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African Journal of Agricultural Research

Full Length Research Paper

Information technology applied to the process of traceability in the wheat supply chain

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The adoption of traceability systems in the food chain is a market differentiator for manufacturing firms. It is appreciated by consumers and is more and more common after the occurrence of certain problems related to food consumption. Legislation, quality standards and best practices now govern the traceability process. The aim of this paper is to address the main regulations that establish procedures for ensuring the safety of food in terms of traceability, and also to present the evolution of information technologies in this area. Although, the latter are still being developed, there are still many opportunities for growth and innovation. As a result, it is possible to see the commonalities between the models of traceability, and also to identify the points of the supply chain of wheat in which the processes are focused.

Key words: Traceability, quality management system, information technology, wheat.

INTRODUCTION

Brazilian market has increased its grain production, from 76 million tons to approximately 184 million tons from 1993 to 2013, respectively. About wheat production, this country is one of the greatest producers, nearly about 6 million tons in 2013 (MAPA, 2013). These statistics reinforce the importance of developments in the control and monitoring of the stages of the wheat production cycle.

Agricultural supply chains include products grown in different ways, regions, periods of the year; transported and stored by different means; all of these being important features that defines the traceability process. In this context, traceability makes it possible to recover the history of production process, from planting to the final product, regardless if the produced wheat for domestic

consumption or for export, since in both cases, traceability is an important tool and contributes to the quality of the final product. The objective is to ensure food safety, guaranteeing the consumer's right to information regarding the origin of the product, and also the right to avoid the consumption of ingredients, for reasons of food intolerance, lifestyle or religion. An example, are the food codes of some religions that rule which animals can be eaten and how they must be slaughtered, prohibiting its consumption and mixing meat with dairy foods.

Traceability is established by rules/regulations that are related to quality and the certification of food products. The purpose of this article is to describe the standardization inherent in food safety, presenting the

evolution of information technology systems that have been applied to the traceability of grain, and more specifically, wheat, in order to meet the requirements of the customer.

THE TRACEABILITY PROCESS

The importance of traceability in the food chain gained momentum in the 1990s, after the occurrence of incidents involving the consumption of beef from animals affected by bovine spongiform encephalopathy (BSE), commonly known as 'mad cow' disease.

Various occurrences of contamination of grain in the food industry are described in the literature. Karnal bunt fungus (Laux and Hurburgh, 2010) was found in wheat by the inspection service in the United States in 1996. This resulted in a reduction in exports, although the area of infection was limited to a small section of the State of Arizona. In this case, the process of traceability made it possible to determine the affected lots, thereby making it unnecessary to recall all wheat production.

According to Eckschmidt (2009), there is a degree of coverage within the production chain that makes it possible to classify traceability requirements. The greater the knowledge about a production chain, the greater the credibility of the information about the final product. Levels of traceability can be classified into 'basic', 'simple' or 'complete'. The first two are represented by information based on the credibility of the supplier, expressed in product labels; the difference between the two categories is that in the 'simple' category numerical codes are used to indicate the origin of the product at any point in the production chain. In 'complete' traceability, the product is followed along the entire chain, making it difficult to falsify information, but also requiring the complete participation of all agents involved at all stages.

The quality of information that is traced is equally as important as the level of information, which is directly related to the degree of confidence that the product will bring to consumers. The recorded data can be informative, validated or certified (Eckschmidt, 2009). When the information is validated or certified, it is confirmed by the authorities and physical evidence is collected, generating greater credibility for the supplier.

An important point, in terms of defining the level of traceability, is the knowledge of the risk points in the production process. The use of quality standards can meet this need because they are geared towards food safety.

The Hazard Analysis and Critical Control Points (HACCP) system (Furtini and Abreu, 2005) has been used by food production companies for the prevention and control of risks inherent to foods, particularly in relation to quality of health.

ISO 22005 (ISO 22005:2007) established standards for implementing a traceability system throughout the entire

production process. In the ISO standards, traceability is the ability to recover the history, application and/or location of an activity or a process, or a product or organization, by registered identification.

Both the HACCP and the ISO standards establish best practices for developing traceability processes, with standards and procedures regarding environmental conditions, facilities, equipment, human resources, technology, control and quality assurance, storage, transportation, etc.

The technologies adopted for the implementation of traceability processes in the grain supply chain were developed following the cited norms, standards and regulations.

REGULATIONS

In 2002, after the occurrence of 'mad cow' disease, the European Union created Regulation 178/2002 (REGULATION, 2002), establishing principles and standards of traceability at all stages of the production, processing and distribution of foodstuffs. In this regulation, traceability is defined as the ability to trace and follow the trail of a foodstuff, animal feed, any animal that produces foodstuffs, or a substance intended to be incorporated into foodstuffs or in animal feed, or expected to be, throughout all the stages of production, processing and distribution.

The regulation cites the need to establish a comprehensive system of traceability within the food sector, to enable withdrawals from the market in an organized and precise manner, or to inform consumers or employees responsible for such control, thereby avoiding unnecessary disruption to the safety of foodstuffs (Regulation (EC) 178/2002).

This regulation became mandatory, stating that all exports to the European community would have to comply with the regulation to ensure the participation of producers on the local, regional and global scale.

In Brazil, Normative Instruction no. 20 of 9/27/2001 (MAPA, 2001) established Integrated Fruit Production. This program established a set of agricultural best practices, necessary for registration, so that producers can be certified. It targeted the monitoring of diseases, pests and environmental conditions in order to guarantee food safety.

In 2008, the program was expanded to the wheat production chain (Tibola and Fernandes, 2009), initiating the integrated production of wheat, which was intended to work towards the quality assurance of that grain. The monitoring of production, from the farming stage onwards, is designed to ensure a product that is distinctive, traceable and of certified quality. The wheat lots are segregated when they are received at the storage unit according to their characteristics of interest, such as variety, business class, or purpose of use (bread,

pasta, cookies, etc.).

Normative Instruction no. 27 of 8/30/2010 (MAPA, 2010) established guidelines for systems that develop integrated programs. The principles are prioritized, based on the search for quality, safety of agricultural products, health, sustainability, certification, traceability and monitoring of the process, through the adoption of field and post-harvest notebooks to record relevant information regarding the stages of the productive chain.

The adoption of an integrated program by producers is not mandatory, but membership allows increased competitiveness in domestic and foreign markets through certifications held by private companies. This certification consists of the audit process in the farms that joined the system; once the requirements are met the farms receive a seal of compliance. The trend has been that there has been an increasing adoption of this system by producers because current demand indicates a prioritization on the part of consumers for products produced in standardized, production systems based on rationalization in the use of inputs, minimizing environmental impact, the optimization of operating costs, in addition to segregation, qualification and assurance of food safety (Tibola, 2007).

regulations that govern and guide implementation of a traceability process are not only applied in public policies related to food safety. Changes in government regulations have been accompanied by a rise in standards in the private sector, which may include rules on equipment, infrastructure, modes of production, quality management; the latter often stipulate stricter requirements than those described in public regulations and laws (Hammoudi et al., 2010). In this context, it is important to cite the GS1, Global Standard (GS1, 2009). which although not a regulation, is a global standard that is recognized worldwide for the identification and coding of products through barcodes, thereby contributing to the evolution of solutions related to traceability.

One of the tools for implementing traceability in the supply chain is the GTS, Global Traceability Standard (GS1, 2009), an independent technology that defines the minimum requirements for a traceability system, which also meet legal requirements. To perform tracking using the GTS standard, it is necessary that the traceable items, as well as the physical locations, have a GS1 identifier from the moment of registration until use. This standard does not compete with other international standards, such as ISO or other certifications of the food chain, but it helps companies and organizations meet these requirements by providing tools to achieve the desired level of traceability.

The technical manual of traceability for the wheat production chain (Fernandes and Tibola, 2009), establishes certain requirements for the implementation of a traceability system according to the GS1 standards. Parts of these requirements are as follows:

1. Registration of those participating in the traceability

- process, such as producers, transporters, storage companies, milling companies and the food industry;
- 2. Registration of lots, as a traceable unit, corresponding to the silo where the product was stored;
- 3 .Registration of the number of the silo where the grain was stored; information about the source and quality from the production phase, and the dates of the processes performed:
- 4. Use of computational tools to facilitate the register and transfer of records and also information regarding traceability.

The technical manual of traceability states that in relation to this last requirement of the process (the use of technologies to facilitate the recording of data) the records can be made by producers or technicians at the farm in spreadsheets that can be scanned in sequence or in electronic equipment with online systems for automatic data transfer. Therefore, some levels of traceability are legal requirements in certain sectors. However, as well as the mandatory requirements of traceability, companies are also developing voluntary processes in order to protect their brand and to ensure that their products are safe, making traceability a vital part of the supply chain. This research will now address the information technology required to meet the demands of traceability processes.

THE EVOLUTION OF TECHNOLOGIES APPLIED TO GRAIN TRACEABILITY

Some applied technologies are described below, which have evolved and which have contributed so that traceability in the supply chain of grain is performed and recognized by consumers and the food industry.

The RIG model - The Integrated traceability of grain

In order to meet standards and protocols regarding the traceability of food and agricultural products, the RIG Model, the integrated traceability of grain, was developed in 2006 (Leonelli, 2006). This model consists of systematic management steps that allow for the promotion of integrated traceability in the grain supply chain. It was initially designed for the soy industry, but it can be used for other grain production chains.

The scope of the RIG model was defined for the activities of selecting inputs, planting, cultivation, harvesting, grain storage, processing and logistics. It is composed of three stages of information: (i) constraints against the adoption of mechanisms for identification and traceability, (ii) planning activities, and (iii) operations management.

The first stage covers regulatory aspects, both mandatory and voluntary, which influence the adoption of traceability mechanisms by the company. These aspects

help to understand the needs of consumers in relation to what will be traced by setting the level of detail. The second stage concerns the planning of operational activities, such as the relevant attributes to be traced, characteristics of the product and its manufacturing process, specifics of where the product was produced, and information related to the labor used in production. Even at this stage, the critical control points regarding the most relevant information are defined. The critical nature of the traceability process can vary according to the size of the production chain, the number of agents involved, and the complexity of the relationship between the links of the production chain. This model allows these critical points to be defined and makes it clear which procedures can be audited.

The third stage systematizes the operations defined in the previous stages, that is, it guarantees the recording of information about inputs, planting and cultivation according to the standards of the product in question, such as harvest, storage and logistics. Of all the processes that are capable of being recorded, storage is one of the most complicated because the object in question is granular and farms do not always have an environment which can ensure the requirements for identification and traceability.

This model contains a proposal for a structured framework for managing the traceability of grain, which assists the implementation of traceability systems.

The RG system - Traceability of grain

The RG system, the traceability of grain (Ceruti, 2007) was developed to evaluate a program for traceability and preserved identity for wheat, documenting the entire process from receiving, drying, storage and processing, and focusing on insect management during storage.

The method traces samples of wheat in silos, during storage, after the use of various types of traps for insects and the application of insecticides. The RG system allows for the logging of data related to the production process, where both the product to be tracked and the entire production and storage process are customizable. Conceptually, the system was designed so that each product to be tracked is an 'entity' that has 'phases' that can be tracked through 'processes', for which required fields are created.

Using software, it is possible to quickly trace the processes in order to retrieve information requested by the industry, solving problems such as the quality of wheat flour. In addition to the modules that allow the registration of the whole process, a database was created with images of insects to aid in their identification. For the development of this system, data surveys were conducted with the help of cooperatives in Paraná State, linking the requirements of traceability with practical experience.

Technical manual of traceability for the wheat supply chain, and the digital field book

The technical manual for the traceability of the wheat supply chain (Fernandes et al., 2009) states that a traceability system includes the selection of varieties, production, transportation, storage, milling, the food industry and marketing (Figure 1).

The need for constant improvement of the traceability process, to supply real and updated information, is justified by the increased domestic production and export of foodstuffs, due to various factors such as demand, higher prices, weather conditions and technological innovations.

The digital field book (Tibola et al., 2009) corresponds to a system of internet traceability and certification of wheat, aligned to the standards of the integrated production system for wheat (PIT). The objective of this system is to maintain records of provenance and practices, from production to the process of post-harvest, such as physical and chemical properties of the soil, planned crop rotation, soil tillage and sowing, seed treatment, fertilizer, weed control and the application of growth regulators, the monitoring and control of diseases and pests, fungicide and insecticide applications, storage units, drying, and the thermometry and aeration of grains.

Using the digital field book, it is possible to integrate information between the agents involved in the production chain, making the certification of integrated production of wheat both agile and reliable. Data entries can be performed quickly via mobile devices, thus ensuring a higher quality traceability process.

Online traceability and qr-code

Some companies are focusing on developing systems for consumers who want quick access to information about the origin of the products they are buying. These are specialized applications to query the data that the company deems most important and of greatest value to the consumer. In the production of soya seeds, the system of online traceability (Gazolla, 2012) permits the storage and interrogation of information related to the seed production process, thereby forming a database with information that can be queried and evaluated whenever necessary.

The data from the 2010 to 2011 harvest were registered, and approximately 15,000 ha of area cultivated with soya were traced. At the UBS, the seed processing unit, seeds were uniquely identified by batches according to the date of receipt, variety, supplier and production area. Identification was performed by QR-Code (Quick Response Code) labels, which provide fast access via mobile devices. When querying the product code via the internet, the client can access the history of production, such as geographic location of the

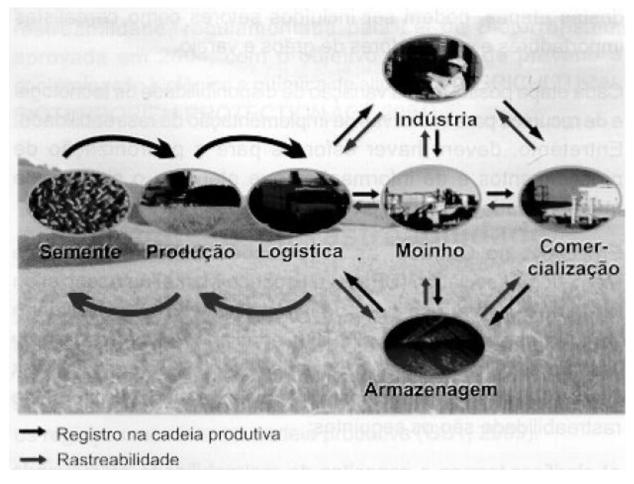


Figure 1. Stages of the traceability system for the wheat supply chain.

farm with photos, technical information on seed treatment, and the results of quality control reviews.

Another project that used the QR-Code Technology, was developed for flour mills in China (Qjan, 2012). The labels were placed on flour packages. Data which can be verified are: company name, product name, date and result of quality parameters. Therefore, the consumer has access to information through a website. Processes such as these add value to the product and bring greater security to the customer. Admittedly, the existence of a traceable process does not guarantee food security, but transparency for the consumer is what can make the difference in a moment of decision between one brand and other.

Traceability of grain and maize flow using radio frequency

In an attempt to reduce the risks of mixing within lots of grains, which can put a traceability system into disrepute, a project was introduced that incorporated the implementation of Radio Frequency Identification

technology, RFID (Rodarte et al., 2011). This technology uses radio signals that access information stored on tags or labels through wireless devices called readers.

RFID technology has advantages over barcodes because the information recorded on the labels need not be in line with the reader in order to be accessed. Thus, it can be incorporated within the packaging or implements, depending on the purpose. The scope and speed of the readers are also attributes that being constantly improved, they can be hundreds of meters away, and thousands of tags can be processed per second (Garcia, and Lunardei, 2011).

In this project, the tags were encapsulated in material, imitating a grain of maize (Figure 2). They were loaded with information specific to the grain producer and the lot, and inserted along with the grains. Experiments have shown a low rate of faults associated with the allocation of tags in the middle of the lot, but the application of RFID in large quantities of grain can be enhanced in relation to range of the reader. Although this experiment was performed with grains of maize, it represents a major breakthrough for future studies and can be used in any grain production chain.

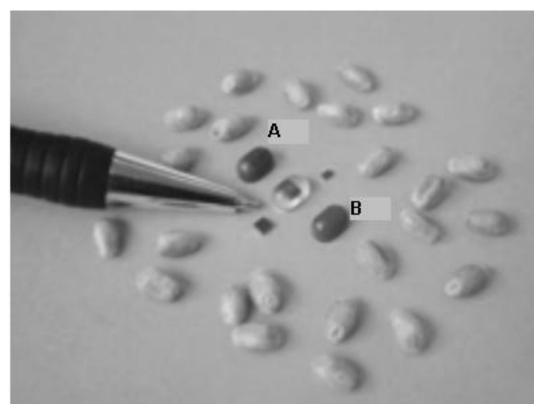


Figure 2. Encapsulated in the form of a grain of maize ('A' and 'B'); RFID Tag $1.6 \times 1.0 \times 0.5$ mm (center). Surrounded by natural grains of maize.

The benefit of using RFID technology is that data collection can be performed automatically using sensors, thereby reducing errors and minimizing the need for manual data entry. Technology like this is currently seen as a market differentiator, but it can be considered to be a necessity for the future.

Use of 'frameworks' for traceability

'Frameworks' is a technology that can reduce the effort involved in developing systems by reusing knowledge that is already structured and which can be applied to similar problems. According to Larman (2004), a 'framework' is a cohesive set of interfaces and classes that collaborate to provide services for the basic and constant part of a logical subsystem. The concept described by Booch (2006) may be used in this context; it corresponds to a standard of architecture, providing an extensive template for applications within a domain.

A traceability system involves the exchange of information between different agents within the production chain. To ensure quality, and access to and interoperability of data, it is important to adopt standards. These standards describe how messages should be built, sent and received. A project called 'TraceFood' was

recently developed to create a framework applied to traceability in the supply chain (Storoy, 2012). This framework contains recommendations for best practice in traceability, common principles for the unique identification of food items, and a standard for the exchange of electronic information related to traceability in the food industry, such as the identification number of the source, and how and when a product was processed, transported and received.

TraceCore eXtensible Markup Language' developed for various food sectors, such as seafood, mineral water, honey and meat. The structure can be applied to the implementation of a traceability system for grain because it has standards and methods that are required in order to model the traceability relationships between the agents involved, in any supply chain. The first step in the construction of the framework is to define the requirements applied to the supply chain. In a systematic approach, 'use cases' can be utilized to represent the relationships between users and the system. TraceFood uses this approach to represent the main requirements of traceability. For example, in the case of registration the following are required: practices of seed development, agricultural practices, management practices and grain storage, practices used in processing, and the authentication of new applications for new

system functionalities. This framework has been tested in food chains in pilot studies in Norway, Spain, France and China and the goal is to turn it into a methodology for the implementation of traceability in food chains. This has already happened with the standard for electronic information exchange in the fish industry, which has been incorporated into this framework, and is currently the ISO standard for the traceability of products from fish farming.

CONCLUSIONS

The traceability of a product is not a novel approach. Laws and registered quality control processes have been market requirements for quite some time. What is changing is the demand of consumers and non-governmental organizations for more detailed information on the origin of foods. The latter are now frequently demanding sustainable production from producers.

Solutions to address the demands of traceability in the food supply sector are of great importance for food quality and safety. When the product to be traced is grain, the degree of difficulty is greater due to the granularity of the product. This particular market requires more specific control and tracing.

Some companies are now structuring themselves to store grain in silos with smaller capacities, to ensure segregation and thus traceability. Thus, the use of standards for the exchange of information between the agents in the chain, and the definition of traceable requirements, contribute to the design and development of a relevant, applicable and quality system. Many laws, regulations, rules, standards and certifications have been created, which have contributed to guidelines on traceability. Technological solutions to implement systems for the different needs of producers and consumers are available and are in constant development.

This article has discussed projects like the model of integrated traceability, software specifically designed for the traceability of grain, digital field books, internet solutions with online information, and the use of more advanced technologies such as RFID and Frameworks. These tools are designed to simplify implementation, to reduce the effort of development, and to improve quality, thus reducing the costs of a project.

A comparative analysis was performed on existing systems and some important considerations can be highlighted. Some systems seek to control the productive tracts, from planting to harvest, but not all agents are part of the production chain, it does not store the information after harvest. In order to make the traceability process effective, particularly in case of any problem, it is necessary to have information on all production stages. Systems that use the QR-Code technology provide some information to the consumer that can be checked. This information is fixed, for instance, farm location, pictures

and quality information. An interesting option for companies that implement a traceability process would be to customize the necessary information to become visible on the Internet.

Regarding prospects for future research, we suggest the development of a framework that can be used for the development of traceability systems in the production process of wheat, aligned to the ISO, HACCP and GS1 quality standards, and the regulations that guide companies in compliance with requirements relating to the tracking of the production process for quality and food safety. In this way, a set of pre-defined patterns can be obtained that can be applied to traceability systems, thus reducing development time and improving the quality of those systems. It is important to emphasize that technological advances should bring innovation because, as well as the legal requirements surrounding traceability, consumers are seeking access to information and this is a motivation for companies looking for solutions to meet consumer demand. The greatest obstacle to the successful implementation of traceability in the food chain organizational, not technical, mav technological solutions are in constant development. Companies need to be motivated to implement new solutions, but there is often resistance, either to changes in operating practices or due to concerns over data security and reluctance to share information.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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Participatory evaluation methods of cassava varieties preferred in the mid-altitude tropical climate conditions of western Kenya

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The advantage of farmers' participation in breeding programmes depends on the effectiveness of the evaluation methods used. There is scarce information on the farmers' evaluation methods and their effectiveness as compared to the breeder's methods. Three districts representing the different agroecologies, inhabiting ethnic communities, cassava production and utilization niches were selected. Focused group discussion was used to determine the variety evaluation methods. The effectiveness of farmers and breeders evaluation methods were tested by evaluating cassava trials consisting of 15 varieties planted in the three districts. High storage roots yields, resistance to diseases and pests were the most preferred traits across the districts. Preference for traits related to plant type and storage roots quality, and variety ranking differed between districts indicating the differences in agro-ecologies, production and utilization niches. Farmers' and breeder's evaluation methods significantly correlated for related traits and elicited differences between varieties for most traits evaluated. Though low cyanide content was among the farmers' preferred traits, their evaluation method did not either elicit genotypic differences between varieties or correlate with the breeder's evaluation. The breeder lacked evaluation method for taste of boiled storage roots despite it being preferred by farmers. Despite the differences in traits preferences and variety ranking between farmers and the breeder and between districts, farmers and the breeder have effective evaluation methods. However, both evaluation methods have deficiency in evaluation of some of the preferred traits indicate the need for participatory variety selection.

Key words: Farmer preferred traits, farmers' qualitative evaluation method, breeder's quantitative evaluation method, participatory variety selection (PVS), variety ranking.

INTRODUCTION

Cassava (Manihot esculenta Crantz) is a clonally propagated crop grown in diverse environments for

diverse utilization within the tropics by small scale farmers mainly for subsistence (El-Sharkawy, 2004). The

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diversity in production environments and utilization methods lead to diversity in variety preferences. These diversities render centralised conventional cassava breeding approach ineffective and the need for target farmers' participation in cassava breeding (Manu-Aduening et al., 2006).

Farmers' participation in breeding programmes gives them an opportunity to select varieties that perform well in their environments with preferred traits (Almekinders and Elings, 2001). Unlike breeders who use quantitative scientific methods, farmers use qualitative indigenous method to evaluate varieties. Breeders therefore are able to identify and accurately measure traits based on their scientific knowledge.

