield.

Table 3. Change in cotton yield according to scenario.

Scenario	Temperature (°C)	Precipitation (%)	Yield change (%)
1	+4	-20	-17.7
2	+4	0	-13.4
3	+4	+20	-10.5
4	0	-20	-4.4
5	0	+20	+2.9

Table 4. Change in income and foreign exchange from 2005 data base.

Scenario	Farmer 'gross income change (%)	Foreign exchange earning change (%)
1	-38	-17.7
2	-29	-13.4
3	-23	-10.5
4	-9	-4.4
5	+6	+2.9

As might be expected, the impact of climate change is very harmful for farmers income and the Burkinabe Faso economy. The findings of Kurukulasuriya and Mendelsohn (2008), Deressa and Hassan (2009), Molua and Lambi (2007), and Gwimbi (2009) also revealed that decreased rainfall or increased temperatures reduce cotton yield, which led to a reduction in net revenues in Africa. These results were also confirmed by the findings in Ouedraogo et al. (2006) in which the Ricardian approach was used to evaluate the relationship between net farm values and climate change. Indeed, they found that if temperature increases by 1°C, revenue will fall by US$ 19.9 ha^{-1} and if precipitation increases by 1 mm/month, net revenue increases by US$ 2.7 ha^{-1}.

Conclusion

In this paper the impact of climate change on cotton production in Burkina Faso was evaluate because cotton production is a major source of foreign revenue and a key sector of the national economy. An econometric analysis enabled the identification of the major factors influencing cotton yields and the evaluation of the likely effects of future climate change. As compared to other studies, this study includes a more comprehensive set of climate and socioeconomic variables.

The results show that cotton yield responds positively to the relative price of cotton. It was also shown that climate variables impact cotton yields. Increasing temperature in the two first growing stages is mostly unfavorable for cotton yield, while it is favorable in the two last stages. Too much rain in the second stage is unfavorable for cotton yield but favorable in the three other stages.

The simulated results regarding the potential impact of future climate change on cotton yield clearly indicate that further increases in global temperature would significantly reduce the yield of cotton. Future changes in rainfall would also affect cotton production, however compared with the effects of temperature, the effects of rainfall is relatively small.

For the last few decades, research in Africa has focused only on drought resistant cultivars. This strategy may not be sufficient according to the results of this study. Therefore, strategies for reducing the impacts of climate change on cotton production should emphasize on the development of heat resistant cultivars rather than drought resistant cotton cultivars in order to mitigate and adapt to the effects of climate change.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Adams RM, Fleming RA, Chang CC, Mc Carl BA, Rosenzweig C (1995). A Reassessment of the Economic Effects of Global Climate Change on US Agriculture. Clim. Change 30:147-167.

Cai R, Bergstrom JC, Mullen JD, Wetzstein ME, Shurley WD (2011). Principal Component Analysis of Crop Yield Response to Climate Change (No. 103947). University of Georgia, Department of Agricultural and Applied Economics.

Chang CC (2002). The potential impact of climate change on Taiwan’s agriculture. Agric. Econ. 27:51-64.

Choi JS, Helmberger PG (1993). How Sensitive are Crop Yields to Price Changes and farm Programs? J. Agric. Appl. Econ. 25:237-244.

Darwin R, Tsigas M, Lewabdrowski J, Raneses A (1995). World Agriculture and Climate Change. Agricultural Economic Report No. 703, US Department of Agriculture, Econ. Res. Service, Washington D.C.

Deressa T, Hassan R (2009) Economic Impact of Climate Change on Crop Production in Ethiopia: Evidence from Cross-section Measures. J. Afr. Econ. 18:529-554.

Deschenes O, Greenstone M (2007). The Economic Impacts of Climate Change: Evidence from Agricultural Output and Random Fluctuations inWeather. Am. Econ. Rev. 97(1):354-385.

Gwimbi P (2009). Cotton Farmers’ Vulnerability to Climate Change in Gokwe District (Zimbabwe): Impact and Influencing Factors. JAMBA: J. Disaster Risk Stud. 2(2).

Hoogenboom G (2000). Contribution of Agrometeorology to the Simulation of Crop Production and its Applications. Agric. For. Meteorol. 103:137-157.

Huang H, Khanna M (2010). An Econometric Analysis of U.S. Crop Yield and Cropland Acreage: Implications for the Impact of Climate Change. Available at SSRN: http://ssrn.com/abstract=1700707

International Food Policy Research Institute (IFPRI) (2009.) Climate change impact on agriculture and costs of adaptations. Food Policy Report

IPCC (2007). Climate change 2007: Synthesis report, Contribution of working groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change [Core writing team, Pachauri R.K., Reisinger A. (eds.)], IPCC, Geneva, Switzerland, 104 p.

Jones JW, Hoogenboom G, Porter CH, Boote KJ, Batchelor WD, Hunt LA, Wilkens PW, Singh U, Gijsman AJ, Ritchie JT (2003) The DSSAT Cropping System Model. Euro. J. Agron. 18:235-265.

Just R, Pope RD (1978) Stochastic Specification of Production Function and Economic Implications. J. Econ. 7:67-86.

Kaufmann RK, Snell SE (1997). A Biophysical Model if Corn Yield: Integrating Climatic and Social Determinants. Am. J. Agric. Econ. 79(1)178-190.

Kurukulasuriya P, Mendelsohn R (2008). A Ricardian Analysis of the Impact of Climate Change on African Cropland. Afr. J. Agric. Res. Econ. 2(1).

Lobell DB, Burke MB, Tebaldi C, Mastrandrea MD, Falcon WP, Naylor RL (2008) Prioritizing climate change adaptation needs for food security in 2030. Sci. 319:607-610.

McCarl BA, Villavicencio X, Wu X (2008). Climate Change and Future Analysis: Is Stationary Dying? Am. J. Agric. Econ. 90(5)1241-1247.

Mendelsohn R, Nordhaus W, Shaw D (1999). The impacts of climate variation on US agriculture. In: Mendelsohn R, Neumann JE (Eds.). The Impact of Climate Change on the United States Economy. Cambridge University Press, Cambridge, Chapter 3, pp. 55-74.

Ministère de l’Agriculture, de l’Hydraulique et des Ressources Halieutiques (MAHRH) (2007). Diagnostic de la filière coton et identification d’aces stratégiques. Burkina Faso, 2007.

Molua E, Lambi C (2007). The Economic Impact of Climate Change on Agriculture in Cameroon. World Bank Policy Research Working Paper 4364:22.

Ouedraogo M, Somé L, Dembele Y (2006). Economic impact assessment of climate change on agriculture in Burkina Faso: A Ricardian Approach. CEEPA Discussion Paper No. 24, Centre for Environmental Economics and Policy in Africa, University of Pretoria

Paeth H, Capo-Chichi A, Endlicher W (2008). Climate change and food security in tropical West Africa: a dynamic-statistical modeling approach. Erdkunde 62:101-115.

Reddy KR, Doma PR, Mearns LO, Hodges HF, Richardson AG, Boone MYL, Kakani VG (2002). Simulating the impacts of climate change on cotton production in the Mississippi delta. Climate Res. 22:271-281.

Rosenzweig C, Parry ML (1994) Potential impact of climate change on world food supply. Nat. 367:133-138.

Schimmelpfennig DE, Chen CC, McCarl BA (2004). Yield Variability as Influenced by Climate: A Statistical Investigation. Climatic Chang. 66:239-61.

Schlenker W, Roberts MJ (2009). Nonlinear Temperature Effects Indicate Severe Damages to U.S. Crop Yields under Climate Change. Proceedings of the National Academy of Sciences, September 15 2009, 106 (37):15594-15598.

Segerson K, Dixon BL (1999). Climate change and agriculture: the role of farmer adaptation. In: Mendelsohn R, Neumann JE (Eds.). The Impact of Climate Change on the United States Economy. Cambridge University Press, Cambridge, Chapter 4:75-93.

Shaw D, Mendelsohn R, Nordhaus W (1994). The Impact of Climate Variation on Agriculture. Sch. For. Environ. Stud. Yale University, New Haven, CT.

Some L, Dembele Y, Ouedraogo M, Some BM, Kambire FL, Sangare S (2006). Analysis of crop water use and soil water balance in Burkina Faso using CROPWAT. CEEPA DP36, University of Pretoria, South Africa

Some L, Sivakumar MVK (1994). Analyse de la longueur de la saison culturale en fonction de la date du début des pluies au Burkina Faso. Compte rendu des Travaux No.1 INERA Burkina Faso ICRISAT Niger

Strzepek K, McCluskey A (2006). District level hydroclimatic time series and scenario analyses to assess the impacts of climate change on regional water resources and agriculture in Africa. CEEPA Discussion Paper No. 13, Centre for Environmental Economics and Policy in Africa, University of Pretoria.

Yingjie L, Erda L (2008). Impact of Climate Warming in the Past 20 Years on Agriculture in Different Regions of China. Adv. Climate Change Res. 4:51-55.

academicJournals

Vol. 12(7), pp. 502 -511, 16 February, 2017
DOI: 10.5897/AJAR2016.12021
Article Number: 2FA9A0662767
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Cryopreservation and effect of lighting conditions and cytokinins on *in vitro* multiplication of *Miconia ligustroides* (DC.) Naudin

Débora de Oliveira Prudente*, Débora Domiciano, Renato Paiva, Fernanda Carlota Nery and Wesley Pires Flausino Máximo

Universidade Federal de Lavras, Lavras, MG 37200-000, Brazil.

Received 29 November, 2016; Accepted 24 January, 2017

Miconia ligustroides **is a species that is native to Brazil and has medicinal and ecological importance. However, the species shows a lack of uniformity and delay in *ex vitro* germination. Thus, this study aimed to establish *in vitro* propagation for the species and to develop a protocol for the cryopreservation of seeds. For *in vitro* germination, activated charcoal (0.0, 0.5, 1.0, and 2.0 g L^{-1}) was tested in Murashige and Skoog (MS) culture medium. Lateral buds excised from the plants were germinated *in vitro* and were encapsulated in an alginate matrix supplemented with 6-benzylaminopurine (BAP), kinetin, and thidiazuron (TDZ: 0.0, 2.0, 8.0, and 16.0 µM). Shoots derived from encapsulated units were inoculated in MS culture medium supplemented with different concentrations of BAP (0.0, 2.5, 5.0, and 10.0 µM) under white or Gro-Lux fluorescent lamps for multiplication. For cryopreservation, the toxicity of the cryoprotectant solution PVS2 (0, 15, 30, 60, 120, and 180 min) was evaluated before the seeds were immersed in liquid nitrogen. The MS culture medium supplemented with 1.0 g L^{-1} of activated charcoal yielded the highest percentage of germination (78%). The encapsulated units presented the largest percentages of regeneration (75%) with 8.0 µM BAP, which assisted in the formation of shoots that were 8.03 cm in length. For shoot production, the highest mean number (3.03 shoots) was obtained in the MS medium containing 5.41 µM BAP. When seeds were subjected to cryopreservation, the immersion time in the PVS2 did not affect the survival of the seeds, which was satisfactory (70%). The protocols developed are considered viable alternatives for use in the conservation of the species, production of seedlings for commercialization purposes, and use in programs of reintroduction in degraded environments.**

Key words: Melastomataceae, *in vitro* conservation, encapsulated units, 6-benzylaminopurine.

INTRODUCTION

The family Melastomataceae is among the most representative of South America and is the sixth largest angiosperm family in Brazil (Romero and Martins, 2002). Representing virtually all plant formations, this family is

*Corresponding author. E-mail: deboraoprudente@outlook.com.

composed of a wide variety of species, ranging from trees to vines and epiphytes (Renner, 1993). In Brazil, this family has 68 genera, of which 10 are endemic, with more than 1500 species spread throughout the country. Among the genera, *Miconia* Ruiz & Pav. is distinguished within the Melastomataceae by its numerous species (Romero and Martins, 2002; Michelangeli et al., 2004).

Among the *Miconia* species considered important from a medical viewpoint is *Miconia ligustroides.* The species occurs in the Cerrado (savanna forest and woodlands), Atlantic Forest, and Caatinga (thorn scrub savanna and forest) regions (Goldenberg et al., 2013) and its medicinal importance is related to its analgesic antimicrobial (Cunha et al., 2003) and trypanocidal actions (Cunha et al., 2006). Another interesting characteristic is the listing since 2001 of *M. ligustroides* as a species recommended for the recovery of riparian forests (SMA 21/01). Although the species exhibits the positive features mentioned, it faces problems related to the maturation of fruits and an unequal number of diasporas (Chaves et al., 2013), culminating in unequal and delayed germination. Such characteristics may be associated with a relatively low percentage of seed germination, approximately 55% (Chaves et al., 2011) which can hinder the propagation and even the survival of this species in the natural environment.

The problems related to the difficulties in germination of the species, however, can be minimized or even overcome through maximization and standardization of germination using *in vitro* cultivation techniques (Reis et al., 2015). Through the correct use of these techniques, regeneration of the meristematic regions (Ferreira et al., 2007) can be established to encapsulate the lateral buds of the material regenerated in an alginate matrix (Sandoval-Yugar et al., 2009). When combined with the use of growth regulators (Jona and Webb, 1978; Bhojwani et al., 1984) and adequate light sources (Rocha et al., 2010, 2013) this technique can generate multiple healthy shoots in a short period from a single plant germinated *in vitro.*

In addition to the *in vitro* germination protocols used to obtain healthy seedlings, seed cryopreservation protocols are also available, which are useful for conserving and preserving endangered species as well as those of commercial interest (Cejas et al., 2012; Prudente et al., 2016). Currently, *M. ligustroides*is stored in conventional germplasm banks, which require large physical spaces and constant maintenance and are subject to contamination and significant losses (Pritchard and Nadarajan, 2008; Engelmann, 2011).

Given the need to overcome these problems inherent germplasm banks and the scarcity of studies on the cryopreservation of this species, new studies focusing on cryopreservation can contribute more effective alternatives to the current methods employed in the conservation of the species.

Studies have suggested that the longevity of cryopreserved seeds (-196°C) can be up to 175 times greater than the longevity obtained at the temperature used in seed banks (Pritchard, 1995; Walters et al., 2004; Cejas et al., 2012). In addition, the storage in cryogenic banks requires a smaller financial investment over time compared to other genetic material conservation systems available (Dulloo et al., 2009).

Given the aforementioned, this study aimed to establish an *in vitro* propagation protocol, through the encapsulation of lateral buds, and a seed cryopreservation protocol for the species *M. ligustroides.*

MATERIALS AND METHODS

Plant and *in vitro* germination

The seeds were extracted from mature fruits with uniform size and state of conservation, and stored in a cold chamber (4°C) for 30 days. The seeds were removed manually with tweezers, washed in distilled water and distributed on two sheets of paper towel. Subsequently, they were taken to the laminar flow chamber, immersed in 70% alcohol for 60 s, in sodium hypochlorite solution (NaOCl) with 1% active chlorine for 10 min and, at the end, washed five times with distilled water autoclaved.

For *in vitro* germination step, different concentrations of activated charcoal (AC) (0.0, 0.5, 1.0, 2.0 g L^{-1}) were tested in MS culture medium (Murashige and Skoog, 1962), plus 30 g L^{-1} sucrose and gelled with 7 g L^{-1} agar (Sigma®). The seeds were inoculated into 90 × 15 mm crystal polystyrene Petri dishes (J. Prolab®, Brazil) containing 25 ml aliquots of culture medium and sealed with polyvinyl chloride (PVC) film (Rolopac®). The seeds were kept in a growth room under photon irradiance of 86 μmol m^{-2} s^{-1} (40 w white fluorescent lamp, Osram, Brazil) at a temperature of 25 ± 2°C and a photoperiod of 16 h. Each treatment consisted of five Petri dishes, with five seeds per plate. After 30 days of *in vitro* cultivation were evaluated the percentage of seed germination, number of leaves, number of roots and length of the shoots (cm).

Encapsulation of lateral buds

Plants from seeds germinated *in vitro* in MS culture supplemented with 1.0 g L^{-1} of activated charcoal (AC) were used as a source of explants for encapsulation of lateral buds. Lateral buds approximately 1.0 mm^2 in length were excised and immersed in MS medium and were supplemented with different concentrations (0.0, 2.0, 8.0, and 16.0 μM) of 6-benzylaminopurine (BAP), kinetin (KIN), and thidiazuron (TDZ) and with sodium alginate (2.5% w/v) added to the medium. Next, with the aid of an automatic pipette, the encapsulated units were individually retrieved and dropped into a calcium chloride solution ($CaCl_2.2H_2O$) (100 mM), where they remained for 20 min for complexation. The encapsulated units, individually formed by a lateral bud covered with a sodium alginate matrix, were subjected to three washes with autoclaved distilled water to remove the excess $CaCl_2{\cdot}2H_2O$. Then, the encapsulated units were immersed in a potassium nitrate solution (KNO_3) (100 mM) for 15 min for decomplexation and, then, were washed again with autoclaved distilled water before inoculation into the basal MS medium. The capsules were placed in a growth chamber with a photoperiod that included 16 h irradiance with 86 μmol $m^{-2}s^{-1}$ (40 W white fluorescent lamp, Osram, Brazil) and at a temperature of 25 ± 2°C. Each treatment comprised five Petri dishes, with five capsules per dish. The variables evaluated were the percentage of regeneration of the encapsulated buds (after 45 days of *in vitro*

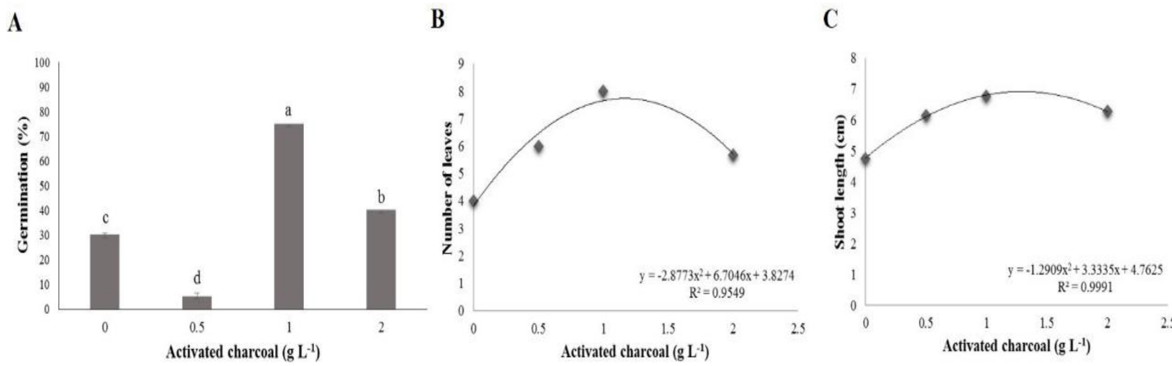


Figure 1. Effect of different concentrations of activated charcoal (0.0, 0.5, 1.0, and 2.0 g L^{-1}) in the MS culture medium on the percentage of seed germination (A), number of leaves (B), and shoot length (D) of *M. ligustroides* seedlings after 30 days of cultivation. Means followed by the same letter in columns in (A) did not differ among themselves, according to the Tukey test at 5% significance.

cultivation), the length of the regenerated shoots (cm), and the number of leaves per shoot (after 60 days of *in vitro* cultivation).

Effect of BAP and light on *in vitro* multiplication of shoots

To evaluate the growth *in vitro,* shoots from encapsulated units germinated *in vitro* after 60 days of cultivation were used, which had been previously inoculated in MS culture medium without the presence of growth regulators for 15 days. After this period, the shoots were standardized at 4.0 cm in length and having three pairs of leaves and were inoculated in MS culture medium supplemented with 30 g L^{-1} sucrose and 7 g L^{-1} agar. The treatments consisted of a bifactorial combination of four concentrations of BAP in the medium (0.0, 2.5, 5.0, and 10.0 µM) and two types of light source: (i) 40 W white fluorescent lamp with a white light with irradiance of 86 µmol m^{-2} s^{-1} (Osram, Brazil) and (ii) Gro-Lux 40 W fluorescent lamp with a red light with irradiance of 94 µmol m^{-2} s^{-1} (Sylvania, Brazil). The explants were grown at 25 ± 2°C in a photoperiod with 16 h of light. The treatments followed a completely randomized design with a 4×2 factorial arrangement (concentrations of BAP × light sources) with 30 replicates. The variables evaluated after 60 days of *in vitro* cultivation were the number of shoots per explant and the length of the shoots (cm).

Cryopreservation of seeds

For the cryopreservation, the initial water content of the seeds was determined according to the oven method at 105 ± 2°C for 24 h (Brasil, 2009), using five samples with 0.5 g of seeds each. The seeds were immersed in Plant Vitrification Solution (PVS2) for different times (0, 15, 30, 60, 120 and 180 min) and immersed in liquid nitrogen (NL) (-196°C) for 90 min. Subsequently, the seeds were thawed in a water bath (37°C) for 3 min and inoculated in MS culture medium plus 1.0 g L^{-1} CA. The seeds were kept in a room of growth under photon irradiance of 86 µmol m^{-2} s^{-1} (white fluorescent lamp 40 w, Osram, Brazil), temperature of 25 ± 2°C and photoperiod of 16 h. Each treatment consisted of five Petri dishes, with five seeds per plate. After 30 days of *in vitro* cultivation, the percentage of germination of the seeds was evaluated.

Statistical analysis

The experimental design was completely randomized for all experiments. Data were submitted to analysis of variance (ANOVA) using the statistical software SISVAR (Ferreira, 2014). According to the results of the ANOVA, data from the qualitative factors were compared by Tukey's test ($P<0.05$), and data from quantitative factors were analyzed by polynomial regression ($P<0.05$).

RESULTS

In vitro germination

The largest percentage of germination (78%) of the seeds was obtained in the MS medium containing 1.0 g L^{-1} activated charcoal (AC) (Figure 1A). The highest mean number of leaves (7.7 leaves) and the largest mean length of seedlings (6.9 cm) were obtained from the inoculation of seeds with the MS medium containing 1.2 and 1.3 g L^{-1} AC, respectively, according to the fit of the data to a quadratic curve (Figure 1B and C). The number of seedling roots was the only variable with no significant difference ($P<0.05$) among the concentrations of AC tested.

Encapsulation of lateral buds

The largest percentages of shoot regeneration from encapsulated lateral buds (75%) were observed at a concentration of 8.0 µM BAP (Figure 2).

A significant interaction ($P≤0.05$) was observed between the cytokinin types and their concentrations for both the length of the shoots and the number of leaves per shoot regenerated from the encapsulated buds.