However, some of the farmer preferred traits are not known to breeders or even if known, breeders more often lack the skills and scientific methods to evaluate them (Morris and Bellon, 2004). Farmer's participation in cassava breeding can only enhance efficiency if they have effective quantitative methods of variety evaluation for preferred traits. Similarly, the breeder's input can enhances the breeding efficiency if the quantitative methods used can effectively evaluate farmer preferred traits.

This study aims at generating information on; the qualitative methods used by farmers to evaluate cassava varieties, the effectiveness the farmers' qualitative and the breeder's qualitative methods of evaluating cassava varieties for preferred traits.

MATERIALS AND METHODS

The study was conducted in three purposefully selected districts in the major cassava growing region of western Kenya. The three districts; Mumias, Teso and Busia were purposefully selected to represent different cassava production systems and utilization methods.

From each district, one active farmer group whose members have long experience in cassava production and utilization was identified. Mumias district was represented by the Development Association Foundation (DAF) farmer group with membership of 33 (14 men, 19 women).

Farmers in this district grow cassava under mixed cropping system and use cassava storage roots after boiling. Naako-Aterait women group with membership of 29 (three men and 26 women) represented Teso district Farmers in Teso district grow cassava as a mono-crop and use storage roots after processing to flour. The flour is used to make; a local brew known as 'Busaa', porridge or local dish (stiff porridge) known as 'Ugali'. Busia district was represented by Agro-farmer group with membership of 31 (19 men and 12 women). Farmers in Busia district grow cassava under both mixed and mono-cropping systems. Both cassava utilization methods; eating after boiling and processing to flour are equally popular in Busia district.

A total of 10 (six landraces and four improved) cassava varieties were used in this study. The varieties were selected and collected from farmers' fields inform of cuttings based on their popularity across the three study districts. Popularity, which was based on secondary data from district agricultural crops production records, knowledge of extension staff and farmers was assumed to indicate the presence of farmer preferred traits and adaptation to the region.

Farmer's selection criteria and indigenous technical knowledge (ITK) methods of evaluating of cassava varieties

Focused group discussion (FGD) involving a total of 222 (73 men and 149 women) farmers, 96 (38 men and 57 women) members of farmer groups and the rest none farmer group members were conducted. During the discussion, farmers enlisted preferred cassava traits and ITK method they use to evaluate them. Farmers' selection criteria were determined by ranking the preferred cassava traits using pairwise ranking method. Farmers also described and defined the ITK method(s) they use to evaluate each preferred traits. Farmers were asked to rank the varieties directly by observing all plants in a plot, both uprooted and none-uprooted plants without considering the scores they awarded for the traits. A correlation analysis between direct variety ranking and traits scores was carried out to confirm the selection criteria.

Farmers' and breeder's variety evaluation

The 15 popular cassava varieties collected were planted under concealed identity by each farmer group in each of the three study districts using 5 x 9 α -Lattice design with three replications. Each plot consisted of three rows of five plants each spaced at 1 x 1 m. The trials were managed by group members. No fertilizer was applied since farmers commonly don't use fertilizers on cassava. At harvest, 12 months after planting (MAP), the 10 varieties were evaluated by farmers and the breeder separately. Only the middle row in each plot was carefully uprooted leaving all storage roots intact and attached to the plant. The uprooted plants were left in their position in the plot for evaluation.

Farmers' variety evaluation

In order to make it easier for farmers' evaluation, the trial plots were blocked into three blocks. Each block comprised of all the 15 varieties. Farmers evaluated each plot for; storage roots yield, disease and pest resistance, plant height, internode length, branching level, cyanide content, taste of boiled storage roots and height to first branching using ITK methods. The farmers scored for each trait on a scale of 0 (trait absent from the variety) to 5 (variety has the trait to farmers' satisfactory level). The farmers were asked to award scores like marks based on how satisfied they were with performance of the variety for each trait. Taste of boiled storage roots was evaluated after cooking two storage roots from each variety.

The storage roots were peeled, chopped into small pieces of 4 to 5 cm long, washed, placed in transparent plastic bags and boiled in a large cooking pot for about 45 min using local cooking methods. The boiled pieces were put on labelled plates on tables for evaluation. Mean scores were computed per site and standardized (Steel and Torrie, 1960). A selection index (SI) formula was developed by weighting the preferred traits based on farmer traits ranking. The standardized mean scores were used in the in SI formula to compute the SI value of each variety. The computed variety SI value was used to rank the varieties. The farmers were also asked to rank the varieties directly without considering the trait scores.

Breeders evaluation

The breeder collected data using scientific qualitative technique in all the three districts on; disease resistance on a score scale of 1 = resistant and 5 = susceptible taken on monthly basis starting 3 MAP and scored per plot; plant height (PT) and height to first branching (HB) (m), dry matter content (DM %) determined by the formula:

| Trait | Dreferred form | | Rank | | | | | |
|--------------------------------|--------------------------------|------|------|---|---------|--|--|--|
| Trait | Preferred form | Т | В | М | Mean | | | |
| Yield of storage roots | High number and size of tubers | 1 | 1 | 1 | 1.0 (1) | | | |
| Cleanliness of plants | Healthy clean leaves and stems | 2 | 2 | 2 | 2.0 (2) | | | |
| Height of plants | Tall plants | 3 | 4 | 4 | 3.7 (3) | | | |
| Bitterness of storage roots | Sweet taste | 8 | 3 | 3 | 4.7 (4) | | | |
| Bushiness of plants | Many branches per plant | 4 | 6 | 6 | 5.3 (5) | | | |
| Length between eyes | Short length between eyes | 5 | 5 | 7 | 5.7 (6) | | | |
| Storage roots taste | Sweet, soft and friable | 7 | 7 | 5 | 6.3 (7) | | | |
| Spearman's rank correlation co | pefficients between Mumias | -0.2 | 8.0 | - | - | | | |
| Spearman's rank correlation co | pefficients between Busia | 0.1 | - | - | - | | | |

Table 1. Farmer preferred traits and their ranking in Teso, Busia and Mumias districts.

DM (%) =
$$\{[Wa/(Wa - Ww)] \times 158.3\} - 142$$

Where; Wa is the mass of roots in air and Ww is the mass of roots in water, cyanide content (PC) taken using picrate score method on a score scale of 1 = low and 9 = high cyanide content, number of storage roots per plant (NR) taken by counting total number of storage roots per plant and fresh storage root yield (t ha⁻¹) (RY) taken by weighing all storage roots per plant, averaged per plot and converted to yield per hectare.

Data were analysed using residual maximum likelihood (REML) procedure in GENSTAT 14th Edition (Payne et al., 2011). Analysis was conducted per site independently and then tested for homogeneity of variance between sites using Bartlett's test before a combined analysis across sites detect variability in performance between genotypes, environments and their interaction.

Variety ranking by the breeder was based on a selection index (SI) formula recommended by Ceballos et al. (2004) with a few modifications. The SI was computed using standardized trait means. The SI formula used was:

$$SI = 10 (RY) - 8 (CMD + PC) + 8(NR + DM) + 3(PT) - 3(HB)$$

Where; SI is selection index; RY = fresh storage roots yield; CMD = cassava mosaic disease reaction score; PC = picrate score; NR = number of storage roots per plant; DR = dry matter content; PT = plant height and HB = height to first branching.

The breeder's and farmers' variety ranking were compared between and within districts using Spearman's rank correlation analysis using farmers' trait mean scores and the related mean agronomic qualitative measurement taken by the breeders.

RESULTS

Farmer's preferred traits and selection criteria

High storage roots yields and clean plants were the most preferred traits in all the three districts ranked first and second, respectively (Table 1). The other traits which can generally be considered as plant and storage roots quality related traits were inconsistently ranked in all districts. Farmer preferences in Mumias and Busia districts highly correlated (Spearman's rank correlation

(rs) of 0.77) where as farmers preferences in Busia and Mumias to Teso districts with rs = 0.61 and 0.54, respectively. Tall plant and high branching level are more preferred by farmers from Teso district, ranked third and fourth respectively, as compared to farmers from Busia and Mumias districts where they were ranked fourth and sixth respectively. Low cyanide content was important in Busia and Mumias districts ranked third, as compared to Teso district where it was ranked last (position eight).

Farmers' indigenous variety evaluation technical knowledge (ITK)

During FGD, farmers described and listed the ITK methods they use to evaluate preferred cassava traits. In this study, the following ITK methods that farmers use to evaluate different cassava preferred traits were revealed:-

- **i. High yield of storage roots:** Is evaluated before and at harvest stages.
- a) Before harvest yield evaluation is either by observing soil cracking around the plant and/or stem thickness at the crown. Large deep cracks radiating from the crown of plant indicate and/or thick stems at the crown indicate high yields and are preferred.
- b) At harvest stage, storage roots yield is evaluation by considering the number and size of storage roots. Many and large storage roots indicate high yields and are preferred.
- ii. Cleanliness of plants: Is evaluated by observing the health of the leaves. Plants with deformed, few, discoloured small leaves are considered susceptible and undesirable. Despite the region being a hot spot for many cassava foliar diseases and pests, most farmers could not identify any of them. They believe the poor health of plants expressed as yellowing, curling and dropping-off of leaves, stunted growth of plants and drying of stems are due to poor soils fertility (soil exhaustion), water stress,

T, B and M = Teso, Busia and Mumias district, respectively.

| Sauras | Mumias | Teso | Busia | Between |
|------------------------------------|--------|-------------------|--------|-------------------|
| Source | n = 33 | n = 29 | n = 31 | districts (MS) |
| Soil cracking (SC) | 77.6** | 106.0** | 60.7** | 1.9 ^{ns} |
| Storage roots number (RN) | 19.3* | 62.2** | 12.3** | 1.1 ^{ns} |
| Storage roots size (RS) | 57.4** | 23.7** | 43.6** | 2.7 ^{ns} |
| Stem thickness (ST) | 84.5* | 41.7** | 34.7* | 1.5 ^{ns} |
| Cleanliness of plants (DR) | 66.8** | 65.5** | 33.2** | 0.5 ^{ns} |
| Height of plant (PT) | 76.2** | 12.9* | 49.1** | 1.3 ^{ns} |
| Bitterness of storage roots (PC) | 27.7* | 5.7 ^{ns} | 21.6ns | 6.3** |
| Bushiness of plants (BL) | 95.1** | 52.9** | 36.1** | 0.9 ^{ns} |
| Length between 'eyes' (IL) | 30.2* | 34.6** | 24.4* | 3.3** |
| Taste of boiled storage roots (TR) | 34.7** | 62.3** | 29.2** | 3.9** |

Table 2. Between districts mean square and within districts Kruskal-Wallis H-values for cassava mean scores evaluated by farmers in Mumias, Teso, and Busia districts.

ns, * and ** = not significant, significant at $P \le 0.05$ and 0.01, respectively; n = number of participating farmers; MS = mean square.

bad environmental conditions, bad omen or witchcraft.

iii. Bitterness of fresh storage roots: Is evaluated by tasting the storage root. Varieties with bitter storage roots are considered to have high cyanide content and are not preferred for chewing raw or boiling. Such varieties are used after drying and processing the storage roots to flour.

iv. Height of plant, length between 'eyes', and bushiness of plant: Are evaluated by direct counting and/or observing the plants.

v. Taste of boiled storage roots: Evaluated by tasting the boiled storage roots.

Farmers' variety evaluation using ITK

The Kruskal-Wallis H-values for trait scores were significant (P < 0.05) for all traits evaluated in all districts except for cyanide content in Teso and Busia districts (Table 2). Significant Kruskal-Wallis H-values indicate significant genotypic differences between varieties and thus the ability of farmers using their qualitative ITK methods to elicit different between varieties. The between districts mean squares (MS) for trait score were significant (P < 0.05) only for cyanide content, internode length and taste of storage roots. The significant MS show either the presence of genotype x environment interaction effect or between districts differences in farmer preferences for these traits.

Due to the differences observed in traits preferences and scoring by farmers between districts (Table 2), selection index (SI) formula was developed for each district separately. Weighting of traits in the SI formula were based on trait ranking by farmers during the FGD. For example out of the seven traits, high storage roots yield (RY) and low cyanide content (PC) were the most and least preferred traits respectively by farmers in Teso district. The SI formula for Teso district, RY and PC are

given weightings of 7 and 1, respectively. The SI formula used for each district was:

$$\begin{split} Teso: & \cdot SI_{ft} = 7(SC+RN+RS+ST)/4 + 6(DR) + 5(PT) + 1(PC) + 4(BL) + 3(IL) + 2(TR) \\ Busia: & \cdot SI_{fb} = 7(SC+RN+RS+ST)/4 + 6(DR) + 4(PT) + 5(PC) + 2(BL) + 3(IL) + 1(TR) \end{split}$$

Mumias:- $SI_{fm} = 7(SC+RN+RS+ST)/4 + 6(DR) + 4(PT) + 5(PC) + 2(BL) + 1(IL) + 3(TR)$

Where: Storage root yield (RY) mean score was considered as a function of soil cracking (SC), storage roots number (RN), storage root size (RS) and stem thickness (ST) mean scores. The SI for each variety in each district was computed using the standardized mean scores.

Farmers' variety ranking

Ebwanatereka in Teso and Mercury, in Mumias and Busia were ranked top by farmers using SI method. Migyera in Teso, Bumba in Mumias and MM96/1871 in Busia were ranked first by farmers using direct ranking method (Table 3). Variety ranking using SI and direct ranking methods correlated significantly (P < 0.05) in all districts with Spearman's' rank correlation coefficient (r_s) of 0.7, 0.6 and 0.8 in Busia, Mumias and Teso district, respectively (Table 4). Significant correlation in variety ranking using the two methods in all the study districts imply either ranking by use of SI or direct ranking can be used. There were no significant (P < 0.05) correlation in variety ranking by farmers between districts except between Mumias using SI method with Busia using both methods with $r_s = 0.7$ and between Mumias' using direct ranking with Teso using both methods with $r_s = 0.5$. Lack of significant correlation in variety ranking by farmers between districts indicates differences in either variety environmental, preferences, and/or genotype environment interaction effects.

Table 3. Cassava variety ranking by farmers and the breeder in Mumias, Busia and Teso districts.

| | Mumias | | | Bu | sia | | Teso | | | cross distri | cts | | |
|------------------|--------|-----|---------|-----|-----|---------|------|------|---------|--------------|----------|----------|--------------|
| /ariety | Fai | mer | Breeder | Far | mer | Breeder | Fa | rmer | Breeder | Far | mer | Dunadas | Overall mean |
| | SI | DT | _ | SI | DT | | SI | DT | | SI | DT | Breeder | |
| Migyera (I) | 3 | 3 | 1 | 5 | 8 | 1 | 3 | 1 | 2 | 3.7(2) | 4.0(1) | 1.3(1) | 1.3(1) |
| Sudhe (L) | 6 | 7 | 12 | 2 | 4 | 11 | 2 | 2 | 3 | 3.3 (1) | 4.3(3) | 8.7(8) | 4.0(2) |
| MM96/1871 (I) | 4 | 2 | 3 | 3 | 1 | 2 | 12 | 14 | 5 | 6.3(5) | 5.7(5) | 3.3(3) | 4.3(3) |
| Mercury (L) | 1 | 5 | 8 | 1 | 3 | 3 | 9 | 8 | 10 | 3.7(2) | 5.3(4) | 7.0(7) | 4.3(3) |
| SS4 (I) | 9 | 6 | 4 | 7 | 2 | 4 | 10 | 9 | 1 | 8.7(9) | 5.7(5) | 3.0(2) | 5.3(5) |
| Bumba (L) | 2 | 1 | 13 | 12 | 9 | 7 | 5 | 3 | 13 | 6.3(5) | 4.0(1) | 11.0(11) | 5.7(6) |
| Nambukaya (L) | 7 | 10 | 5 | 4 | 7 | 8 | 8 | 4 | 7 | 6.3(5) | 7.0(8) | 6.7(6) | 6.3(7) |
| MM96/4684 (I) | 10 | 4 | 2 | 8 | 10 | 6 | 4 | 6 | 11 | 7.3(8) | 6.7(7) | 6.3(5) | 6.7(8) |
| Ebwanatereka (L) | 8 | 9 | 14 | 6 | 5 | 10 | 1 | 7 | 4 | 5.0(4) | 7.0(8) | 9.3(9) | 7.0(9) |
| MM96/3972 (I) | 12 | 11 | 6 | 10 | 14 | 5 | 11 | 13 | 6 | 11.0(12) | 12.7(14) | 5.7(4) | 10.0(10) |
| CK9 (L) | 11 | 12 | 7 | 9 | 15 | 13 | 7 | 10 | 9 | 9.0(10) | 12.3(13) | 9.7(10) | 11.0(11) |
| Kaleso (L) | 15 | 14 | 9 | 15 | 11 | 12 | 15 | 11 | 12 | 15.0(15) | 12.0(11) | 11.0(11) | 12.3(12) |
| Opongi (L) | 14 | 8 | 15 | 14 | 12 | 14 | 6 | 5 | 8 | 11.3(13) | 8.3(10) | 12.3(14) | 12.3(12) |
| Sifros (L) | 5 | 15 | 11 | 11 | 6 | 15 | 14 | 15 | 15 | 10.0(11) | 12.0(11) | 13.7(15) | 12.3(12) |
| Serere (L) | 13 | 13 | 10 | 13 | 13 | 9 | 13 | 12 | 14 | 13.0(14) | 12.7(14) | 11.0(11) | 13.0(15) |

L, landraces; I, improved varieties; SI and DT, farmers' ranking using selection index and direct ranking, respectively.

Breeder's evaluation

Variety MS were significant (P < 0.01) for the traits evaluated by the breeder (Table 5) indicating significant genotypic differences between the varieties. MS for districts and variety x district interaction effects were significant (P < 0.01) for all traits except for resistance to foliar diseases and number of storage roots per plant. These imply there are significant differences in test environments and the presence of significant genotype x environment interaction effect, respectively for all traits except resistance foliar diseases and number of storage roots per plant.

Migyera in Mumias and Busia districts, SS4 in Teso district were ranked first while MM96/4684,

MM96/1871 and Migyera were ranked second in Mumias, Busia and Teso district, respectively by the breeder (Table 3). Generally, all the varieties ranked top by the breeder are improved varieties. There were no significant (P < 0.05) correlation in breeder's variety ranking between districts except between Mumias and Busia districts ($r_s = 0.70$) indicating differences in environments and the presence of cross-over effects resulting from genotype x environment interaction effect.

Farmers' qualitative versus breeder's quantitative evaluation

There were significant (P < 0.05) correlation

between qualitative scoring by farmers for preferred traits and quantitative data taken by the breeder on all related agronomic traits taken except picrate content score versus bitterness taste scores, number of storage roots per plant versus stem thickness and storage roots yield versus size of storage roots taken by the breeder and farmers respectively in all districts (Table 6). There was negative correlation between the breeder's disease score and farmers score for cleanliness of the plant implying farmers awarded high scores for plants that the breeder had low scores and vice-versa. This means farmers preferred varieties resistant to foliar diseases. Correlation between branching level taken by the breeder and bushiness of the plants scored by

Table 4. Spearman's rank correlation coefficients between farmers variety ranking using selection index (SI), direct ranking and the breeder's ranking in Teso, Busia and Mumias.

| | ВВ | BFD | BFS | MB | MFD | MFS | ТВ | TFD |
|-----|--------|--------|--------|-------|-------|------|------|--------|
| BFD | 0.40 | 1.00 | | | | | | |
| BFS | 0.55* | 0.70** | 1.00 | | | | | |
| MB | 0.70** | 0.13 | 0.39 | 1.00 | | | | |
| MFD | 0.70** | 0.46 | 0.48 | 0.30 | 1.00 | | | |
| MFS | 0.49 | 0.66** | 0.67** | 0.15 | 0.61* | 1.00 | | |
| TB | 0.45 | 0.43 | 0.59* | 0.33 | 0.39 | 0.14 | 1.00 | |
| TFD | 0.15 | 0.07 | 0.27 | -0.06 | 0.54* | 0.30 | 0.34 | 1.00 |
| TFS | 0.09 | 0.09 | 0.39 | -0.12 | 0.51* | 0.28 | 0.26 | 0.81** |

BB, MB and TB, breeder's ranking in Busia, Mumias and Teso district respectively; BFD, MFD and TFD, farmers' direct ranking in Busia, Mumias and Teso district, respectively; BFS, MFS and TFS, farmers ranking using selection index in Busia, Mumias and Teso district, respectively.

Table 5. Mean square values of important agronomic and farmer preferred traits evaluated by the breeder across three districts, Mumias, Busia and Teso.

| Source of variation | df | DR | DM (%) | BL | PC | PT (m) | NR | RY (t ha ⁻¹) |
|---------------------|----|-------------------|---------|-------|-------|------------|-------------------|--------------------------|
| Variety (V) | 14 | 8.3** | 45.4** | 9.2** | 5.5** | 3310.9** | 37.6** | 8.6** |
| District (D) | 2 | 0.5 ^{ns} | 102.6** | 2.8** | 1.0** | 100061.9** | 4.4 ^{ns} | 6.6** |
| V x D | 28 | 0.4 ^{ns} | 42.8** | 0.8** | 3.6** | 1628.9** | 4.3 ^{ns} | 2.2** |

df, degrees of freedom; DR, foliar disease resistance scored on a 1= Resistant to 5 = susceptible; DM, dry matter content (%); BL, branching levels PC, picrate score on a 1 = no cyanide to 9 = high levels of cyanide; PT, plant height (m); NR, number of storage roots per plant; RY, fresh storage roots yields (t ha $^{-1}$); *, Significant at P \geq 0.01; ns, non-significant.

Table 6. Correlation coefficients between qualitative and quantitative evaluation of cassava varieties by farmers and the breeder for related traits in Mumias, Teso and Busia districts.

| Traita correlated | | District | |
|---------------------------------|--------|----------|--------|
| Traits correlated | Mumias | Teso | Busia |
| DR by breeder vs DR by farmers | -0.8** | -0.8** | -0.5* |
| B L by breeder vs BL by farmers | - | -0.6** | 0.4* |
| IL by breeder vs IL by farmers | -0.8** | -0.9** | -0.7** |
| PC by breeder vs PC by farmers | -0.1 | 0.2 | -0.2 |
| PT by breeder vs PT by farmers | 0.6** | 0.4* | 0.3* |
| NR by breeder vs NR by farmers | 0.4* | 0.4* | 0.8** |
| NR by breeder vs RS by farmers | -0.3 | -0.6** | -0.4* |
| NR by breeder vs ST by farmers | 0.2 | -0.3 | 0.1 |
| RY by breeder vs NR by farmers | 0.4* | 0.2 | 0.8** |
| RY by breeder vs RS by famers | 0.1 | 0.2 | 0.3 |
| RY by breeder vs ST by famers | -0.3 | 0.4* | 0.2 |

farmers correlated negatively in Teso but positively in Busia. These imply farmers in Teso prefer plants that branch at a lower height while farmers in Busia prefer plants that branch at high heights. This may reflect the differences in cropping systems used in Teso and Busia.

There were no significant (P < 0.05) correlation between the breeder's and farmers' variety ranking using

both in all districts except in Busia district between the breeder and farmers' using direct ranking method (rs = 0.55) (Table 6). Three out of the top 5 ranked varieties by the breeder were improved varieties in all districts (Table 5). This ranking is opposite to farmers ranking where at least 3 out of the top 5 ranked varieties were landraces. Three varieties, Migyera, MM96/1871 and SS4, all

improved varieties were ranked among the top 5 by the breeder in all districts. Migyera was ranked among the top 5 by farmers in all districts while Mercury and MM96/8171 were ranked among the top 5 by farmers in Mumias and Busia only. Farmers in Teso district tend to prefer landraces as compared to the other two districts. Migyera and MM96/1871 in Mumias district, MM96/1871 and Mercury in Busia district and Migyera in Teso were ranked among the top 5 varieties by both the breeder and farmers using both ranking methods. Migyera was the only variety ranked among the top 5 by the breeder and farmers from all districts.