For the length of the shoots, the analyses of the cytokinin types within each concentration demonstrated no significant differences between BAP, KIN, and TDZ only at the concentration of 2.0 µM. For the concentration of 8.0 µM, BAP was shown to be the most suitable cytokinin, leading to the formation of shoots with 8.03 cm

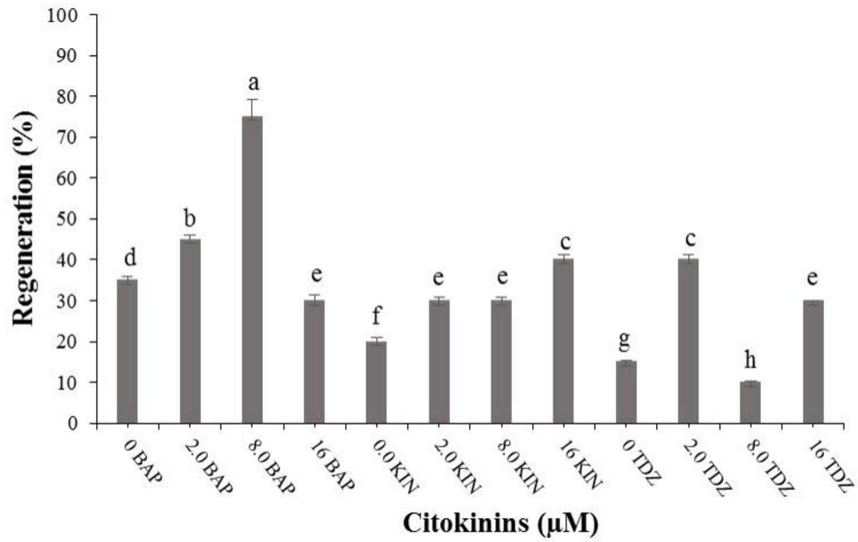


Figure 2. Percentage of shoot regeneration from lateral buds encapsulated in different types and concentrations of cytokinins: BAP, KIN, and TDZ-(0.0, 2.0, 8.0, and 16.0 µM). Means followed by the same lowercase letter within each concentration did not differ among themselves according to the Tukey test (P≤0.05).

lengths (Figure 3C). Finally, at the concentration of 16.0 µM, both KIN and TDZ were more efficient than BAP, influencing the generation of shoots 6.17 and 6.14 cm in length, respectively (Figure 3E). In the analysis of concentrations within each cytokinin, all presented a significant difference (P≤0.05). In the MS culture medium containing BAP, the largest length of shoots was 8.0 cm when using 7.9 µM of this cytokinin (Figure 3B). In the media containing TDZ, the largest mean length of the shoots was 6.51 cm in the presence of 11.39 µM of the plant growth regulator (Figure 3F). For both BAP and TDZ, the data fit a negative quadratic trend, unlike the data for the length of the shoots inoculated in medium containing KIN (positive quadratic trend). In the latter case, the smallest length (4.05 cm) was obtained for 7.72 µM KIN (Figure 3D).

For the leaf number of shoots, analysis of the cytokinin types within each concentration showed a significant difference (P≤0.05) between cytokinins (Figure 4A) only at a concentration of 8.0 µM, with BAP being the most efficient (mean of 4.57 leaves per shoot) (Figure 5). The analysis of the concentrations within each type of cytokinin showed that only BAP showed significant differences among the concentrations used, with the largest mean number of leaves (4.26 leaves) being observed with 8.70 µM, according to the fit of the data to the quadratic curve (Figure 4B).

Effects of BAP and light on *in vitro* shoot multiplication

A significant interaction (P≤0.05) was observed between the concentrations of BAP and the light source types only for the leaf number per shoot. For the variables number of shoots and length of shoots, only the concentrations of BAP showed a significant difference at the 5% significance level.

The largest mean number of shoots (3.03 shoots) was obtained in the MS culture medium containing 5.41 µM BAP (Figure 6A). The largest length of shoots was 7.48 cm in the presence of 10.0 µM BAP (Figure 6B).

For the leaf number per shoot, an analysis of the types of light within each concentration of BAP showed that only 5.0 µM BAP generated a significant difference (P ≤0.05) between the two types of light source used, white and Gro-Lux fluorescent lamps, where the Gro-Lux lamps were more efficient in generating shoots with more leaves (mean of 9.2 leaves) than white fluorescent lamps (mean of 6.5 leaves) (Figure 6C). In the analysis of concentrations of BAP within each type of light source, only the Gro-Lux lamp generated a significant difference at the 5% significance level between the concentrations of BAP, with the highest mean leaf number per shoot (8.63 leaves) being obtained in the presence of 5.73 µM cytokinin, according to the data fit to a quadratic curve (Figure 6D).

Cryopreservation of seeds

The water content of the seeds was 11%, and the immersion time in the cryoprotectant solution PVS2 did not affect the survival process. After immersion in liquid nitrogen (LN), the seeds germinated satisfactorily in MS medium containing 1.0 g L^{-1} AC, reaching a maximum of

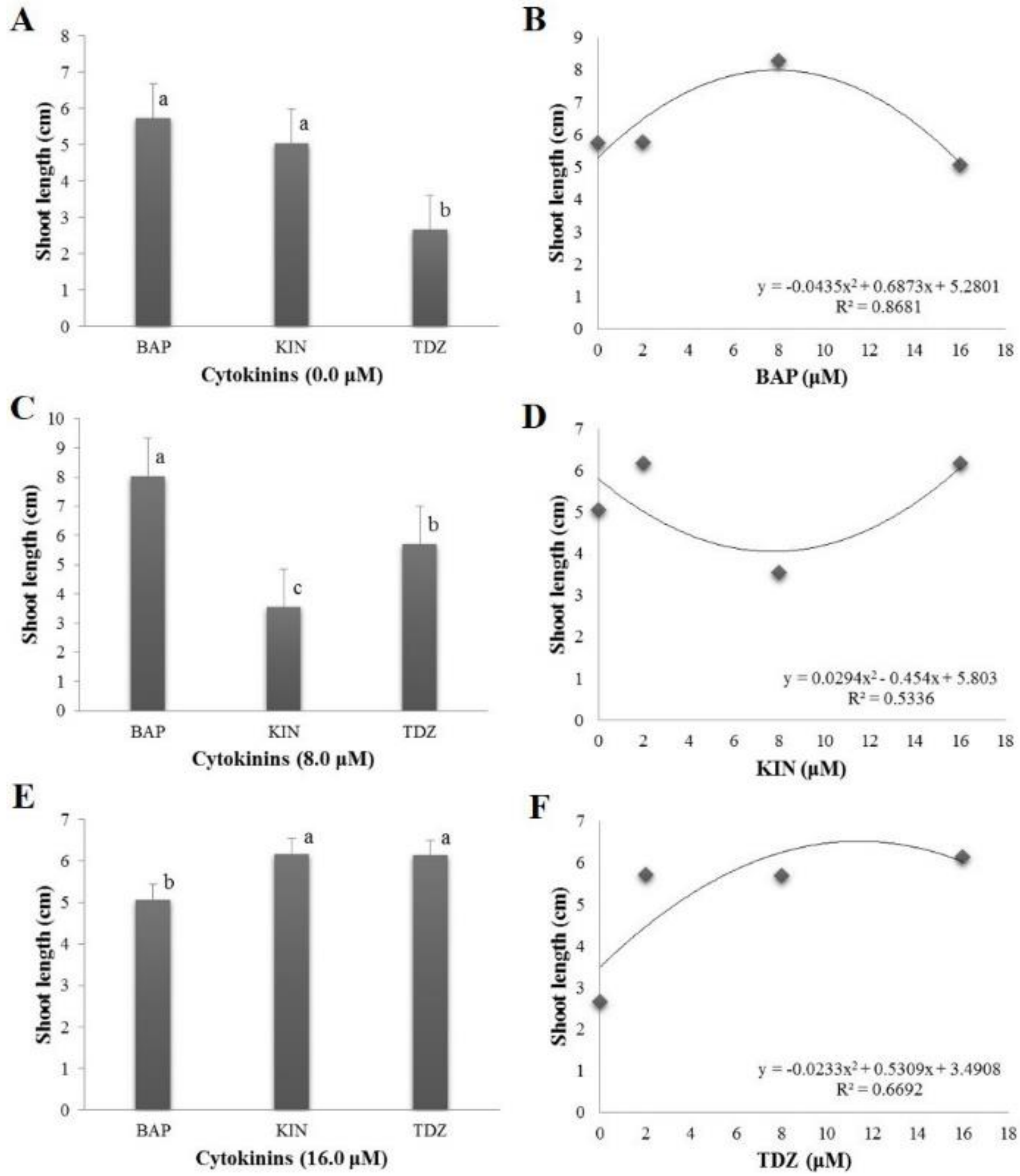


Figure 3. Effect of 0.0 µM (A), 8.0 µM (C), and 16.0 µM (E) cytokinins (BAP, KIN, and TDZ) and different concentrations (0.0, 2.0, 8.0, and 16.0 µM) of BAP (B), KIN (D), and TDZ (F) on the length of the shoots from the encapsulated buds regenerated in MS culture medium after 60 days of cultivation. Means followed by the same letter in columns (A), (C), and (E) do not differ among themselves, according to the Tukey test at 5% significance.

70% germination after 120 min in PVS2 (Figures 7 and 8).

DISCUSSION

Based on the results obtained in this study, we report that AC presented a significant positive effect on the *in vitro* germination of *M. ligustroides.* This effects related to the physical characteristics of AC, namely its tiny pores that cover a wide area of the culture medium, thus providing a large adsorption capacity (López-Pérez et al., 2015). As a result, a drastic reduction occurs in the oxidation of the phenolic compounds that many species, including *M. ligustroides*, exude (Thomas, 2008). In agreement with the results obtained in this study, *Elaeis guineensis*

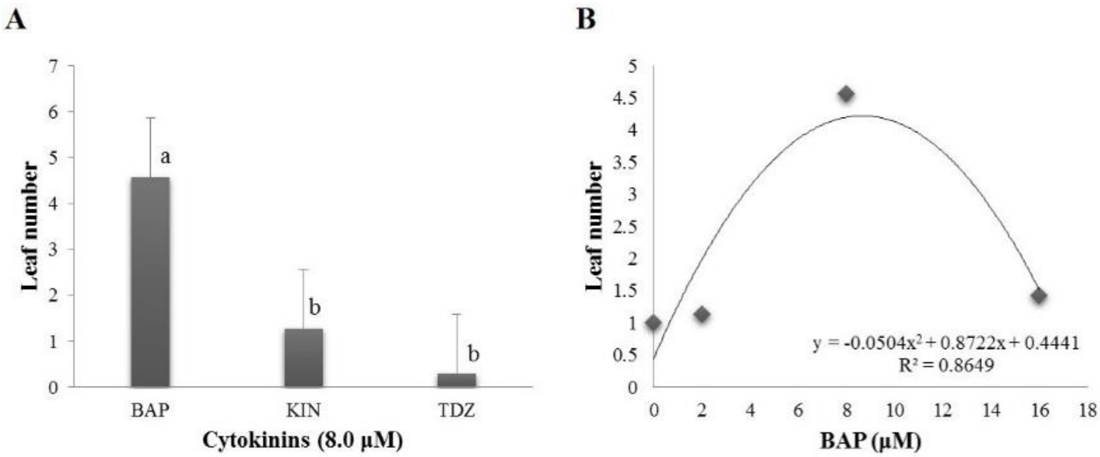


Figure 4. Effect of 8.0 µM BAP, KIN, and TDZ (A) and different concentrations of BAP (0.0, 2.0, 8.0, and 16.0 µM) (B) on the leaf number of shoots from the encapsulated buds regenerated in the MS culture medium after 60 days of cultivation. Means followed by the same letter in columns in (A) do not differ among themselves, according to the Tukey test at 5% significance.

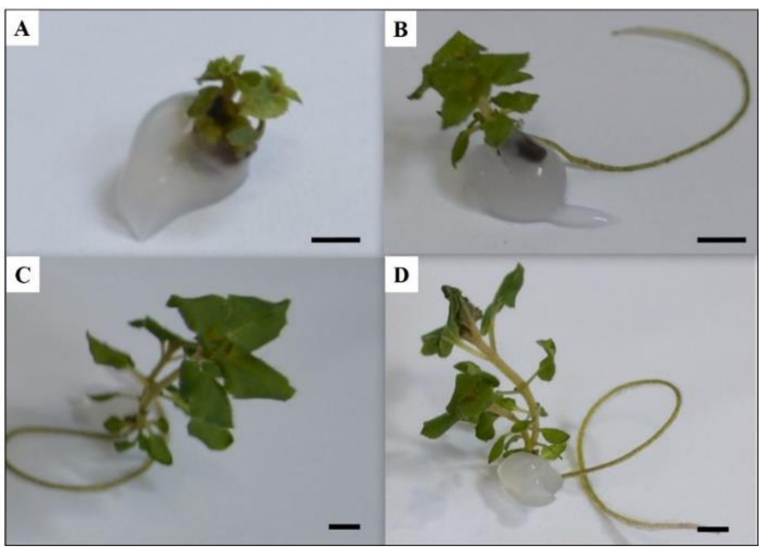


Figure 5. Visual appearance of the lateral buds encapsulated in a sodium alginate matrix in MS culture medium supplemented with 8.0 µM BAP at 30 days (A) bar = 0.5 cm; 45 days, bar = 0.5 cm (B); 60 days, bar = 1.0 cm (C); and 75 days of cultivation *in vitro,* bar = 1.0 cm (D).

embryos grown in MS culture medium supplemented with 2.0 g L^{-1} AC presented increased development (97%) in comparison to those embryos grown in media without AC (Suranthran et al., 2011).

Once a culture medium that maximizes the germination rate is obtained, the cryopreservation of the plant material in the form of seeds can be tested and widely used (Silva et al., 2013). The effect of the cryoprotectant PVS2 at different immersion times was evaluated after immersion in LN through the vitrification technique; however, no significant difference was observed in the survival of the seeds. The seeds of *M. ligustroides* have hard coats (Chaves et al., 2013), which may have affected the absorption of the cryoprotectant. The cryoprotectant solution cannot only influence the vitreous state of the internal solutes but can also lead to chemical toxicity (Prada et al., 2015) accelerating the loss of water and causing lethal damage during cryopreservation, mainly for species as *M. ligustroides* which has orthodox seeds and presents low water content (Engelmann and Gonzalez-Arnao, 2013). Therefore, the vitrification technique, without the need for prior immersion in PVS2,

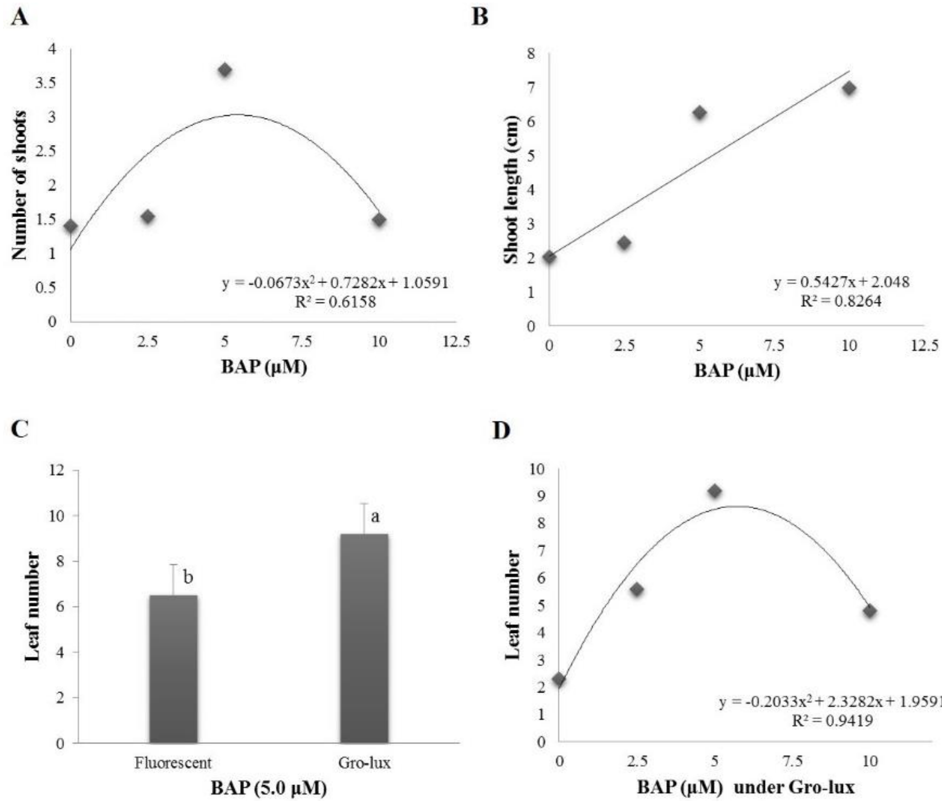


Figure 6. Effect of different concentrations of BAP (0.0, 2.5, 5.0, and 10.0 µM), regardless of the light source type used, on the number of shoots (A) and the length of shoots (B) of *M. ligustroides* after 60 days of *in vitro* cultivation in MS culture medium. Effect of the light source type – white or Gro-Lux fluorescent lamps – in the presence of 5.0 µM BAP on the leaf number per shoot (C) and the influence of different concentrations of BAP (0.0, 2.5, 5.0, and 10.0 µM) in the presence of the Gro-Lux light source on the leaf number (D). Means followed by the same letter in columns in (C) did not differ among themselves, according to the Tukey test at 5% significance.

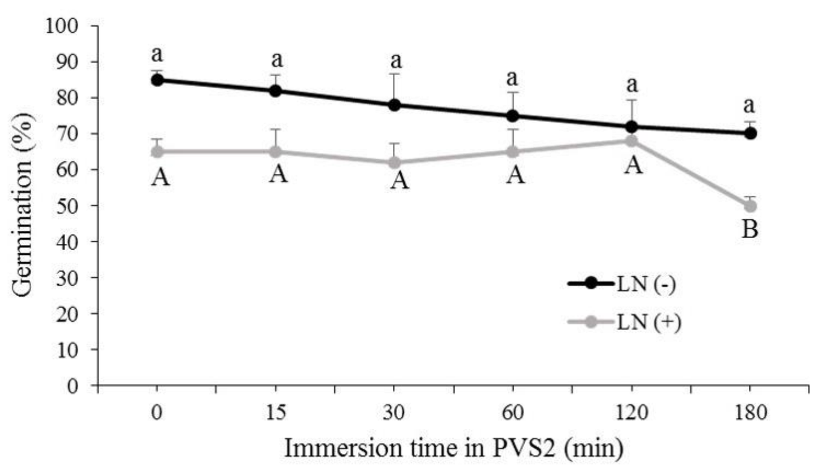


Figure 7. Percentage of germination of cryopreserved (LN +) and non-cryopreserved (LN -) seeds at 30 days of *in vitro* cultivation after exposure to the cryoprotectant solution PVS2 for different times (0, 15, 30, 60, 120, and 180 minutes). Means followed by the same lowercase letter in LN (-) or uppercase in LN (+) do not differ among themselves, according to the Tukey test at 5% significance.

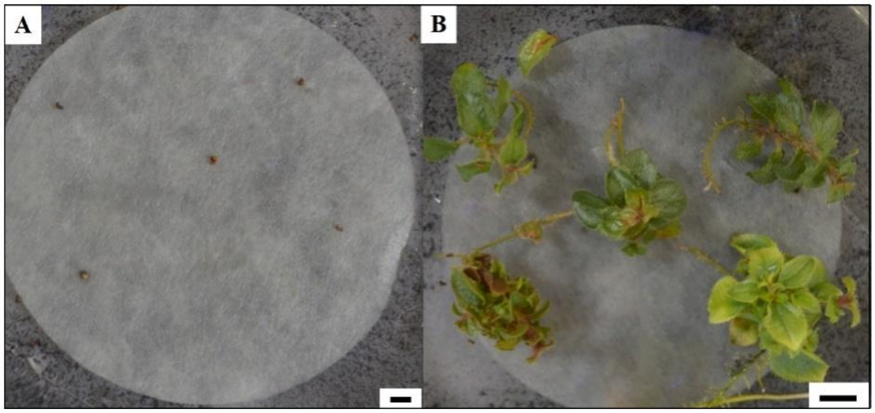


Figure 8. Visual appearance of *M. ligustroides* seeds recently removed from the cryopreservation process (bar= 0.5 cm) (A) and after 60 days of *in vitro* cultivation (bar= 0.5 cm) (B). The seeds were exposed to the cryoprotectant solution PVS2 for 120 min.

presented a high *in vitro* survival rate and can be used in future applications for the species.

Recently, the use of the encapsulation of plant explants in alginate matrix has become a beneficial alternative for *in vitro* multiplication (Fatima et al., 2013). The sodium alginate serves as artificial endosperm and must provide nutrients for the growth of the encapsulated explants (Kumar et al., 2005; Piatczak and Wysokinska, 2013). The addition of growth regulators can improve the development of plant material because the capsules provide a direct contact area with the explant with the small contact surface in test tubes or Petri dishes (Etienne and Berthouly, 2002; Polzin et al., 2014), thus increasing the desired effects.

Among the plant growth regulators used to supplement the culture medium, cytokinins act by inducing the proliferation of auxiliary buds, acting directly on cell division, elongation, and differentiation (Zhang et al., 2011). However, the type and concentration of the cytokinin varied in the *in vitro* multiplication process for each species. In this study, the cytokinin BAP, at a concentration of 8.0 µM, provided the best condition for the growth of encapsulated lateral buds, enabling rupture of the capsules and shoot development at a higher frequency.

The cytokinin TDZ, used in positive regenerative responses in a wide variety of species (Hosseini-Nasr and Rashid, 2002; Yancheva et al., 2003; Matand and Prakash, 2007), acts systematically in responses linked to somatic embryogenesis (Guo et al., 2011; Faisal et al., 2014). The cytokinin KIN presents various biological properties, such as promotion of transcription, cell cycle control, calcium flux, and antioxidant activity (Barciszewski et al., 1999; Verbeke et al., 2000), however, kinetin showed no positive effects for the variables evaluated during the development of *M. ligustroides* lateral buds.

For the shoot multiplication, an interaction was observed between BAP and the different types of fluorescent light (white or Gro-Lux), with the Gro-Lux red light bulbs generating larger values for the afore-mentioned characteristics. Such results could be related to a higher carbon gain through increased net photosynthetic rate and/or indirectly caused by plant hormonal changes resulting from changes in the light spectrum, which is important for the accumulation of starch and consequently greater investment in young sink organs (Folta, 2004; Rocha et al., 2010). Thus, several authors have studied this wavelength in order to ensure the maximization of the growth and development of plants cultivated *in vitro (Tanaka et al., 1998; Galdiano Júnior et al., 2012).*

Conclusions

The protocols developed here are considered suitable alternatives for the conservation of *M. ligustroides* and production of seedlings for commercialization purposes and use in programs of reintroduction in degraded environments.

ACKNOWLEDGEMENTS

The authors are grateful to Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq; Brasília, DF, Brazil), Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES; Brasília, DF, Brazil) for support in the form of a doctoral fellowship.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Barciszewski J, Rattan SI, Siboska G, Clark BF (1999). Kinetin- 45 years on. Plant Sci 148:37-45.

Bhojwani SS, Mullins K, Cohen D (1984). *In vitro* propagation of *Pyrus pyrifolia*. Sci. Hortic. (Amsterdam) 23:247-254.

Cejas I, Vives K, Laudat T, González-Olmedo J, Engelmann F, Martínez-Montero ME, Lorenzo JC (2012). Effects of cryopreservation of *Phaseolus vulgaris* L. seeds on early stages of germination. Plant Cell. Rep. 31:2065-2073.

Chaves IDS, Alvarenga AAD, Dousseau S, Soares GCM, Souza EDS, Artur MAS (2011). Germination of *Miconia ligustroides* (Melastomataceae) diaspores submitted to different treatments for dormancy overcoming. Rev. Bras. Bot. 34:335-341.

Chaves IDS, Alvarenga AA, Dousseau S, Souza EDS, Artur MAS, Lara TS (2013). Morphological characterization of fruits, diaspores and germination of *Miconia ligustroides* (DC.) Naudin (Melastomataceae). Acta Sci. Biol. Sci. 35:93-98.

Cunha WR, Crevelin EJ, Arantes GM, Crotti AEM, Silva, ML, Furtado NA, Ferreira DDS (2006). A study of the trypanocidal activity of triterpene acids isolated from *Miconia* species. Phyther. Res. 20:474-478.

Cunha WR, Silva MLA, Turatti ICC, Ferreira DS, Batarello HL (2003). Avaliação da atividade analgésica de *Miconia ligustroides* (Melastomataceae) utilizando o teste de contorção abdominal em camundongos. Rev. Bras. Farmacogn. 84:47-49.

Dulloo ME, Ebert A W, Dussert S, Gotor E, Astorga C, Vasquez N, Omondi C (2009). Cost Efficiency of cryopreservation as a long-term conservation method for coffee genetic resources. Crop Sci. 49:2123-2138.

Engelmann F (2011). Use of biotechnologies for the conservation of plant biodiversity. Vitr. Cell. Dev. Biol- Plant 47:5-16.