DISCUSSION

Farmers evaluate varieties intuitively by considering many factors (Sunwar et al., 2006) as compared to breeders who evaluate varieties using quantitative measurements based on scientific knowledge and expertise. Collaboration between farmers and breeder in variety evaluation has been observed to increase breeding efficiency and effectiveness (Witcombe et al., 2006). However, farmer participation in breeding has been seen to increase breeding costs and can only add value if the quantitative methods they use to evaluate varieties are valid and effective (Witcombe et al., 2006).

In this study, farmers across the three districts prefer cassava varieties that have clean plants (resistant to pests and diseases) and produce high yield of storage roots. However, preferences of traits related to plant type (bushiness, length between nodes and height of plants) and quality of storage roots (taste of boiled storage roots and bitterness of storage roots) differed between districts. The differences in preferences for traits related to plant type between the districts can be attributed to the differences in cropping systems between the districts. Farmers in Mumias and Busia districts predominantly grow cassava under mixed cropping system and therefore would prefer non-bushy varieties as compared to farmers in Teso districts who predominantly grow cassava as a mono-crop (EARRNET, 2004).

Highly branched and short plants prevent sun light penetration which affects the performance of the other crop in the mix. Differences in preferences of storage roots quality can be attributed to the differences in cassava utilization methods between the districts. Low bitterness in storage roots is more preferred by farmers from Busia and Mumias districts, than in Teso district. Farmers in Teso district farmers predominantly use cassava after processing to flour while Busia and Mumias district farmers predominantly eat cassava after boiling (EARRNET, 2004). Cassava processing has been shown to reduce cyanide content (Mkumbira et al., 2003).

Farmers' cassava variety evaluation techniques for all the preferred traits are qualitative and based on observation without making any measurements. Using these techniques, farmers' evaluation just like the breeders' evaluation drew-out genotypic differences between varieties for most of the preferred traits. evaluation using these techniques also Farmers' significantly correlated to the breeder's evaluation for related traits. These imply the farmers' qualitative methods are as effective as the breeder's quantitative methods. However, there were no significant differences between varieties when evaluated for cyanide content by farmers. Number of storage roots per plant and storage roots yield evaluated by the breeder did not correlate significantly with, bitterness of storage roots, stem thickness and size of storage roots taken by farmers, respectively in all districts. Though taste of boiled storage roots was among the farmers' preferred traits, the breeder lacked quantitative method to evaluate it. These results show that though farmers' and breeder's evaluation methods are effective, they are deficient and may not be effective for some traits. There is need for participatory approach where farmers qualitative and breeder's quantitative evaluation methods augment each other.

Farmers' and breeders' variety ranking did not significantly correlate within and between districts while the breeder's analysis revealed; lack of significant correlation in variety ranking between districts, swooping of variety ranks when grown in different districts and significant genotype x environment mean squares. These indicate either differences between farmers from different districts in preferences or the presence of genotype x environment interaction (GEI) effects. Though GEI effect has been reported in cassava on most agronomic traits, further experiments to determine whether these differences are as a result of the differences in preferences between the groups should be carried out. GEI effects can be overcome either by breeding for either specific or broad adaptation (Cach et al., 2006).

Breeding for specific adaptation requires demarcation of the cassava growing area into uniform production and utilization niches (Lin and Binns, 1988). Varieties are then selected and promoted in the specific niche where they perform well.

Conclusions

In this study, each district can be considered as separate niches. The most preferred varieties are; Bumba in Mumias, Mercury and MM96/1871 in Busia and Sudhe and Migyera in Teso district. These varieties are adapted to the specific districts and have traits that satisfy the preferences of farmers from the districts.

Demarcating cassava growing region into specific niches may not be feasible since cassava is grown by small scale farmers under diverse production environments which may vary from farm to farm (El-Sharkawy, 2004). Breeding for broad adaptability requires

selection of varieties with above average performance across the growing area (stable varieties) (Finlay and Wilkinson, 1963). It is only Migyera which was ranked among the top five varieties across all the three districts by both the farmers and the breeder. Migyera is therefore a stable variety which is resilient to changes in production niches (Lin and Binns, 1988).

Though farmers' qualitative method of variety evaluation is equally effective as the breeder's quantitative methods, they both were ineffective in evaluating some of the preferred traits. The differences in traits preference, variety ranking between districts by farmers and the breeder and the presence of GEI show that, farmers from difference districts may prefer and adopt different varieties. There is therefore need to reorient cassava breeding strategy and adopt decentralized participatory breeding. Variety evaluation should be conducted in each production niche where both farmers' qualitative and breeder's quantitative evaluation methods are used and supplement each other.

Conflict of Interests

The authors have not declared any conflict of interests.

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Determinants of smallholder farmers' hybrid maize adoption in the drought prone Central Rift Valley of Ethiopia

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This paper examines the factors influencing smallholder farmers' adoption decision of hybrid maize in drought prone Central Rift Valley (CRV) of Ethiopia. The analysis is based on data collected through a questionnaire survey of 277 randomly selected maize grower farm household heads. Binary response Logit model was employed in the data analysis. Hybrid maize adoption in the CRV was found to be influenced by age, years of formal education, farmland size, the proportion of farmland allocated to maize, frequency of extension visit, grain market distance and altitude. Extension visit was negatively associated with hybrid maize adoption contrary to the prevailing beliefs and the earlier findings. Extension workers have been promoting open pollinated varieties (OPVs) maize since the OPVs have been more common in drought prone area though the farmers in the CRV also producing hybrids. Even though the prevailing maize hybrids have essentially been released for high potential areas, the production of these hybrids is currently expanding in the CRV where it is grown by 30% of the farmers. Likewise, hybrid maize adoption in this area offers better opportunity for private seed companies' involvement in the seed provision of adapted hybrids. A paradigm shift in the maize breeding efforts and extension service on hybrid for drought prone areas is commendable in order to enhance the food security of smallholder farmers in the CRV.

Key words: adoption, Central Rift Valley, hybrid maize, drought prone.

INTRODUCTION

Maize is one of the most important food crops produced and consumed in Ethiopia. Among cereal crops, it stands first both in production and productivity and second after teff in area coverage. It is grown by slightly more than nine million farm households (CSA, 2012). It contributes about a quarter of the daily calorie requirements of the Ethiopians. Increasing and sustaining maize productivity

is a vital concern in Sub-Saharan African countries in enhancing a household food security principally in drought induced risk prone areas such as Central Rift Valley (CRV) of Ethiopia. Ethiopia has been pursuing agricultural production intensification approach to boost crop productivity through the application of modern agricultural inputs primarily improved varieties fertilizers

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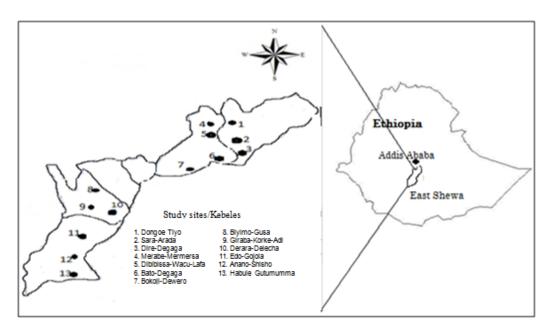


Figure 1. Map of the study area.

and improved agronomic practices focusing on high potential areas. Accordingly substantial amount of resources were devoted to the development and dissemination of improved maize varieties (Alemu et al., 2008). So far, about 43 improved maize varieties including the hybrids and open pollinated varieties (OPVs) have been released and disseminated though farmers adopted a few. Limited information has been documented aboutthe factors influencing adoption decision of maize growing farmers particularly in drought prone areas.

In Ethiopia, about 40% of area of maize production is located in moisture stress environments and contributes to less than 20% of maize production. Low maize production in drought stressed areas in relation to its land area is partly attributed to low research attention given to this agro-ecology since the beginning of maize research and afterwards. There have been substantial progresses made under African Maize Stress (AMS), Drought Tolerant Maize for Africa (DTMA) and Sustainable Intensification of Maize-Legume Cropping Systems for Food Security in Eastern and Southern Africa (SIMLESA) projects in research and dissemination of improved open pollinated maize varieties for drought prone areas of the country.

Adoption of technological innovations in agriculture has attracted a considerable attention of socio-economic researchers and policy makers in developing countries since its population drive most of their livelihood from agriculture (Feder and Umali, 1993). Moreover, a new technology is believed to offer an opportunity to boost production and income substantially. However, the introduction of technological innovations have met with

only partial successes, as measured in observed rates of adoption.

Several studies have been devoted to analyzing the influencing the adoption of agricultural technologies such as improved varieties and documented different results. Those studies considered human and physical resource endowments in the analysis. There are no, however, conclusive evidences from the factors considered in adoption study where some essential factors such agro-ecological conditions were barely considered which are critical in crop production decision in the study area. Moreover, the studies conducted focused on high potential areas for crop production. This paper tries to capture agro-ecology by considering altitude since altitude is a salient feature influencing both temperature and precipitation in Ethiopia. In general, identifying factors that hinder or facilitate the adoption of improved maize varieties such as hybrids in stress environments provide valuable information in improving the efficiency of maize research, extension services, and food security concerns to smoothen enhanced technological changes. The objective of this paper is, therefore, to analyze factors influencing the adoption of hybrid maize in the drought prone CRV of Ethiopia. It takes into account hybrid maize in the analysis based on a cross-sectional data obtained from farm household survey.

METHODOLOGY

The study area and methodological considerations

The study was conducted in East Shewa Zone, CRV of Ethiopia (Figure 1). The area is characterized by drought prone agro-

ecology where effective crop growing period lasts for three months per annum. Moreover, there is a significant crop harvest stress once every 3 to 4 years. In most of the months, the precipitation is in short of evapotranspiration. The rainfall of the area by low and erratic rainfall, averaging between 600 and 800 mm per year. The minimum temperature ranges from 8 to 17°C and the maximum from 24 to 32°C. The major cereal crops produced in the study areas include maize, teff, wheat and barley while common bean is the main pulse crop produced under rainfed condition.

Sampling and data collection method

Data were obtained from farm household survey conducted in 2011, using multi-stage sampling approach. East Shewa Zone of Oromia Regional State of Ethiopia was purposively selected for its importance in drought prone maize producing area, presence of well-organized maize research and extension services. Accordingly, four districts: Adama, Boset, Dugda and Adami-Tulu-Jido-Kombolcha (ATJK) were selected based on their importance in maize production. From those districts, thirteen maize producing Kebeles were randomly selected (Figure 1). Kebele is the lowest administrative entity in the Ethiopian administrative tiers and also known as Peasant Association (PA). Finally, 277 maize farmers were randomly selected from agricultural land use taxpayers lists obtained from the respective district offices of the revenue. Structured and pretested questionnaires were administered to the farmers to solicit information through face-to-face interview.

Data management and analysis

Data collected from maize grower farm household heads on selected factors were coded, entered, cleaned then analyzed using descriptive statistics and Logit model. SPSS 17.0 was used in the data management while both the SPSS and StatalC 10 Softwares were used in the data analysis.

Conceptual framework and analytical considerations

The conceptual framework for the analysis is based on partial adoption of agricultural innovation (Feder et al., 1985). Adoption of agricultural innovation was hypothesized to be a result of an elaborate set of inter-technology choice made by a farm household head among available alternative production inputs. Many of the adoption studies were preoccupied with analysis of the impact of factors such as credit, information availability, risk, and farm size. The factors explaining adoption of improved technology can be estimated using Logit, Probit or Tobit models. The choice of improved technology adoption can be assumed as a dichotomous outcome of adoption or non-adoption. Hence, a farmer's choice was represented by a dichotomous dependent variable measured in nominal dummy variables1 (adoption) or 0 (absence of adoption). Selection between Logit and Probit models is sticky since both models provide equally efficient parameter (Demaris, 1992). However, when continuous independent variables are included in the model, Logit model is well suited for explaining and testing the hypotheses about the relationships between a categorical outcome variable and one or more categorical or continuous variables (Peng et al., 2002). Thus, in this study, Logit model chosen since the explanatory variables are constructed from both categorical and continuous variables while the dependent variable is categorical.

Model specification and description of the variables

Logit model of binary response (Menard, 2002) was employed to

test the factors influencing the adoption of hybrid maize. The model is specified as:

$$\log \left(\frac{P_i}{1 - P_i}\right) = \sum \beta_j X_{ij} + \varepsilon_i$$

The probability that a farmer adopts the technology is denoted as P= P[y_i = 1] while the probability for adopters is 1 and P = P[y_i = 0] for non-adopters. This binary adoption variable has a probability function $f(y) = P^y(-P)^{1-y}$ where y=0 or 1. Thus, P_i stands for the conditional probability that a farmer adopts hybrid maize while (1-P_i) represents the conditional probability that a farmer does not adopt hybrid maize; $\beta_i s$ are coefficients to be estimated about an explanatory variable; $X_{\!S}$ are the explanatory variables and \mathcal{E}_i is the error term. The explanatory variables of hybrid maize adopter (HYMA) in the CRV is specified and explained as follows.

HYMA = β_0 + β_1 GND+ β_2 AGE+ β_3 HHSIZ+ β_4 EDC+ β_5 FRMSZ+ β_6 MZAP+ β_7 OX+ β_8 EXTN+ β_9 FLD+ β_{10} TMKT + β_{11} DRGHT + β_{12} ALTI.

A number of working hypotheses were employed and literatures were consulted in constructing the model since there is no firm econometric theory that dictates the choice of independent variables in adoption studies. The factors influencing the adoption of hybrid maize were roughly categorized into three: human and physical resource endowments, institutional and agro-ecologic factors. The human and physical resource includes gender (GND), age (AGE), household size (HHSIZ), formal education of the household head (EDC), farmland size (FRMSZ), percent of farmland allocated to maize (MZAP) and number of oxen owned (OX) were captured.

In agricultural technology adoption, behavioral differences between genders is presumed since female-headed households are usually poorer in developing countries and their access to information and innovation limited that may negatively affect their improved technology adoption. Age is a proxy for farming experience of a farmer that can erode or generate confidence to try a new technology. Accordingly, younger farmers are expected to be adventurous or risk takers; and thus, test an improved technology and adopt it. Likewise, household size accounts for household farm labor since an intensive field management for optimum production usually accompanies the adoption of improved varieties. Hence, larger household size is presumed to provide ample labor for the intensive field management in improved variety such as hybrid maize cultivation. Year of formal education is expected to enhance farm household likelihood of the adoption of improved technology since education boosts the capacity of a farmer in acquiring, processing and utilizing new information.

Farmland and livestock (particularly oxen) are the major farm household assets in pursuing crop production. A farmer's possession of those resources is anticipated to enhance a farmer's hybrid maize adoption. Hence, a well-off farmer is supposed to allocate part of his or her farmland to improved varieties, and be able to purchase inputs that are accompanying them. Likewise, farmers who own sufficient numbers of oxen are expected to perform land preparation on time hence adopt hybrid maize to enhance their maize production.

The second set of explanatory variables of hybrid maize adoption comprise extension visit (EXTN), field day participation(FLD), time taken to the nearest grain market (TMKT) which are related to institutional factors. Extension visit is presumed to be a major means for a farmer's access to reliable agricultural information. Hence, it is expected to influence the adoption of improved variety. A farm household head participation on field days of improved maize variety demonstration is presumed to develop farmers'

| Variable | Definition/unit | Expected sign |
|----------------------------------|---|---------------|
| Gender of the household head | 1 if the farmer is male; 0 otherwise | + |
| Age of the household head | Years completed since birth | +/- |
| Household size | Number of household members | + |
| Years of formal education | Number of years formal education completed by household head | + |
| Farmland size | Total farmland area in hectare owned by household | + |
| Proportion of maize area | Per cent area of the household planted to maize | + |
| Oxen owned | Number of oxen owned by household | + |
| Extension visit | Frequency of extension contact per month | + |
| Field day participation | 1 if the farmer participated on improved maize demonstration field day; 0 otherwise | + |
| Time to the nearest grain market | Walking time (minutes) to the nearest grain market | + |
| Drought encounter frequency | Frequency of drought that caused substantial maize yield stress | _ |
| Altitude | Altitude of the area in meters above sea level (masl) | + |

Table 1. Definition of explanatory variables and their expected signs in hybrid maize adoption in the CRV.

confidence and facilitate improved variety adoption. Distance to the nearest grain market is a proximity to market accessibility. A farmer residing close to a grain market place is expected to have up-to-date information about agriculture inputs and outputs availability and their prices then the farmer is expected to use improved variety in order to enhance grain production.

In the third category of explanatory variables of hybrid maize agro-ecological characteristics were adoption. Environment, namely altitude (ALT) and drought (DRGHT) are expected to shape crop production thereby influence improved variety adoption. In Ethiopia altitude is the outstanding feature that alter both temperature and rainfall consequently crop production. Hence, raise in altitude is expected to positively influence hybrid maize adoption in the CVR of Ethiopia. Recurrent drought is a major challenge of crop production in Ethiopia particularly in drought prone areas. Once every ten years a severe drought occurs in the country (Nigussie et al., 2002) and it recurs once in three to four years in the CRV of Ethiopia. Therefore, to incorporate the risk associated with maize production, the number of severe drought encountered (drought caused a significant maize harvest stress) was included to examine farmers' behavior in such circumstances (Table 1).

RESULTS

Description of hybrid maize producing farm households

Female-headed households constituted 14.5% of the 277 respondents. On average, a household was composed of seven persons headed by 41 years-old adult with a low level of formal education (about two years). Farmers had long experience (about 24 years) in maize production with mixed crop and livestock production systems. The land holding was 2.36 ha per household. The major livestock were found to be cattle, goat, sheep and donkey. Maize receives 46% of farm area allocated to crops and covered 60% of the total production of the area. Oxen were the critical source of draught power for crop production. On average basis, farmers in the study

area had slightly less than a pair (1.8) of oxen.

Maize varieties produced in the Central Rift Valley of Ethiopia

A number of different maturity groups of maize varieties were found to be cultivated in the CRV of Ethiopia. Medium maturing improved OPVs and hybrid were dominantly grown. OPVs have been recommended for commercial production in the area. Among the OPVs, the varieties released by MARC were the dominant ones. OPVs are preferred because the seed can be recycled, and their seeds production cost is cheaper than that of hybrid. The popular OPVs in the study area showed variable maturity dates. Some varieties were very early and can escape peak season and terminal drought stresses.

The OPVs had disadvantages of lower yield than that of the hybrids; and showed non-uniform performances among individual plants in the population. Using high inputs for the production of OPV maize can reduce farmers' profit margin due to its lower yielding potential. On the other hand, hybrids were found to be high yielders than OPVs. Under suitable conditions, hybrids show uniform grain color, maturity, and other plant characteristics. Hence, hybrids usually have better market advantage when sold with recommended quality standards. The empirical studies showed that there is a significant yield differences between hybrids and OPVs under farmers' condition. The current study showed highly significant yield differences (P < 0.001) between OPVs and hybrids. Average yield obtained from OPV was 2.3 tons per hectare while that of hybrid was 3.7 tons per hectare, indicating that hybrids had more than 50% yield advantage over the OPVs. Over the last ten years, an average of 2 to 3 varieties (minimum 1 and maximum 7) were grown by individual farmer while a total of 37

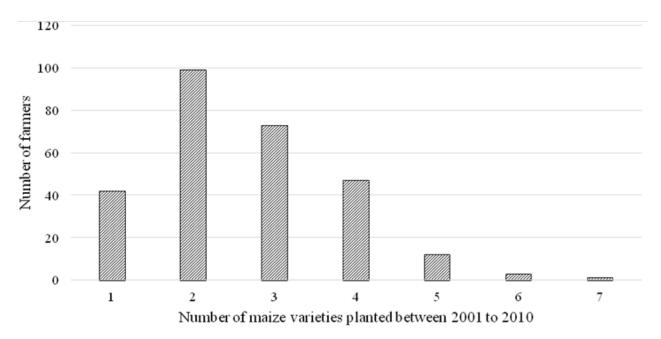


Figure 2. Number of maize varieties grown by farm households in the Central Rift Valley of Ethiopia between 2001 and 2010

Table 2. Characteristics of maize varieties grown in the Central Rift Valley of Ethiopia.

| Variety | Release Year | Altitude (m)('00) | Rainfall (mm)('00) | Days to maturity | Average yield on research station (t/ha) | Per cent of farmer who has grown the variety in past 10 years ‡ |
|-----------------------|-----------------|----------------------|-----------------------|---------------------|---|--|
| Hybrids | | | | | | |
| BH-540 | 1995 | 10-20 | 10-20 | 145 | 9.0 | 25 |
| BH-543 | 2005 | 10-20 | 10-12 | 151 | 9.8 | 7 |
| BH-660 | 1993 | 16-23 | 10-15 | 160 | 10.5 | 1 |
| Zama | 2009 | 6-17 | 4.5-12 | 135 | 11.0 | 3 |
| Wolel (Phb30v53) | 2006 | 10-20 | 8-12 | 163 | 9.0 | 3 |
| Open pollinated varie | ties | | | | | |
| Awassa-511 | 1973 | 10-19 | 6–9 | 142 | 5.6 | 31 |
| Bukuri | na | na | na | 135 | 3.2 | 1 |
| Ghibe | 2001 | 10-17 | 9-12.5 | 145 | 7.4 | 3 |
| Guto | 1988 | 10-17 | 8-12 | 126 | 4.0 | 2 |
| Hararghe | na | na | na | 150 | 3.7 | 7 |
| Hizbawi nuro | na | na | na | 150 | 3.5 | 3 |
| Katumani | 1974 | 12-17 | 5-7 | 110 | 3.1 | 27 |
| Limat | na | na | na | 150 | 3.4 | 14 |
| Marid | na | na | na | 150 | 4.0 | 6 |
| Melkassa-1 | 2001 | 5-16 | 4.5-7.5 | 85 | 4.5 | 7 |
| Melkassa-2 | 2004 | 12-17 | 6-8 | 130 | 5.0 | 25 |
| Melkassa-4 | 2006 | 10-16 | 5-7 | 105 | 4.0 | 2 |
| Melkassa-6Q | 2008 | 10-17.5 | 5-8 | 120 | 5.0 | 3 |
| Milisha | na | na | na | 150 | 3.6 | 21 |
| Shaye | na | na | na | 150 | 3.6 | 27 |
| Sinde | na | na | na | 120 | 3.0 | 5 |

na= data not available; ‡ the figure in the column do not add up to 100% since one farmer plant more than variety over this period.

0.00

51.79

| Variables | All respondents (n=277) | Non-adopters (n=193) | Adopters (n=84) | F | p-value |
|---|-------------------------|-------------------------|--------------------|-------|---------|
| Gender of the household head (% male) | 0.84(0.36) ^a | 0.81(0.39) | 0.92(0.28) | 4.80 | 0.03 |
| Age of the household head (year) | 41.1(12.7) | 42.6(12.6) | 37.8(12.2) | 8.56 | 0.00 |
| House hold size (persons) | 6.7(3.2) | 6.3(2.8) | 7.6(3.7) | 9.49 | 0.00 |
| Formal education (year) | 1.9(3) | 1.1(2.3) | 3.6(3.5) | 45.90 | 0.00 |
| Farmland size (ha) | 9.4(5.8) | 8.9(5) | 10.7(7.3) | 5.97 | 0.02 |
| Maize area proportion (%) | 0.46(0.22) | 0.44(0.2) | 0.57(0.25) | 39.41 | 0.00 |
| Oxen owned (number) | 1.8(1.3) | 1.5(1.2) | 2.3(1.5) | 17.43 | 0.00 |
| Extension visit (number per month) | 2.4(1.6) | 2.7(1.6) | 1.8(1.2) | 19.73 | 0.00 |
| Field day participation (% yes) | 0.2(0.4) | 0.2(0.4) | 0.3(0.4) | 3.86 | 0.05 |
| Time to the nearest grain market (minute) | 94(51) | 100(53) | 82(44) | 7.46 | 0.01 |
| Drought encounter (frequency in 10 years) | 2.7(1.4) | 2.9(1.4) | 2.3(1.3) | 9.34 | 0.00 |

1530(127)

1561(118)

Table 3. Descriptive summary of explanatory variables included in hybrid maize adopters and non-adopters.

Altitude (masl)

varieties including both improved varieties and land races were grown over the past ten years. About 85% of the farmers grew more than one variety (Figure 2). At the time of this study, the majority of the farmers were producing improved varieties and the total number of varieties grown by the farmers' were 26. Table 2 shows major maize varieties grown in the CRV of Ethiopia during the study period.

Explanatory variables of hybrid maize adoption in CRV

Table 3 presents the explanatory variables in hybrid maize adoption. Variables that showed a significant difference in the descriptive statistics included in the Logit model in the estimation of hybrid maize adoption. The rate of hybrid maize adopters was 30%. Significant differences were found between hybrid maize adopters and non-adopters by gender, age, year of formal education completed by the household heads when resource endowments were Households headed by male, younger and the more literate person were found to be more adopters than households headed by female, older and illiterate counterparts.

Hybrid maize adopters leave close to grain market center as compared to non-adopters. There was also a significant difference in extension visit between adopters and non-adopters that the frequency of visit was considerably less among adopters as compared to nonadopters. Moreover, hybrid maize adopters were clearly distinguishable by higher proportion of farmland allocated to maize production. Adopters tend to live in relatively higher altitude areas as compared to non-adopters; and hence, altitude was strongly associated with hybrid

Multicollinearity analysis adoption. among the independent variables using the Variance Inflation Factor (VIF) showed absence of multicollinearity problem as the highest VIF value was 1.7, which was far less than the threshold level of 10 (Table 3).

1631(38)

Logit model estimate of hybrid maize adoption in the CRV of Ethiopia

The variables included in the Logit model showed the expected signs, except for the frequency of extension visit (Table 4). Six variables found to be statistically highly significant (at 5% or less) while two variables were marginally significant (at 10% or less) in explaining farmer adoption of hybrid maize variety.

The model was highly significant as indicated by likelihood ratio χ^2 statistics of 144.08 (P <0.001). The likelihood ratio χ^2 statistic test signifies the dependence of adoption of hybrid maize on the explanatory variables captured in the model. The Logit model estimation has shown that the independent variables simultaneously associated with the log odds of hybrid maize adoption. The null-hypothesis that all the variables can be dropped from the model is rejected at less than 1% level of significance since the Wald'sx2statistic is 62.38 (P<0.01).

Regarding human and physical resource endowment, gender did not show significant association with the adoption of hybrid maize. Age had the expected negative association with log odds of hybrid maize adoption and it had significant influence on hybrid maize adoption (Table 4). Hybrid maize adopters were distinguishable by their age since the adopters found to be significantly younger than non-adopters.

The size of farmland owned and percent of farm area

^aFigures in the bracket indicate standard deviations.

Table 4. Logit model estimates for hybrid maize adoption in drought prone areas of CRV of Ethiopia (N=277).

| Variables | β | Wald's χ ² | Ехр (β) | ∆% adoption |
|---|----------|-----------------------|------------------------------|-------------|
| Constant | 20.52*** | 21.18 | 0.000 | |
| Gender | 0.05 | 0.01 | 1.05 | |
| Age | -0.03* | 3.63 | 0.97 | -3.3 |
| Household size | 0.06 | 0.82 | 1.06 | |
| Formal Education | 0.13* | 3.26 | 1.14 | 13.6 |
| Farmland size | 0.08* | 3.64 | 1.08 | 7.9 |
| Maize area percent | 0.04*** | 22.20 | 1.05 | 4.5 |
| Oxen owned | 0.22 | 1.76 | 1.25 | |
| Extension visit | -0.39*** | 7.23 | 0.68 | -32.3 |
| Field day participation | 0.50 | 1.12 | 1.64 | |
| Time to the nearest grain market | -0.01** | 6.37 | 0.99 | -1.0 |
| Drought encounter frequency (in 10 years) | -0.13 | 0.80 | 0.88 | -12.2 |
| Altitude | 0.01*** | 20.01 | 1.01 | 1.2 |
| Likelihood ratio χ ² (12) | | 144.08 | Cox and Snell R ² | 0.40 |
| Wald's χ^2 (12) | | 62.38 | Nagelkerke R ² | 0.57 |
| Overall Percentage prediction | | 83.8 | | |

^{***, **, *} indicate statistical significance at 1%, 5% and 10%, respectively.

allocated to maize production were positively associated with log odds of hybrid maize adoption. The likelihood of adoption of hybrid maize increase with the proportion of farmland allocated to maize that was statistically highly significant (P<0.001). Accordingly, the log odds was in favor of adopting hybrid maize increase by 4.5% for every 1.0% increase in the farmland allocated to maize among non-adopters. The average walking time required to get to the nearest grain market was negatively associated with log odds of hybrid maize adoption. In this aspect, improving rural road and transportation means can help in enhancing hybrid maize adoption. Extension visit showed negative association with log odds of hybrid maize adoption which was statistically highly significant (P =0.04). Altitude found to be highly influential in hybrid maize adoption. Every one-meter raise in altitude is associated with 2% increase in log odds of hybrid adoption.

DISCUSSION

Adoption of an improved crop variety is a dynamic process of a farmers 'decision to include an innovation in the existing farming system (Feder and Umali, 1993). The decision process considers a number of factors. Several studies were devoted to analyzing the factors influencing the adoption of agricultural innovation such as improved crop varieties and production information. The studies considered human and physical resource endowments, institutional and environmental factors in their analysis. Beginning with gender, Tura (2010) from Ethiopia and Ouma (2002) from Kenya pointed out a negative

association between a female headed household and improved maize variety adoption in high potential areas (area with more than 900 mm rainfall). On the other hand, Morris (1999b) and others (Morris et al., 1999a; De Groote et al., 2002) did not observe a significant influence of gender on improved maize variety adoption.

Although age is an important factor in the adoption of improved varieties, previous studies showed inconsistent results of its effects. Some investigators reported nonsignificant influence of age on adoption of improved variety (Paudel and Matsuoka, 2008; Cavane, 2009; Alene et al., 2000) while Ensermu et al. (1998) and Kaliba (1998) found strong positive association between age and improved variety adoption. In line with the current study, Morris et al. (1999a) and Fufa and Hassan negative influence of age on (2006) report found adoption. A year increase in age of a farmer from average was accompanied by a 3.3% decrease in the log-odds of hybrid maize adoption. Hybrid adopters tend to be younger, and have longer year of formal education suggesting that the young and literate farmers have a tendency to take more risk, seek and utilize innovations than the older farmers. This indicates that younger farmers try to maximize their harvest by growing high yielding varieties such as hybrids. Moreover, in the near future, demand for hybrid is expected to increase in Ethiopia, as more number of young and educated people will join crop farming while the land holding size is squeezing per head as the population is growing at an alarming rate (2.6% per year).

Education of the farm household head tends to significantly influence hybrid maize adoption. This result is in line with the finding of Tura et al. (2010), Paudel and

Matsuoka (2008) and Feleke and Zegeye (2005), who reported that there was significant influence on improved maize variety and related innovation adoption. The result, however, in contrast with the finding of Fufa and Hassan (2006) and Kaliba et al. (1998) who find no meaningful influence of years of formal education on improved crop variety adoption. According to the present study, every single year of additional formal schooling increased hybrid maize adoption by 13.6% keeping other variables constant at their means.

Likewise, hybrid maize adoption found to be influenced by the distance of a farmer's residence from local grain market center. For every 10 min increase in walking time to the nearest grain marketplace, the log odds of hybrid maize adoption decreased by 10%. The result supported the theoretical hypothesis and the findings of Feleke and Zegeye (2005). The present study is in contrast with empirical results of Tura et al. (2010) and Cavatassi et al. (2010).

Hybrid maize adopters were clearly distinguishable by the proportion of farmland they allocate to maize suggesting that the adopters tendency of specialization in maize production, particularly the hybrid varieties. The adopters allocated 57% of their farmland to maize as compared to only 44% by the non-adopters. Likewise, every 1.0% increase in the land allocated to maize was associated with a 4.5% increase in the hybrid maize adoption. This suggested production specialization and market orientation of hybrid maize adopters. Landholding size is a wealth indicator of a farm household among physical resources; and supposed to have a positive influence on improved variety adoption. However, the exiting evidences provide mixed results from those which show absence of considerable influence of landholding (Fufa and Hassan, 2006; Gemeda et al., 2001; Degu et al., 2000; Ntege-Nanyeenya et al., 1997) to those who substantiated positive and meaningful influence on improved variety adoption (Alene et al., 2000; Tura et al.,

Ox is an essential farm household asset and draught power source in Ethiopia. The influence of oxen ownership on improved variety adoption is mixed. Like many previous studies (Gemeda et al., 2001; Tura et al., 2010; De Groote et al., 2002; Feleke and Zegeye, 2005), the present work found no meaningful influence of oxen on improved maize variety adoption. On the other hand, Degu et al. (2000) on the other hand established a positive and significant influence of oxen ownership on hybrid maize adoption. The apparent reason for the absence of meaningful influence of oxen on adoption might be attribute to availability of nearly a pair of oxen that is sufficient to plough the average farmland size of about two and a half hectares owned by a farm household.

Extension visit believed to provide direct and meaningful influence on crop technology adoption. The result of extension visit provides contrasting results from

negative result though absence of significant influence to positive influences. A number of studies (Paudel and Matsuoka, 2008; Feleke and Zegeye, 2005; De Groote et al., 2002; Alene et al., 2000) proved a positive and meaningful influence of extension visit on crop variety adoption.

Some investigators, for instance, Gemeda et al. (2001) found that extension visit had positive influence on adoption of hybrid maize variety but negative effect on OPV adoption in Western Ethiopia. Ntege-Nanyeenya et al. (1997) find no significant association between extension visit and adoption of agricultural innovations.

The present study also in support of the group those who reported negative association but with no significant association. Differing from the widely held beliefs and empirical literature, extension visits had negative and statistically significant influence on hybrid maize adoption.

The plausible explanation is that extension workers in the study area do not provide advice on hybrid maize production as the hybrids were mainly recommended for high moisture areas. For drought prone areas of the CRV, OPVs were recommended; and hence, extension workers have been focusing on the promotion of those varieties. This is further confirmed by the fact that most varieties released by Melkassa Agricultural Research Center, the main maize breeding center for drought prone areas, are OPVs.

In the current study, field day participation did not have influence on hybrid maize adoption. This result is in agreement with the work of Ouma et al. (2002) who reported effective influence of field day participation on improved variety demonstration enhances variety adoption. This result is in contrast with the findings Gemeda et al. (2001) which established direct and strong relationship between maize adoption and field day participation.

Hybrid maize adopters were found to reside in mid to high altitude areas as compared to non-adopters. This attributes to the increase in moisture availability in higher altitudes as compared to the lowland areas. The available hybrids require higher moisture to express the potential productivity.

Likewise, every meter rise in an altitude is associated with 0.9% increase in the log odds of adoption of hybrid maize. This result agrees with the findings of Kaliba et al. (1998) who reported a negative association between hybrid maize adoption and low altitude and low rainfall areas in Tanzania. Contrary to the current findings, Cavatassi et al. (2010) reported that agro-ecology does not show any influence on the adoption of modern sorghum varieties.

The adoption rate of hybrid maize in the study area was substantial (30%), suggesting that maize breeding and extension works for drought prone areas should give a due attention to the development and dissemination of high yielding and well adapted hybrid maize varieties to

benefit smallholder maize producers in the drought prone areas of CRV of Ethiopia.

CONCLUSION AND IMPLICATIONS

This paper examined the factors influencing farmers' hybrid maize adoption in CRV of Ethiopia. The study was conducted in drought prone areas where improved variety adoption study is rarely exist and hybrid maize variety development received little attention. The factors influencing adoption of hybrid maize varieties were discussed considering farmers' resource endowment, institutional and environmental factors.

Different factors found influencing the adoption of hybrid maize. Age of farm household head, year of formal education of household head and distance from the local grain market substantially influenced adoption of hybrid maize. Contrary to the commonly held beliefs and many empirical results, extension visit found to have negative influence on adoption of hybrid maize. This could be because extension workers in the study area were promoting the production of OPVs rather than hybrids. A reasonable proportion -about one-third of the farm household grew hybrid maize in drought prone environments of the CRV of Ethiopia though research and extension services tend to be reluctant in the development and promotion of hybrid maize in the area lest of drought influences. However, a range of maize varieties are produced including the hybrids and farmers who produce hybrids tend to specialize in maize farming as they allocate larger proportion of their farm plots to maize. Likewise, hybrid production will give an opportunity for private seed sector involvement in seed supply system in the drought prone areas, who had been reluctant to get involved in seed provision of OPVs. Similarly, farmer research group approach that has been used and found effective for promotion of OPV maize and other crops can be employed to enhance the dissemination and adoption of hybrid maize in the CRV of Ethiopia. Considering the level of acceptance and of the factors affecting the adoption of improved maize varieties, maize research, maize seed production and maize extension need to react and pursue proactive measures in providing hybrid maize varieties to smallholders' farmers.

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Full Length Research Paper

Evaluation of neem leaves-based preparations as insecticidal agents against the green peach aphid, *Myzus persicae* (Sternorrhyncha: Aphididae)

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Myzus persicae Sulzer, 1776 (Sternorrhyncha: Aphididae) is an insect pest of several crops. Chemical control against pests causing harmful effects, so it is necessary to find alternative methods. In this context, insecticidal activity of water and hydroethanol extracts of neem leaves was investigated against the aphid M. persicae through biological performance and feeding behavior assessment. Water and hydroethanol extracts of neem leaves are prepared by soaking ground leaves in water or 10% ethanol overnight. Both water and hydroethanol extracts of neem leaves at 1, 5 and 10% in artificial diet significantly reduced (p < 0.05) the survival of nymphs, leading to more than 95% mortality. Both extracts at 0.1% significantly reduced (p < 0.05) the survival rate and fecundity of M. persicae adults. The feeding behavior of aphids was studied with a dual-choice assay and using the electrical penetration graph (EPG) technique. Dual-choice assays revealed the aphid rejection of the water extract at 10% and the hydroethanol extract at 1 and 5% concentrations. EPG monitoring showed enhanced duration of probing and ingestion on artificial diet containing the water extract at 10%. On plants sprayed with water extract at 10%, EPG monitoring showed reduced duration of probing, delayed phloem access, and reduced salivation and ingestion phases. Our results showed that neem leaves-based preparations are insecticidal agents and effective to control M. persicae.

Key words: Neem leaves extracts, *Myzus persicae*, survival, feeding behavior.

INTRODUCTION

Crop damages caused by aphids (Sternorrhyncha: Aphididae) is one of the most serious problems in

agriculture. These sap suckers may directly alter plant metabolism through removal of phloem sap and injection

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of salivary secretions inducing morphological changes, sap modification and various local as well as systemic symptoms (Giordanengo et al., 2010) like discoloration of organs, necrosis and windings of leaves. However, the most important damages caused by aphids are indirect. There stem from many phytopathogens transmitted to plants causing severe yield losses (van Emden and Harrington, 2007). Distributed worldwide, the peach potato aphid *Myzus persicae* (Sulzer) is a highly polyphagous species capable of infecting plants in more than 40 different plant families including many economically important plants like peach, potato and cabbage on which it can vector up to 100 phytoviruses (van Emden and Harrington, 2007).

To date, control of aphids mainly relies on the use of pesticides but the rise of environmental concerns (Devine and Furlong, 2007) and the emergence of insecticide resistance risk (Foster et al., 1998; Anstead et al., 2005) have led to search of alternative strategies to control population outbreaks.

Insecticidal properties of neem (Azadirachta indica A. Juss, Meliaceae) are traditionally used in cultural practices from several thousand years (Philogène et al., 2003). Neem compounds present various effects ranging from repellency to toxicity against a wide spectrum of pests including Orthoptera, Lepidoptera, Coleoptera, Diptera and Hemiptera (Schmutterer, 1990; Isman, 2006; Siddiqui et al., 2009; Degri et al., 2013; Shannag et al., 2014). These biological properties are mediated by different groups of compounds among which limonoids and particularly azadirachtin mainly present in the neem seeds are considered the most active components responsible of both antifeedant and insecticidal effects (Isman, 2006). Neem-based insecticides have low environmental impact because of a rapid degradation in plants and in the soil (Isman, 2006) and low effects on beneficial insects (Tang et al., 2002; Haseeb et al., 2004; Defago et al., 2011). Moreover, azadirachtin has been proved non-toxic to vertebrates (Mordue, 2004; Isman, 2006) and therefore neem extracts represent a valuable tool to control population outbreaks in integrated pest management programs.

The problem is that several commercial formulations containing azadirachtin are available on the world market for insect control (Boursier et al., 2011) but refined products are too costly for developing countries (Isman, 2006; Boursier et al., 2011). Aqueous extract of seeds is traditionally used in malian cotton fields to fight Hemiptera pests and the pathogens they vectored (Boursier et al., 2011). Despite two fructification periods per year, their discontinuous availability limits the use of seed-based preparations. Interestingly, numerous active compounds including limonoids have also been found in neem leaves (Siddiqui et al., 2000; Afshan, 2002) which extracts were shown to exert insecticidal effects (Brunherotto et al., 2010; Egwurube et al., 2010).

Insecticidal activity of water and hydroethanol extracts prepared from roughly ground neem leaves has been

confirmed to protect cabbage against Lepidoptera (Akantétou, 1990) and Sternorrhynca pests in fields in Togo (Mondédji, 2010). Owing to such potential of neem leaves-based preparations to control insect populations, our hypothesis is that these preparations affect *M. persicae* survival (antibiosis) after ingestion and/or are antifeedant for it by changing its feeding behavior (antixenosis). We have focused our study on two main objectives: 1) to evaluate aphicidal (antibiosis) efficiency of neem leaves extracts and 2) identify their nature (antixenosis) to assess the process of preparing neem leaves extracts to enable producers to produce by themselves and throughout the year.

MATERIALS AND METHODS

Plants and insects

Potato plants, *Solanum tuberosum* L. (Solanaceae) cv. Désirée, were grown from tubers in 9 cm plastic pots filled with peat mossbased potting medium in a growth room maintained at $20 \pm 1^{\circ}$ C, 65 \pm 5% H.R. and L16:D8 photoperiod. The peach potato aphid, *M. persicae* Sulzer, colony was started from a single virginiparous female collected in early summer 1999, from a potato field near Loos-en-Gohelle, France (50°27'27'N, 2°47'30'E). Aphids were reared in a separate growth room on potted *S. tuberosum* plants at $20 \pm 1^{\circ}$ C, $65 \pm 5\%$ H.R. and L16:D8 photoperiod.

Elaboration of leaves extracts and diets

Neem, *A. indica* A. Juss. (Meliaceae), leaves were collected on neem trees in the Lomé University campus. Extracts were obtained by soaking 1 kg of grinded fresh leaves in 1.5 L water or 10% hydroethanol solution overnight at 25 to 30°C. The preparations were then filtrated through a Whatman No 1 filter paper to obtain the crude neem leaves water or hydroethanol extract (hereafter referred as W or H, respectively).

An artificial diet adapted for *M. persicae* was used as a carrier for neem extracts dilution and as a negative control (C). Artificial diet added with 10% ultrapure water (CW) or 10% hydroethanol solution at 10% (v/v with ultrapure water) (CH) was used as positive control. The diet was prepared as described by Febvay et al. (1988) and modified by Down et al. (1996). W or H was incorporated to the artificial diet to obtain diets W0.1, H0.1, W1, H1, W5, H5, W10 and H10, containing respectively 0.1, 1, 5 and 10% (v/v) of W or H. After preparation, the diets were passed through a 0.2 µm filter (Millipore Corp., Bedford, Massachusetts, USA). Parafilm® pouches (80 µl) were prepared under aseptic conditions.

Effects of neem leaves extracts on aphid demographic parameters

Five nymphs younger than 24 h were transferred to a new pouch of each diet (C, CW, CH, W0.1, H0.1, W1, H1, W5, H5, W10 and H10) and maintained under the same rearing conditions as described in the section "Plants and insects". Diet pouches were changed every second day. Ten replicates were carried out for each diet. Nymphal survival, prereproductive period (that is, the period of time from birth until onset of reproduction), adult emergence Le Roux et al. (2004), reproduction and survival were recorded every 2 days according to the Jackknife method (Meyer et al., 1986) was used to evaluate the variance of the intrinsic rate of natural increase $(r_{\text{m}}, \sum_{e^{-r_{\text{m}}x}, lx, mx = 1})$

with the DEMP 1.5 software (Giordanengo, 2010).

The effects of neem leaves extracts on nymphal survival were analysed with Pearson's χ^2 test. One-way analysis of variance (ANOVA) was carried out to test the effects of the diets on aphid demographic parameters. Significantly altered demographic parameters were further analyzed with Fisher's positive least significant difference test (PLSD). To determine the LD₅₀, dose-mortality relationship was determined using WIN DL (CIRAD-CA/MABIS, Montpellier, France), based on probit analysis (Finney, 1971).

Effects of neem leaves extracts on aphid feeding behavior

Dual-choice assay

The dual-choice assay was realized using the device described by Sauvion et al. (2004). It consisted of Plexiglas® built-in cylindrical boxes (7.2 mm diameter, 6 mm high) closed by two pouches of diet (20 μ l) diametrically opposite. One wingless adult aphid aged less than 24 h was inserted in the device and aphid position relative to the pouches was recorded after 6 h. The experiment was conducted in the dark in a growth room maintained at 20 \pm 1°C. The diets (that is, CW, CH, W0.1, H0.1, W1, H1, W5, H5, W10 or H10) and the control (C) were alternated and the device cleaned with TFD4 detergent (Franklab, St-Quentin en Yvelines, France) between two replications. Thirty six replicates were done for each treatment. For each experiment, the distribution of responding aphids was analyzed using a Wilcoxon test (Z) for paired samples. We used a Kruskal-Wallis test (H) to compare the percentages of non-responding aphids between experiments.