Engelmann F, Gonzalez-Arnao MT (2013). Introducción a la conservación ex situ de los recursos genéticos vegetales. In: Crioconservación plantas en América Lat. y el Caribe. Available on: https://www.researchgate.net/publication/259763161_Crioconservacion_de_recursos_geneticos_de_tuberculos_y_raices_andinos_en_el_Peru#page=38.

Etienne H, Berthouly M (2002). Temporary immersion systems in plant micropropagation. Plant Cell Tissue Organ Cult 69:215-231.

Faisal M, Alatar A.A, Hegazy AK, Alharbi SA, El-Sheikh M, Okla MK (2014). Thidiazuron induced in vitro multiplication of *Mentha arvensis* and evaluation of genetic stability by flow cytometry and molecular markers. Ind. Crops Prod. 62:100-106.

Fatima N, Ahmad N, Anis M, Ahmad I (2013). An improved *in vitro* encapsulation protocol, biochemical analysis and genetic integrity using DNA based molecular markers in regenerated plants of *Withania somnifera* L. Ind. Crops Prod. 50:468-477.

Ferreira DF (2014). Sisvar: a Guide for its Bootstrap procedures in multiple comparisons. Ciênc. Agrotec. 38:109-112.

Ferreira EA, Pasqual M, Rezende JC (2007). 2, 4-D and kinetin in callogenesis of *Ficus carica* L. Acta Hortic pp. 433-436.

Folta KM (2004). Green light stimulates early stem elongation, antagonizing light-mediated growth inhibition. Plant Physiol. 135:1407-1416.

Galdiano Júnior RF, Mantovani C, Pivetta KFL, Lemos EG de M (2012). Crescimento *in vitro* e aclimatização de *Cattleya loddigesii* Lindley (Orchidaceae) com carvão ativado sob dois espectros luminosos. Ciência Rural 42:801–807.

Goldenberg R, Almeda F, Caddah MK, Martins AB, Meirelles J, Michelangeli FA, Weiss M (2013). Nomenclator botanicus for the neotropical genus *Miconia* (Melastomataceae: Miconieae). Phytotaxa 106:1.

Guo B, Abbasi BH, Zeb A, Xu, LL, Wei YH (2011). Thidiazuron: A multi-dimensional plant growth regulator. African J. Biotechnol. 10:8984-9000.

Hosseini-Nasr M, Rashid A (2002). Thidiazuron-induced shoot-bud formation on root segments of *Albizzia julibrissin* is an apex-controlled, light-independent and calcium-mediated response. Plant Growth Regul. 36:81-85.

Jona R, Webb KJ (1978). Callus and axillary-bud culture of *Vitis vinifera* “Sylvaner Riesling.” Sci. Hortic. (Amsterdam) 9:55-60.

Kumar MBA, Vakeswaran V, Krishnasamy V (2005). Enhancement of synthetic seed conversion to seedlings in hybrid rice. Plant Cell Tissue Organ Cult. 81:97-100.

López-Pérez AJ, Carreño J, Martínez-Cutillas A, Dabauza M (2015). High embryogenic ability and plant regeneration of table grapevine cultivars (*Vitis vinifera* L.) induced by activated charcoal. VITIS - J. Grapevine Res. 44:79.

Matand K, Prakash CS (2007). Evaluation of peanut genotypes for *in vitro* plant regeneration using thidiazuron. J. Biotechnol. 130:202–207.

Michelangeli FA, Penneys DS, Giza J, Soltis D, Hils MH, Skean JD (2004). A preliminary phylogeny of the Tribe Miconieae (Melastomataceae) bsed on nrITS Sequence data and its implications on inflorescence position. Taxon 53:279.

Murashige T, Skoog F (1962). A revised medium for rapid growth and bio assays with tobacco tissue cultures. Physiol. Plant 15:473-497.

Piatczak E, Wysokinska H (2013). Encapsulation of *Centaurium erythraea* Rafn – an efficient method for regeneration of transgenic plants. Acta Biol. Cracov. Ser. Bot. 55:37-44.

Polzin F, Sylvestre I, Déchamp E, Ilbert P, Etienne H, Engelmann F (2014). Effect of activated charcoal on multiplication of African yam (*Dioscorea cayenensis-rotundata*) nodal segments using a temporary immersion bioreactor (RITA®). Vitr. Cell Dev. Biol. Plant 50:210-216.

Prada JA, Aguilar ME, Abdelnour-Esquivel A, Engelmann F (2015). Cryopreservation of Seeds and embryos of *Jatropha curcas* L. Am. J. Plant Sci. 6:172-180.

Pritchard HW (1995). Cryopreservation of Seeds. In: Cryopreservation and Freeze-Drying Protocols. Humana Press, New Jersey pp. 133-144

Pritchard HW, Nadarajan J (2008). Cryopreservation of Orthodox (Desiccation Tolerant) Seeds. In: Plant Cryopreservation: A Practical Guide. Springer New York, New York, NY pp. 485-501

Prudente de OD, Nery FC, Paiva R, dos Reis MV, de Oliveira Paiva PD, Nery M C (2016). Cultivo *in vitro* de *Miconia ligustroides* (DC.) Naudim. Plant Cell Cult. Micropropagation 12:13-19.

Reis ES, Pinto JEBP, Rosado LDS, Corrêa RM (2015). Influência do meio de cultura na germinação de sementes *in vitro* e taxa de multiplicação de *Melissa officinalis* L. Ceres 55:160-167.

Renner SS (1993). Phylogeny and classification of the Melastomataceae and Memecylaceae. Nord J. Bot. 13:519-540.

Rocha PSG, Oliveira RP, Scivittaro WB (2013). Sugarcane micropropagation using light emitting diodes and adjustment in growth-medium sucrose concentration. Ciênc. Rural 43:1168-1173.

Rocha PSG, Oliveira RP, Scivittaro WB, Santos UL (2010). Diodos emissores de luz e concentrações de BAP na multiplicação *in vitro* de morangueiro. Ciência Rural 40:1922-1928.

Romero R, Martins AB (2002). Melastomataceae do Parque Nacional da Serra da Canastra, Minas Gerais, Brasil. Rev Bras. Botânica 25:19-24.

Sandoval-Yugar EW, Dal Vesco LL, Steinmacher DA, Stolf EC, Guerra MP (2009). Microshoots encapsulation and plant conversion of *Musa* sp. cv. “Grand Naine.” Ciênc. Rural 39:998-1004.

Silva CL, Paiva R, Swennen R, Andrè E, Panis B (2012). Shoot-Tip Cryopreservation by Droplet Vitrification of *Byrsonima intermedia* A. Juss.: A woody tropical and medicinal plant species from Brazilian Cerrado. CryoLetters 34:338-348.

Suranthran P, Sinniah U, Subramaniam S, et al. (2011). Effect of plant growth regulators and activated charcoal on *in vitro* growth and development of oil palm (*Elaeis guineensis* Jacq. var. Dura) zygotic embryo. Afr. J. Biotechnol. 10:10600-10606.

Tanaka M, Takamura T, Watanabe H, Endo M, Yanagi T, Okamoto K (1998). *In vitro* growth of *Cymbidium* plantlets cultured under superbright red and blue light-emitting diodes (LEDs). J. Hortic. Sci. Biotechnol. 73:39-44.

Thomas TD (2008). The role of activated charcoal in plant tissue culture. Biotechnol. Adv. 26:618-631.

Verbeke P, Siboska GE, Clark BFC, Rattan SIS (2000). Kinetin inhibits protein oxidation and glycoxidation *in vitro*. Biochem. Biophys. Res. Commun. 276:1265-1270.

Walters C, Wheeler L, Stanwood PC (2004). Longevity of cryogenically stored seeds. Cryobiology 48:229-244.

Yancheva SD, Golubowicz S, Fisher E, Lev-Yadun S, Flaishman MA

(2003). Auxin type and timing of application determine the activation of the developmental program during *in vitro* organogenesis in apple. Plant Sci. 165:299-309.

Zhang W, To JP, Cheng CY, Eric Schaller G, Kieber JJ (2011). Type-A response regulators are required for proper root apical meristem function through post-transcriptional regulation of PIN auxin efflux carriers. Plant J. 68:1-10.

academicJournals

Vol. 12(7), pp. 512-523, 16 February, 2017
DOI: 10.5897/AJAR2016.11807
Article Number: ECBBF8462771
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Physical and physiological attributes of black oat seeds produced in southern Brazil

Roberto Carbonera[1]*, Ubirajara Russi Nunes[2], Alessandro Dal'Col Lúcio[2], José Antônio Gonzalez da Silva[1] and Lorenzo Ghisleni Arenhardt[1]

[1]Department of Agrarian Studies, Regional Northwest University of Rio Grande do Sul (UNIJUÍ), 3000 Comércio Street, Ijuí, RS, 98700-000, Brazil.
[2]Department of Crop Science, Federal University of Santa Maria (UFSM), University Campus - Rural Science Center Building 77, 97105-900 - Santa Mari, a - RS - Brazil.

Received 9 October, 2016; Accepted 18 January, 2017

Black oat crop occupies the largest area during winter. It is important for animal production, crop rotation and dry matter production of approximately 3.8 million hectares in Rio Grande do Sul State. To ensure sowing, seeds are produced which must have high quality standards and must be evaluated by seed testing laboratories. Given this, the present study evaluated the physical and physiological quality of oat seeds analyzed by the Seed Analysis Laboratory of UNIJUI from 2006 to 2014. 2,910 samples were evaluated; 2,229 were evaluated with seed production process; 357 were evaluated with seed analysis of own use and 324 were evaluated with tetrazolium analysis. The samples obtained through seed production process were analyzed in terms of their physical and physiological aspects, while the own seed and tetrazolium test used were evaluated only in terms of their physiological attributes, following the methodology described in the Seed Analysis Rules. The data were analyzed through descriptive statistics for each variable studied per year, and the averages, maximum and minimum standard deviation and coefficient of variation were identified. The data were also evaluated based on their dispersion, and compared to weather occurrences and national standards, in order to estimate the percentages of samples approved. Seeds produced according to the National Seed system had high levels of physical and physiological quality from 2006 to 2010. However, between 2011 and 2014, 14.0 and 14.5% of the samples were above the standard levels for seeds of other cultivated species and harmful prohibited species respectively. The own used seeds showed greater variability and dispersion, with 18.1 and 31.7% samples below the standards for germination in the years 2006 to 2010 and 2011 to 2014, respectively, while the samples analyzed through the tetrazolium test showed approved levels of 19.4 and 12.5%, respectively. The major physiological qualities were obtained in 2008, 2010 and 2011 and the lowest in 2009, 2012 and 2014. It is noteworthy that the seed quality is related to years with levels of rainfall and appropriate temperatures for vegetative development, physiological maturity and harvest.

Key words: *Avena strigosa* (Schreb.), purity, germination, tetrazolium.

INTRODUCTION

The use of seeds with high genetic, physical, physiological and health quality constitutes a decisive element for the implementation of crops with the potential to maximize the cultivars' performance. The seeds have a great deal of responsibility for agricultural development in normal times as well as for its recovery after the occurrence of shocking events such as droughts, floods and epidemics (Oliveira et al., 2013; Carvalho and Nakagawa, 2012; Peske et al., 2006).

Livestock production in the Northwest Region of Rio Grande do Sul was stimulated by the introduction of forage species, due to the wheat and soybean binomial crisis in the 70s and 80s. From this introduction, research that enabled the domain of seeds production technology was performed (Medeiros, 1976, 1987; Souza et al., 1992).

The knowledge generated led to the increase in seed production, either by producers, cooperatives or companies. The planting of introduced cultivars seeds remained for decades, thanks to natural selection processes and exchange of seeds between regions. This enabled natural crossings, causing the genetic basis of cultivated species such as annual ryegrass (*Lolium multiflorum* Lam.), oat (*Avena strigosa* Schreb.), white clover (*Trifolium repens* L.), red clover (*Trifolium pratense* L.), vetch (*Vicia* spp.), birdsfoot trefoil (*Lotus corniculatus* L.) and alfalfa (*Medicago sativa* L.) (Maia, 2013).

Currently, forage cultivation is the basis for feeding dairy and plays an important role in the development of this activity in RS Northwest Region. According to Agricultural Census of 2006, there were 204,000 establishments producing 2.7 billion liters of milk annually in the RS, and the North-west Region accounted for over 60% of production (Trennepohl, 2011). Currently, the state produces four billion liters of milk per year, the second Brazilian state production (Mello, 2013).

For a long period, there was great informality in the forage species seed sector, with few cultivars suitable for cultivation and multiplication (Pereira, 2013). This condition has been changing in recent years with greater organization and formalization of this industry, from the approval and implementation of national seed and seedling system (Brazil, 2003).

Among the cultivated species, oat occupies a prominent place in the state of Rio Grande do Sul. According to Del Duca et al. (2004), it is the species that has been occupying the largest growing area in winter, with an area estimated at 3,850,000 ha in 2013/14 crop (Abrasem, 2014). The recommended seeding rate ranges from 30 to 60 kg ha^{-1} (Flaresso et al., 2001; Debiasi et al., 2007), which can generate an effective demand for seeds to 231,000 tons (Abrasem, 2014).

The oat is a species of temperate climate, is rustic and resistant to small droughts; it has excellent ability of tillering, green mass production, is tolerant to animal trampling, and resistant to pests and diseases. With the evolution of tillage, crop rotation and training straw were used, with benefits to the successor species (Carvalho et al., 2010; BortolinI et al., 2000).

In support of seed production system, studies have been carried out to assess the physical and physiological quality in order to clarify the standards presented by the seeds produced. Nakagawa et al. (2004) showed that seed oats originating from soil with lower fertility had less storage capacity, especially in natural laboratory environment with zero percent germination after 48 months. The seeds produced in the most fertile soil germinated above 90% at 60 months, when stored in air-conditioned environments and greater storage capacity was enhanced in less favorable storage conditions.

Work performed by Fonseca et al. (1999) showed that only 47% of ryegrass seed samples showed standards of purity and germination according to the RS standards; 81% had standards for germination and 54% met the physical standards of purity.

Study on ryegrass seeds carried out in 2005 to 2007, in the state of Paraná showed that in the harvests of 2005 and 2006 most of the samples of pure seed had 86 to 96% below the National Standard, which is 97%. In 2007, 100% of the samples of Italian ryegrass were within the standard (Ohlson et al., 2008).

The same study showed in the germination test, that 46% of the samples had lower germination percentage than the standard in 2005 and 64% of the samples of Italian ryegrass had lower germination percentage than the standard in 2006. In 2007, 100% of the samples of ryegrass had germination percentage above the standard, which is 70%.

Studies performed in 2008 to 2010 concluded that from 50 to 100% depending on the year and cultivar, the samples of ryegrass were below the standard for the parameter of pure seeds. Regardless of the reporting year, the predominant cultivar was ryegrass (66 to 100%), and all samples belonging to the category S2 (Ohlson et al., 2011a).

Colonião grass analyses showed that all samples of cultivar Tanzania were below the standard in pure seed parameter; for the cultivar, Mombasa, from 50 to 82% depending on the season, they did not reach the standard. The same was observed in cultivar Aruana in 2007 season, when only 50% samples were framed in this parameter. In 2007, 100% of pearl millet samples of BRS 1501 cultivar were below the standard for pure seed in the State of Paraná (Ohlson et al., 2010).

In *Brachiaria brizantha,* Ohlson et al. (2011b) showed that from 97 to 100%, depending on the year and cultivar, the samples were class S2 (not certified seed of the second generation); and from 55 to 100%, depending on

*Corresponding author. E-mail: carbonera@unijui.edu.br.

Table 1. Number of samples of oats seeds analyzed by the Seed Analysis Laboratory, Agronomy Course, Department of Agrarian Studies, Universidade Regional do Noroeste do Estado do Rio Grande do Sul, from 2006 to 2014, Santa Maria, 2016.

Category	2006	2007	2008	2009	2010	2011	2012	2013	2014	Total
Seeds S1 and S2	256	281	260	394	547	116	139	184	52	2229
Own use	44	44	25	47	55	39	40	38	25	357
Tetrazolium analysis	84	42	42	46	38	31	20	11	10	324
Total	384	367	327	487	640	186	199	233	87	2910

S1; S2, Not certified seed, first and second generation, respectively.

the year, the samples reached the minimum for pure seeds in Paraná State.

In the analyses of seeds carried out with 106 samples of *B. brizantha* 11 *Brachiaria humidicola* and 38 samples of *Panicum maximum* in the State of Rondônia, 85.9, 72.7 and 94.7%, respectively were below the standards, and the main reason for the low quality was impurity (Parmejiani et al., 2014).

As it is observed, there is a great variability in the quality of forage seeds produced, a fact that may impact the establishment of appropriate plant stand and the productive potential of these species. Thus, it is important to characterize and categorize the physical and physiological attributes of the seeds produced and associate them with national quality standards in order to indicate the use of seeds with high quality standards.

Given this, the present study has the general objective of evaluating the physical and physiological quality of black oat seeds produced in Southern Brazil, and specifically, to characterize the physical purity levels presented in the samples, the percentage of pure seeds, other seeds and inert material, as well as studying the occurrence of other seeds. The study aimed also to associate the quality of seeds to the production profile either by companies or for their own use, and the possible effects caused by meteorological factors.

MATERIALS AND METHODS

This work was carried out by using Seed Analysis database from the Laboratory of Seeds Analysis – Agronomy Course, Agrarian Studies (DEAg), Universidade Regional do Noroeste do Estado do Rio Grande do Sul (UNIJUÍ), located in the city of Ijuí, RS. The department was accredited in 1995 by the Ministry of Agriculture, Livestock and Supply.

The analysis results of black oat seeds were evaluated (Avena strigosa Schreb.), belonging to 2006 to 2014 crops (Table 1). 2,910 samples were considered. Out of these, 2,229 were members of the seed production process and all samples belonged to the categories S1 and S2 (not certified seed from first and second generations, respectively). It was also studied the results of 357 seed samples intended for their own use, as the current legislation assures Brazil (1997) and Brazil (2003) and 324 seed analysis evaluated by the tetrazolium test. The number of samples, for example, accounted for 5.1% of the total of 96,086.72 tons of oats seeds produced in the state of Rio Grande do Sul in 2013 (Rio Grande Do Sul, 2014).

Analyses of physical and physiological quality of the seeds were carried out following the methodology recommended by the Seed Analysis Rules (BRAZIL, 1992, 2009). Purity analysis determines the percentage composition by weight and the identity of the different species of seed and inert material present in the sample. The working sample is separated into three components: pure seed, other seeds and inert material, which are indicated in percentage by weight of the working sample. Purity analysis was performed on a sample of 100 g to 2010 and, from this year, in a sample of 50 g. This change was promoted due to the entry of Instruction No. 33/2010 in force, Ministry ... (2010), which abolished the Ministerial Order No. 381/1998 (Ministry, 1998).

The determination of harmful species, complementing the purity analysis was carried out until the year 2010, according to Ministerial Order No. 381/1998 (Ministry, 1998, 1992). This determination was changed to the determination of other seeds by number, including other cultivated species, wild and harmful tolerated and prohibited species, because the Instruction No. 33/2010 was in force (Ministry, 2010). These measurements were performed in samples of 500 g.

The completion of the germination test also followed the recommendations for the species. Pre-drying was done at 35°C for seven days. After that, the seeds were sown on roll paper, with four replications of 100 seeds and placed in a chamber at 20°C temperature. In the evaluation, the percentage of normal seedlings, abnormal seedlings and dead seeds was determined (Brazil, 1992, 2009). Work done by Grzybowski et al. (2015) showed that pre-drying may be carried out for five days. The tetrazolium test which determines the feasibility of seeds also followed the criteria established in the rules for seed testing.

Samples of seeds originating in the production process following the national system of seed production and seedlings were analyzed for purity and germination, while the samples of own use of seeds were analyzed only with germination test. The tetrazolium analysis is used in the seed production process, as previous analysis, or own use of seeds, with an effective test for the rapid assessment of the viability of oats seeds (Souza et al., 2009).

The standards used to compare the results of analyses carried out by the year 2010 are as follows: Minimum germination of 75%, minimum 95% of pure seeds, maximum of 50 seeds from other cultivated species, 40 seeds of wild species, 40 seeds of tolerated harmful species and zero of harmful prohibited species seeds, according to Ministerial Order No. 381/1998 (MINISTRY, 1998). From the year 2010, for seeds belonging to S1 and S2 categories, not certified seeds from first and second generation, respectively, the following standards were used: Minimum germination of 80%, minimum of 97% of pure seeds, maximum 1% of other seeds, up to 16 seeds of other cultivated species or 20 from the oat species, and a maximum of 48 seeds for the category S2, up to 20 wild seeds, six harmful tolerated seeds and zero prohibited harmful seeds as stated in Instruction No. 33/2010 for black oat (Ministry, 2010).

The results were submitted to descriptive statistical analysis based on the years studied and we identified the outliers from the mean, standard deviation and coefficient of variation, using the GENES program (Cruz, 2013). There was also the presentation of data in scatter plots, with the assistance of the Office Excel

Table 2. Descriptive statistics (ED), number of samples (N), mean (x̄), standard deviation (SD), maximum (Mmax), minimum (Mmin), pure seeds (%), inert material (%) other seeds (%), number of other seeds by number, number of cultured species seeds, number of wild species seeds, number of harmful tolerated species and harmful prohibited in black oat seeds produced in crops from 2006 to 2014, by the national system of seed production, LAS / UNIJUI. Santa Maria, RS, 2016.

Variable	ED	Years/seed purity									
		2006	2007	2008	2009	2010	2011	2012	2013	2014	x̄
Pure seeds (%)	N	256	281	260	394	547	116	139	184	52	247.7
	x̄	99.3	98.9	99.3	99.4	99.5	99.4	99.5	99.7	99.7	99.4
	DP	0.59	0.73	0.76	0.45	0.45	0.44	0.43	0.21	0.28	0.48
	Mmáx	100	100	100	100	100	100	100	100	100	100
	Mmin	96.6	96.7	96.3	97.5	96	97.8	97.0	99	98.8	97.3
		Purity decomposition									
Inert matirial (%)	x̄	0.60	0.92	0.45	0.44	0.42	0.48	0.43	0.23	0.25	0.47
	DP	0.49	0.72	0.44	0.28	0.40	0.37	0.43	0.18	0.24	0.39
	Mmáx	3.2	3.3	2.3	1.9	3.1	2.1	3.0	1.0	1.0	2.32
	Mmin	0	0	0	0	0	0	0	0	0	0
Other seeds (%)	x̄	0.13	0.13	0.27	0.17	0.04	0.09	0.03	0.32	0.04	0.13
	DP	0.24	0.21	0.46	0.24	0.11	0.16	0.05	0.79	0.13	0.26
	Mmáx	1.1	1.10	2.30	2.20	0.90	0.80	0.20	0.50	0.90	1.11
	Mmin	0	0	0	0	0	0	0	0	0	0
		Purity decomposition, other seeds by number									
Number of other seeds by number	x̄	10.5	8.5	14.6	8.6	6.3	32.9	7.5	9.5	5.2	11.5
	DP	19.6	12.2	24.4	9.7	39.6	55.1	5.87	12.9	7.3	20.74
	Mmáx	128.0	92.0	142.0	51.0	315.0	238.0	33.0	75.0	50.0	124.8
	Mmin	0	0	0	0	0	0	0	0	0	0
Cultured species	x̄	8.3	7.3	13.5	7.4	5.9	27.7	3.3	7.5	3.2	9.3
	DP	15.9	11.0	24.0	9.2	29.6	51.0	4.5	12.9	6.8	18.3
	Mmáx	108.0	91.0	139.0	50.0	315.0	230.0	22.0	73.0	44.0	119.1
	Mmin	0	0	0	0	0	0	0	0	0	0
Wild species	x̄	1.20	0.74	0.87	1.10	0.23	0.31	0.00	0.00	0.02	0.5
	DP	3.48	1.73	1.80	1.79	0.68	1.89	0.00	0.00	0.14	1.27
	Mmáx	26	10	16	15	6	19	0	0	1	10.3
	Mmin	0	0	0	0	0	0	0	0	0	0
Harmful tolerated species	x̄	0.99	0.42	0.20	0.18	0.11	4.82	4.2	2.00	1.98	1.7
	DP	2.66	1.33	0.84	1.01	0.55	5.99	4.44	2.25	1.74	1.71
	Mmáx	28.0	14.0	10.0	14.0	5.0	31.0	21.0	13.0	6.0	15.7
	Mmin	0	0	0	0	0	0	0	0	0	0
Harmful prohibited species	x̄	0.01	0	0	0	0	0	0	0	0	0.00
	DP	0.06	0	0	0	0	0	0	0	0	0.01
	Mmáx	1.00	0	0	0	0	0	0	0	0	0.11
	Mmin	0.00	0	0	0	0	0	0	0	0	0.00

application program, and the percentage of samples outside the recommended standards was determined, and we compared the physiological quality data with weather occurrences of maximum temperatures and rainfall.