Stylet activities

Stylet penetration activities during aphid feeding behaviour were studied using the DC electrical penetration graph (EPG) (Tjallingii, 1988). A 2 cm gold wire (20 µm in diameter) was pasted on the aphid's dorsum by conductive silver glue. For *in planta* experiments, three-weeks old potted plants were treated using a hand-held trigger spray (Pulsar 1 L, Tecnoma, Epernay, France) to apply 6 ml of W (PW), 10% W diluted with distilled water (v / v) (PW10) or distilled water as control (PC) to each plant. A treated plant (PW, PW10 or PC) was connected to the system via a copper electrode stuck in the potting medium. Each connected aphid was carefully placed on the abaxial surface of the fourth fully expanded leaf of the potato plant and monitored for 8 h. For *in vitro* experiments, the second electrode was inserted through the Parafilm® into a pouch for 4 h

The feeding behavior of eight wingless adult aphids aged less than 24 h, each feeding on a separate plant or pouch, were monitored simultaneously using a Giga 8[™] amplifier with 10⁹ Ω input resistance (EPG-systems, Wageningen, The Netherlands) in an electrically grounded Faraday cage to shield the setup from external electrical noise. All experiments were carried out in a growth room maintained at 20 ± 1°C. At least 18 aphids were monitored for each treatment. The recordings consistently started around 10:00 h (±30 min). Data acquisition and analyses were done with PROBE 3.5 software (EPG-systems, Wageningen, The Netherlands), according to Tjallingii and Hogen Esch (1993). Eighteen to 21 replications were performed for each series. Waveforms A, B, C, and pd (potential drop, that is, brief intracellular puncture) were grouped and labelled as 'C pattern' corresponding waveforms, E1 (salivation into the sieve elements), E2 (passive to the advancement of the stylets in the plant tissues. Other ingestion of phloem sap), G (active ingestion of xylem sap), and F (derailed stylet mechanics) were also identified. Electropenetrography parameters were calculated using EPG-CALC 4.9 software (Giordanengo, 2014).

Fifteen parameters organized into five classes (Table 3) were chosen to describe the effects of plant treatments on the probing behavior of *M. persicae*. The 'general probing behavior' class included the mean number of probes, the total duration of probing and the time from the beginning of the monitoring to the first stylet insertion. The 'pathway phase' class corresponded to probing activities in non-vascular tissues and included the mean number and the total duration of pathway phases. The 'phloem salivation' class included the number and the total duration of salivation bouts with a distinction between single phloem salivation periods and phloem salivation periods followed by sap ingestion, and duration from the first probe before the first salivation. The 'phloem ingestion phase' class included the mean number and the total duration of phloem sap ingestion bouts, and the duration from the first probe to the first ingestion. Fifth class, 'other parameters' included the total duration of xylem ingestion and stylets derailment bouts.

For in vitro experiments, only C, E1 and G waveforms were recorded. The latter corresponding to the active ingestion of the artificial diet (Sauvion and Rahbé, 1999) consistently differed from that observed for aphids feeding on plants since passive phloem sap ingestion (E2) is forced by the pressure in the sieve tubes (Miles, 1999; Tjallingii and Cherqui, 1999). To describe the effects of W10 diet on the probing behaviour of M. persicae, we selected 9 parameters organized into three classes (Table 4). The 'general probing behavior' class included the mean number of probes, the total duration of probing and the time from the beginning of the monitoring to the first stylet insertion. The 'salivation phase' class included the number and the total duration of salivation bouts and the total duration from the first probe before the first salivation. The 'ingestion phase' class included the mean number and the total duration of diet ingestion bouts, and the duration from the first probe to the first ingestion. Pair-wise comparison between each treatment and its respective control was performed with a Mann-Whitney *U*-test to analyze the effects of plant treatment or diet on the EPG data. All statistical analyses were performed using STATISTICA 6 software (StatSoft, Tulsa, OK, USA).

RESULTS

Effects of neem leaves extracts on aphid demographic parameters

Nymph survival (Figure 1) was not affected by adding 10% water (CW: p > 0.05) or 10% hydroethanol solution (CH: p > 0.05) (positive controls) in the artificial diet, when compared with the artificial diet alone (C) (negative control). Whatever the dose, both water (W0.1: p < 0.002; W1: p < 0.001; W5: p < 0.001; W10: 3, p < 0.001) and hydroethanol (H0.1: p < 0.01; H1: p < 0.001; H5: p < 0.001; H10: p < 0.001) extracts added to the diet significantly increased M. persicae nymph mortality. LD_{50} as a percentage of crude hydroethanol or water extract was determined at 0.37 and 0.43% (Table 1) with equations of mortality linear regression Y = 0.41 + 0.95X and Y = 0.33 + 0.92X respectively.

Demographic parameters were only calculated for aphids reared on W0.1 and H0.1 diets on which adults were obtained (Table 2). Significantly extended prereproductive period and reduced daily fecundity and

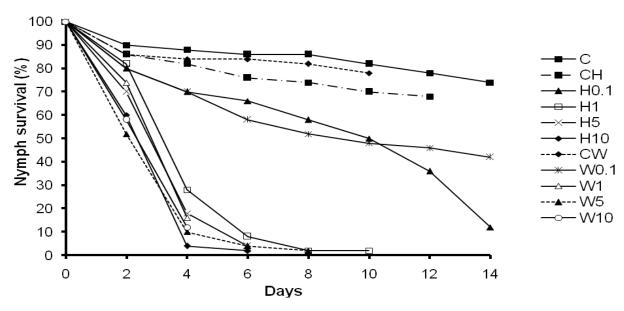


Figure 1. Survival of *Myzus persicae* nymph reared on artificial diet (C) as negative control or artificial diet added with hydroethanol (H) or water (W) neem leaves extract at 0.1 (H0.1 and W0.1), 1 (H1 and W1), 5 (H5 and W5) or 10 (H10 and W10)% (v / v).

Table 1. Dose-mortality relationship with LD50 with inferior and superior limits of the 95% confidence interval for water and hydroethanol extracts of neem leaves delivered via ingestion route to *Myzus persicae*.

| Extracts | Inf lim < LD50* < sup lim | |
|--------------|---------------------------|--|
| Water | 0.011 < 0.434 < 1.085 | |
| Hydroethanol | 0.008 < 0.371 < 0.961 | |

^{*}LD50 is expressed as a ratio of crude neem leaves water or hydroethanol extract diluted artificial diet (v / v).

oviposition period were observed for adults whatever water or hydroethanol extracts added to the diet. Intrinsic rate of natural increase (r_m) and finite ratio of increase (λ) were therefore significantly reduced.

Effects of neem leaves extracts on aphid feeding behavior

Dual-choice assay

A blank test (control vs. control, p > 0.05) confirmed the absence of any bias in the dual-choice setup. Whatever water or hydroethanol dose tested, the percentage of non-responding aphids was not statistically different between experiments (p > 0.05). No positive control (CW, p > 0.05 and CH, p > 0.05) influenced aphid distribution (Figure 2). Neither 0.1, 1 nor 5% concentration of neem water extract in the artificial diet led to a non randomly distribution (W0.1, p > 0.05; W1, p > 0.05; W5, p > 0.05). Rejection of neem water extract occurred at 10%

concentration (W10, p < 0.001). No preference for the 0.1 or 10% neem hydroethanol extract added diets was shown (H0.1, p > 0.05; H10, p > 0.05). *M. persicae* significantly avoided the pouches containing 1 and 5% of neem hydroethanol extracts (H1, p < 0.05; H5, p < 0.05).

Stylet activities

In vitro monitoring: General probing behavior of *M. persicae* was affected on diet added with 10% neem water extract (W10 diet), aphids showing significantly enhanced duration of probing (parameter 2) (Table 3). None of the parameters describing the salivation phase was significantly affected. Ingestion phases were altered on W10 diet as aphids spent more time ingesting diet (parameter 8).

In planta monitoring: On W10 sprayed potato plants (PW10) the total duration of probing, the number of single salivation phases and the total duration of both single and

Table 2. Average (±SE) of demographic parameters for *Myzus persicae* reared on control diet (C) or on diet containing 0.1% (v / v) neem leaves water (W0.1) or hydroethanol (H0.1) extract.

| Demographic parameter | C n=39 | W0.1 n=19 | H0.1 n=14 | F | P |
|---|-------------------------|-------------------------|-----------------------|-------|---------|
| Pre-reproductive period (days) | 10.3 ± 0.1 ^a | 17.4 ± 1.0 ^b | 14.3 ± 0.7^{c} | 43.38 | < 0.001 |
| Daily fecundity (nymphs per female per day) | 0.34 ± 0.03^{a} | 0.06 ± 0.02^{b} | 0.187 ± 0.76^{b} | 11.90 | < 0.001 |
| Oviposition period (days) | 7.53 ± 0.50^{a} | 0.86 ± 0.3^{b} | 1.9 ± 0.8^{b} | 49.32 | < 0.001 |
| Adult survival (days) | 11.3 ± 0.8^{a} | 8.8 ± 0.8^{b} | 6.3 ± 1.0^{b} | 7.83 | < 0.001 |
| r _m (females per female per day) | 0.088 ± 0.006^{a} | -0.064 ± 0.024^{b} | 0.042 ± 0.003^{c} | 28.37 | < 0.001 |

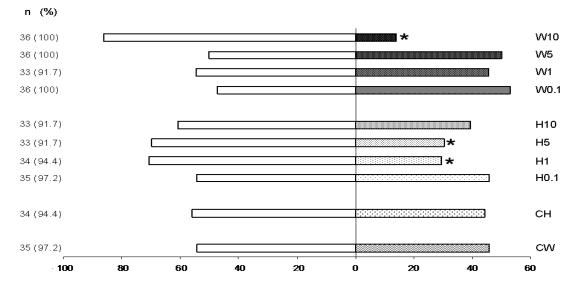


Figure 2. Distribution of *M. persicae* adults in the dual-choice setup after 6h experiment C: negative control (artificial diet); CW: artificial diet added with 10% ultrapure water (positive control); CH: artificial diet added with 10% hydroethanol solution (positive control); H0.1, H1, H5, H10: artificial diet added with hydroethanol neem leaves extract at 0.1, 1, 5 or 10%; W0.1, W1, W5, W10: artificial diet added with water neem leaves extract (v / v).

fraction salivation bouts were reduced (Table 4). When monitored on those plants, *M. persicae* showed a delayed first salivation bout. All the parameters linked to phloem ingestion were

modified: the number and the duration of phloem ingestion phases were reduced and phloem ingestion delayed. Stylets derailment and xylem ingestion were not affected.

DISCUSSION

Both water and hydroethanol extracts intoxication through ingestion route showed dose-dependent

Table 3. Feeding behavior of *Myzus persicae* during 4 h access to standard diet (C) or standard died added with 10% (v / v) neem leaves water extract (W10).

| | С | W10 diet |
|--|---------------|----------------|
| EPG classes and related parameters | n=18 | n=21 |
| General probing behavior | | |
| 1. Number of probes | 16.8 ± 2.9 | 22.1 ± 2.5 |
| 2. Total duration of probing | 137.5 ± 19.0 | 332.6 ± 33.6* |
| 3. Time from start of recording to first probe | 8.0 ± 4.5 | 7.5 ± 3.1 |
| Salivation phase | | |
| 4. Number of salivation phases | 1.9 ± 0.4 | 1.9 ± 0.5 |
| 5. Total duration of salivation phases | 11.0 ± 8.6 | 13.1 ± 7.0 |
| 6. Time from first probe to first salivation | 36.6 ± 11.6 | 40.1 ± 9.3 |
| Ingestion phase | | |
| 7. Number of ingestion phases | 11.6 ± 1.9 | 15.6 ± 2.0 |
| 8. Total duration of ingestion phases | 112.3 ± 14.4 | 239.1 ± 33.5* |
| 9. Time from first probe to first ingestion | 33.3 ± 14.4 | 18.9 ± 18.0 |

Means (\pm SE) followed by * are significantly different from control plants (Mann-Whitney *U*-test: P < 0.05). n, number of aphids; times and durations are expressed in min.

Table 4. Feeding behavior of *Myzus persicae* during 8 h access to leaves of *Solanum tuberosum* cv Désirée sprayed by 10% (v / v) neem leaves water extract diluted in ultrapure water (PW10) or ultrapure water as control (PC).

| | Control (PC) | PW10 |
|---|------------------|-----------------|
| EPG classes and related parameters | n=22 | n=21 |
| General probing behavior | | |
| 1. Number of probes | 46.9 ± 4.5 | 40.6 ± 7.8 |
| 2. Total duration of probing | 303.4 ± 19.1 | 214.8 ± 23.8* |
| 3. Time from start of recording to first probe | 12.0 ± 2.7 | 40.6 ± 13.7 |
| Pathway phase | | |
| 4. Number of pathway phases | 52.8 ± 4.7 | 43.3 ± 6.0 |
| 5. Total duration of pathway phases | 188.2 ± 13.3 | 167.8 ± 21.0 |
| Phloem salivation phase | | |
| 6. Number of single salivation periods | 3.9 ± 0.6 | $1.2 \pm 0.4^*$ |
| 7. Total duration of single salivation phases | 18.2 ± 3.5 | $9.0 \pm 3.8^*$ |
| 8. Number of fraction salivation phases | 2.1 ± 0.5 | $1.0 \pm 0.4^*$ |
| 9. Total duration of fraction salivation phases | 11.0 ± 2.5 | $3.5 \pm 0.6^*$ |
| 10. Time from first probe to first salivation | 128.3 ± 29.1 | 269.9 ± 36.8* |
| Phloem ingestion phase | | |
| 11. Number of phloem ingestion | 1.9 ± 0.4 | $0.8 \pm 0.3^*$ |
| 12. Total duration of phloem ingestion | 30.9 ± 11.5 | $4.6 \pm 1.6^*$ |
| 13. Time from first probe to first phloem ingestion | 228.9 ± 37.9 | 342.9 ± 31.8* |
| Others parameters | | |
| 14. Total duration of stylet derailment | 13.7 ± 6.1 | 2.2 ± 0.4 |
| 15. Total duration of xylem ingestion | 44.7 ± 11.4 | 32.3 ± 5.4 |

Means (\pm SE) followed by * are significantly different from control plants (Mann-Whitney *U*-test: P < 0.05). n, number of aphids; times and durations are expressed in min.

effects of neem leaves extracts leading to more than 95% *M. persicae* nymphs mortality for 1, 5 and 10% doses. A drastic reduction of nymph survival caused by

azadirachtin or neem seed extracts was reported on several aphids species (Lowery and Isman, 1996; Tuncer and Aliniazee, 1998; Tang et al., 2002; Pavela et al.,

2004) among which M. persicae (Lowery and Isman, 1996). Such mortality of immature stages may result, at least partly, from the molting inhibition property of azadirachtin (Mordue and Blackwell, 1993; Tang et al., 2002; Isman, 2006) as surviving M. persicae nymphs grown on diet added with neem leaves extracts were smaller and most of them did not develop into adult. Amtul (2014) reported A. indica derived compounds as inhibitors of digestive alpha-amylase in insect pest Tribolium castaneum. When M. persicae nymphs developed into adults (that is, only for 0.1% neem leaves water or hydroethanol extracts treatments), enhanced pre-reproductive period was observed as reported on the filbert aphid Myzocallis coryli (Tuncer and Aliniazee, 1998) and the brown citrus aphid Toxoptera citricida (Tang et al., 2002) intoxicated with neem seed extracts.

Adult intoxicated with 0.1% neem leaves water or hydroethanol extracts exhibited drastically reduced fecundity (that is, both offspring per aphid per day and oviposition period) and survival. Alteration or even failure of offspring production of aphids exposed to neem seed oil or azadirachtin (Nisbet et al., 1994; Lowery and Isman, 1996; Tuncer and Aliniazee, 1998; Tang et al., 2002; Pavela et al., 2004) was ascribed to embryos mortality just before parturition (Nisbet et al., 1994; Lowery and Isman, 1996). For the 0.1% water extract (W0.1) the negative intrinsic rate of population increase ($r_{\rm m}$) would result in population extinction.

Besides these antibiosis effects, dual-choice assays showed aphids rejection of 10% concentration of neem water extract, and 1 and 5% concentration of neem hydroethanol extracts. Drastic antifeedant effects of neem-treated leaves were reported on various grazing insects such as Lepidoptera (Blaney et al., 1990; Simmonds et al., 1990; Ma et al., 2000), Coleoptera (Streets, 1976), Orthoptera (Mordue et al., 1998) and on aphids (Koul, 1999). Aqueous plant extracts of A. indica showed feeding deterrence on the cabbage butterfly Pieris brassicae (Sharma and Gupta, 2009) and the diamondback moth Plutella xylostella (Charleston et al., 2005), and Singh et al. (1987) reported such antifeedant effect in leaf water extracts on P. brassicae. In vitro EPG monitoring showed enhanced duration of probing and ingestion when aphids fed on artificial diet added with 10% water extract. Ingestion of neem-seed oil does not immediately inhibit feeding (Morgan, 2009). As enhanced duration of ingestion phase monitored by EPG can not be directly linked with an increased amount of ingested diet it could be the consequence of difficulties to ingest as seemed to indicate enhanced probing duration. Such behavior recorded during 4 h would thus lead to reject the intoxicated diet as observed after 6 h in the dual-choice assays. This previous hypothesis is supported by in planta EPG monitoring which showed reduced duration of probing and delayed phloem phase (salivation and ingestion phases), and reduced salivation and ingestion phases indicating repellent properties perceived within

the plant tissues and antifeedant properties perceived in the phloem sap, respectively. Azadirachtin has been shown to act systemically (Isman, 2006; Morgan, 2009), however, taken together *in vitro* and *in planta* monitoring also suggest a deterrent effect perceived on the plant surface by chemosensory sensillae on the proboscis as reduced probing duration was only observed on sprayed plants. Such deterrency of surface treatment to aphids appears highly variable depending on the targeted species (West and Mordue, 1992; Lowery and Isman, 1993; Koul et al., 1997; Koul, 1999) and the nature of the sprayed extract (Koul, 1999).

Though neem leaves are reported a very poor source azadirachtin (Morgan, 2009) numerous active compounds including limonoids have been identified in neem leaves (Siddiqui et al., 2000; Afshan, 2002). The composition of the extracts we dealt with was not investigated but whatever their water or hydroethanol nature, their effects on aphids survival and their LD₅₀ were very similar. Compare to an azadirachtin-based commercial product Boursier et al. (2011) attributed the similar performance of the traditional seed-based water extraction to the presence of other components among which terpenoids. Such easy to made botanical extract with a good insecticidal performance can be prepared throughout the year by the producers themselves in developing countries, where synthetic insecticides are not affordable to growers.

Further works are needed to evaluate storage capacity as plant extracts may lose their activity when having a too long contact with water (Schmahl et al., 2010) and chemical degradation of azadirachtin and derivative limonoids increases with temperature (Barrek et al., 2004) and sunlight exposure (Caboni et al., 2009). However, this work underlines that a well-known botanical insecticide that could be part of integrated pest management strategies in developing countries needs that researchers refocus their attention on its development and application.

Conclusion

Neem leaves extracts tested, reduced survival and reproductive potential of the green peach aphid *M. persicae*. There induced mortality of nymphs throughout ingestion. These extracts showed interesting aphicide properties to *M. Persicae* with dose - response relationships well correlated which were observed. Dualchoice assays showed aphids rejection of neem extracts at some concentrations. EPG monitoring showed delayed phloem phase (salivation and ingestion phases), and reduced salivation and ingestion phases indicating repellent properties perceived within the plant tissues and antifeedant properties perceived in the phloem sap. This work reports that neem leaves-based preparations showed aphicidal (antibiosis) and antifeedant behavior

(antixenosis) properties and was efficient to protect vegetable against Sternorrhynca pest *M. persicae*.

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Conflict of Interests

The author(s) have not declared any conflict of interests.

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Full Length Research Paper

Antibacterial potential of white crystalline solid from red algae *Porteiria hornemanii* against the plant pathogenic bacteria

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This study was investigated by analyzing the potent bioactive compounds in the chloroform extract of *Porteiria hornemanii* using gas chromatography–mass spectrometry (GC-MS) analysis. The crude 50 μ g/ml of white crystalline solid were tested *in vitro* for their antibacterial effect against *Xanthomonas axonopodis* pv. *citri* and *Xanthomonas axonopodis* pv. malvacearum using paper disc diffusion technique showing 10.00 mm and 12.00 zone inhibition respectively, whereas streptomycin sulphate showed 15.00 mm zone and inhibition. The white crystalline solid mainly contained 1,2–Benzenedicarboxylic acid, diisoctyl ester (C_{24} H_{38} O_4) (48.21%) and 1,2–Benzenedicarboxylic acid, bis (2-methylpropyl) ester (C_6 H_{22} O_4) (19.67%).

Key words: Gas chromatography-mass spectrometry (GC-MS), *Porteiria hornemanii*, white crystalline solid, seaweeds.

INTRODUCTION

Intensive application of synthetic pesticides in agriculture caused damage to the ecological state of the agricultural system (Abetz and Young, 1983). Pesticides of biological origin are generally less toxic, affect only the target pest and closely related organisms and are effective in very small quantities which decompose quickly. Published literature reports on the diverse bioactivities of seaweeds, but the antibacterial efficacy of seaweeds against plant pathogens are comparatively a new concept and a few attempts have been made in this regard (Kumar et al., 2008; Kulik, 1995; Arunkumar et al., 2000, 2005; 2010; Arunkumar and Sivakumar, 2012; Manimala and Rengasamy, 1993). Harder (1917) was the first to observe antimicrobial substance in seaweeds. Then until 1970s no large scale screening of antimicrobial activity

was carried out (Welch, 1962; Hornsey and Hide, 1974; Henriquez et al., 1979). The Seaweeds are bestowed with varied source of bioactive natural products that exhibits antimicrobial properties against plant pathogens (Arunkumar et al., 2012; Kulik, 1995; Ara et al., 1998; Kumar et al., 2008).

Seaweeds have been identified as a rich source of bioactive compounds (Arunkumar et al., 2010). Seaweeds constituting an important renewable marine resource occur generally on the rocky substratum in the intertidal and subtidal regions of the coastal waters. Ocean has been recognized as a storehouse of fine chemicals.

Several works have been undertaken on crude and purified compounds obtained from seaweeds for

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evaluating their bioactive potential (Faulkner, 1992). A promising strategy for the replacement of chemical pesticides has been the implementations of chemical pesticides and biological control. The recent development in the commercialization of biological control products has accelerated this approach (Fravel et al., 2003). Algae are one of the chief biological agents that have been studied for the control of fungi plant pathogens (Hewedy et al., 2000; Abdel-Kader, 1997). Hence the present investigation was carried out to determine the possible phyto components from *Porteiria hornemanii* and to analyze the potent bioactive white crystalline solid by GC-MS as the first report among the seaweeds.

MATERIALS AND METHODS

Collection of seaweed

About 1 kg of live, healthy and disease free matured seaweed of red alga *P. hornemanii* occurring along the coast of Pamban near Rameswaram, Gulf of Manner, Tamilnadu, India collected during the post–monsoon season (February) in the year 2005 in spring tide was washed thoroughly in seaweeds followed by tap water to remove extraneous materials and sand particles. Identification of the seaweed was carried out at CMFRI, Mandapam, Tamilnadu.

Seaweeds were collected during post monsoon season because Arunkumar and SivaKumar (2012) reported that this season maximum antibacterial activity are found in seaweeds followed by those collected during monsoon, pre-monsoon and seaweeds followed by those collected during monsoon, pre-monsoon and the summer season. The plant pathogenic bacteria *Xanthomonas axonopodies pv.citri* and *Xanthomonas axonopodies pv.citri* and *Xanthomonas axonopodies pv.malvacearum* causing canker in citrus and angular spot in cotton, respectively used in the present were isolated from the parts of the plant.

Extraction of seaweed

Shade dried algae was pulverized and ground as fine powder. The powdered 100 g of algal sample was extracted with 500 ml chloroform: methanol (1:1v/v) in air tight 1 L Erlenmeyer conical flask at room temperature in dark for 1 month and shaken at intervals daily. Extract was filtered through Whatmann No.1 filter paper. 50 mg of anhydrous MgSO $_4$ was added and shook vigorously for 5 min continuously.

Isolation and crystallization

Then the crude extract was filtered using Whatmann No.1 filter paper and kept for evaporation of the solvent under aseptic dark condition in the laboratory without any disturbance for 2 weeks.

During the course of the 2 weeks time colorless crystals starts forming in the bottom of the flask with thick reddish viscous mass. The thick reddish viscous mass was decanted and 16 mg of white solid crystals were obtained and stored at 0°C until for bioassay and GC-MS study.

Antibacterial activity of crystalline compound

A crystal weighing 1 mg dissolved in chloroform was used for bioassay. The antibacterial activity was conducted through agar disc diffusion technique.

Antibacterial assay

Antibacterial activity was determined against the selected plant pathogens using paper disk assay (Potato textrose agar) method (El-Masry et al., 2000). Control disk also maintained for each extract by impregnate respective organic solvent alone. Nutrient Agar (NA) plates (90 mm) were prepared and overnight broth culture (1.2×108 cfu / ml) of test pathogens were inoculated uniformly. Triplicates were maintained for each test pathogen. The plates were incubated at 37°C for 48 h. The zone of inhibition was measured and expressed in mm in diameter. Streptomycin sulphate was used in bioassay study for comparison.

GC-MS analysis

GC-MS analysis on chloroform extract of red algae P. honemannii, a white crystalline solid sample was carried out in Indian Institute of Crop Processing Technology, Thanjavur, Tamilnadu, India. GC Clarus 500 Perkin Elmer system comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument was used, employing the following conditions: Column Elite – 5MS fused silica capillary column (30 x 0.25 mm 1 D x 1 μ Mdf, composed if 100% Dimethyl polysiloxne), Operating in electron impact mode at 70 ev, helium (99.999%) was used as carrier gas at a constant flow of 1 ml/min and an injection volume of 2 μl was employed (Split ratio of 20:1) injector temperature 250°, ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min), with an increase of 10°/min, to 200°C, then 5°C / min to 280°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5 s and fragments from 45 to 450 Da. Total GC running time is 36 min. The white crystalline solid was dissolved in chloroform and analyzed in GC-MS for different components.

Identification of components

Interpretation of GC-MS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, mole under weight and structure of the components of the test materials were ascertained.

RESULTS AND DISCUSSION

GC-MS: White crystalline components in chloroform extract of *P. hornemanii* by GC-MS report

The presence of chemical components in chloroform extracts of *P. hornemannii* is tabulated (Table 1) and represented by graphical method. The GC–MS analysis resulted in the identification of a total six components in *P. hornemannii*. The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) are found. The prevailing compound was 1,2 – Benzene dicarboxyllic acid, diisoctyl ester (48.21%) and 1,2 – Benzene dicarboxylic acid, bis (2 methyl propyl) ester (19.67%). The zone of inhibition

| Table 1. Antibacterial activity of crystalline solid of red algae <i>Porteria hornemai</i> | Table 1. Antibacterial a | activity of crystalline | e solid of red algae | Porteria hornemanii. |
|---|--------------------------|-------------------------|----------------------|----------------------|
|---|--------------------------|-------------------------|----------------------|----------------------|

| Bacteria | Extract (µg/µl) | Disc diffusion inhibition zone (mm) |
|--|-----------------|-------------------------------------|
| Gram negative Xanthomonas campestries pv.citri | 50 | 12.00 |
| Xanthomonas campestries pv. malvacearum | 50 | 10.00 |
| Streptomycin sulphate (positive control) | 50 | 15.00 |

Table 2. Phyto components in chloroform extract of white crystalline solid from red algae Porteria hornemannii by GC-MS.

| S/No | RT | Name the compound | Molecular formula | MW | Peak area % |
|------|-------|--|---|-----|-------------|
| 1. | 9.55 | Octadecane, 3 – ethyl 5 (2 – ethyl butyl) | C ₂₆ H ₅ O ₄ | 366 | 0.30 |
| 2. | 15.70 | 1,2 - Benzene dicarboxylic acid, bis (2-methyl propyl) ester | $C_{16}H_{22}O_4$ | 278 | 19.67 |
| 3. | 17.24 | N – Hexadecanoic acid | $C_{16} H_{32} O_2$ | 256 | 6.36 |
| 4. | 25.51 | Tert – Hexadecanethiol | $C_{16}H_{34}S$ | 258 | 9.68 |
| 5. | 26.08 | 1,2 - Benzenedicarboxylic acid, diisooctyl ester | $C_{24}H_{38}O_4$ | 390 | 48.21 |
| 6 | 26.96 | Heptacosane | $C_{27}H_{56}$ | 380 | 15.78 |

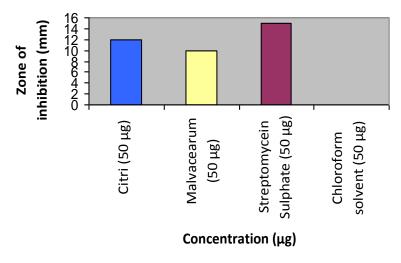


Figure 1. Antibacterial activity of white crystalline solid extract of *Porteria hornemanii* against two plant pathogens.

shown in Figure 1, GC-MS spectrum of chemical compounds shown in Figure 2 and the corresponding chemical shift peaks of the spectrum were shown, whereas the chemical structure is shown in Figure 3.

The chloroform extract, the white crystalline solid of 50 µg/ml showed inhibitory effect on *X. campesteries pv. citri* with inhibition zone of 10.0 mm and *X. campesteries pv. malvacearum* with inhibition zone of 12.00 mm (Table 2). Standard Streptomycin sulphate of 50 µg/ml showed 15.0 mm zone of inhibition. No inhibitory effect was showed by chloroform. This is the first report in the red algae. Marine algae are a rich source of novel bioactive compounds which may find several applications (Aziz et

al., 2003; Delattre et al., 2005; Chandia and Mastsuhiro, 2008). Paulert et al. (2007) also found that methanol extracts have *in vitro* activity against the plant pathogenic bacteria *Erwinia carotovora* and *X. cam*pestris as well as human dermatophyte fungus (*Trichrphton mentagrophytes*).

The thin layer chromatography purified fraction of green seaweed *Cladophora glomerata* was subjected to GC-MS analysis of the chemical constituents against gram negative human pathogenic bacteria (Yuvaraj et al., 2011). The GC-MS analysis revealed the presence of hydrocarbons, fatty acids and cholesterol. Olecic acid (1 g. 58%) and n – hexandecanonic (acid 24.73%) and

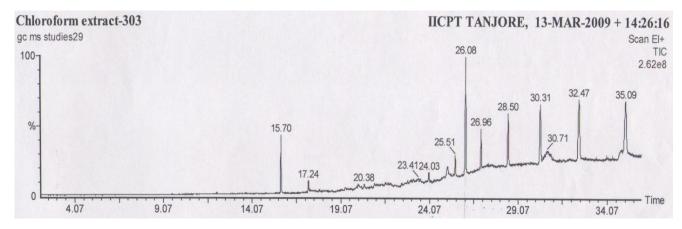
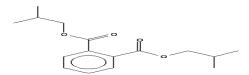


Figure 2. GC-MS analysis spectrum of the photochemical white crystalline solid from the chloroform extract of the red algae *Porteria hornemannii*.

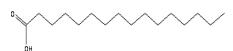
Octadecane, 3 - ethyl 5 (2 - ethyl butyl)



1,2 - Benzene dicarboxylic acid, bis (2-methyl propyl) ester



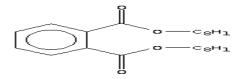
N – Hexadecanoic acid



Tert - Hexadecanethiol



1,2 - Benzenedicarboxylic acid, diisooctyl ester



Heptacosane

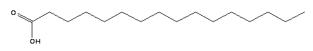


Figure 3. Structures of the white crystalline solid compound.

aromatic dicarboxylic acid were the major components of *Acanthophora spricifera* (Zakaria et al., 2011). Benzenedicarboxylic acid bis (2-ethylhexyl) phthalate has been isolated from a marine alga, *Sargassum weightii*, and apart from its plasticizing ability it was also found to have antibacterial effect on a number of bacteria (Sastry and Rao, 1995).

Conclusion

From the present study, the extraction of major phyto components was observed in chloroform extract of crystalline solid as 1,2 Benzendicarboxylic acid diisooctyl ester (48.21%) of rT 26.08 and 1,2 Benzen dicarboxylic acid, bis (2-methyl propyl) ester (19.37%) rT (15.70), Heptacosane (15.78) rT 26.96 and the components showed potent antibacterial activity against the plant pathogenic bacteria for bio control. This is the frist phytochemical analysis of volatile components performed by GC-MS reported for *P. hornemannii*.

In this study, the crude crystalline compounds and its antimicrobial mechanisms were known and thus further research should be made to identify the single active compound, which would be helpful to the agricultural society.

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Conflict of Interests

The authors have not declared any conflict of interests

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Phenotypical correlations between agronomical characters in Prata type bananas (*Musa*) and its implications on yield estimate

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This research aims to estimate correlations between agronomic traits and yield in Pome bananas, contributing to the elaboration of models for estimation of harvest. Agronomic traits were evaluated in 98 plants of 'Dwarf Prata' and 96 'BRS Platina', considered replicates and sampled at random. Correlations between the masses of the bunch, and hands and plant height; pseudostem perimeter at ground level, at 30 cm and at 100 cm in height; number of green leaves; mass of the bunch; number of hands and fruits per bunch; mass of the hands; mass, length and diameter of the stem, and mass of the fruit of the external row from the first to the tenth hand were estimated. The highest correlation coefficient between yield and vegetative traits for 'Dwarf Prata' and 'BRS Platina', was obtained for pseudostem perimeter at ground level, indicating its greater potential to compose harvest estimation models. The correlations between the masses of the bunch and hands with yield traits, for both genotypes, showed a higher potential for predicting the number of hands and fruits. Appropriate correlation estimates are associated with the fourth and fifth bunch, for the 'Dwarf Prata' and 'BRS Platina', respectively, using the mass of the fruit of the external row.

Key words: Banana, phenotypical descriptors, statistics, associations, genotypes.

INTRODUCTION

The banana crop (*Musa*) has a social and economical importance in over 80 countries, especially in small properties (Silva et al., 2002). Brazil is the fifth main banana producer in the world, with 6.9 thousand tons produced in 481 thousand hectares, resulting in

anaverage yield of 14.3 ton ha⁻¹ (FAO, 2013).

Among the most planted cultivars nationwide, the Prata, Dwarf-Prata, Pacovan and Common-Prata types can be highlighted, since they represent almost 80% of the area cultivated with bananas (Silva et al., 2008).

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Despite the importance of these genotypes, all of them are susceptible to the main bananas pests and diseases, and, in the last years, the Panama disease has become a serious problem. It is causing limitations in important producing areas of the country, especially in some properties with sandier soils (Rodrigues et al., 2011).

In the light of these facts, it becomes evident the importance of developing breeding programs aiming to obtain cultivars with adequate height, adapted to different ecosystems, and resistant to the main diseases and pests, as well as improving fruit quality (Donato et al., 2006a), in order to supply the increasingly demanding consumer and producer markets..

According to Vencovsky and Barriga (1992), this kind of studies provide important information for genetic breeding, such as the possibility of identifying the proportion of phenotypic correlation that occurs due to genetic causes; verify if the selection in one character affects the other: quantify indirect increases due to selection made in correlated characters; and evaluate the complexity of the characters. Knowledge about the association between characters is very important, especially if the selection of one of them is difficult, due to low heritability or difficult measuring and identification problems.stimates of phenotypical correlations involving production bananas, yield components and other important characters in bananas can be found in several articles (Arantes et al., 2010; Donato et al., 2006a; Lima Neto et al., 2003; Soares et al., 2012; Tenkouano et al., 2002). However, for same character, it can be identified the estimate of positive, negative or null values. This can be explained by the fact that those correlations were estimated in different genotypes and environments, using different methods. Hence, if there is a possibility, new correlations should be estimated to help the conduction of a breeding program (Vencovsky and Barriga, 1992). Still, a correlation study can be employed to predict values related to harvest using significant variables that associate highly with production.

Therefore, it is important to identify variables associated with high productivity for use in mathematical modeling to estimate the harvest, based on agronomic traits measured during the crop cycle. In this case, the mathematical model, provided that it is adjusted in a functional way, allows those involved with the bananas crop, whether a researcher and/or a producer, planning and organization of pre- and post-harvest operations. Still, it is noteworthy that the regression models generated a posteriori may indicate the need for subsequent management adjustments to enable correction of characteristics that show unfavorable at any given time, especially for the next crop cycle.

Thus, the analysis of behavior and expression of the plants is of wide interest to those engaged in plant production research, being very useful for proper planning of agricultural activities. This way, several statistical studies, linear and nonlinear, are developed with the purpose of obtaining information and describe

future plant growth over time (Hernández et al., 2007; Maia et al., 2009; Scarpari and Beauclair, 2009; Soares et al., 2012), from the analyses of the correlation coefficients among the plant variables.

As an outcome of the above, this work was carried out with the objective of estimating the correlations between agronomic characters and yield in type Prata bananas, so that it can contribute to the elaboration of models to estimate harvest.

MATERIALS AND METHODS

The experiment was established in a Red-Yellow Latosol (Hapludox), medium texture, hypoxerophilous Caatinga (thorn-shrub savannah) phase, with flat to moderate topography. The area is located at the Instituto Federal Baiano, in the Guanambi Campus, BA, 14°13'30 "S, 42°46'53" W, 545 m of altitude, average annual precipitation of 663.69 mm, average temperature of 26°C and relative humidity of 64%. The local climate is Aw, according to Köppen's classification.

Micropropagated bananasplantlets were used, in a 3.0×2.5 m spacing and irrigated with fixed micro-sprinklers. Installation and cultivation followed the recommendations for the crop, and fertilizers were applied based on analysis of soil and leaves. The two evaluated genotypes were: Dwarf Prata, triploid (AAB), susceptible to yellow and black Sigatoka and Panama disease; and the BRS Platina hybrid, tetraploid (AAAB), resistant to yellow Sigatoka and Panama disease, derived from the cross between 'Dwarf Prata' (AAB) and M53 diploid (AA), formerly known in pre-release as PA42-44.

Measurements were made at the time of harvest. Each plant, the basic unit, was considered as a replicate. Therefore, to assess vegetative and yield characteristics, the plants were sampled at random into the two genotypes with different numbers of replicates, 98 for 'Dwarf Prata' and 96 for 'BRS Platina'.

The measurements consisted of phenotypic vegetative descriptors plant height, pseudostem perimeter at ground level, 30 and 100 cm in height, number of green leaves at harvest. Bunch yield characteristics bunch weight, number of hands and fruits per bunch, hands mass and stem mass, length and diameter were determined, as well as hands yield characteristics number of total fruits and fruits per bunch, and mass, internal and external length and diameter of the central fruit on the external and internal rows were determined. The measured values were obtained according to the methodological proposal contained in catalogs of standard morphological descriptors for bananas (Descriptors for banana, 1996).

For each evaluated genotype, 'Dwarf Prata' and 'BRS Platina', phenotypic correlations were estimated from the associations between the masses of the bunch and hands with the evaluated yield and vegetative characteristics, based on Pearson correlation (Steel et al., 1997). The data came from observations of individual replicates of each genotype for establishing associations between characteristics.

RESULTS AND DISCUSSION

In order to characterize the 'Dwarf Prata' and 'BRS Platina' bananas, Tables 1 and 2 show the averages and standard deviations of the evaluated vegetative and yield characteristics. Those allowed the correlation statistical procedure and implications in harvest estimates.

Table 1. Averages and standard deviations of the vegetative characteristics of 'Dwarf Prata' and 'BRS Platina' bananas.

| Genotypes | ¹ PH (cm) | ² PPGL (cm) | ³ PP30 (cm) | ⁴ PP100 (cm) | ⁵ NGH (un) |
|--------------------|----------------------|------------------------|------------------------|-------------------------|-----------------------|
| Dwarf Prata | 335 ± 35 | 113 ± 14 | 99 ± 14 | 78 ± 9 | 14 ± 2 |
| BRS Platina | 348 ± 34 | 108 ± 10 | 93 ± 9 | 72 ± 8 | 11 ± 2 |

¹PH, Plant height; ²PPNS, pseudostem perimeter at ground level; ³PP30, pseudostem perimeter at 30 cm height; ⁴PP100, pseudostem perimeter at 100 cm height; ⁵NGH, number of green leaves at harvest time.

Table 2. Averages and standard deviations of the yield characteristics of 'Dwarf Prata' and 'BRS Platina' bananas.

| Characteristics | Genótipos | | | |
|-------------------------|------------------|------------------|--|--|
| Characteristics | Dwarf Prata | BRS Platina | | |
| Bunch mass (kg) | 27 ± 6 | 24 ± 5 | | |
| Hands mass (kg) | 24 ± 5 | 21 ± 5 | | |
| Number of hands (un) | 11 ± 1 | 8 ± 1 | | |
| Average hands mass (kg) | 2.16 ± 0.39 | 2.58 ± 0.50 | | |
| Stem mass (kg) | 2.83 ± 0.39 | 2.55 ± 0.55 | | |
| Stem length (cm) | 38.61 ± 8.94 | 43.07 ± 8.04 | | |
| Stem diameter (mm) | 73.27 ± 8.15 | 68.51 ± 6.68 | | |
| Number of fruits (un) | 183 ± 26 | 128 ± 21 | | |

The average values and the variability of yield and vegetative characteristics (Tables 1 and 2) measured in the bananas 'Dwarf Prata' and 'BRS Platina' are in agreement with data obtained in the same cultivation area for the same genotype (Donato et al., 2006, 2009).

Analyzing Table 1, it is seen that the genotypes Dwarf Prata and BRS Platina present proximity of vegetative characteristics values. Several authors confirm the size similarity of the genotypes (Donato et al., 2006, 2009; Ledo et al., 2008; Oliveira et al., 2008; Marques et al., 2011). However, these studies observed the superiority of the progenitor compared to the hybrid, regarding the pseudostem perimeter and number of green leaves at harvest. However, Donato et al. (2009) found similarities between these genotypes also for these latter characteristics.

The 'Dwarf Prata' has larger values for characteristics number of hands and fruits compared to the hybrid, while the 'BRS Platina' is more expressive for the value of the average weight of the bunches (Table 2). In this context, several studies confirm these differences between 'Dwarf Prata' and 'BRS Platina', which indicate that these are varietal differences that happen regardless of environmental and management conditions (Donato et al., 2006; Ledo et al., 2008; Donato et al., 2009; Marques et al., 2011). Thus, considering that bananas are commercialized upon classification, which includes fruit dimensions, both genotypes presented fruits that would be well ranked (Table 2), with an advantage for the 'BRS Platina'.

The correlations between the masses of the bunch and hands and the vegetative characteristics plant height, pseudostem perimeter at ground level, at 30 and 100 cm in height, and number of green leaves at harvest for the 'Dwarf Prata' and 'BRS Platina' bananas were significant and positive for all variables (Table 3). This indicates the direct variation of the yield variables masses of the bunch and hands with the vegetative variables analyzed in this work.

Thus, the genotypes 'Dwarf Prata' 'and 'BRS Platina' showed positive and significant correlations between the average bunch and hands masses with plant height. This association denotes the direct variation of these bunch and hands masses with the size of the plant. However, the magnitude of this correlation was the lowest among the vegetative variables evaluated for hybrid BRS Platina (Table 3). Soares (2013) stated the same for the cultivar tropical. However, Siqueira (1984) found, from clones of 'Prata' bananas, that it is possible to select smaller genotypes without production impairing, as plant height, in most of the evaluated clones, did not correlate with any yield-associated character.

From the analysis of the association between masses of bunch and hands and the pseudostem perimeter measured at ground level, at 30 and 100 cm from ground, satisfactory correlation estimates can be observed. However, the association with the pseudostem perimeter at ground level expressed better response, which suggests its use as an effective variable for prediction (Table 3). Despite being measured at harvest time, the

| | Geno | types | Genotypes | | |
|--|-------------|-------------|-------------|-------------|--|
| Vegetative characteristics | Bunch mass | | Hands | s mass | |
| | Dwarf Prata | BRS Platina | Dwarf Prata | BRS Platina | |
| Plant height | 0.63** | 0.49** | 0.63** | 0.47** | |
| Pseudostem perimeter at ground level | 0.75** | 0.75** | 0.75** | 0.73** | |
| Pseudostem perimeter at 30 cm height | 0.69** | 0.64** | 0.68** | 0.63** | |
| Pseudostem perimeter at 100 cm height | 0.68** | 0.74** | 0.67** | 0.72** | |
| Number of green leaves at harvest time | 0.35** | 0.52** | 0.35** | 0.52** | |

Table 3. Correlation coefficients between the masses of the bunch and hands, with the vegetative characteristics of Prata type bananas, 'Dwarf Prata' and 'BRS Platina'.

pseudostem perimeter would present the same dimensions as if it were assessed at flowering, as this characteristic remains constant after that, since at this time, the bananas tree ceases the development of roots and leaves, beginning senescence of these organs and culminating with bunch ripening (Robinson and Galán Saúco, 2010). Additionally, to compose the prediction equations, the variables must be easily measurable and of simple practical implementation, allowing the determination in advance of the phenomenon to be predicted about 120 to 150 days before bananas harvest bananas (Donato et al., 2006, 2009).

Different studies correlating vegetative and reproductive characteristics in bananas report a significant correlation between pseudostem perimeter and bunch production (Lima Neto et al., 2003; Arantes et al., 2010). In addition, Siqueira (1984) found, from clones of 'Prata' bananas, that among the vegetative growth related characters, the pseudostem perimeter was the most positively correlated with the production characters.

However, this result does not agree with Donato (2006a) and Soares (2013), who state that bunch weight and pseudostem perimeter correlation is not significant and positive, which may indicate that, regardless of pseudostem thickness, the bunch mass may have a satisfactory value. However, genotypes with higher and lower height showed, respectively, higher and lower pseudostem perimeter as well. In addition, Lima Neto (2003) found that the correlation between bunch mass and pseudostem diameter in 'Gros Michel' bananas is not usually influenced by environmental factors.

In this work, the values found were statistically significant and positive for the association bunch and hands masses with the number of green leaves at harvest for 'Dwarf Prata' and 'BRS Platina' (Table 3). However, the highest correlation estimate was expressed by the hybrid, but for the two genotypes the correlation was low. This result can be attributed to the high number of leaves present at harvest time, with an average of 14 and 11 leaves, respectively, for genotypes Dwarf Prata and BRS Platina. Thus, the 'BRS Platina' had fewer leaves, but higher correlation coefficient (0.52) than the

'Dwarf Prata' (0.35), which had more leaves (Table 3). In a study conducted in northern Minas Gerais, the 'Dwarf Prata' bunches were heavier when the plant was maintained with a minimum of 12 leaves. Therefore, the amount of leaves recorded in this study meets the minimum requirement to normally fill banana bunches in 'Prata' type bananas (Rodrigues et al., 2009), which explains the low correlation. Additionally, the evaluation was done at harvest time, and not at flowering time, when it could be expected a higher correlation between these variables.

However, some authors do not share these same results, since those genotypes showed non-significant and positive values for the association number of leaves at harvest time and bunch yield, in both production cycles (Lima Neto et al., 2003; Donato et al., 2006a). Nevertheless, Siqueira (1984) noted significant correlations between the referred characters. According to the exposed by the correlations in this study and in others reviewed, it can be inferred that these characters do not show strong correlation, and that may cause low reliability for harvest prediction if used as a model component.

In this context, Soares (2013) observed that, despite being significant and positive, the relation bunch mass and number of green leaves showed a correlation coefficient of low magnitude. This indicates that the bunch mass varies in a direct way in relation to the number of green leaves, but, if the minimum demands of number of leaves is met, this influence is not expressive.