RESULTS AND DISCUSSION

The average percentage of pure seed was 99.4% studied in nine years (Table 2). In every year there were samples with 100% pure seed. The standard deviation of the average was 0.48. These data indicate higher levels of quality than those obtained by Fonseca et al. (1999) in ryegrass. The data also show superiority when compared to data from other similar works with analysis of forage species seed (Ohlson et al., 2008, 2011a; Parmejiani et al., 2014). This is due probably to the efforts and investment made by the seed companies in order to obtain seeds with a high degree of physical purity.

The presence of inert material reached an average of 0.47% over nine years, with a maximum of 3.3% in 2007. These data, combined with an average standard deviation of 0.39, indicate a low degree of presence of these impurities, meaning that the seeds were harvested and processed efficiently in removing this material. In inert material, straw, soil particles, dust, part of caryopses, palea, lemma and culture remained was found.

The average presence of other seeds was 0.13% and the standard deviation was 0.26 (Table 2). This variable also has a low average value, since the maximum is 1% of the sample (Ministry, 2010). As for the presence of other seeds by number, the average was 11.5 seeds and the average standard deviation was 20.74 (Table 2). The data indicate that there was a great variability in the samples analyzed for this variable. Out of these, 9.3 seeds per sample were seeds of other cultivated species, wild seeds 0.5, 1.7 seeds harmful tolerated species and zero of harmful prohibited.

The presence of grown species seeds was on average 9.3 seeds per sample and a high standard deviation of 18.3 (Table 2). Data maximum numbers indicate samples with up to 315 seeds, in 2010, for a maximum permitted up to 16 seeds of other cultivated species and 20 seeds of the oat species (Ministry, 2010). The high seed values of other cultivated species was mainly due to the presence of *Lolium mulflotum* Lam presenting limitations of specific products for their management and control in areas of black oat.

Wild species of seeds occurred, on average, 0.5 seeds per sample, standard deviation of 1.27. The highest data occurred in 2006 with 1.2 seeds on average per sample (Table 2). The occurrence of wild species seed was low, since it is allowed up to 20 seeds per sample (Ministry, 2010).

The average presence of harmful species tolerated seeds was 1.7 seeds per sample, occurring more sharply in the years 2011 and 2012 in which 4.8 and 4.2 seeds occurred per sample, respectively. The average standard deviation was 1.71. The increased presence of harmful tolerated species of seed was given mainly due to the reframing of *Avena barbata* Pott ex Link that went from wild to harmful tolerated, and allowed only six seeds per sample (Ministry, 2010).

The presence of harmful prohibited species seeds was observed in a sample in 2006. This indicates that seeds produced had no seeds of harmful prohibited species expressing a high degree of purity regarding this physical attribute.

Figure 1 shows the samples of scatter plots for purity analysis, containing the percentage of purity, inert material and other seeds, the number of other seeds by number, the number of crop species seeds, the number of wild seed and the number of seeds of harmful tolerated species in oat seed samples produced in the seed production process following the national system of seeds and seedlings.

Analysis of the physical attributes found a high degree of physical purity over the years, as shown in Figure 1A. There were slight increases in levels of the percentage of inert material in 2007 and 2010 (Figure 1B), and percentage of other seeds in 2006, 2008 and 2009 (Figure 1C).

Regarding the number of other seeds by number, a significant increase in rates in 2010 and 2011 can be observed (Figure 1D). In these years, there was an increase in the number of other seeds of cultivated species, especially ryegrass (Figure 1E). A trend to reduce the number of seeds of wild species and the increase of seeds of harmful tolerated species from the year 2010 may be noted, also. This is due mainly to the changing frame of the species *A. barbata Pott* ex Link that went from wild species to harmful tolerated species, based on Instruction No. 33/2010 (Ministry, 2010).

The results of physiological analysis of black oat seeds evaluated by germination test are shown in Table 3. The average germination was 90.3% over the nine years of analysis, with mean maximum of 98.7%, average minimum of 59.3% and a standard deviation of 6.0. In 2008, 2010 and 2011 averages above 92% of normal seedlings were obtained, and in the years 2009, 2012 and 2014 the averages were below 90%.

Abnormal seedlings had an average of 5.6%, with 0.5% average of the minimum and 18.6% of average maximum and a low average standard deviation of 3.6. There was percentage of abnormal seedlings in the year 2012; an average of 8.2% (Table 3).

Dead seeds showed an average of 4.1%, ranging from zero to 30.5%, average of the maximum and average standard deviation of 3.7 (Table 3). The percentage of dead seeds was higher in 2014, with 7.5%, and this year was considered a year with a low average germination.

The physiological attributes of seeds, evaluated by the germination test, can be seen in Figure 2. The same shows that there was less dispersion of normal seedlings in the years 2011 and 2013 and increased dispersion in the year 2009, which was a year with lower germination means (Figure 2A).

Abnormal seedlings rates exhibited higher dispersion in the years 2007, 2009 and 2012 (Figure 2B) and dead seedlings indexes showed the highest dispersion in the years 2009 and 2014 (Figure 2C).

The results of the analysis of own use seeds germination are shown in Table 4. The average germination was 83.2%, the average maximum was 98.2%, average minimum was 26.2% and a standard deviation was 15.4. The averages were higher in 2007 and 2013, whereas in the years 2012 and 2014 averages were below 80%. The percentage of abnormal seedlings was 5.9%, while the percentage of dead seeds was 10%.

By comparing the data analysis of the germination

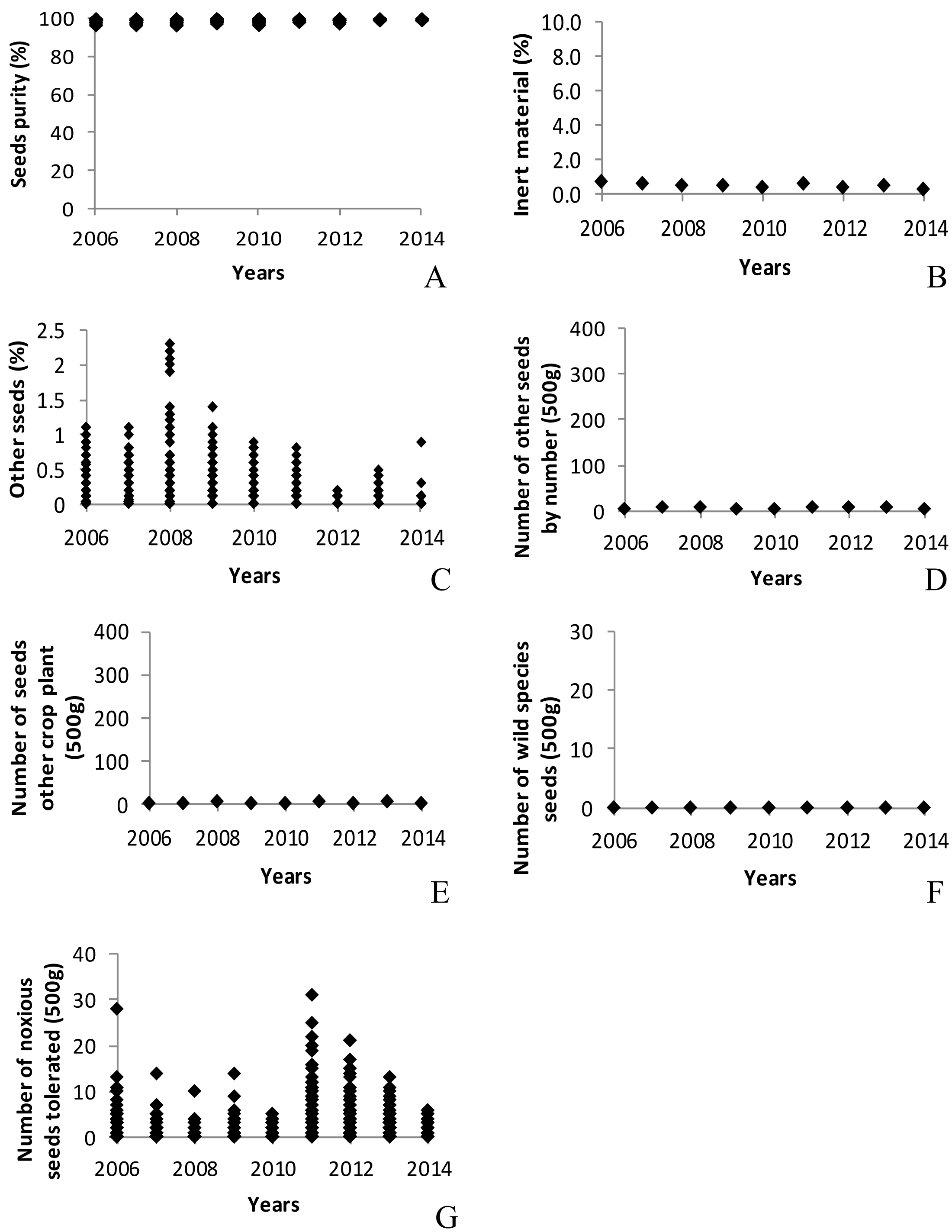


Figure 1. Scatter plots for physical purity (%), inert materials (%), other seeds (%) number of other seeds by number, other crop species, wild and harmful tolerated species in samples of black oat seeds, analyzed between 2006 and 2014, LAS/UNIJUI. Santa Maria, RS, 2016. A, Seeds purity (%); B, inert material (%); C, other seeds (%); D, number of other seeds by number; E, number of seeds other crop plant; F, number of wild species seeds; G, number of harmful tolerated seeds.

seeds produced in the seed production process with own use seeds, a reduction in average percentage of 90.3% germination was observed for 83.2%, increase in deviation standard from 6.0 to 15.4 and increase of the percentage of dead seeds from 3.7 to 10%.

The data presented above indicate that the physiological quality of own use seeds were lower than seed produced in the seed production process, possibly

Table 3. Descriptive statistics (ED), number of samples (N), mean ($\bar{x}$), Standard Deviation (SD), maximum (Mmax), Minimum (Mmin) to Normal Seedlings (%) Abnormal Seedlings (%) Dead Seeds (%) in oat seeds analyzed between 2006 and 2014, LAS / UNIJUI. Santa Maria, RS, 2016.

Variable	ED	Years/ germinated seeds									
		2006	**2007**	**2008**	**2009**	**2010**	**2011**	**2012**	**2013**	**2014**	**x̄**
Normal seedlings (%)	N	256	281	260	394	547	116	139	184	52	247.7
	x̄	91.2	91.6	92.0	88.6	92.1	92.1	87.5	90.6	87.6	90.3
	DP	9.40	5.08	4.85	5.9	5.16	3.88	5.2	6.01	8.67	6.0
	Mmáx	99	99	99	99	99	98	97	99	99	98.7
	Mmin	3	69	66	46	61	82	65	77	65	59.3
Abnormal seedlings (%)	x̄	3.2	5.9	4.1	5.9	5.8	5.9	8.2	6.3	4.9	5.6
	DP	2.16	3.99	2.83	5.94	2.27	3.69	3.60	4.53	3.64	3.6
	Mmáx	13	20	19	24	20	16	23	18	15	18.6
	Mmin	0	0	0	0	0	1	2	1	1	0.5
Dead seeds (%)	x̄	5.6	2.9	3.8	5.4	3.0	1.9	3.9	3.1	7.5	4.1
	DP	9.08	2.64	3.70	4.17	3.01	1.47	3.07	2.42	3.83	3.7
	Mmáx	94	26	33	24	32	8	19	11	28	30.5
	Mmin	0	0	0	0	0	0	0	0	0	0.0

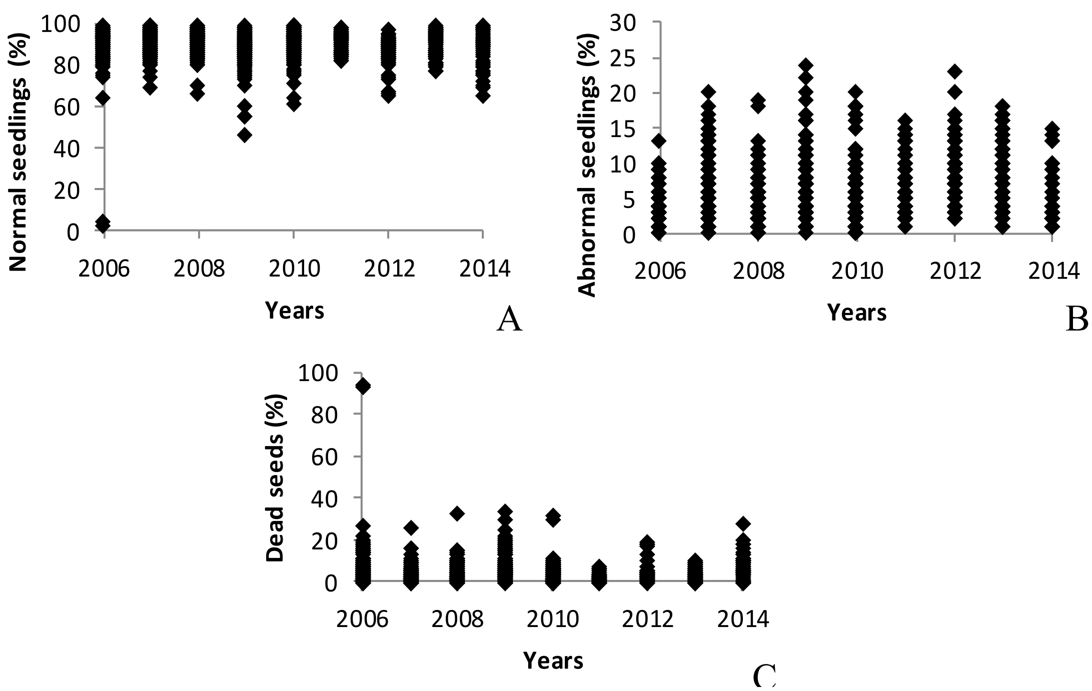


Figure 2. Scatter plots for normal seedlings (%), abnormal seedlings (%) and dead seeds (%) in samples of oats seeds, analyzed between 2006 and 2014, LAS / UNIJUI. Santa Maria, RS, 2016. A: Normal seedlings (%); B: Abnormal seedlings (%); C: Dead seeds (%).

because the seed producers can manage, harvest and process the seeds, making batches with better physiological standards.

Whereas own use seeds probably receive lower levels of investment in crops management, minor care in harvesting and processing. For both categories of seeds in the years 2012 and 2014, lower results were obtained in terms of physiological quality, due to adverse environmental conditions.

In Figure 3, it is presented the scatter plots for normal

Table 4. Descriptive statistics (ED), number of samples (N), mean (x̅), Standard Deviation (SD), maximum (Mmax), minimum (Mmin) to normal seedlings (%) abnormal seedlings (%) dead seeds (%) in own use seeds in black oat analyzed between 2006 and 2014, LAS / UNIJUI. Santa Maria, RS, 2016.

Variable	ED	Years/germination own use seeds									
		2006	**2007**	**2008**	**2009**	**2010**	**2011**	**2012**	**2013**	**2014**	**x̅**
Normal seedlings (%)	N	44	44	25	47	55	39	40	38	25	39.7
	x̅	83.9	87.4	84.9	83.1	82.9	84.2	79.7	86.4	76.5	83.2
	DP	15.4	13.3	18.1	12.4	15.1	13.8	20.0	10.1	20.2	15.4
	Mmáx	99	100	98	98	98	98	98	98	97	98.2
	Mmin	41	44	39	38	6	35	8	57	6	26.2
Abnormal seedlings (%)	x̅	4.3	4.9	5.4	6.7	7.3	7.6	9.0	6.9	8.5	5.9
	DP	3.6	4.6	4.2	6.2	5.8	7.0	5.3	4.9	5.7	5.3
	Mmáx	15	17	15	31	19	41	29	23	22	23.5
	Mmin	0	0	1	1	1	1	2	1	2	1.0
Dead Seeds (%)	x̅	11.8	7.7	10.0	10.1	9.7	8.2	11.3	6.6	14.9	10.0
	DP	14.2	12.0	14.5	9.7	11.9	10.5	12.2	6.8	18.7	12.3
	Mmáx	57	54	47	55	75	57	85	24	88	60.2
	Mmin	0	0	0	0	0	0	0	0	1	0.1

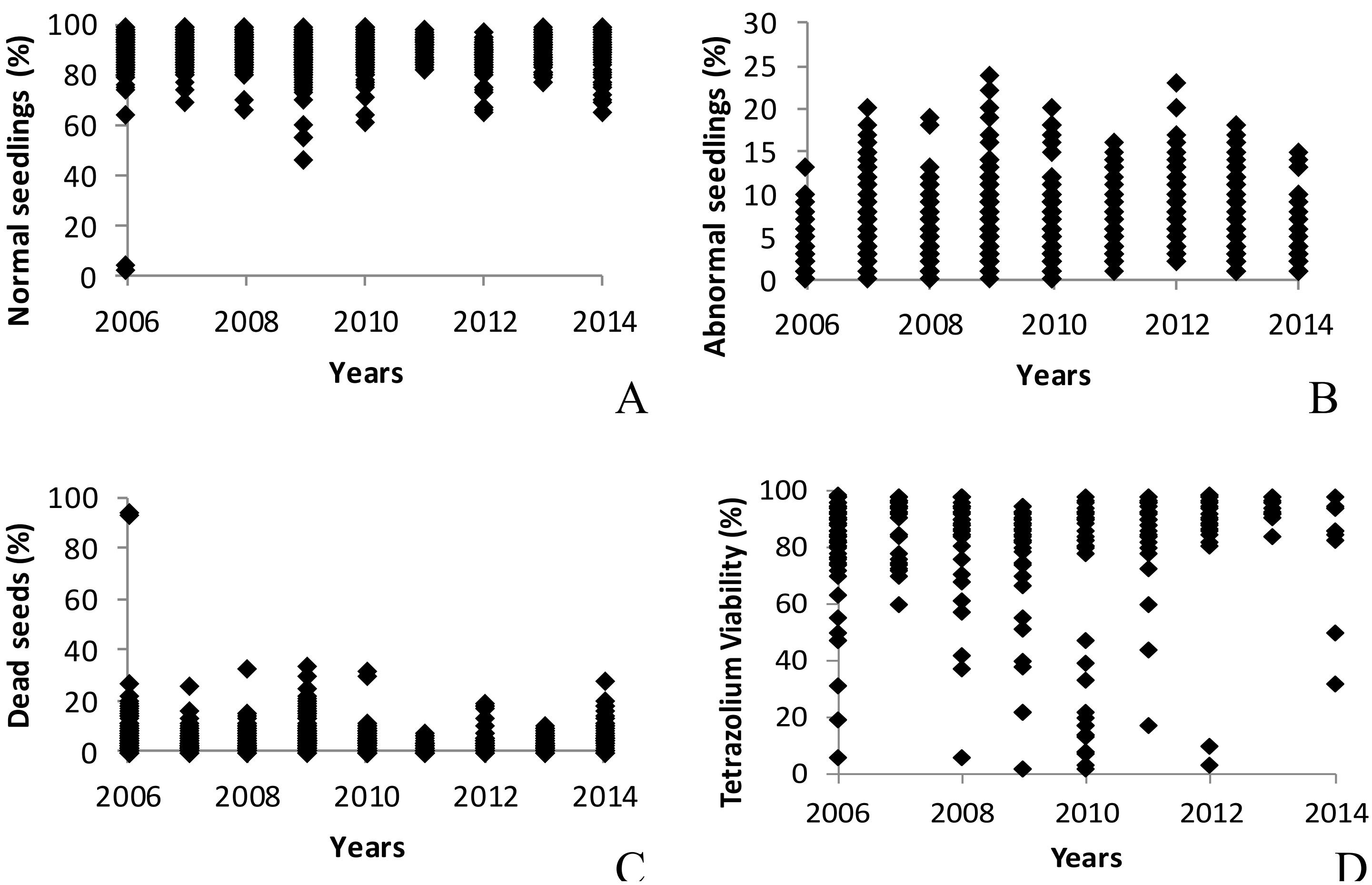


Figure 3. Normal seedlings for scatter plots (%), abnormal seedlings (%) and dead seeds (%) in samples of own use seeds and viability (%) of seeds evaluated by the tetrazolium test in black oat seeds analyzed between 2006-2014, LAS / UNIJUI. Santa Maria, RS, 2016. A, Normal seedlings; B, abnormal seedlings; C, dead seeds; D, tetrazolium viability.

Table 5. Descriptive statistics (ED), number of samples (N), mean (x̄), standard deviation (SD), maximum (Mmax), Minimum (Mmin) to viable seeds through tetrazolium test in black oat seeds analyzed between 2006 and 2014, LAS / UNIJUI. Santa Maria, RS, 2016.

Variable	ED	Years/viability seeds through tetrazolium test									
		2006	2007	2008	2009	2010	2011	2012	2013	2014	x̄
Viability (%)	N	84	42	42	46	38	31	20	11	10	36.0
	x̄	83.1	89.9	84.3	75.9	60.7	85.3	83.3	93.4	81.4	81.9
	DP	16.5	9.5	18.9	23.9	36.4	17.1	26.7	4.0	22.3	19.5
	Mmáx	99.0	98.0	98.0	95.0	98.0	98.0	99.0	98.0	98.0	97.9
	Mmin	6.0	60.0	6.0	2.0	2.0	17.0	3.0	84.0	32.0	23.5

Table 6. Percentage of samples classified as below the minimum standard required by the legislation in oats seeds analyzed between 2006 and 2014, LAS / UNIJUI. Santa Maria, RS, 2016.

Variable	Seeds S1;S2		Own use		Tetrazolium	
	2006\10	2011\14	2006\10	2011/14	2006\10	2011/14
Physical putity (%)	0.0	0.0	-	-	-	-
Other seeds (%)	1.8	0.0	-	-	-	-
Others cultured species	3.1	14.0	-	-	-	-
Wild species	0.0	0.0	-	-	-	-
Harmful tolerated	0.0	14.5	-	-	-	-
Harmful Prohibited	*	0.0	-	-	-	-
Germination (%)	0.7	3.8	18.1	31.7	19.4	12.5
Total evaluated samples	1738	491	215	142	252	72

-, Test not performed; *Only one non-standard sample.

seedlings, abnormal seedlings and dead seeds in samples of own use seeds and viability (%) of seeds evaluated by the tetrazolium test.

As for the own use seeds, there is less dispersion of results for the variable normal seedlings in the years 2008, 2011 and 2013, being considered the best years in terms of physiological quality. However, a greater dispersion of data occurred for this indicator in 2006, 2009, 2012 and 2014.

In terms of abnormal seedlings, data point to the years 2009 and 2012 with higher dispersions, meaning loss of physiological quality. In terms of dead seeds, the data dispersions were higher in the years 2006, 2010, 2012 and 2014.

The results of the samples analyzed by the tetrazolium test are shown in Table 5. The average viability was 81.9%. In 2010, the average viability was only 60.7% for a 80% minimum standard, according to Normative Instruction No. 33/2010 (Ministry, 2010). In this year, there were significant number of samples with low levels of viability, probably due to the introduction of a practice of pre-harvest desiccation that interfered with the seeds germination (Figure 3D). The highest average was 93.4%, obtained in 2013, considered the best year for this category of seeds.

The standard deviation was the highest in terms of physiological quality, with 19.5. This means that there was great variability in the samples analyzed, which can be seen by observing the dispersion shown in Figure 3 D, in which the years 2006, 2008, 2009 and 2010 stood out. It should be noted that the tetrazolium test is widely accepted and used for previous analysis of seed samples produced according to the national seed system, as well as for own use seed evaluation, to be an effective test for the rapid assessment of the viability of oats seeds (Souza et al., 2009).

The results of the percentage of samples classified as below the minimum standard required by the legislation in black oat seeds, both for seeds obtained in the seed production process, identified as seeds, as well as proper use of seeds and seeds analyzed through the tetrazolium test are shown in Table 6.

In Table 6, it can be observed that no sample was below the standard for physical purity of the seeds, that is, all samples were within the standards required by law, with a minimum of 95% by 2010 and 97% based on Instruction No. 33/2010 (Ministry, 2010).

From 2006 to 2010, 1.8% of the samples were below the standard for variable percentage of other seeds, that is, 98.2% of the samples were within the required standards (Table 6). In 2011 to 2014, no sample was approved in this parameter which indicates also that the seeds showed a high degree of physical purity.

From 2006 to 2010, 3.1% of the samples were rejected for the presence of other seeds from cultivated species, while from 2011 to 2014, 14% of the samples was rejected (Table 6). The elevation of disapproval percentage in the last period is due in part to the inclusion of this analysis in the determination of other seeds by number, in the sample of 500 grams (before it was performed the purity sample in 100 g), and reduction 50 to 16 seeds as limit from other seeds of cultivated species present, as stated in Instruction No. 33/2010 for oat (Ministry, 2010).

No sample was outside the standards for the presence of seeds of wild species over the nine years of study.

From 2006 to 2010, no sample was approved for the presence of seeds of harmful tolerated species; however, from 2011 to 2014, 14.5% of the samples were exposed. There was increased failure percentage in the last period, due, in part, to a reduction from 40 to six seeds as a limit on the sample of 500 g and the inclusion of wild species in harmful tolerated ratio, *A. barbata* L., based on Instruction No. 33/2010 for oat (Ministry, 2010).

The physical purity parameters indicate high degrees of purity of the samples analyzed, except for the number of cultivated species of seed and the number of seeds of harmful tolerated species reached levels of 14 and 14.4%, respectively, from 2011 to 2014. The increase in disapproval level of samples occurred largely because of the changes in the patterns introduced by the Normative Instruction No. 33/2010 (Ministry ..., 2010).

In Table 6, it is observed that only 0.7% of the samples did not meet the standard for germination from 2006 to 2010, and 3.8% were below the minimum from 2011 to 2014. These data point to high levels of physiological quality of the seeds analyzed, and the increase of seeds that have not reached the standard, was due, in part, to raising the minimum germination from 75% to 80%, as determined by the Normative Instruction No. 33/2010 (MINISTRY, 2010).

The data obtained for the analysis of seeds obtained in the seed production process indicate high physical and physiological quality conditions of the desired plant population per area (Krzyzanowski, 2013; Marcos-Filho, 2013).

The index of samples that did not reach the minimum germination standard in own use seeds was 18.1% from 2006 to 2010 and increased to 31.7% from 2011 to 2014 (Table 6). That is, close to one third of the samples analyzed for the purpose of own use did not provide the standard of viability to be used for sowing.

For the samples analyzed by the tetrazolium test, 19.4% were below the standard in the years from 2006 to 2010, decreasing to 12.5% from 2011 to 2014. In this test, prior analyses are carried out in the seed production process and/ or own use. This probably explains the high data samples below standard, although there was a reduction in the last period, unlike what happened with the own used seeds.

The changes introduced by legislation with regard to increasing the minimum percentage of germination and the decrease in the number of seeds of other cultivated species and the number of seeds of harmful tolerated species increased considerably the number of samples approved.

The years 2009, 2012 and 2014 suffered negative effect from meteorological factors of maximum temperatures and rainfall being that intense rainfalls which occurred during the vegetative growth favor leaching and nutrient losses and heavy rains during the physiological maturity and harvest interfere negatively on physiological seed quality. This is because harsh environmental conditions may change the preferential metabolic drain, wherein the vegetative plant parts and roots become preferred drains to the detriment of reproductive organs or seeds, affecting the yield and quality of seeds (Pedó et al., 2013). In addition to environmental conditions, the composition of the seeds can affect the quality thereof, as identified in soybean seeds by Castro et al. (2016), who find that seeds that had higher lignin content in the seed coat had lower percentages of damage from moisture and better physiological qualities.

The physiological attributes were enhanced when it comes to performance in 2008, 2010 and 2011, due to the levels of rainfall and appropriate temperatures for their vegetative development, physiological maturity and harvest (Figure 4).

Conclusion

The black oat seeds produced between 2006 and 2010, according to the National Seed System, had high levels of physical and physiological quality. However, 14.0 and 14.5% of the samples exhibited seeds levels from other cultivated species and harmful prohibited species above the standards, respectively, from 2011 to 2014. Own used seeds showed wide variability in results, with 18.1 and 31.7% of samples below standard for germination from 2006 to 2010 and 2011 to 2014, respectively, while the samples analyzed through the tetrazolium test showed disapproval levels of 19.4 and 12.5%, respectively.

The major physiological qualities were obtained in 2008, 2010 and 2011 and the lowest in 2009, 2012 and 2014. It is noteworthy that the seed quality is related to the years with levels of rainfall and temperatures appropriate for the vegetative development, physiological maturity and harvest.

CONFLICTS OF INTERESTS

The authors have not declared any conflict of interests.

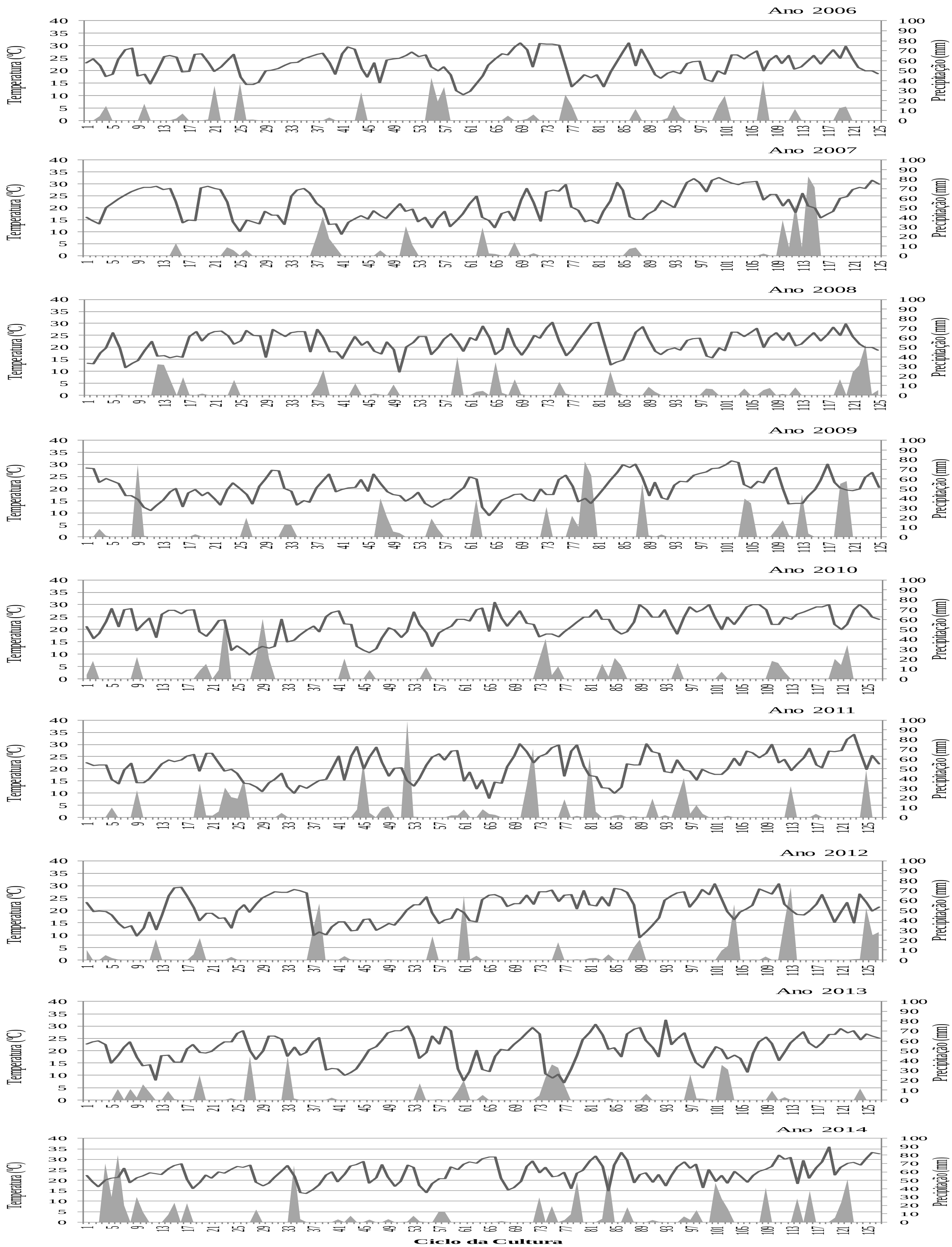


Figure 4. Maximum temperature (°C) and daily precipitation (mm) occurred during the crop cycle of black oat (Avena strigosa Schreb.), From June to September in the years from 2006 to 2014, Weather Station, Regional Institute of Rural Development Department of Agrarian Studies, UNIJUÍ, Augusto Pestana, RS, Santa Maria, in 2016.

REFERENCES

ABRASEM (2014). Anuário 2014: Estatística da produção e comercialização de sementes no Brasil. ABRASEM, Brasília.

Bortolini CG, Silva PRF da, Argenta G (200). Efeito de resíduos de plantas jovens de aveia preta em cobertura de solo no crescimento inicial do milho. Pesquisa Agropecuária Gaúcha 6(1):83-88.

Brazil (1997). Lei nº 9.456: Institui a Lei de Proteção de Cultivares e dá outras providências. Brasília: Presidência da República, 25/4/1997.

Brazil (2003). Lei n^{o} 10.711: Dispõe sobre o Sistema Nacional de Sementes e Mudas e dá outras providências. Brasília: Presidência da República, 5/8/2003.

Brazil (1992). Ministério da Agricultura, Pecuária e Abastecimento. Regras para análise de Sementes. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Brasília, DF: Mapa.

Brazil (2009). Ministério da Agricultura, Pecuária e Abastecimento. Regras para análise de sementes. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Brasília, DF: Mapa; ACS, 2009.

Carvalho NM, Nakagawa J (2012). Sementes: ciência, tecnologia e produção. 5. ed. Jaboticabal: Funep.

Carvalho PC de F, Santos DT de, Gonçalves EN, Moraes A de, Nabinger C (2010). Forrageiras de Clima Temperado. In: Fonseca DM. de, Martuscello JA. Plantas forrageiras. Viçosa: UFV pp. 494-537.

Castro E de M, Oliveira JA, Lima AE de, Santos HO dos, Barbosa JIL (2016). Physiological quality of soybean seeds produced under artificial rain in the pre-harvesting period. Journal of Seed Science, Viçosa, ABRATES 38(1):14-21.

Cruz CD (2013). GENES - a software package for analysis in experimental statistics and quantitative genetics. Acta Sci. 35(3):271-276.

Debiasi H, Martins JD, Missio EL (2007). Produtividade de grãos e componentes do rendimento da aveia preta (*Avena strigosa* Schreb.) afetados pela densidade e velocidade de semeadura. Ciência Rural, Santa Maria 37(3):649-655.

Del Duca L de JA, Fontaneli RS, Dalla Lana B, Nascimento Junior A do, Cunha GR da, Rodrigues O, Guarienti EM, Miranda MZ de, Costamilan LM, Chaves MS, Lima MIPM (2004). Experimentação de trigo e outros cereais de inverno para duplo propósito no Rio Grande do Sul, em 2003. Documentos Online 41. Passo Fundo: Embrapa Trigo 21p.

html. (Embrapa Trigo. Documentos Online; 41). Disponível em: http://www.cnpt.embrapa.br/biblio/do/p_do41.htm.

Flaresso JA, Gross CD, Almeida DX (2001). de Época e Densidade de Semeadura de Aveia Preta (Avena strigosa Schreb.) e Azevém (Lolium multiflorum Lam.) no Alto Vale do Itajaí, Santa Catarina. Rev. Bras. de Zootecnia 30(6S):1969-1974.

Fonseca MG da, Maia M de S, Lucca-Filho OA (1999). Avaliação da qualidade de sementes de azevém-anual (*Lolium multiflorum* Lam.) produzidas no Rio Grande do Sul. Revista Brasileira de Sementes, Campinas 21(1):101-106.

Grzybowski CR de S, Farinácio R, Panobianco M (2015). Reduction in the period for evaluation of the physiological quality of newly harvested black oat seeds. J. Seed Sci. Viçosa, ABRATES 37(4):244-248.

Krzyzanowski FC (2013). Controle de qualidade e a produção de sementes de alta qualidade. Informativo ABRATES 23(2):17.

Maia MS (2013). A pesquisa em sementes de espécies forrageiras de clima temperado e subtropical no Brasil. Informativo ABRATES 23(2):38.

Marcos-Filho J (2013). Importância do potencial fisiológico da semente de soja. Informativo ABRATES 23(1):21-24.

Medeiros RB de (1987). Introdução. Efeitos das pastagens nas rotações agrícolas. In: Dhein, R. A.; Carbonera, R.; Medeiros, R. B. de. Resultados de experimentação e pesquisa*:* Centro de Treinamento Cotrijuí – CTC – 1976-1986. Ijuí: Cotrijuí.

Medeiros RB de (1976). Formação e manejo de pastagens para a Região do Planalto Médio e Missões*.* Porto Alegre: Secretaria da Agricultura do RS.

Mello J (2013). Vantagens competitivas e comparativas da produção de leite no RS. Cruz Alta: CCGL.

Ministério Da Agricultura (1998). Pecuária E Abastecimento. Mapa. Portaria nº 381, de 05/08/1998.

Ministério Da Agriculturan (2010). Pecuária E Abastecimento. Mapa. Instrução Normativa nº 33, de 4/11/2010.

Nakagawa J, Cavariani C, Castro MM (2004). Armazenamento de sementes de aveia preta produzidas em solos de diferentes fertilidades. Rev. Bras. de Sementes, Londrina 26(2):07-14.

Ohlson O de C, Souza CR de, Panobianco M (2008). Levantamento da qualidade de sementes de azevém comercializadas no Estado do Paraná. Informativo Abrates, Londrina 18(1,2,3):18-22.

Ohlson O de C, Souza CR de, Panobianco M (2010). Qualidade física e fisiológica de sementes de capim-colonião e milheto, comercializadas no Estado do Paraná. Informativo Abrates*,* Londrina 20(1,2):30-36.

Ohlson OC, Grzybowski CRS, Da Silva BA, Nogueira JL, Panobianco M (2011a). Análise exploratória de dados: qualidade de sementes de azevém comercializadas no Estado do Paraná. Informativo Abrates, Londrina 21(3)47-51.

Ohlson OC, Grzybowski CRS, Da Silva BA, Nogueira JL, Panobianco M (2011b). Informações sobre a qualidade de sementes de *Brachiaria brizantha* comercializadas no Estado do Paraná. Informativo Abrates, Londrina 21(3):52-56.

Oliveira AL, Moreira AA, Sampaio GV, Oliveira EAS, Santos ES, Dourado VV (2013). Produção de sementes em comunidades tradicionais da agricultura familiar. In: Schuch LOB, Vieira JF, Rufino C de A, Júnior J de SA. Sementes: produção, qualidade e inovações tecnológicas. Pelotas: Editora e Gráfica Universitária pp. 483-495.

Parmejiani RS, Silva RB da, Mello R de A (2014). Qualidade física e fisiológica de sementes de forrageiras comercializadas no estado de Rondônia: safra 2012/2013. Informativo Abrates*,* Londrina 24(3):112-117.

Pereira S (2013). Sistema de produção de sementes de espécies forrageiras tropicais na visão da Sulpasto: pesquisa, comercialização e fiscalização. In: Informativo Abrates 23(2):40.

Peske ST, Lucca Filho OL, Barros ACS (2006). Sementes: fundamentos científicos e tecnológicos. 2. ed. Pelotas: Ed. Universitária UFPel.

Pedó T, Martinazzo EG, Aumonde TZ, Villela FA (2013). Princípios fisiológicos na produção de sementes. In: Schuch L. OB, Vieira JF, Rufino C de A, Júnior J de SA. Sementes: produção, qualidade e inovações tecnológicas. Pelotas: Editora Gráfica Universitária.

Rio Grande Do Sul (2014). Comissão de Sementes e Mudas. Catálogo de produtores de sementes e mudas do Rio Grande do Sul 2013/2014. Passo Fundo: Comissão de Sementes e Mudas do RS.

Souza CR de, Ohlson O de C, Panobianco M (2009). Avaliação da viabilidade de sementes de aveia preta pelo teste de tetrazólio. Revista Brasileira de Sementes*,* Londrina. 31(3):057-062.

Souza JM, Medeiros RB, Zambra JEG, Gieseler P, Guth O (1992). Introdução e avaliação preliminar de gramíneas tropicais e subtropicais no Noroeste do Rio Grande do Sul. In: Carbonera R, Pereira TFP, Severo, JLP, Viau LVM. Pesquisa no Centro de Treinamento Cotrijuí. Ijuí: Cotrijuí pp. 143-156.

Trennepohl D (2011). Avaliação de potencialidades econômicas para o desenvolvimento regional. Ijuí: Ed. Unijuí.

academicJournals

Vol. 12(7), pp. 524-534, 16 February, 2017
DOI: 10.5897/AJAR2016.12068
Article Number: 4A6831462774
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Artificial neural networks in whole-stand level modeling of *Eucalyptus* plants

Mirella Basileu de Oliveira Lima, Ilvan Medeiros Lustosa Junior, Elian Meneses Oliveira, Jéssica Cristina Barbosa Ferreira, Kálita Luis Soares and Eder Pereira Miguel*

Department of Forest Engineering, University of Brasília, Brasília, DF, 70910-900, Brazil.

Received 12 December, 2016; Accepted 19 January, 2017

Forestry production is traditionally predicted using mathematical modelling, where whole-stand models are prominent for providing estimates of growth and production per unit area. However, there is a need to perform research that adopts innovative tools, such as artificial Intelligence techniques. The objective of this study was to train and evaluate the efficiency of Artificial Neural Networks (ANN) in the modeling process of growth and production of Whole-Stand Level, in "equineanean" forests of the *Eucalyptus* genus clones. For the training of the networks, the supervised method was adopted. There were 100 networks trained, of which, for each output variable, 5 networks were selected. The criteria used to verify the quality of the training and validation of the networks were: Student's test "T", graphical analysis of the dispersion of residues, standard error of the relative estimate (Syx%), Pearson's correlation coefficient (*r*) between the observed and estimated values, and aggregate difference (AD). The ANN selected in the training process, which estimated the volume variables, site index and future production, when validated did not differ statistically by the Test "T" and presented adequate statistical accuracy values to the modeling process, with satisfactory correlation values (*r*) between the observed and estimated values, low values of standard error of the relative estimate (Syx%) and aggregate difference (AD). Artificial neural networks of the multilayer perceptron type are precise and efficient in the entire modeling process for whole-stand level of *Eucalyptus* plantations, therefore its use is recommended.

Key words: Artificial intelligence, prognosis, site classification, Brazil.

INTRODUCTION

Planted forests correspond to 7% of the world's total forest areas, having increased by more than 110 million hectares since 1990 (FAO, 2015). With only 7.74 million hectares, corresponding to 0.9% of the national territory, the Brazilian sector of planted trees accounts for 91% of all the wood produced for industrial purposes in the country - the remaining 9% comes from native forests legally managed.

Eucalyptus is the most planted tree in the tropics (Epron et al., 2013), in addition of being the main genus

*Corresponding author. miguelederpereira@gmail.com. Tel: +55 61 3107-5629.

cultivated in Brazil. Within this scenario, based on the year 2014, eucalyptus plantations occupied 5.56 million hectares in the country, representing 71.9% of the total area of planted trees (Ibá, 2015).

Soil and climatic characteristics of Brazil provide ideal conditions for the growth of species belonging to the genus *Eucalyptus*. The productivity of these stands in Brazil is ten times higher than the forest production of countries like Finland, Portugal, the United States and even Australia, the country of origin of the species (Cib, 2016).

As a result of the potentiality of the genus in question, the focus of forest management is to provide quality timber products, in an efficient manner, by estimating the dendrometric parameters and the area in which they are inserted (Curto et al., 2016). The study of forest growth and production has, as the most widespread technique, the adjustment of linear and nonlinear regression models. Among the applications of the models, it is worth mentioning the recommendation of management regimes, by species and site, besides making possible the planning of the harvest and elaboration of the supply plan, starting with the prognosis of the production (Acerbi et al., 2002).

The production of “equineanean” or “inequineanean” forests is predicted using models that describe natural dynamics and simulates production over time, considering different management and exploitation regimes (Vanclay, 1994). The forest growth and production models can be classified according to the level of detail of the information in: diametric distribution models (DDM), individual tree level models (ITM) and stand level models (SLM), also called global models (Campos and Leite, 2013).

Stand level modeling provides estimates of growth and production per unit area, serves efficiently most modelers, and is currently the most used category by Brazilian forest companies (Azevedo et al., 2016; Campos and Leite, 2013; Castro et al., 2013).

Generally associated to SLM, Clutter's model (1963) is the most used by forestry companies. In this model, the production is obtained according to the initial basal area, initial stand age, site index and future age. It can be a conventional production model for current conditions, when the future age is equal to the current age, and at the same time a model of projection or growth for future conditions (Santana et al., 2005).

The use of regression models is consolidated in forest modeling, however, there is a need to carry out research that adopts innovative tools and increases the accuracy of estimates, reducing processing difficulties and minimizing costs. Based on this presumption, artificial intelligence techniques, such as Artificial Neural Networks (ANN), have been increasing their importance in the forest environment.

Artificial Neural Networks (ANN), an interconnected set of simple elements, units or nodes, called artificial neurons, whose functionality is based on the biological neuron, are able to solve complex problems, being characterized by adaptive learning, capacity for self-organization, and efficient learning (Gurney, 1997). These networks are tolerant to discrepant data or outliers; they can model different variables and their non-linear relationships, and enable modeling with quantitative and qualitative variables (Kuvendziev et al., 2014, Özçelik et al., 2010).

In the last years, ANN have gained prominence in the forest environment, with applications in the estimation of the trees volume (Binoti et al., 2014; Miguel et al., 2016; Silva et al., 2009) growth and production (Binoti et al., 2015), *taper* (Diamantopoulou, 2005; Leite et al., 2010), individual tree volume (Castro et al., 2013), total tree height (Binoti et al., 2013), and diametric distribution (Binoti et al., 2012), however further studies are still needed on the subject.

Based on this assumption, the objective was to train and evaluate the efficiency of artificial neural networks (ANN) in the modeling of growth and production of whole-stand level in “equineanean” forests of *Eucalyptus* genus clones.

MATERIALS AND METHODS

The data used are from the continuous forest inventory (CFI) carried out in a clonal stand of *Eucalyptus urophylla*, located in the municipality of Niquelândia, State of Goiás. The soil of the region is classified as Dystrophic Red-Yellow Latosol (Embrapa, 2013) and the predominant climate, according to the Köppen classification, is Aw type, with an annual average temperature of 25ºC and precipitation of 1700 mm.