Correlations between bunch mass and other yield descriptors measured at the harvest time were positive and significant at a 1% level of probability, except for stem length in the genotype BRS Platina, which was non-significant (Table 4). This way, the bunch mass varied in the same way as the other yield characters considered. Nevertheless, in prediction studies, it is desirable that the variable that composes the model show high correlation coefficient and also easy measuring in the field.

The bunch mass in relation to the hands mass expressed the highest correlation, 0.99 for both genotypes, Dwarf Prata and BRS Platina, as expected,

^{**}Significant at 1% level of significance.

| Yield characteristics of the bunch = | Genotypes | | | |
|--------------------------------------|--------------------|--------------------|--|--|
| rieid characteristics of the bunch | Dwarf Prata | BRS Platina | | |
| Hands mass | 0.99** | 0.99** | | |
| Stem mass | 0.82** | 0.71** | | |
| Stem length | 0.35** | 0.08 ^{ns} | | |
| Stem diameter | 0.69** | 0.68** | | |
| Number of hands | 0.70** | 0.60** | | |
| Number of fruits | 0.74** | 0.74** | | |
| Average hands mass | 0.86** | 0.89** | | |

Table 4. Phenotypic correlation coefficients between bunch mass and other yield characteristics in Prata type bananas, 'Dwarf Prata' and 'BRS Platina'.

because the hands mass is the main component of the bunch, only without the rachis (Table 4). The general behavior of this correlation is similar to that obtained by other authors (Jaramilo, 1982; Lima Neto et al., 2003; Donato et al., 2006a). However, although this is a high magnitude and significant association, it presents a limitation as a variable to be used in harvest prediction, since it assumes the condition of response variable, as well as the bunch mass.

The correlation between masses of bunch and stem, for the studied genotypes, showed statistically significant and positive values (Table 4). However, Donato et al. (2006a) found varied correlations for the approached genotypes, being significant and positive the correlation for 'Dwarf Prata'. However, for the BRS Platina hybrid, it was found a significant and negative correlation. Nevertheless, the variable stem mass, to estimate harvest, is considered as a destructive and hard to measure sample, because to quantify its mass, it is necessary to detach the whole bunch. These reasons, coupled with its measurement period, which coincides with the measurement period of the phenomenon that is intended to be predicted, make the variable of little practical application for harvest prediction.

The association between bunch mass and stem length, despite being significant and positive for 'Dwarf Prata', showed the lower estimate, 0.35, and for 'BRS Platina', it was non-significant, with a value of 0.08 (Table 4). Donato et al. (2006a) did not observe stable correlations for the relation bunch mass and stem length during the production cycles for the studied genotypes. Soares (2013) states that the bunch mass does not depend of the stem length. Thus, it can be suggested that it is not a consistent and reliable variable to compose a harvest prediction equation, although it is an easily measured and non-destructive variable.

Stem diameter is an easily measurable variable in the field, with significant, positive and relatively high correlation, 0.69 and 0.68 (Table 4), respectively, for 'Dwarf Prata' and 'BRS Platina'. Similar results were obtained by Soares (2013), with the Tropical cultivar

which denotes a general pattern of straight variation of bunch mass with the stem diameter, an expected result. This happens because bunches are composed of hands (fruits) and stem (rachis), and the bigger bunches have longer rachises (Jaramillo, 1982), although the percentual participation of the rachis in the bunch mass decreases with the increase on its mass. Such behavior was proved by Jaramillo (1982) for Cavendish type cultivars and Azevedo et al. (2010) for Prata type cultivars, with tall stature.

The bunch mass shows large variation among bunches with the same number of hands and usually increases with the number of hands. However, it may change this variation pattern in function of variations in mass, diameter and length of fruits, number of fruits per bunch and fluctuations in the mass of the rachis (Jaramillo, 1982). Donato et al. (2008) found, for 'Tropical' bananas, the same average bunch mass, 14.32 and 14.85 kg, for the first and second cycle, respectively, despite the number of hands have increased from 5 to 7. The absence of increments between cycles, for the bunch mass, was due to the decrease observed in mass, length and diameter of the fruit from 158.07 to 103.74 g, 15.88 to 14.39 cm, and 40, 77 to 34.13 mm, respectively.

However, in this study, the correlations between bunch mass and number of hands were significant and positive, with the highest correlation coefficient associated to the genitor 'Dwarf Prata' (0.70). The hybrid BRS Platina showed a lower correlation coefficient (0.60) for the same variables (Table 4). According to Donato et al. (2006a), for the evaluated genotypes, this association was stable. positive and with a high magnitude. Moreover, it is noteworthy that the variable number of bunches has a discrete quantitative nature, being easy to apply in practice. It can be obtained at the flowering stage and about 120 to 150 days before harvesting the 'Dwarf Prata' and 'BRS Platina' bunches. That, associated to the age marking of the bunch technique as a criterion for harvesting point (Lichtemberg et al., 2008), allows predicting harvest time and yield with greater accuracy, which qualifies its use in a harvest prediction model.

^{**} and ^{ns}: significant at 1% level of significance and non-significant, respectively.

| | Geno | Genotypes | | Genotypes | | |
|------------------------------------|--------------------|--------------------|--------------------|--------------------|--|--|
| Bunch yield characteristics | Mass o | f bunch | Mass o | ass of hands | | |
| | Dwarf Prata | BRS Platina | Dwarf Prata | BRS Platina | | |
| Mass of the first hand | 0.61** | 0.60** | 0.62** | 0.57** | | |
| Mass of the second hand | 0.62** | 0.67** | 0.63** | 0.65** | | |
| Mass of the third hand | 0.86** | 0.80** | 0.87** | 0.80** | | |
| Mass of the fourth hand | 0.85** | 0.85** | 0.86** | 0.85** | | |
| Mass of the fifth hand | 0.88** | 0.83** | 0.89** | 0.82** | | |
| Mass of the sixth hand | 0.84** | 0.79** | 0.86** | 0.78** | | |
| Mass of the seventh hand | 0.82** | 0.81** | 0.84** | 0.80** | | |
| Mass of the eighth hand | 0.85** | 0.62** | 0.87** | 0.59** | | |
| Mass of the ninth hand | 0.83** | 0.69** | 0.83** | 0.70** | | |

0.64*

Table 5. Phenotypic correlation coefficients between bunch and hands masses and individual masses from the first to the tenth hand in Prata type bananas, 'Dwarf Prata' and 'BRS Platina'.

0.83**

As well as the number of bunches, the number of fruits can be measured in the field. The correlation between bunch mass and number of fruits was statistically significant, positive and with appropriate magnitude. The estimate of 0.74 for both genotypes, Dwarf Prata and BRS Platina (Table 4), was higher, also, for the two genotypes than the correlation between bunch mass and number of hands. Lima Neto et al. (2003) and Soares (2013) obtained positive and significant correlation estimates for most of the studied genotypes. Donato et al. (2006a) found significant positive correlations, with values close to the present study, for 'BRS Platina' and positive and non-significant for 'Dwarf Prata'. However, these authors reported negative and significant correlation estimates between bunch mass and number of fruit for Calipso and Preciosa genotypes, and positive and significant between bunch mass and average fruit mass, indicating that the bunches with higher mass can have a lower number of fruits, yet with larger fruits. In this sense, the variable number of fruits anticipates the quantitative characteristics of the bunch at the harvest time, being, therefore, highly reliable for harvest estimate, as well as being easy to measure in field conditions.

Mass of the tenth hand

Similarly, correlation between bunch mass and fruits mass were found significant values, positive, only with higher magnitudes for 'Dwarf Prata' (0.86) and for 'BRS Platina' (0.89) (Table 4). However, the high values for this correlation have a difficult application in a harvest prediction equation for practical reasons, because it is a destructive and hard to measure variable. Thus, it would not be an interesting variable to compose a harvest prediction equation.

Significant and positive correlations were found between the bunch mass and the hands mass, from the first to the tenth hand, for both evaluated genotypes (Table 5). The highest correlation coefficients, both for the bunch mass and for the hands mass, were observed in association with the mass of the fifth hand for the 'Dwarf Prata', and with the mass of the fourth hand for 'BRS Platina'. For that reason, the characteristics of these bunches have the potential to compose the harvest prediction models.

0.67*

0.85**

So, considering the analysis of Table 5, it can be suggested that the fifth and fourth hands, respectively, for 'Dwarf Prata' and 'BRS Platina', show higher reliability to compose the harvest prediction equation, both to estimate bunch mass, and the hands mass. However, these are considered destructible samples, since the hand must be extracted to obtain the mass.

Aiming to minimize the losses when sampling the variables due to removal of the hands, a correlation study was carried out with the unitary descriptors of the central fruit of the external and internal rows, due to the lower impact of removing those to measure the bunch. For that, evaluations were made of the number of fruits per hand and the length, diameter and mass of the fruits, both from the external and internal rows of the hands. For such measurements, the hands from the first to the tenth position were used. The correlations between bunch mass and the mass of the fruit from the external row, from the first to the tenth hand, were significant and positive, and with higher values when compared to the other descriptors (Table 6).

Correlations between bunch mass and the masses of the fruits from the external row of the ninth and tenth hands (0.80) for the 'Dwarf Prata', and the mass of the fruits from the external row of the eighth (0.73) and ninth (0.79) hands for the 'BRS Platina', have the higher correlation coefficient values (Table 6). However, these hands are not recommended to compose the harvest prediction model due to the limitation of the number of hands in the bunch, as bunches of these genotypes in field conditions usually have 7 to 13 hands. This way, the prediction equation would be limited to predicting

^{**} and ^{ns}: significant at 1% level of significance and non-significant, respectively.

Table 6. Phenotypic correlation coefficients between bunch and hands masses and the individual masses of the central fruits from the external row from the first to the tenth hand in Prata type bananas, 'Dwarf Prata' and 'BRS Platina'.

| · | Geno | types | Geno | otypes |
|---|-------------|--------------------|--------------------|--------------------|
| Hands yield characteristics | Bunch mass | | Hands mass | |
| | Dwarf Prata | BRS Platina | Dwarf Prata | BRS Platina |
| Mass of the central fruit on the external row of the first hand | 0.73** | 0.64** | 0.75** | 0.63** |
| Mass of the central fruit on the external row of the second hand | 0.67** | 0.60** | 0.70** | 0.59** |
| Mass of the central fruit on the external row of the third hand | 0.67** | 0.56** | 0.70** | 0.55** |
| Mass of the central fruit on the external row of the fourth hand | 0.73** | 0.61** | 0.75** | 0.61** |
| Mass of the central fruit on the external row of the fifth hand | 0.70** | 0.67** | 0.71** | 0.66** |
| Mass of the central fruit on the external row of the sixth hand | 0.70** | 0.66** | 0.71** | 0.65** |
| Mass of the central fruit on the external row of the seventh hand | 0.68** | 0.65** | 0.70** | 0.64** |
| Mass of the central fruit on the external row of the eighth hand | 0.71** | 0.73** | 0.73** | 0.73** |
| Mass of the central fruit on the external row of the ninth hand | 0.83** | 0.79** | 0.83** | 0.80** |
| Mass of the central fruit on the external row of the tenth hand | 0.80** | 0.40 ^{ns} | 0.82** | 0.38 ^{ns} |

^{**} and ^{ns}: Significant at 1% level of significance and non-significant, respectively.

bunches with more than 8 hands for the 'BRS Platina' and more than 9 hands for 'Dwarf Prata'. However, for those considerations, it can be suggested to include in the prediction model the masses of the fruit from the external row of the fourth (0.73) and fifth (0.67) hands, for 'Dwarf Prata' and 'BRS Platina', respectively (Table 6). An additional reason is the fact that the masses of the fourth and fifth hands show higher correlation coefficients with the masses of bunch and hands, which directed to estimates of models involving unitary characteristics of these hands.

Similar correlations, in magnitude and significance, were also found between hands mass and mass of the fruit on the external row, from the first to the tenth hand (Table 6). In this case, they were also considered the higher correlation coefficient values, coupled with the practical aspects discussed, as the number of hands normally found for the evaluated genotypes. Thus, the association between hands mass and mass of the fruit on the external row of the fourth (0.75) and fifth (0.66) hands, for 'Dwarf Prata' and 'BRS Platina' (Table 6), respectively, show higher application to compose the harvest prediction model, based on yield characteristics.

Conclusion

Higher correlation coefficient between yield and

vegetative characteristics, for 'Dwarf Prata' and 'BRS Platina', was obtained for the pseudostem perimeter at ground level, which indicates higher potential to compose the harvest estimate. The correlations between bunch and hands masses with the yield characteristics, for both genotypes, presented higher prediction potential for number of hands and fruits. Adequate correlation estimates are associated to the fourth and fifth hands, for the 'Dwarf Prata' and 'BRS Platina', respectively, using the mass of the fruit from the external row.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Limestone and phosphogypsum effects on soil fertility, soybean leaf nutrition and yield

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Quantifying soil and plant response to surface applied limestone and phosphogypsum (PG) will help promote no-tillage soybean production. Our objective was to determine effects of annual surface application of phosphogypsum and limestone on soil profile chemistry, fertilizer use efficiency, soybean leaf nutrient concentrations, seed yield, and yield components under a no-tillage system (NTS). The study was conducted in the Brazilian Cerrado under field conditions during three growing seasons on an acidic clay loam soil. Four soil remediation treatments [(limestone, lime + phosphogypsum, phosphogypsum and control (no soil correction)] with four fertilizer levels (0, 50, 100 and 150% of recommended P and K fertilizer for soybean) were arranged in a randomized design using a 4 x 4 factorial design. Treatments involving lime application improved soil chemical characteristics of the soil, resulted in significant increases in plant nutrient concentrations, and increased soybean grain yield. In contrast, application of phosphogypsum did not improve plant development conditions and resulted in no significant changes in soil properties or soybean grain yield. Using increased fertilizer application rates for 3 years also produced significant increases in soybean grain yield. Our results imply that, for acidic soils liming NTS is the best practice to increase fertilizer use efficiency.

Key words: Calcium carbonate, Glycine max, no-tillage system (NTS), mineral nutrition, soil acidity.

INTRODUCTION

Brazil is the second largest producer of soybean in the world, surpassed only by the U.S.A. (FAO, 2013). This crop is widely planted in soils managed under the notillage system (NTS) (Embrapa Soja, 2010; Nascente and Crusciol, 2012). Due to the benefits provided (increased levels of soil organic matter (SOM), reduction of soil erosion, increased soil moisture conservation and improvement of soil fertility), NTS use has grown significantly throughout the world, and almost 117 million

ha worldwide are cultivated using this system (FAO, 2013). The use of this system has increased in Brazil since 1970s. Currently, an estimated area of more than 25 million ha is cultivated using the NTS (Nascente et al., 2013).

The success and continuity of the NTS over the years is achieved through management planning of the soil fertility over the depth profile, which includes a soil acidity correction. Soil acidity limits crop production in many

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areas of the world due to the toxicity caused by aluminum, phosphorus deficiency and low base saturation (Caires et al., 2003). Tropical soils such as those of the Brazilian Cerrado or the African savannas are naturally acidic because high levels of rainfall over many years have leached basic ions, resulting in reduced soil fertility (Caires et al., 2008). According to Fageria et al. (2011), tropical soils are highly weathered and are dominated by 1:1 clay minerals such as kaolinite and iron oxides (hematite and goethite) and Al (gibbsite), which have a high P adsorption capacity. In these areas, satisfactory cash crop grain yields are dependent on proper liming and soil fertilization (Caires et al., 2001; Alleoni et al., 2009; Souza et al., 2011).

In Brazil, the most common material used for acidity correction is limestone, which effectively increases soil pH, calcium and magnesium content and base saturation reduces the levels of exchangeable aluminum in the soil (Caires et al., 2004). In most grain-producing areas under the NTS, the soil acidity is corrected by applying limestone to the soil surface without incorporation (Soratto and Crusciol, 2008a; Alleoni et al., 2009). However, the reactions produced by limestone are limited the location generally to at which application/incorporation occurs due to the low mobility of limestone in soil. The results of field studies show that the movement of lime to depth varies according to the timing and rate of liming, the form of lime applied, the soil type, the weather conditions, the addition of acidic fertilizers and the cropping system (Gascho and Parker, 2001; Convers et al., 2003; Tang et al., 2003; Caires et al., 2008; Soratto and Crusciol, 2008a, b; Churka Blum et al., 2013). According to Caires et al. (2003), liming, whether applied to the surface or incorporated into the soil, provided more intense soil acidity correction in the superficial layer (0 to 0.5 m). On the other hand, a stronger reaction also occurred in the 0.05 to 0.10 m and 0.10 to 0.20 m layers when lime was incorporated into the soil. Caires et al. (2005) added that the effects of surface liming on all three acidity-related variables (pH, Al, and basic cations) were significant at depths of 0 to 0.05 m and 0.05 to 0.10 m from 1 year onward and at a depth of 0.10 to 0.20 m from 2.5 years onward. As a result, the reactions of lime in the soil are subject to further delays, especially at times relatively close to the time of application (Caires et al., 1998; Fageria and Baligar, 2008; Soratto and Crusciol, 2008b).

The application of lime with phosphogypsum (PG) appears to be an excellent alternative for improving the chemical conditions of the soil profile in the Cerrado region because the application of lime alone cannot provide a better environment in the deeper layers of the soil; these conditions may limit the development of the plant root system to the surface layers. As a result, the root systems exploit only a small soil volume, thus limiting crop productivity, especially in locations where periods without rain are frequent (Caires et al., 2005; Soratto and Crusciol, 2008b).

Phosphogypsum can be used to improve the environment for root growth in the deep soil layers. This product is a soil conditioner and has high mobility in the soil profile. It can provide Ca²⁺ and SO₄²⁻ in solution in the soil and can be leached, enriching the deep layers with these nutrients and reducing Al3+ at depth. Thus, the use of phosphogypsum may allow the development of deep roots, increasing the volume of soil that can be explored and improving the drought tolerance of the plant. The successful use of phosphogypsum to improve the root environment has been far-reaching in the Cerrado region of Brazil (Sousa and Lobato, 2004). Approximately, 80% of the area of this region is subject to problems of soil subsurface acidity and frequent dry spells, especially in the months of January and February, the critical time for the development of summer crops (Ramos et al., 2006; Caires et al., 2008).

The identification of alternatives that allow the correction of soil acidity at depth in the NTS using methods of surface application without incorporation may facilitate the persistence and success of this system. Information is lacking about reactions at depth involving lime and phosphogypsum applied on the soil surface under NTS. Moreover, there is need for additional information about the effect of this approach on soil chemical characteristics, plant mineral nutrition and crop production (Franchini et al., 2003; Caires et al., 2006b; Soratto and Crusciol, 2008b, c). Little is known about the behavior of correctives that can act effectively through the soil profile when they are applied superficially without incorporation. This information is important because the permanence of the system for long periods after the implementation of the NTS may not be feasible in many cases as there are persistent factors contributing to soil acidification, such as rainfall and uptake of nutrients by crops. Accordingly, this study is based on the following hypotheses: a) Due to the higher mobility of phosphogypsum to the deep layers of the soil, the application of this amendment after liming could promote the enrichment of bases in the soil profile more rapidly than that obtained with only limestone; and b) The use of lime and phosphogypsum can significantly increase soybean leaf nutrition and grain yield. The objective of the study was to determine the effect of the annual application of phosphogypsum and lime on the soil surface without incorporation on the correction and the fertilization efficiency through the soil profile as well as its effects on leaf nutrition, yield components and grain yield and on grain nutrient accumulation and content in soybean grown under the NTS.

MATERIALS AND METHODS

Site description

The experiment was conducted for three growing seasons (2010/2011, 2011/2012 and 2012/2013) at Capivara Farm, located in the city of Santo Antonio de Goias, GO, Brazil. The geographical

0 - 0.20

0.20 - 0.40

| D (1 /) | Ca | Mg | Al | H+AI | K | CEC ¹ | pH (H₂O) |
|---|------|------|------|------|------|------------------|----------|
| Depth (m)cmol _c dm ⁻³ | | | | | | | |
| 0 - 0.20 | 0.72 | 0.37 | 0.38 | 6.5 | 0.16 | 7.78 | 5.0 |
| 0.20 - 0.40 | 0.53 | 0.17 | 0.42 | 7.1 | 0.14 | 7.94 | 4.8 |

P (Mehlich)

6.0

2.3

Zn

3.1

2.1

Cu

-mg dm⁻`

2.1

1.3

Table 1. Chemical characteristics of the soil in the experimental area before the beginning of the experiment, November 2010.

SOM3 (g dm⁻³)

18.3

17.0

coordinates of the site are 16° 28′ 00″ S, 49° 17′ 00″ W. The altitude of the site is 823 m. The climate is tropical savanna, considered Aw according to the Köppen classification. There are two well-defined seasons: usually, the dry season extends from May to September (autumn/winter) and the rainy season from October to April (spring/summer). The historic average annual rainfall ranges from 1500 to 1700 mm. The historic average annual temperature is 22.7°C, ranging annually from 14.2 to 34.8°C. Additionally, the daily average temperature and precipitation during the experiment were monitored (Figure 1).

V2 (%)

16.0

10.6

The experimental area was cultivated for 5 years under the NTS, with corn (Zea mays L.) grown in summer and common bean (Phaseolus vulgaris L.) irrigated in winter. The soil was classified as a clay loam (kaolinitic, thermic Typic Haplorthox). Before the experiment began, the chemical characteristics of the soil were determined (at depths of 0 to 0.20 m and 0.20 to 0.40 m) to characterize the soil in the experimental area to calculate the requirements for liming and phosphogypsum (Table 1). The soil analysis was performed according to Claessen (1997). The soil pH was determined in a 0.01 mol L-1 CaCl2 suspension (1:2.5 soil/solution). Exchangeable Ca, Mg, and Al were extracted with neutral 1 mol L⁻¹ KCl in a 1:10 soil/solution ratio and determined by titration with a 0.025 mol L-1 NaOH solution. Phosphorus and exchangeable K were extracted with a Mehlich 1 extracting solution (0.05 M HCl in 0.0125 M H₂SO₄). The extracts were colorimetrically analyzed for P, and flame photometry was used to analyze K. The base saturation values were calculated using the results of exchangeable bases and total acidity at pH 7.0 (H + Al). Micronutrients (Fe, Zn, Cu and Mn) were determined in Mehlich 1 extract by atomic absorption, and organic matter was determined by the method of Walkley and Black (Walkley and Black, 1934).

Experimental design and treatments

The experimental design was a randomized complete block layout arranged in a 4 × 4 factorial design with four replications. The treatments consisted of four types of soil amendment [limestone, limestone + phosphogypsum, phosphogypsum and control (no amendment application)] with four fertilization rates [0, 50, 100 and 150% of the fertilizer recommended for soybean (Sousa and Lobato, 2004)]. The recommended fertilization corresponded to the application of 80 kg ha⁻¹ of P_2O_5 (55% of superphosphate and 45% of triple superphosphate) and 80 kg ha⁻¹ of K_2O (potassium chloride). The lime application rate (5.0 Mg ha⁻¹) was calculated to increase the base saturation to 70% at a depth of 0 to 0.20 m. The amount of phosphogypsum (gypsum) used (2.5 Mg ha⁻¹) was determined by the clay content of the soil (500 g kg⁻¹) at a depth of 0.20 to 0.40 m, as recommended by Sousa and Lobato (2004).