The CFI was performed in 21 permanent plots of 500 m² with ages of 3, 4, 5 and 6 years. The spacing between trees was 3.1 × 2 m having as obtained variables the diameter with a bark height up to1.30 m above ground (DBH) of all trees with DBH greater than 4 cm; a total height (Ht) of the trees and dominant height (Hdom), considering the average of the heights of the five trees of greater diameter of each plot (Assmann, 1970).

With data of the average Hdom, per plot and age, ANNs were tested (Table 1) to generate site index curves by the guide curve method. Then, site classification was performed, adopting 6 years as reference age and with amplitude between classes of 2 m. In the rigorous counting process, 36, 60, 67 and 62 trees were slaughtered at the ages of 3, 4, 5 and 6 years, respectively. The individual volume (m³/tree) was obtained according to the methodology proposed by Smalian (Husch et al., 1982).

In the training of the networks for the different stand variables, during the modeling process, the supervised method was adopted, where input and output variables were indicated for the networks. This method is a *feed forward* type and uses the unidirectional data flow algorithm, without cycles (Haykin, 2001).

Initially, the weights of all the networks were randomly generated (Heaton, 2011). Consecutively the individual update value evolved during the learning process, based on the error function.

For the cubing data, ANNs were generated to estimate individual volume with bark, in which the input and output variables are shown in Table 1. For the network training process, 70% of the database was used, and other 30% was used for its validation. Since the validation process was employed for the ANNs it resulted in a better statistics precision.

Table 1. Description of the network configurations adopted in the training of ANNs.

Parameter	Type of network	Input variable	Output variable	Nº of tested neurons	Activation function (input and output)	Nº of trained networks	Nº of validated networks
Volume	MLP	DBH (cm); Ht (m)	Volume (m^3)	1 to 9	Logistic	100	5
Dominant height	MLP	Age (years)	Hdom (m)	1 to 6	Logistic	100	5
Prognosis	MLP	$1/S_1$; $1/I_2$; I_1/I_2*LnG_1; $1-I_1/I_2$; S_1*1-I_1/I_2	Ln(V2) (m^3 ha^{-1})	1 to 11	Logistic	100	5

Where: MLP = Multilayer Perceptron; DBH = Breast height diameter; Ht = Total height; I1 = current age; I2 = future age; S1 = site index; G1 = basal area; Hdom = mean of dominant heights per plot; V2= volume referring to age II.

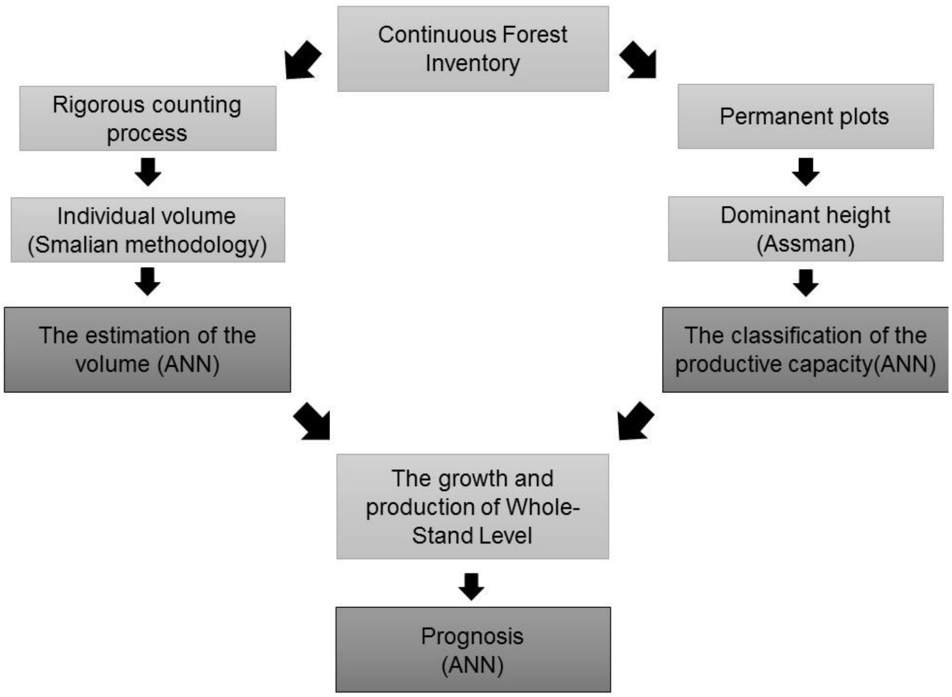


Figure 1. Flowchart of the methodology used in the modelling of whole-stand level.

Statistica software version 8.1 was used (Statsoft Inc, 2011) in all processes involving ANN. There were tested 24 different network configurations, all with Multilayer Perceptron (MLP) architecture. For the volume estimation, 12 network configurations were tested, with input variables DBH and Ht, and as the output variable or volume (V); other 12 configurations for the classification of the productive capacity of the site where age (I), the input variable and dominant height (Hdom), and the output variable (Table 1).

In total, 100 networks were trained, of which 5 networks were selected for each output variable. This selection was based on the standard error of the relative estimate (Syx%), Pearson's correlation coefficient between the observed and estimated values, the graphs of the observed variable relationship and predicted variable, percentage residues and classes of errors.

The network training persevered until the error rate was reduced to an acceptable range between the predicted values and the actual values provided to the network, known as the delta rule, or until reaching the maximum number of times or cycles (Shiblee et al., 2010). The adopted learning rate was 0.001, being this parameter able to assume values in the range of 0 to 1, whose main objective is to maintain the stability of the process by minimizing errors.

With the data obtained in the estimation of the volume and the site classification, the ANN technique was again applied to evaluate the projection of growth and production at the whole-stand level. The input variables were site index (S), current age (I1), future age (I2), stand basal area at current age (B1), and stand basal area at future age (B2); and the stand volume at future age (V2) was defined as the output variable. A flowchart of the whole process can be visualized in Figure 1.

Regarding the architecture of the trained neural networks, the number of neurons in the hidden layer varied according to the synaptic weights established by the intelligent problem solver tool. The activation function was logistic and the training algorithm was the resilient propagation (Riedmiller and Braun, 1993).

For the validation process of ANNs, the "t" test obtained 5% of significance between the observed and estimated values, all the three graphics, standard error of the relative estimate (Equation 1), Pearson's correlation coefficient (Equation 2) and the aggregate

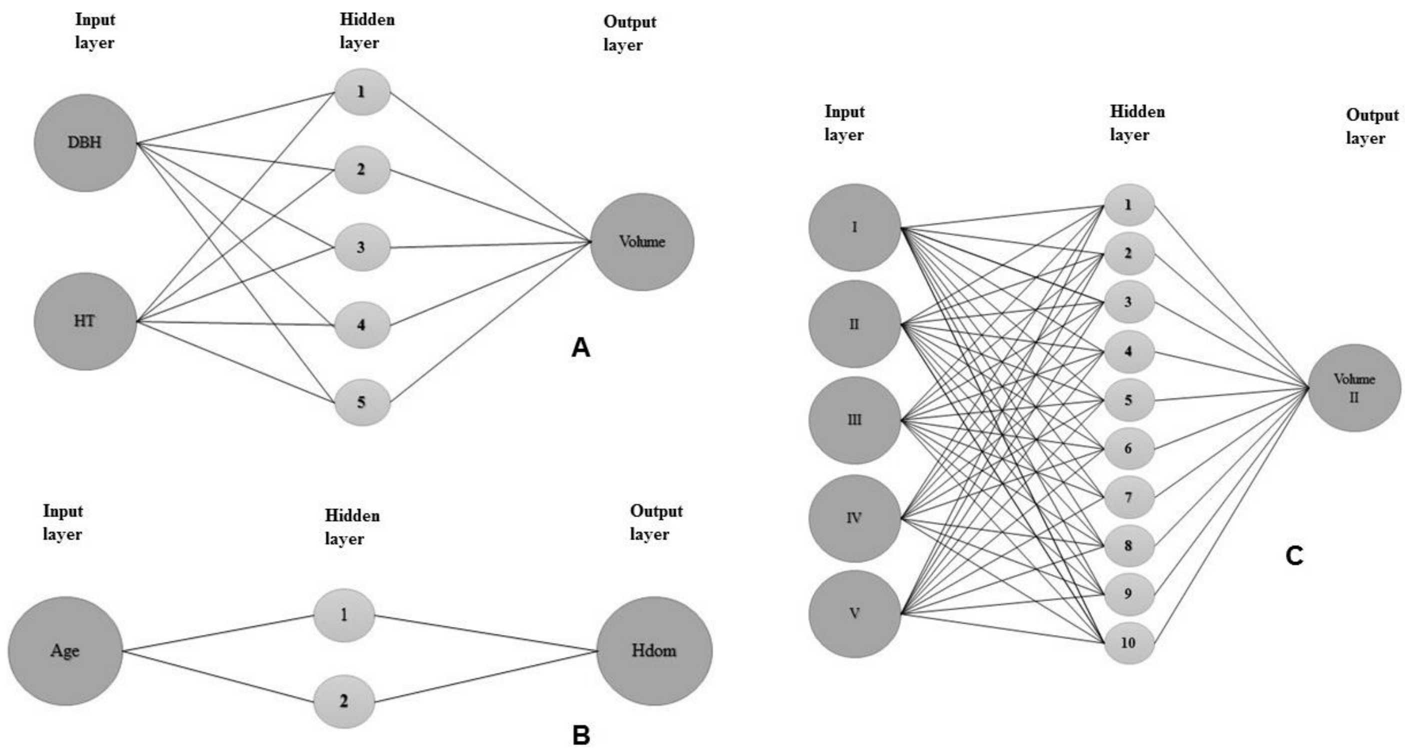


Figure 2. ANNs' architectures selected for validation. I = 1/site index; II = 1/future age; III = (current age/future age) *Ln (basal area); IV = 1- (current age/future age); V = site index* (1- (current age/future age)).

and percentage difference (Equation 3).

$$\boldsymbol{Sxy}(\%) = \mathbf{100} * \sum_{i=1}^{n} \frac{Y_i - \hat{Y}_i}{Y_i} \quad (1)$$

$$r = \frac{Cov\,(Y_i, \hat{Y}_i)}{\sqrt{V(Y_i)V(\hat{Y}_i)}} \quad (2)$$

$$DA(\%) = 100 * \left(\frac{\sum_{i=1}^{n} Y_i - \sum_{i=1}^{n} \hat{Y}_i}{\sum_{i=1}^{n} Y_i}\right) \quad (3)$$

Where, Y_i = variable observed, $\hat{Y}_i$ = variable estimated, V = variance; Cov = coefficient of variation.

RESULTS AND DISCUSSION

Volume estimates

The five best ANN recollected in the training process, which estimated the volume variables, site index and future production, presented adherence to the data and statistics of appropriate adjustment and precision to the modeling process, with satisfactory correlation values (r) between the observed and estimated values, and low values of standard error of the estimate (Syx%).

The ANNs' architectures that presented the best quality statistics, both for volume (A), site classification (B) and prognosis of production (C), are presented in Figure 2.

The volume variable, when estimated by ANN2, behaved appropriately according to the observed data once it presented slightly higher statistics, both in the training process and in the validation of ANNs (Table 2).

The aggregate difference (Da) is a statistical parameter used as an adjustment index for models. It is the difference between the sum of the observed values and the sum of the estimated values, therefore, it is used as an indicator criterion of under and overestimates, that can also be obtained in percentage form for better visualization (Leal et al., 2015).

For the data used in the network validation phase the DA% resulted in low and positive values, varying from 2.19 to 2.22 indicating that the networks showed a behavior to estimate volume values ($m^3.ha^{-1}$) slightly lower to the real values, and that can be used effectively in estimating the individual volume of *Eucalyptus* trees, not differing statistically (t 0.095857 <0.05). Miguel et al. (2015) found similar results when testing ANNs, in which the aggregate difference presented positive values for total wood volume and stem volume (3.41 and 8.58%, respectively).

In the graphical analysis of the residues and the correlation between observed and estimated ANN2 values, there was little difference in the quality of the trained and validated network, both obeying the randomization of errors. As observed in the frequency histogram of percentage errors of ANN2 estimates, 73% of the data were wrong in the range of -10 to 10% when

Table 2. Precision characteristics and statistics of the five best neural networks selected for volume estimation (m³/ha).

ANN	Architecture			Training		Validation		
	Input	Hidden	Output	*r*	Syx%	r	Syx%	DA%
1	2	5	1	0.9934	8.50	0.9823	12.93	2.220
2	2	7	1	0.9934	8.48	0.9824	12.90	2.202
3	2	8	1	0.9933	8.53	0.9821	12.96	2.213
4	2	8	1	0.9933	8.54	0.9821	12.99	2.222
5	2	5	1	0.9933	8.54	0.9822	12.94	2.195

submitted to training, while in the validation process for the same interval the data missed 63% (Figure 3).

Considering the results found in the present work, it can be verified that the artificial neural networks are efficient to estimate the individual volume variable in *Eucalyptus* forest stands, also corroborated by the studies developed by Silva et al. (2009), Binoti et al. (2015) and Miguel et al. (2016).

Site classification

The dominant height (Hdom) is the variable that has the least influence in response to silvicultural treatments, and that best expresses the quality or productive capacity of the place, besides showing a significant correlation with the total volume production in the stand (Machado and Figueiredo Filho, 2006).

In order to estimate this dendrometric variable, the five neural networks recollected also presented good precision statistics, where ANN4 stood out with slightly higher estimates and greater simplicity in its architecture - number of neurons in the layers (Table 3). According to Bullinaria (2016), they should be preferred, as long as they are suitable for training the database, in order to avoid over fitting, when the network memorizes the training data, and optimize the training time of the network.

The graphical analysis of the residues and the correlation between the observed and estimated values indicated a slight overestimation of the dominant heights at the age of 4 years and a stabilization of the residual errors in the posterior ages. According to the histogram of percentage frequencies of the percentage errors of ANN4 estimates, most of the errors were concentrated between 0 and 5% in the network training, proving the efficiency of the method (Figure 4).

The classification by site indexes (site), determined by the average dominant height of the stands at a specific age (age-index), is considered a practical, quantitative and consistent method for assessing site quality, since all environmental factors are reflected in an interactive way in height growth, which is directly related to volume (Campos and Leite, 2013).

In this sense, the productive capacity of the settlement was established in three classes, according to the sites indexes generated based on the dominant mean heights estimated by the artificial neural networks (Table 4). In the present study, the classification of productive units was considered satisfactory and superior to that found by Consenza et al. (2015), since it was not necessary to add other input variables as soil type and spacing, facilitating the classification process of the productive units.

The local index curves (Figure 5) were obtained by the guide curve method, which maintains a constant growth rate for height independent of the site (Scolforo, 2006).

Prognosis of forest production

In the evaluation of the prognosis of forest production, through the quality statistics of the recollected five artificial neural networks, where the correlation (r), the standard error of the estimate (Syx%) and the graphs of observed and predicted values, residues and histogram of errors, resulted in adequate values. Despite having the same architectural configuration (numbers of input, hidden and output layers), ANN3 generated slightly higher statistics in the training process, in contrast, in the validation process of ANNs, ANN2 presented the lowest standard error of the estimate (Syx%), as well as low value in the aggregate difference (DA%), being this a priority statistic when compared to the correlation (Table 5). These low values found in Syx% and DA% as well as the non-significant result of the "t" test (0.1044> 0.05) between observed and predicted values. However, all the selected networks show the adhesion in the projection of the volume of wood in *Eucalyptus* stands.

Compact and homogeneous residues, and distribution of errors in class intervals close to zero, are desirable in the validation process independent of the modeling technique, since they demonstrate the predictive capacity of the models. As shown in the training, the ANN presented efficiency in the projection of the volume of wood per unit area, demonstrating adherence, homogeneity and compaction of residues. In addition, approximately 90% of the errors were concentrated between -5 and 5% for training and validation of data, which proves the efficiency in the use of artificial intelligence for this purpose (Figure 6).

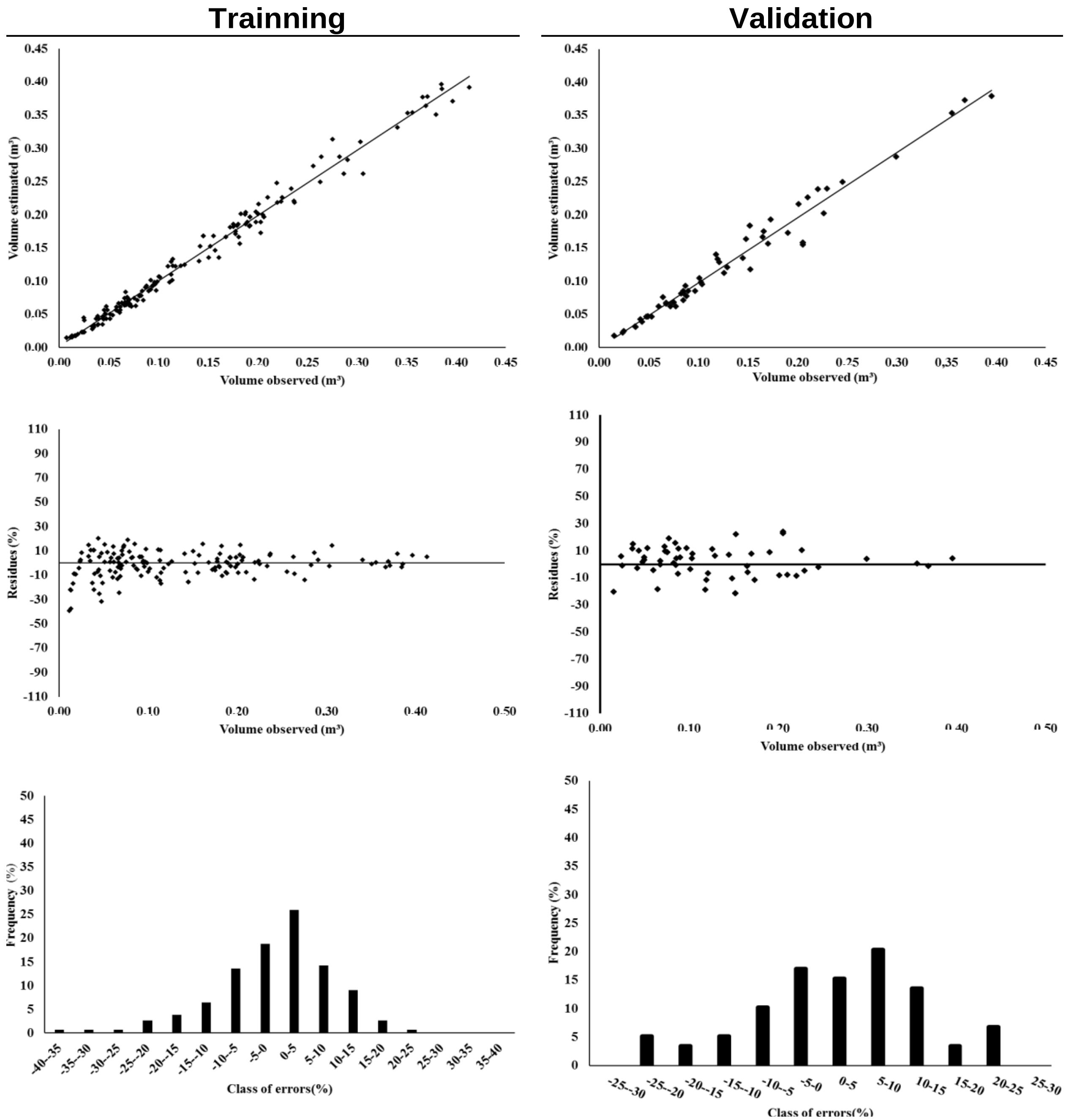


Figure 3. The relationship between observed and estimated values of volume, dispersion of residues and histogram of dispersion of errors, of training and validation, by the ANN2.

Table 3. Precision characteristics and statistics of the five best neural networks selected for estimating dominant heights (m).

ANN	Architecture			Training	
	Input	Hidden	Output	*r*	Syx%
1	1	6	1	0.9728	4.36
2	1	3	1	0.9729	4.35
3	1	8	1	0.9729	4.35
4	1	2	1	0.9729	4.35
5	1	7	1	0.9729	4.35

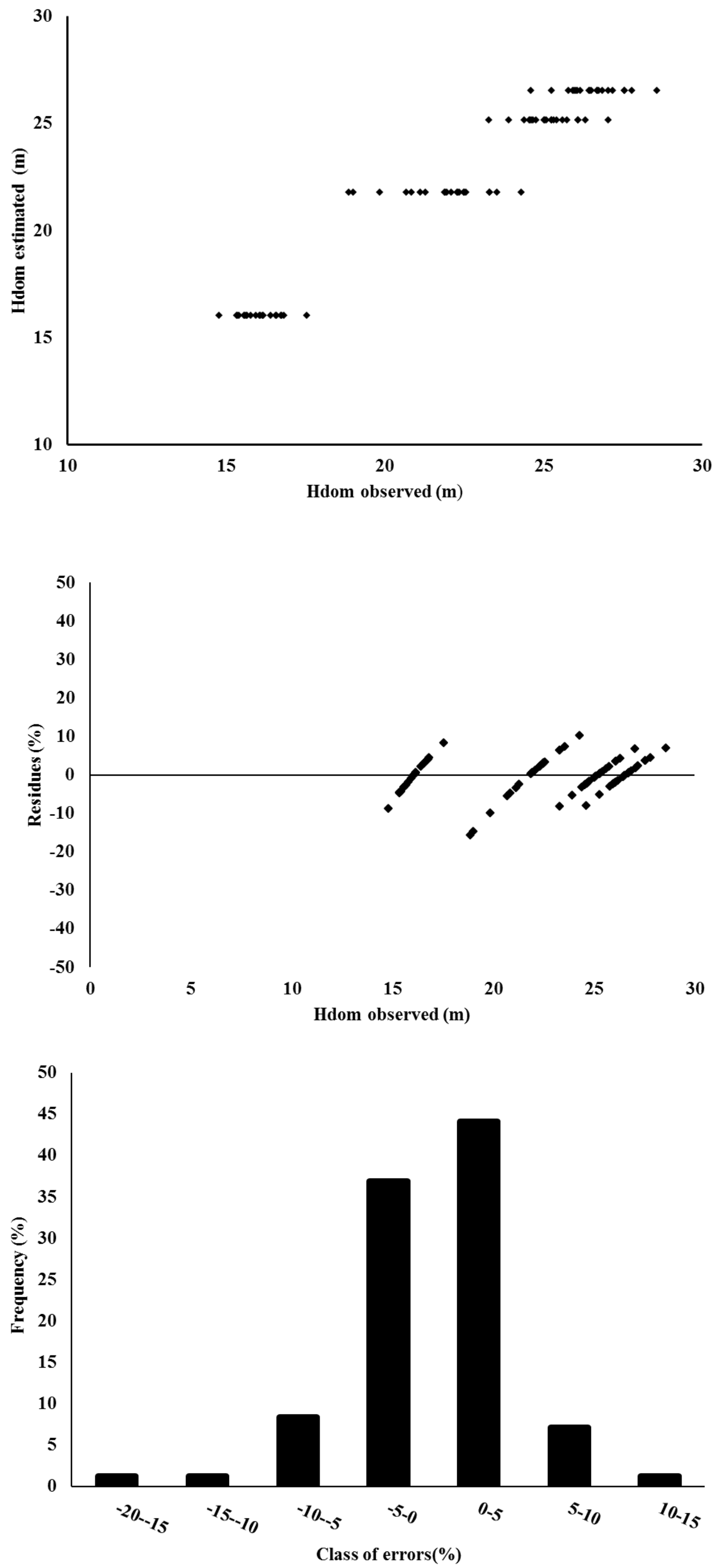


Figure 4. Graph of the relationship between observed and estimated values of dominant height (Hdom), dispersion of residues and histogram of dispersion of errors, of the ANN4 training.

Table 4. Classification of sites by the ANN4.

Site classes	Intervals of Hdom (m) classes	Site indexes
I	30│---28	29
II	28│---26	27
III	26│---24	25

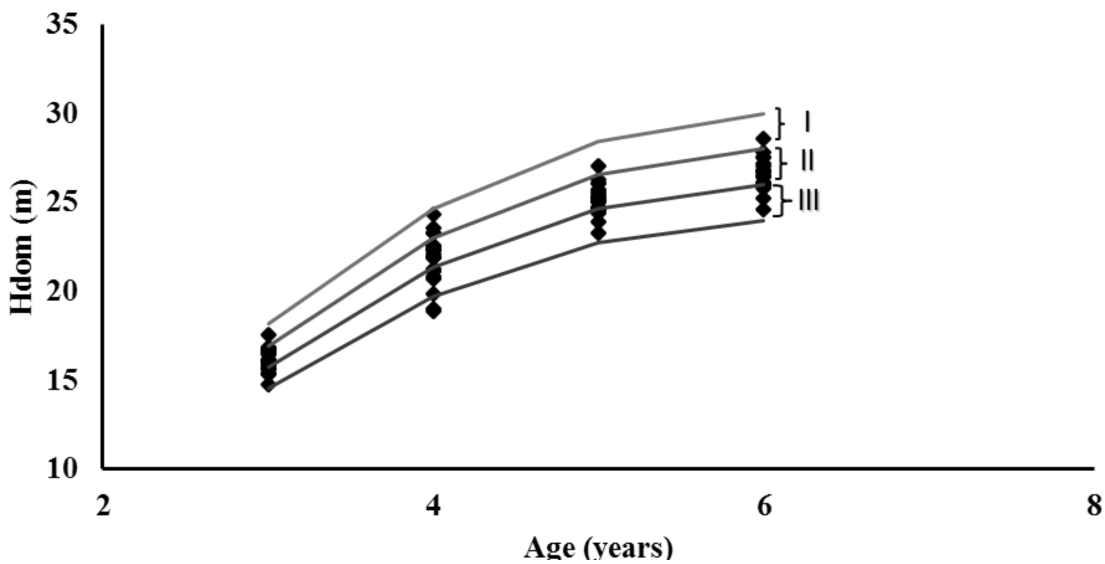


Figure 5. Site index curves generated by the ANN4 from the guide curve method.

Table 5. Precision statistics and characteristics of the five best neural networks selected for projection of stand production.

ANN	Architecture			Training		Validation		
	Input	Hidden	Output	*r*	Syx%	*r*	Syx%	DA%
1	5	3	1	0.9863	5.31	0.9823	3.63	2.70
2	5	10	1	0.9946	3.36	0.9897	3.11	2.20
3	5	10	1	0.9949	3.25	0.9908	3.18	2.22
4	5	5	1	0.9946	3.34	0.9898	3.24	2.21
5	5	10	1	0.9944	3.41	0.9903	3.20	2.24

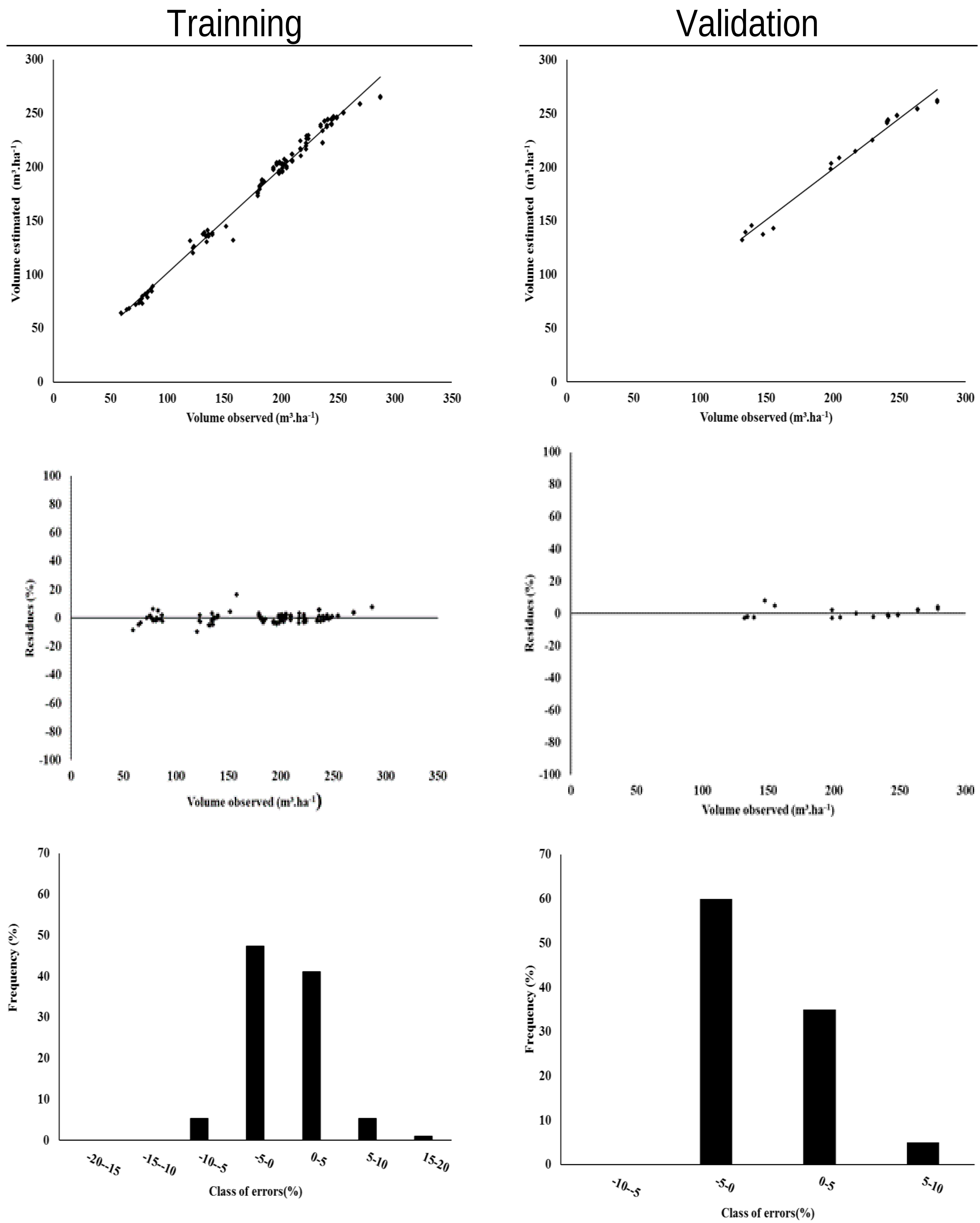
It should be emphasized that the results obtained in this research are specific to the local conditions of the stand; however, new research should be done addressing other aspects related to the genus *Eucalyptus*, as well as different architectural networks configurations using this technique. As a suggestion, the insertion of different clones, spacing, and edaphic characteristics as categorical variables in the input layer is an interesting alternative, and may result in a single ANN capable of accurately modeling a future volume projection for a range of species of the genus *Eucalyptus*.

Conclusion

Artificial neural networks of the multilayer perceptron type are precise and efficient in the entire modeling process for whole-stand level of Eucalyptus plantations, so their use is recommended.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.


Trainning
Validation
Volume estimated (m³.ha⁻¹)
Volume observed (m³.ha⁻¹)
Residues (%)
Frequency (%)
Class of errors(%)
-20--15
-15--10
-10--5
-5-0
0-5
5-10
15-20

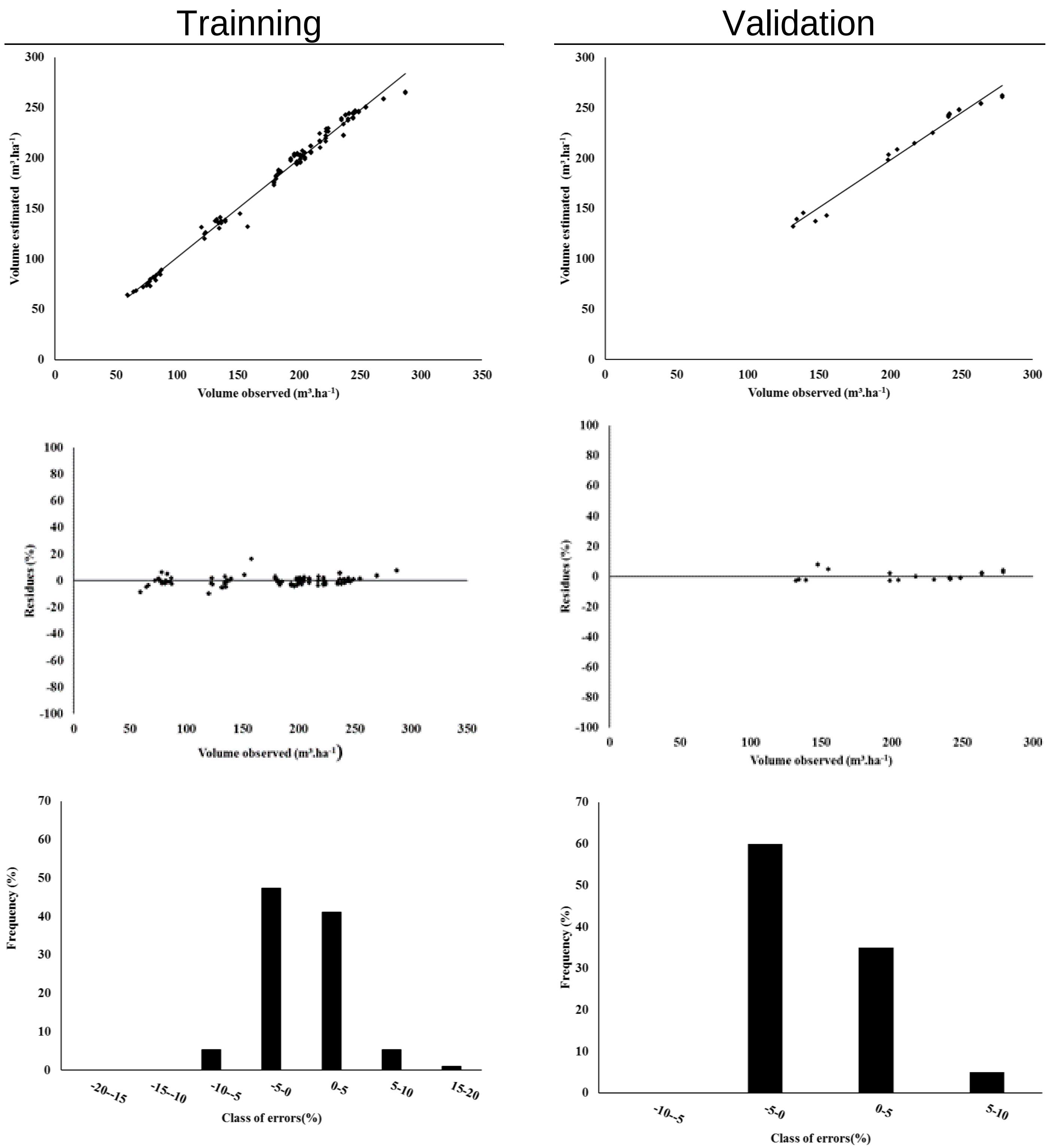


Figure 6. Relationship between observed and estimated values of volume, dispersion of residues and histogram of dispersion of errors, of training and validation, by ANN2.

REFERENCES

Assmann E (1970). The principles of forest yield study. Oxford: Pergamon Press 506 p.

Acerbi JRFW, Scolforo JRS, Oliveira AD, Maestri R (2002). Modelo para prognose do crescimento e da produção e análise econômica de regimes de manejo para *Pinus taeda* L. Rev. Árvore 26(6):699-713.

Azevedo GB, Oliveira EKB, Azevedo GTOS, Buchmann HM, Miguel EP, Rezende AV (2016). Modelagem da produção em nível de povoamento e por distribuição diamétrica em plantios de eucalipto. Sci. For. 44(110):383-392.

Binoti DHB, Binoti MLMS, Leite HG, Silva A (2013). Redução dos custos em inventário de povoamentos equiâneos. Agrária 8(1):125-129.

Binoti DHB, Binoti MLMS, Leite HG, Silva A, Santos ACA (2012). Modelagem da distribuição diamétrica em povoamentos de eucalipto submetidos a desbaste utilizando autômatos celulares. Rev. Árvore 36(5):931-939.

Binoti DHB, Binoti MLS, Leite HG (2014). Configuração de redes neurais artificiais para estimação do volume de árvores. Ciênc. Madeira 5(1):58-67.

Binoti MLMS, Leite HG, Binoti DHB, Gleriani JM (2015). Prognose em nível de povoamento de clones de eucalipto empregando redes neurais artificiais. Cerne 21(1):97-105.

Bullinaria JA (2016). Introduction to neural computation. Archieved at http://www.cs.bham.ac.uk/~jxb/inc.html.

Campos JCC, Leite HG (2013). Mensuração florestal: perguntas e respostas. 4ª edição. Viçosa: Editora UFV. 605 p.

Castro RVO, Soares CPB, Leite HG, Souza AL, Nogueira GS, Martins FB (2013). Individual growth model for *Eucalyptus* stands in Brazil using artificial neural network. ISRN Forestry 2(1):1-12.

Clutter JL (1963). Compatible growth and yield for loblolly pine. For. Sci. 9(3):354-371.

CIB–Conselho de Informações sobre Biotecnologia (2016). Guia do eucalipto. Oportunidades para um desenvolvimento sustentável. Archieved at http://cib.org.br/wp-content/uploads/2011/10/Guia_do_Eucalipto_junho_2008.pdf.

Consenza DN, Leite HG, Marcatti GE, Binoti DHB, Alcântara AEM, Rode R (2015). Classificação da capacidade produtiva de sítios florestais utilizando máquina de vetor de suporte e rede neural artificial. Sci. For. 43(108):955-963.

Curto RA, Mattos PP, Braz EM, Péllico Netto, S, Zachow, R (2016). Wood density of Araucaria angustifolia from overstocked strand. PFB. 36:51-59.

Diamantopoulou MJ (2005). Artificial neural networks as an alternative tool in pine bark volume estimation. Comput. Electronics Agric. 10(3):235-244.

Embrapa (2013). Sistema Brasileiro de Classificação de Solos. 3ª edição revisada e ampliada. Brasília: Embrapa, 353 p.

Epron D, Nouvellon Y, Mareschal L, Moreira RME, Koutika LS, Geneste B, Delgado-Rojas JS, Laclau JP, Sola G, Gonçalves JLDM, Bouillet JP (2013). Partitioning of net primary production in *Eucalyptus* and *Acacia* stands and in mixed-species plantations: Two case-studies in contrasting tropical environments. For. Ecol. Manage. 301(1):102-111.

Food and Agriculture Organization of the United Nations – FAO (2015). The Global Forest Resources Assessment. 1ª edição. Roma: CFRQ, 253p.

Husch B, Miller CI, Beers TE (1982). Forest mensuration. 3ª edição. New York: John Wiley & Sons. 402p.

Gurney K (1997). An Introduction to Neural Networks. Londres: UCL Press, 317 p.

Haykin S (2001). Redes neurais: princípios e prática. Porto Alegre: Bookman. 900p.

Heaton J (2011). Programming Neural Networks with Encog3 in Java. 2ª edição. St. Louis: Heat Research Incorporated. 240p.

Indústria Brasileira de Árvores – IBÁ (2015). Relatório IBÁ – 2015. Indicadores de desempenho do setor nacional de árvores plantadas referentes ao ano de 2014. Archieved at: http://www.iba.org/images/shared/iba_2015.pdf.

Kuvendziev AS, Lisichkova K, Zekovic Z, Marinkovski M (2014). Artificial neural network modelling of supercritical fluid CO_2 extraction of polyunsaturated fatty acids from common carp (*Cyprinus carpio* L.) viscera. J. Supercrit. Fluids 92(6):242-248.

Leal FA, Miguel EP, Matricardi EAT, Pereira RS (2015). Redes neurais artificiais na estimativa de volume em um plantio de eucalipto em função de fotografias hemisféricas e número de árvores. Rev. Bras. Biom. 33(2):233-249.

Leite HG, Silva MLM, Binoti DHB, Fardin L, Takizawa FH (2010). Estimation of inside-bark diameter and heartwood diameter for *Tectona grandis* Linn. trees using artificial neural networks. Eur. J. For. Res. 130 (2):263-269.

Machado AS, Figueiredo Filho A (2006). Dendrometria. 2ª edição. Guarapuava: Unicentro. 316 p.

Miguel EP, Mota FCM, Téo SJ, Nascimento RGM, Leal FA, Pereira RS, Rezende AV (2016). Artificial intelligence tools in predicting the volume of trees within a forest stand. Afr. J. Agric. Res. 11(21):1914-1923.

Miguel EP, Rezende AV, Leal FA, Matricardi EAT, Vale AT, Pereira RS (2015). Redes neurais artificiais para a modelagem do volume de madeira e biomassa do cerradão com dados de satélite. Pesq. Agropec. Bras. 50(9):829-839.

Özçelik R, Diamantopoulou MJ, Brooks JR, Wiant Jr.H (2010). Estimating tree bole volume using artificial neural network models for four species in Turkey. J. Environ. Manage. 91(3):742-753.

Riedmiller M, Braun H (1993). A direct adaptive method for faster backpropagation learning: therporp algorithm. Int. Conf. Neural Netw. 1(7):586-591.

Santana C, Mello AA, Eisfeld RL, Sanquetta CR (2005). Sistema de equações para simulação do crescimento e da produção em povoamentos de *Eucalyptus grandis* Hill ex Maiden. Sem desbaste baseado no modelo de Clutter. Ambiência 1(2):239-256.

Scolforo JRS (2006). Biometria Florestal: Modelos de crescimento e produção florestal. Lavras: UFLA/FAEPE. 393 p.

Shiblee MD, Chandra B, Kalra PK (2010). Learning of geometric mean neuron model using resilient propagation algorithm. Expert Syst. Appl. 37(12):7449-7455.

Silva MLM, Binoti DHB, Gleriani JM, Leite HG (2009). Ajuste do modelo de Schumacher e Hall e aplicação de redes neurais artificiais para estimar volume de árvores de eucalipto. Rev. Árvore 33(6):1133-1139.

Statsoft. STATISTICA (Sistema de Análise de Dados): versão 8.1, 2011. Archieved at http://www.statsoft.com.

Vanclay JK (1994). Modeling forest growth and yield: applications to mixed tropical forests. Copenhagen: CAB International. 312p.

academicJournals

Vol. 12(7), pp. 535-541, 16 February, 2017
DOI: 10.5897/AJAR2016.11607
Article Number: 08C829462773
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Agronomical and morphological diversity of the accessions of cassava in Central African Republic

Kosh-Komba, E.[1,2]*, Aba-Toumnou L.[1], Semballa, S.[1], Zinga, I.[1], Yandia, P.[1], Atato, A.[2], Kadekoy-Tigague, D.[4], Wabolou, F.[3], Kongbo Dembo, E.[3], Batawila, K.[2] and Akpagana, K.[2]

[1]Laboratory of Biological and Agronomical Sciences for Development, Faculty of Sciences, University of Bangui, Central African Republic.
[2]Laboratory of Botanical and Ecological Plant, Faculty of Sciences, University of Lome, Togo.
[3]High Institute of Rural Development, University of Bangui, Central African Republic.
[4]Central African Republic Institute of Agronomical Researches (ICRA), Bangui, Central African Republic.

Received 26 August, 2016; Accepted 20 October, 2016

Cassava (*Manihot esculenta* Crantz) is an important subsistence food crop in Central African Republic. Data collections for the agronomical and morphological characterization provided 59 accessions of Cassava and were subjected to multivariate analysis to discriminate groups of accessions according to their morphological features. The Eigenvalues of the principal axes extracted from the multivariate analysis indicated that the first two factors explained 7% of the total variability. Generally, morphological variation of the pool of accessions grown in different cassava production sites largely covers the overall variability and therefore there is no structuring in relation to agro-morphological characters from the sites. The dendrogram established on 59 accessions in relation to agronomical and morphological character is divided into two groups. Group A is the smallest with 12 accessions, while Group B has 47. Most of 59 accessions were different on all 44 descriptors. However, some accessions reported under different names, such as "ICRA and six months", "Boots and Assa", were identical on all the characters. In addition, some accessions collected in different places under one name, such as "Six months," have a likeness of all phenotypic traits.

Key words: Agronomical, cassava, diversity, morphological.

INTRODUCTION

Cassava (*Manihot esculenta* Crantz) (*Euphorbiaceae*) is the third largest source of carbohydrates for man in the world and is one of the most important crops in Africa (FAO, 2009). It is efficient in carbohydrate production, adapted to a wide range of environments and tolerant to drought and acidic soils (Fermont et al., 2007).

A sustainable agricultural system requires that components of diversity be used in a way and at a rate that will not lead to a long term decline of diversity, thus maintaining its potential to meet the needs and

*Corresponding author. E-mail: koshkomba2002@yahoo.fr.

aspirations of present and future generations (Alves, 2002). Genetic diversity is however threatened by the introduction and adoption of modern high yielding varieties (Kosh-Komba, 2013). A dramatic increase in the use of small number of highly selected accessions has led to loss of valuable genetic resources. The proportion of genetic diversity accessed by the popular varieties has often not been determined yet it is critical to the sustainable use of cassava genetic resources in CAR (Duval, 2008). Since cassava is predominantly vegetative propagated, over reliance on a few varieties which may also share a common ancestry may minimize the on farm diversity and thus increase the risks posed by such co-evolving biotic factors as pests and diseases to cassava farming (Fresco, 1986; Fauquet and Fargette, 1990).

Uncoordinated planting and lack of information related to genetic diversity of cassava is one of the factors for low quality cassava starch (Bellon, 1996). Comprehensive study related to various types of cassava in CAR, cultivated or out in the wild was infrequent. This research aims to elucidate the agronomical and morphological diversity of the accessions of cassava in CAR to derive an appropriate breeding strategy.

MATERIALS AND METHODS

Site description

The experimental plot for the agronomical and morphological characterization was located in the village Kapou (Figure 1). Kapou was chosen to represent a range of environments and management practices in cassava-based cropping systems in the mid-altitude zone of CAR. Main soils in the region include ferric and orthic Acrisols and orthic and haplic Ferralsols; soils that are derived from strongly weathered granite or sedimentary parent material (Boulvert, 1986). The climate in all sites is sub-humid with a bimodal rainfall distribution. This allows for the production of most annual crops during both the long (March-August) and the short rains (September-October). Altitude ranges between 1200 and 1500 masl. Cassava is planted in the first 2 months of the short or long rains and remains in the field for about a year. Agricultural systems are diverse with farmers growing 4-6 main crops on average (Conaway et al., 2012). In addition, Kapou is also Experimental Station of PRASAC project where all accessions of cassava in CAR are planted.

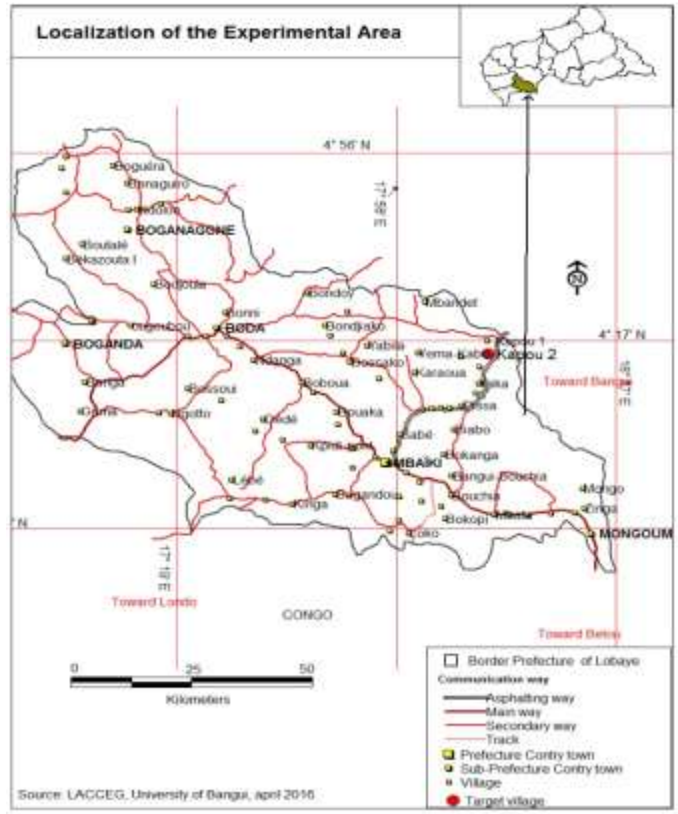


Figure 1. Location of the Kapou in Central African Republic.

Data collection

Data collection for the agronomical and morphological characterization of the accessions was made over a period of twelve months from 45 descriptors of cassava (Fukuda et al., 2010; Emperaire et al., 2003). The data were collected in four steps the third month after planting. First step of characterization had two (2) descriptors (3 months): the color of apical leaves; the pubescence of apical leaves.

Second step of characterization had thirteen (13) descriptors (6 months): leaf retention; the shape of the central leaves, the color of the petiole; the color of leaves; the number of lobes; the lobe length; the lobe width; the lobe of the margin; the length of the petiole; color of the midrib; orientation of the petiole; Flower; pollen. Third step of characterization had nine (9) descriptors (9 months): leaf scars; the color of the cortex of the shaft; the color of the skin of the stems; color of the outer shaft; the length of the internodes; the shape of the stem; the color of the branches of the adult plant; the length of stipules; margin of states.

Four step of characterization had 20 the descriptors (12 months): fruit; seed; plant height; branch level; plant habit; branching angle; form of the plant; number of tubers/plant; number of marketable tubers; length of root stalk; constriction of the root; form of tubers; external color of the tubers; color of the root pulp; color of the root cortex; cortex: fitness for peeling; texture of the epidermal root; taste roots; average weight of tubers.

Observations on the vegetative were made on a sample of cuttings of 10/20 (10 clones) set collection at each elementary plot. For each accession, cuttings were taken from a sample of three (3) clones to harvest in 12 months to calculate the average number of tubers and the average weight per accession.

Statistical analysis

The data was subjected to multivariate analysis to discriminate groups of accessions according to their morphological features using the software community analysis package Version 2.15 (Henderson and Seaby, 2002).

The factorial analysis of correspondence of morphological descriptors was conducted using the software Cap (Hill, 1979). This analysis project accessions on a plane whose axes are defined as new independent variables composites. Each axis (composite variable) is a combination of morphological descriptors weighted by their level of explanation of the overall variability of the system.

Table 1. Eigenvalues and variance percentage.

	A1	A2	A3	A4
Eigenvalues	0.0587	0.0195	0.0071	0.0048
Variance (%)	0.048	0.021	0.0015	0.0011
Cumulated (%)	0.048	0.042	0.0030	0.1

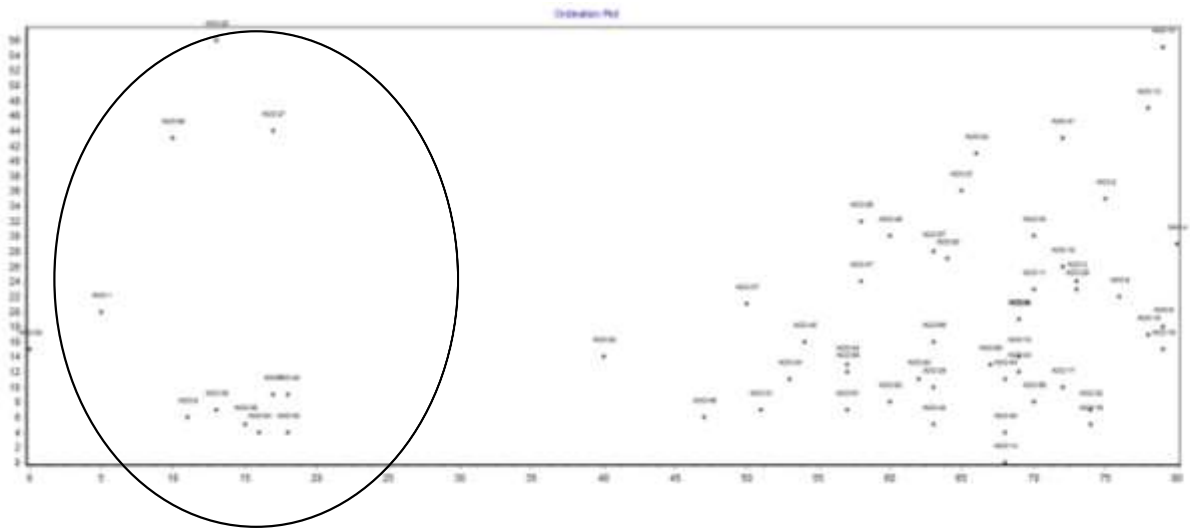


Figure 2. Graphic representation of the 59 accessions from the four axes of the factorial analysis of the correspondences gotten from the 45 morphological describers.

RESULTS

Typology of diversity of cassava

The Eigenvalues (Table 1) of the principal axes extracted from the multivariate analysis indicated that the first two factors explained 7% of the total variability. Generally, morphological variation of the pool of accessions grown in different cassava production sites largely covers the overall variability and therefore there is no structuring in relation to agro-morphological characters from the sites (Figure 2).

Cluster analysis

The dendrogram established on 59 accessions (Table 2) in relation to agro-morphological character is divided into two groups (Figure 3). Group A is the smallest with 12 accessions, while Group B has 47 accessions. Most of 59 accessions were different on all 44 descriptors. However, some accessions reported under different names, such as "ICRA and six months", "Boots and Assa" are identical on all the characters. In addition, some accessions collected in different places under one name, such as "Six months," have a likeness of all phenotypic traits.

Each group is characterized by a number of descriptor. Group A is distinguished by the color of apical leaves (gray and purple); sheets (light green); the length of which is short stipules; of the absence of fruit and seed; port of the plant which is erect and the cylindrical root form. Group B is characterized by the color of the petiole (green) of the main ridge (green). Note the presence of flowers, pollen, fruits and seeds. All other phenotypic traits used in this study have not formed discriminated groups are scattered on all pools.

Cassava variability for root yield traits

Twenty accessions have 7 to 8 root number (Figure 4). There was no correlation between the mean number and the mean weight of roots.

DISCUSSION

The Eigenvalues of the principal axes extracted from the multivariate analysis indicated that the first two factors explained 7% of the total variability. Generally, morphological variation of the pool of accessions grown in different cassava production sites largely covers the

Table 2. Local and gout of accessions.

Groups	Number of accessions	Accessions' local name	Bitter/sweet accessions
A	acc1	ASSA	Bitter
	acc35	B	Sweet
	acc55	Ligbia	Sweet
	acc5	Boumba	Sweet
	acc36	Andjete	Bitter
	acc54	JPN	Bitter
	acc7	Casano Nigeria	Sweet
	acc30	Zaoro-mbissé III	Bitter
	acc38	Babouche	Sweet
	acc40	Bamasson	Bitter
	acc27	Yambolo	Bitter
	acc28	Zaoro-mbissé	Bitter
B	acc2	ASSA	Bitter
	acc15	ICRA	Sweet
	acc9	Claire	Sweet
	acc12	Danzi	Bitter
	acc17	Icra rouge	Sweet
	acc43	Boda	Bitter
	acc14	Giodofondo	Bitter
	acc23	Rendre	Bitter
	acc56	Mondélépacko	Sweet
	acc16	Icra blanc	Sweet
	acc32	Zeteyabongo	Bitter
	acc19	Mboumba	Bitter
	acc39	Babouche	Bitter
	acc44	Cimetière	Bitter
	acc48	Gabon	Bitter
	acc52	Icra	Sweet
	acc50	Gozo-Bangui	Bitter
	acc53	JPN	Bitter
	acc59	Ombella	Bitter
	acc3	Bambari	Bitter
	acc8	Casano Nigeria	Sweet
	acc6	Bozizé	Bitter
	acc11	Claire	Sweet
	acc29	Zaoro-mbissé	Bitter
	acc26	Yaclaire	Bitter
	acc57	Nakowara	Bitter
	acc22	Pipom	Sweet
	acc31	Zaoro-mbissé III	Bitter
	acc49	Gabon	Bitter
	acc45	Cimetière	Bitter
	acc34	Aboundou	Sweet
	acc42	Batamolengué	Bitter
	acc51	Icra	Sweet
	acc58	Ombella	Bitter
	acc4	Bambari II	Bitter
	acc18	Kessembin	Bitter
	acc24	Rendre III	Bitter
	acc10	Claire III	Sweet

Table 2. Contd.

acc13	Garouaboulaye	Bitter
acc20	Mboumba (6 mois)	Sweet
acc25	Séssè	Bitter
acc46	Dongo	Bitter
acc21	Yinfin	Sweet
acc37	Adou	Bitter
acc47	Gabon	Bitter
acc33	Abandou	Sweet
acc41	Bambari	Bitter

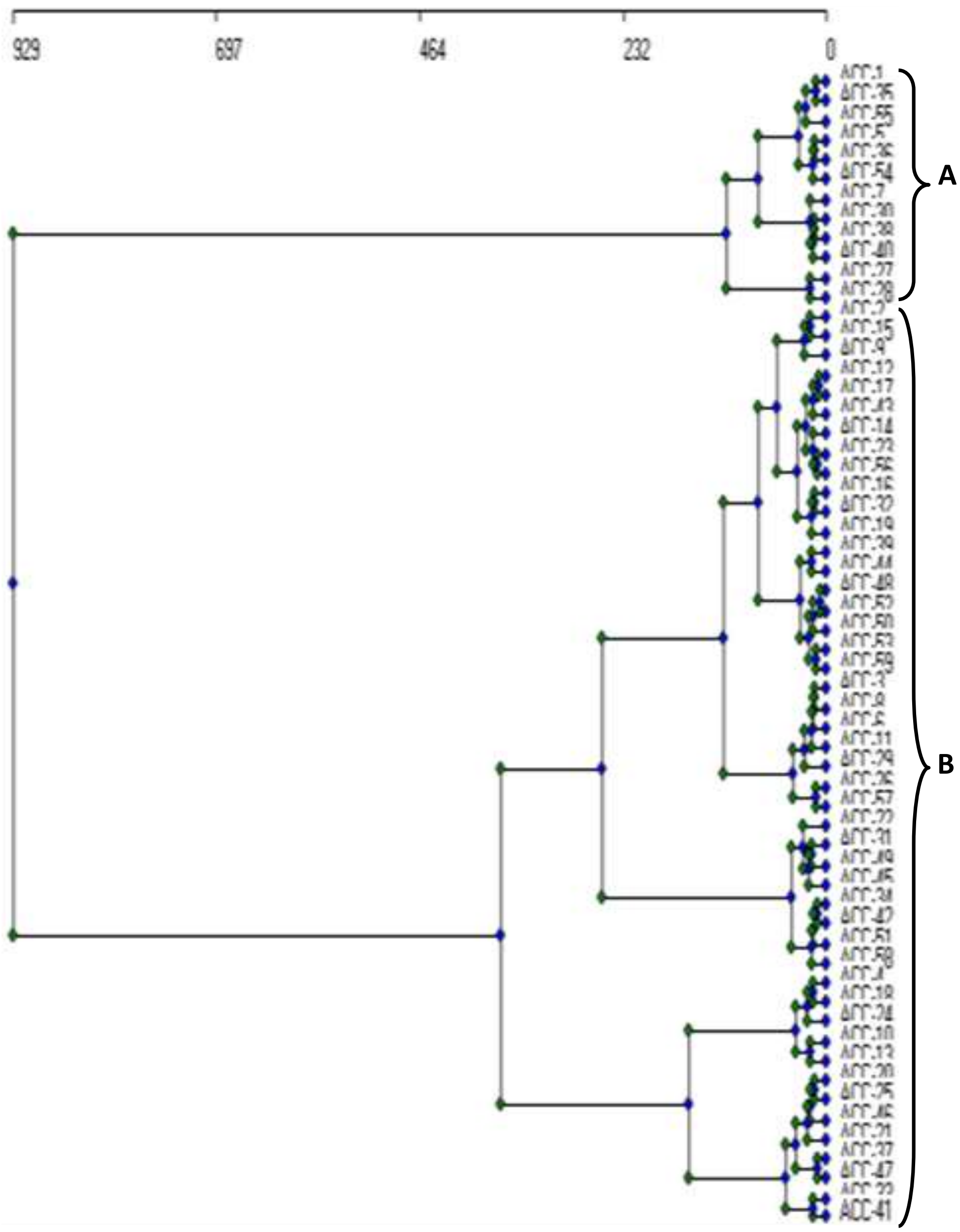


Figure 3. Dendrogram showing hierarchical accessions of 59 cassava phenotypes based on qualitative characters.

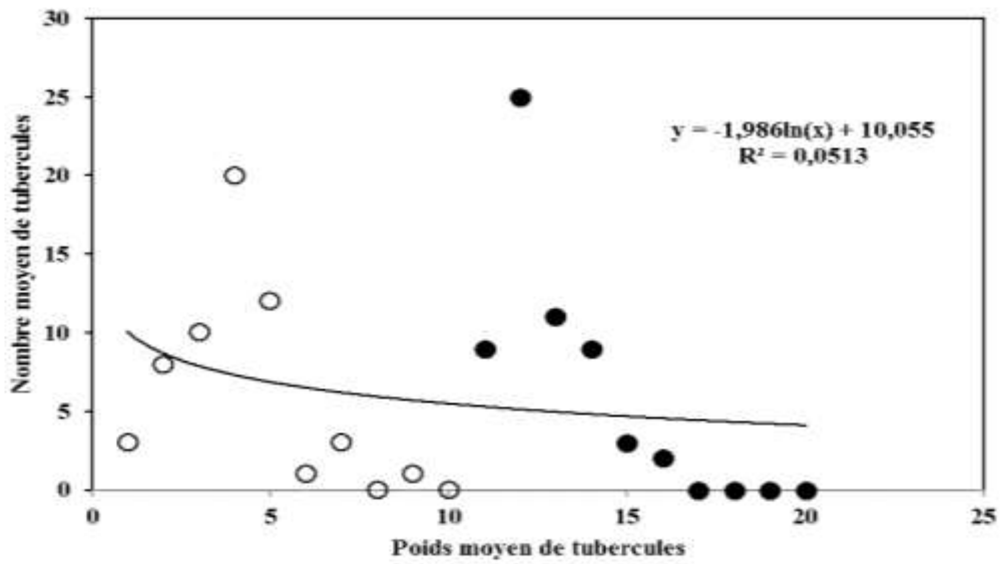


Figure 4. Cassava variability for root yield traits.

overall variability and therefore there is no structuring in relation to agro-morphological characters from the sites.

The dendrogram established on 59 accessions in relation to agronomical and morphological characters is divided into two groups (Figure 3). Group A is the smallest with 12 accessions, while Group B has 47 accessions. The 59 accessions are different on all 44 descriptors. However, some accessions reported under different names, such as "ICRA and six months", "Boots and Assa", are identical on all the characters. In addition, some accessions collected in different places under one name, such as "Six months," have a likeness of all phenotypic traits.

Each group is characterized by a number of descriptor. Group A is distinguished by the color of apical leaves (gray and purple); Sheets (light green); the length of which is short stipules; of the absence of fruit and seed; port of the plant which is erect and the cylindrical root form. Group B is characterized by the color of the petiole (green) of the main ridge (green). Note the presence of flowers, pollen, fruits and seeds. All other phenotypic traits used in this study did not form discriminated groups and are scattered on all pools.

However, similar study in agronomical and morphological diversity of cassava reported that the total diversity which was given by pubescence on apical leaves, petiole color, color of leaf vein, length of leaf lobe, ratio of lobe length to lobe width of central leaf lobe (Cooper et al., 1992; Alves, 2002).

An agronomical and morphological diversity of cassava is elucidated from pollen and flowers characters, from color of stem cortex, color of stem epidermis and color of stem exterior, from number of leaf lobe, presence of pollen and flowers character (Kosh-Komba, 2013; Asare, 2011).

In Indonesia, an accession 576 of cassava accession from Papua, an agronomical and morphological diversity elucidated that only five characters from nineteen characters give positive effect that is bristle in character, that is, pubescence on apical leaves, petiole color, color of leave vein, length of leaf lobe and ratio between width and length in central leaf lobe (IITA, 2005; Etudaiye, 2009). Based on the result of cluster analysis, cassava accession has diversity length of Euclidian length 1-17. The result of main component analysis shows a relatively high contribution value in 181 cassava accession based on nineteen morphological characters. The result of biplot analysis in 181 accession shows that the deployment of 181 cassava accession is very extensive, there are six groups in two quadrants which are formed relatively far between their groups. It shows that the potential of cassava in Indonesia has an extensive diversity considering the geographical condition of Indonesia that is quite extensive. A geographical effect naturally gives the diversity marker in cassava accession itself (Odoemenem and Otanwa, 2011; Adesehinwa et al., 2011).

An agronomical and morphological diversity of cassava was elucidated for accessions of Manihot low entropy for the descriptors: sinuosity of leaf lobe, flowering, pollen and leaf color developed. It should also be noted that, featuring cassava germplasm found low entropy for the following: stem growth habit, flowering, texture of the epidermis of the root and the root constriction (Nuwamanya et al., 2009; Asare et al., 2011).

Cassava germplasm has larger entropies for the external color of the stem, petiole color, shape and color of the central lobe of the apical leaf descriptors. In the study of *Manihot* germplasm, the largest entropies were found for petiole color, shape of the central lobe, outside color of stem and number of lobes descriptors (Emperaire et al., 2003; Singh, 1981).

Another work emphasized that the distribution of the variance is associated with the nature and number of characters used in the analysis and focuses on the first principal components used only with a few descriptors of agronomic interest or a group (plant, flowering, fruit and agronomic) (Pereira et al., 1992).

Conclusion

Two distinct grouping were made out of the 59 different cassava accessions based on their similarity level with respect to their agronomical and morphological characters. Group A is the smallest with 12 accessions, while Group B has 47 accessions. Generally, morphological variation of the pool of accessions grown in different cassava production sites largely covers the overall variability and therefore there is no structuring in relation to agro-morphological characters from the sites. The recommendation from this study is that since the study has succeeded in grouping 59 different cassava accessions into two groups, the farmers need to grow only two out of the 59 accessions of cassava, one from each group and have almost all the benefit of rowing all the 59 accessions at a time.

Information on accession diversity, morphology and agronomy, may be used as comprehensive database of local cassava diversity in CAR for further research.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENT

This work was funded by grants from the Regional Pole of Applied Research to Development Farming Systems (PRASAC).

REFERENCES

Adesehinwa OK, Obi OO, Makanjuola BA, Oluwole OO, Adesina MA (2011). Growing pigs fed cassava diet supplemented with or without farmazyme 3000 proenx: effect on growth, carcass and blood parameters. Afr. J. Biotechnol. 10(14):2791-2796.

Alves AAC (2002). Cassava botany and physiology. In: Hillocks, R.J., Thresh, J.M., Bellotti, A.C. (Eds.), Cassava, Biology, Production and Utilization. CABI Publishing, Wallingford.

Asare PA, Galyuon IKA, Sarfo JK, Tetteh JP (2011). Morphological and molecular based diversity studies of some cassava (*Manihot esculenta* Crantz.) germplasm in Ghana. Afr. J. Biotechnol. 10(63):13900-13908.

Bellon MR (1996). The dynamics of crops infraspefic diversity: A conceptual framework at the farmer level. Hum. Ecol. 50(1):26-39.

Boulvert Y (1986). Carte Phytogéographique de la République Centrafricaine (feuille Ouest- feuille Est) à 1:1000000. ORSTOM. 31 p.

Conaway JL, Ouedraogo AK, Coneff J (2012). Activité de zonage plus de moyens d'existence de la République centrafricaine. USAID (United States Agency International Development), Bangui, Centrafrique 41p.

Cooper D, Velve R, Hobbelink H (1992). Growing diversity - Genetics Resources and local food security. GRAIN, IT Publications, London 166 p.

Duval MF (2008). Appui à l'étude de la diversité du manioc en République Centrafricaine, CIRAD. 60 p.

Emperaire L, Santos Mühlen G, Fleury M, Robert T, Mckey D, Pujol D, Elias M (2003). Approche comparative de la diversité génétique et de la diversité morphologique des maniocs en Amazonie (Brésil et Guyanes). Actes du BRG 4:247-226.

Etudaiye HA, Nwabueze TU, Sanni LO (2009). Quality of *fufu* processed from Cassava Mosaic Disease (CMD) resistant varieties. Afr. J. Food Sci. 3(3):061-067.

FAO (2009). Perspectives de l'alimentation. Analyse des marchés mondiaux. Point de mire. Division du commerce international et des marchés (EST) 113 p.

Fauquet C, Fargette D (1990). African cassava mosaic virus: etiology, epidemiology, and control. Plant Dis. 74:404-411.

Fermont AM, Obiero HM, van Asten PJA, Baguma Y, Okwuosa E (2007). Improved cassava varieties increase the risk of soil nutrient mining: an exante analysis for western Kenya and Uganda. In: Bationo, A., Waswa, B., Kihara, J., Kimetu, J. (Eds.), Advances in Integrated Soil Fertility Management in sub- Saharan Africa: Challenges and Opportunities. Springer, Dordrecht pp. 511-520.

Fresco L (1986). Cassava in shifting cultivation: A systems approach to agricultural technology development in Africa. Royal Tropical Institute, Amsterdam 290 p.

Fukuda WMG, Guevara CL, Kawuki R, Ferguson MME (2010). Selected Morphological and Agronomic and Agronomic Descriptors for the Caracterization of Cassava. IITA, Abadan (Nigéria) 19 p.

Henderson PA, Seaby RMH (2002). Community Analysis Package. Version 2.15. Pisces Conservation LTD. WWW.irchouse.demon.co.uk.

Hill MO (1979). Decorana fortran program for detrented correspondence Analysis and reciprocal averaging.

IITA (2005). Growing cassava commercially in Nigeria. Cassava illustration guide book. International Institute of Tropical Agriculture, Ibadan, Nigeria. pp. 21-22.

Kosh-Komba E (2013). Gestion paysanne, diversité agromorphologique et génétique du manioc (*Manihot esculenta* Crantz, Euphorbiaceae) cultivé dans trois zones agroclimatiques en Centrafrique. Thèse, Université de Lomé, Togo, 136 p.

Nuwamanya E, Baguma Y, Emmambux N, Taylor J, Patrick R (2009). Physicochemical and functional characteristics of cassava Starch in Uganda varieties and their progenies. J. Plant Breed. Crop Sci. 2(1):001-011.

Odoemenem IU and Otanwa LB (2011). Economic analysis of cassava production in Benue State, Nigeria. Curr. Res. J. Soc. Sci. 3(5):406-411.

Pereira AV, Vencovsky R, Cruz CD (1992). Selection of agronomical and botanical descriptors for the characterization of cassava (Manihot esculenta Crantz.) germplasm. Braz. J. Genet. Ribeirão Preto 15:115-124.

Singh D (1981). The relative importance of characters affecting genetic divergence. Indian J. Genet. Plant Breed. New Delhi 41:237-245.

African Journal of Agricultural Research

Related Journals Published by Academic Journals

- *African Journal of Environmental Science & Technology*
- *Biotechnology & Molecular Biology Reviews*
- *African Journal of Biochemistry Research*
- *African Journal of Microbiology Research*
- *African Journal of Pure & Applied Chemistry*
- *African Journal of Food Science*
- *African Journal of Biotechnology*
- *African Journal of Pharmacy & Pharmacology*
- *African Journal of Plant Science*
- *Journal of Medicinal Plant Research*
- *International Journal of Physical Sciences*
- *Scientific Research and Essays*

academicJournals