These amendments were applied by scattering on the surface of the soil without incorporation. The treatments were divided into three applications and used as follows: on 13/11/2010 (2 Mg ha⁻¹ of lime and 1.0 Mg ha⁻¹ phosphogypsum), on 11/11/2011 (2 Mg ha⁻¹ of lime and 1.0 Mg ha⁻¹ of phosphogypsum) and on 19/10/2012 (1 Mg ha⁻¹ of lime and 0.5 Mg ha⁻¹ of phosphogypsum). The limestone used contained 26% Ca and 7.2% Mg. Its effective neutralizing value was 86.56%. The phosphogypsum contained 21.8% Ca and 17.4% S.

Fe

58.7

38.6

Mn

11.7

6.5

Soybean cultivation (Summer 2010/11, 2011/12 and 2012/13)

The sowing of soybean cultivar BRS 7860 RR was performed mechanically on 19/11/2010, 10/11/2011 and 01/11/2012 using notill seeding (Semeato, model Personale Drill 13, Passo Fundo, RS, Brazil) with a row spacing of 0.45 m and a density of 20 pure live seeds m⁻¹. Each plot consisted of 10 rows, each 5 m long. The usable area consisted of the 8 central rows, with 0.5 m on each side disregarded. A corridor 2 m in width was left between the plots. The soybean seeds were inoculated with *Bradyrhizobium japonicum*. Seedling emergence occurred at 8, 9 and 5 days after sowing in 2010, 2011 and 2012, respectively. Crop management was performed according to the crop recommendations (Embrapa, 2010).

Offseason crops

In May, 2011 and May, 2012, common bean was cultivated in the same area and fertilized with 0, 60, 122 and 183 kg ha $^{\text{-}1}$ of N (ammonium sulfate and urea), 0, 100, 200 and 300 kg ha $^{\text{-}1}$ P_2O_5 (simple superphosphate) and 0, 70, 140 and 210 kg ha $^{\text{-}1}$ of K_2O (potassium chloride) in the plots corresponding to soybean fertilization rates of 0, 50, 100 and 150%, respectively.

Soil characterization

A galvanized steel auger 4.5 cm in diameter was used for sampling at depths of 0 to 0.20 m and 0.20 to 0.40 m in November 2012, 24 months after the first liming. For each soil horizon, 15 random subsamples were collected from underneath the soybean plant rows (5 subsamples) and from the middle of the inter-row spaces (0.225 m from the plant row, 10 subsamples) from each plot. For each location (in-row and between rows), the 15 subsamples were combined to form a composite sample.

The composite samples were air-dried and sieved (2 mm mesh) and later analyzed to determine the pH (CaCl₂ 0.01 mol L⁻¹),

¹Cation exchange capacity; ²Percent base saturation; ³Soil organic matter.

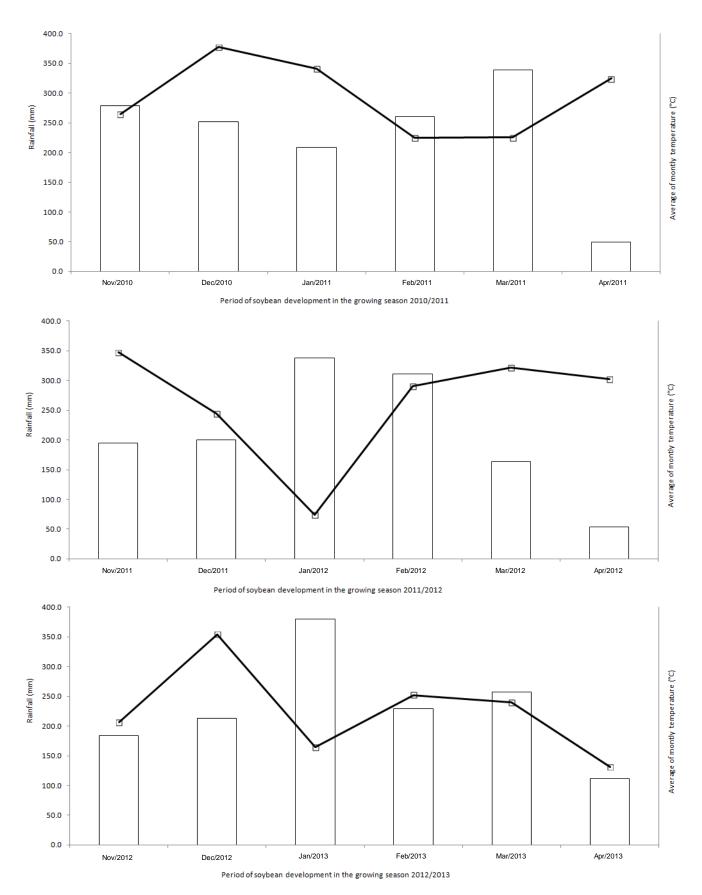


Figure 1. Temperature and rainfall in the experimental area during the trial (November, 2010 to March, 2013).

organic matter content, potential acidity (H + Al), Al, Ca, Mg and exchangeable K, and percent base saturation (V) was calculated. The content of Fe, Zn, Mn and Cu was also analyzed. Soil analyses were performed following the methods described above.

Soybean leaf diagnosis

Soybean leaf samples were collected from the upper third trifoliate at the R_2 growth stage (Fehr and Caviness, 1977). Petioles from 30 plants per plot were collected, washed and then dried under forcedair circulation at 65°C for 72 h before grinding and analyzing the samples for chemical composition. The concentrations of N, P, K, Ca, Mg and S were determined using methods described by Malavolta et al. (1997). Nitrogen was extracted with H_2SO_4 , and the other nutrients were extracted with a nitro-perchloric solution. The N concentration in the digested solution was determined by Kjeldahl analysis. The P, K, Ca, Mg and S concentrations were determined by atomic absorption spectrophotometry.

Yield components and grain yield

Soybeans were harvested on 02/04/2011, 22/03/2012 and 12/03/2013 from the usable area (8 central rows, with 0.5 m on each side disregarded) using a mechanical harvester. The soybean seed was weighed, and the yields were corrected to a moisture content of 130 g kg⁻¹ and converted to kg ha⁻¹. Agronomic characteristics, including the final plant population (calculated per ha from the number of plants in the two 4 m rows in the center of each plot), plant height (in cm), number of pods per plant and number of seeds per pod, were evaluated for 10 randomly chosen plants per plot, along with the 100-seed weight (calculated from 8 random samples per plot, adjusted to a moisture content of 130 g kg⁻¹).

Efficiency of fertilizer use

To calculate the efficiency of fertilizer use, we summed the average soybean seed yields in each corrective treatment (control, lime, lime + phosphogypsum, phosphogypsum) from the three growing seasons (2010/2011, 2011/2012, 2012/2013) and divided this sum by the amount of fertilizer used on the soybean crop during the three growing seasons.

Statistical analyses

An analysis of variance and F-test were performed for all variables. The soil corrective and level of fertilizer were considered fixed effects. Blocks, years and all interactions were considered random effects. A comparison of means was performed with a Turkey test (p \leq 0.05). To analyze the efficiency of fertilizer use, we used least significant difference (LSD) test at p \leq 0.05. A regression analysis was used for quantitative data (fertilizer levels). These analyses were performed using SAS statistical software (SAS Institute, 1999).

RESULTS

Soil attributes

Twenty-four months after the first application, liming significantly affected the following soil attributes at a

depth of 0 to 0.10 m: pH, Ca, Mg, Al, H + Al, CEC, V and Mn (Tables 2 and 3). pH, Ca and base saturation were greater in the treatments with lime (with or without phosphogypsum), and Mg was highest in the treatment with lime alone. Aluminum and H + Al were lowest in the treatments with lime. The highest value of cation exchange capacity (CEC) was achieved by the treatment with phosphogypsum alone. Treatment effects at a depth of 0.10 to 0.20 m were observed only for the levels of Mg, Al and Fe. The highest values of Mg and the lowest values of Al were obtained in the treatments with lime. The highest values of Fe were obtained in the treatments with phosphogypsum, which differed from the control. At a depth of 0.20 to 0.40 m, there was no effect of lime or phosphogypsum on soil attributes.

The levels of fertilizer affected pH, Ca, Mg, K, P, Cu, Fe, Mn and Zn at a depth of 0 to 0.10 m (Figures 2 and 3). The pH data fitted with a linear regression, and all nutrients with quadratic regressions. At a depth of 0.10 to 0.20 m, significant effects were observed only for the levels of P, Zn and Cu (Figure 3), which fitted with quadratic regressions. At a depth of 0.20 to 0.40 m, effects were observed on Ca, Mg, Al, K, V, P, Zn and Mn (Figure 4). With the exception of Zn, which fitted a linear equation, all the effects fitted with quadratic regressions.

Nutrient content in soybean leaves

During the first growing season (2010/2011), lime showed a significant effect on the leaf content of Ca, Mg and S (Table 4), with the lowest values occurring in the treatment with lime alone. The rates of fertilizer application affected the levels of P, Ca and Mg (Figure 5). Phosphorus and Ca fit quadratic adjustments, and Mg fit a linear equation. During the second growing season (2011/2012), the amendments affected the leaf content of N, K, Ca and S. N and K were highest in the treatment with lime alone. In contrast, the values of Ca and S were lowest in this treatment. The fertilization rates did not produce changes in the nutrient content. During the third growing season (2012/2013), the amendment did not alter the leaf content of nutrients. The fertilization rates affected the levels of N, P, Ca and S. N and S fit quadratic adjustments, and Ca and P fit linear adjustments.

Yield components and seed yield

The liming and phosphogypsum application produced significant changes in the 100-seed weight and seed yield of the soybeans evaluated during the first growing season (2010/2011) (Table 5). Liming produced the highest 100-seed weight, which differed from that found in the treatment with phosphogypsum. The seed yield was greater in the treatments with liming with or without

Table 2. Effects of the application of lime (L) and/or phosphogypsum (PG), fertilization rates and soil depth layers on the values of pH, Ca, Mg, Al, H + Al, K, cation exchange capacity (CEC) and base saturation (V) in the soil 24 months (November, 2012) after the first application of the soil correction. Crop managed under the no-tillage system (NTS).

| Soil corrective | | | | Depth 0 | - 0.10 m | | | |
|----------------------|-------------------|-------------------|-------------------|--------------------|---------------------------------|-------------------|------------------|-------------------|
| Soil corrective | рН | Ca | Mg | Al | H + Al | K | CEC | ٧ |
| | | | | cmo | l _c dm ⁻³ | | | % |
| Control | 5.3 ^{b1} | 1.09 ^c | 0.48 ^c | 0.16 ^a | 5.6 ^a | 0.26 ^a | 7.5 ^b | 24.6 ^b |
| Limestone | 5.7 ^a | 1.81 ^a | 1.14 ^a | 0.01 ^b | 4.1 ^b | 0.27 ^a | 7.4 ^b | 43.9 ^a |
| L + PG | 5.7 ^a | 1.75 ^a | 0.86 ^b | 0.04 ^b | 4.5 ^b | 0.24 ^a | 7.3 ^b | 39.5 ^a |
| Phosphogypsum | 5.3 ^b | 1.36 ^b | 0.43 ^c | 0.18 ^a | 5.9 ^a | 0.25 ^a | 8.0 ^a | 25.8 ^b |
| Fertilization rates | | | | | | | | |
| 0 | 5.7 | 1.35 | 0.68 | 0.10 | 5.0 | 0.25 | 7.3 | 32.2 |
| 0.5 | 5.5 | 1.66 | 0.80 | 0.09 | 5.0 | 0.24 | 7.7 | 35.5 |
| 1.0 | 5.5 | 1.58 | 0.76 | 0.09 | 5.0 | 0.23 | 7.5 | 34.5 |
| 1.5 | 5.4 | 1.42 | 0.66 | 0.11 | 5.2 | 0.30 | 7.6 | 31.6 |
| Source of variation | | | | F prob | oability | | | |
| Soil corrective (SC) | < 0.001 | < 0.001 | < 0.001 | < 0.001 | < 0.001 | 0.3983 | 0.0300 | < 0.001 |
| Fertilization (F) | 0.0027 | 0.0021 | 0.0496 | 0.8010 | 0.8889 | 0.0022 | 0.3256 | 0.3263 |
| SC x F | 0.7639 | 0.7241 | 0.5051 | 0.8200 | 0.7520 | 0.4399 | 0.8831 | 0.6169 |
| Soil corrective | | | | Depth 0.1 | 0 - 0.20 m | | | |
| Control | 5.1 ^a | 0.75 ^a | 0.29 ^b | 0.29 ^a | 6.0 ^a | 0.17 ^a | 7.2 ^a | 16.8 ^a |
| Limestone | 5.2 ^a | 0.94 ^a | 0.48 ^a | 0.18 ^b | 5.4 ^a | 0.13 ^a | 6.9 ^a | 22.7 ^a |
| L + PG | 5.3 ^a | 1.01 ^a | 0.45 ^a | 0.17 ^b | 5.4 ^a | 0.15 ^a | 7.0 ^a | 22.8 ^a |
| Phosphogypsum | 5.1 ^a | 0.95 ^a | 0.28 ^b | 0.26 ^{ab} | 6.0 ^a | 0.16 ^a | 7.4 ^a | 18.8 ^a |
| Fertilization rates | | | | | | | | |
| 0 | 5.3 | 0.77 | 0.33 | 0.25 | 5.9 | 0.15 | 7.2 | 17.5 |
| 0.5 | 5.2 | 0.97 | 0.43 | 0.23 | 5.6 | 0.15 | 7.1 | 22.2 |
| 1.0 | 5.2 | 0.98 | 0.41 | 0.19 | 5.5 | 0.15 | 7.1 | 21.9 |
| 1.5 | 5.1 | 0.92 | 0.32 | 0.23 | 5.8 | 0.16 | 7.2 | 19.4 |
| Source of variation | | | | F prob | oability | | | |
| Soil corrective (SC) | 0.0883 | 0.0838 | <0.001 | 0.0051 | 0.0554 | 0.2544 | 0.3389 | 0.0322 |
| Fertilization (F) | 0.0638 | 0.1306 | 0.0659 | 0.5168 | 0.4924 | 0.8582 | 0.9446 | 0.1570 |
| SC x F | 0.9237 | 0.9693 | 0.9515 | 0.8905 | 0.9129 | 0.7262 | 0.4727 | 0.9920 |
| Soil corrective | | | | Depth 0.2 | 20 - 0.40 m | | | |
| Control | 5.1 ^a | 0.61 ^a | 0.17 ^a | 0.29 ^a | 5.0 ^a | 0.11 ^a | 5.9 ^a | 15.1 ^a |
| Limestone | 5.1 ^a | 0.68 ^a | 0.23 ^a | 0.28 ^a | 5.2 ^a | 0.09 ^a | 6.2 ^a | 16.5 ^a |
| L + PG | 5.1 ^a | 0.66 ^a | 0.22 ^a | 0.26 ^a | 5.0 ^a | 0.10 ^a | 5.9 ^a | 16.9 ^a |
| Phosphogypsum | 5.2 ^a | 0.75 ^a | 0.20 ^a | 0.26 ^a | 4.9 ^a | 0.10 ^a | 5.9 ^a | 18.0 ^a |
| Fertilization rates | | | | | | | | |
| 0 | 5.1 | 0.52 | 0.16 | 0.33 | 5.2 | 0.09 | 6.0 | 12.9 |
| 0.5 | 5.2 | 0.61 | 0.20 | 0.27 | 5.0 | 0.09 | 5.9 | 15.6 |
| 1.0 | 5.1 | 0.79 | 0.24 | 0.23 | 4.7 | 0.09 | 5.8 | 19.5 |
| 1.5 | 5.1 | 0.80 | 0.23 | 0.26 | 5.2 | 0.12 | 6.3 | 18.6 |
| Source of variation | | | | F prob | oability | | | |
| Soil corrective (SC) | 0.7121 | 0.3670 | 0.1361 | 0.7395 | 0.6554 | 0.2141 | 0.6843 | 0.4173 |
| Fertilization (F) | 0.5837 | 0.0015 | 0.0278 | 0.0456 | 0.2413 | 0.0029 | 0.3628 | 0.0017 |
| SC x F | 0.9790 | 0.9718 | 0.9970 | 0.6190 | 0.9018 | 0.2453 | 0.9113 | 0.9210 |

 $^{^{1}}$ Means followed by the same letter in a column do not differ by the Tukey test at p < 0.05.

Table 3. Effects of the application of lime (L) and/or phosphogypsum (PG), fertilization rates and soil depth layers on the values of SOM, P, Cu, Zn, Fe and Mn in the soil 24 months (November 2012) after the first application of the soil correction. Crop managed under the no-tillage system (NTS).

| | | | Depth 0 | - 0.10 m | | |
|----------------------|---------------------|-------------------|-------------------|---------------------|--------------------|--------------------|
| Soil corrective | МО | Р | Cu | Zn | Fe | Mn |
| | g dm ⁻³ | | | mg dm ⁻³ | | |
| Control | 29.5 ^a 1 | 16.2 ^a | 1.34 ^a | 5.51 ^a | 23.4 ^a | 11.2 ^c |
| Limestone | 30.7 ^a | 14.1 ^a | 1.13 ^b | 5.38 ^a | 23.4 ^a | 15.9 ^a |
| L + PG | 30.0 ^a | 14.6 ^a | 1.13 ^b | 4.66 ^a | 23.2 ^a | 13.6 ^b |
| Phosphogypsum | 30.0 ^a | 17.0 ^a | 1.28 ^a | 4.84 ^a | 25.8 ^a | 14.0 ^{ab} |
| Fertilization rates | | | | | | |
| 0 | 29.9 | 5.9 | 1.18 | 2.71 | 24.0 | 11.6 |
| 0.5 | 31.5 | 14.0 | 1.15 | 4.89 | 24.4 | 15.3 |
| 1.0 | 29.8 | 13.5 | 1.19 | 5.89 | 21.4 | 14.7 |
| 1.5 | 28.9 | 28.4 | 1.36 | 6.89 | 25.9 | 13.1 |
| Source of variation | | | F prob | ability | | |
| Soil corrective (SC) | 0.6312 | 0.8023 | <0.001 | 0.5060 | 0.2154 | <0.001 |
| Fertilization (F) | 0.0561 | <0.001 | 0.0013 | <0.001 | 0.0235 | <0.001 |
| SC × F | 0.0521 | 0.9999 | 0.9615 | 0.7805 | 0.7864 | 0.7299 |
| Soil corrective | | | Denth 0.1 | 0 - 0.20 m | | |
| Control | 28.6ª | 12.2ª | 1.43 ^a | 5.41 ^a | 21.2 ^b | 8.1 ^a |
| Limestone | 27.4 ^a | 18.0 ^a | 1.51 ^a | 6.94 ^a | 23.5 ^{ab} | 9.8 ^a |
| L + PG | 27.3 ^a | 16.3 ^a | 1.49 ^a | 6.21 ^a | 22.7 ^{ab} | 8.6 ^a |
| Phosphogypsum | 29.2 ^a | 16.5 ^a | 1.53 ^a | 6.67 ^a | 24.2 ^a | 10.3 ^a |
| Fertilization rates | | | | | | |
| 0 | 29.5 | 5.7 | 1.38 | 2.01 | 22.8 | 8.0 |
| 0.5 | 28.8 | 12.7 | 1.47 | 5.98 | 23.3 | 10.4 |
| 1.0 | 27.9 | 18.2 | 1.48 | 7.73 | 21.8 | 9.9 |
| 1.5 | 26.4 | 29.5 | 1.46 | 9.51 | 23.7 | 9.9 8.6 |
| 1.5 | 20.4 | 29.5 | | | 23.7 | 0.0 |
| Source of variation | | | - | oability | | |
| Soil corrective (SC) | 0.0999 | 0.5553 | 0.6705 | 0.8779 | 0.0100 | 0.1517 |
| Fertilization (F) | 0.0680 | <0.001 | 0.0420 | 0.0036 | 0.1773 | 0.0936 |
| SC × F | 0.3586 | 0.9299 | 0.5098 | 0.6456 | 0.6842 | 0.9795 |
| Soil corrective | | | _ | 20 - 0.40 m | | |
| Control | 24.2 ^a | 2.1 ^a | 1.40 ^a | 2.47 ^a | 20.1 ^a | 6.3 ^a |
| Limestone | 25.0 ^a | 2.4 ^a | 1.41 ^a | 2.86 ^a | 21.3 ^a | 8.4 ^a |
| L + PG | 23.9 ^a | 2.7 ^a | 1.46 ^a | 2.69 ^a | 21.5 ^a | 6.4 ^a |
| Phosphogypsum | 26.0 ^a | 2.4 ^a | 1.46 ^a | 2.60 ^a | 21.9 ^a | 7.8 ^a |
| Fertilization rates | | | | | | |
| 0 | 25.4 | 1.3 | 1.38 | 1.58 | 21.7 | 5.8 |
| 0.5 | 24.8 | 2.1 | 1.46 | 2.17 | 20.6 | 6.5 |
| 1.0 | 23.9 | 3.1 | 1.40 | 3.32 | 20.7 | 8.1 |
| 1.5 | 24.9 | 3.2 | 1.51 | 3.56 | 21.8 | 8.4 |
| Source of variation | | | F prob | ability | | |
| Soil corrective (SC) | 0.0548 | 0.7126 | 0.6285 | 0.8560 | 0.1745 | 0.0792 |
| Fertilization (F) | 0.3078 | 0.0004 | 0.1490 | 0.0002 | 0.3058 | 0.0256 |
| SC × F | 0.6677 | 0.6407 | 0.9797 | 0.9698 | 0.9978 | 0.8265 |

 $^{^{1}}$ Means followed by the same letter in a column do not differ by the Tukey test at p < 0.05.

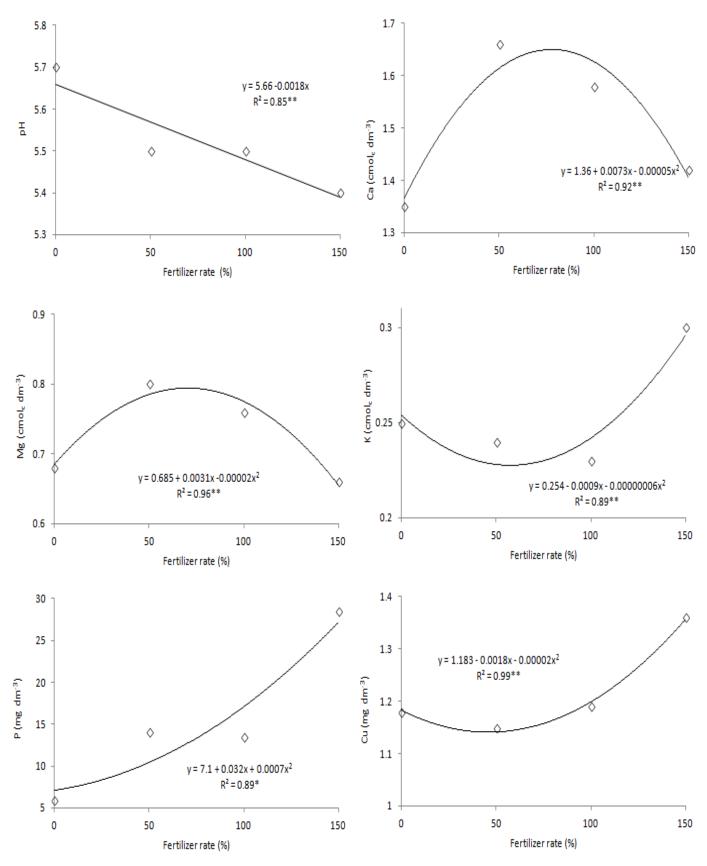


Figure 2. Chemical attributes of the soil (pH, Ca, Mg, K, P and Cu) in the depth 0 - 10 cm as a function of fertilization rates. Soil measured 24 months after first application of lime and phosphogypsum under the no-tillage system.

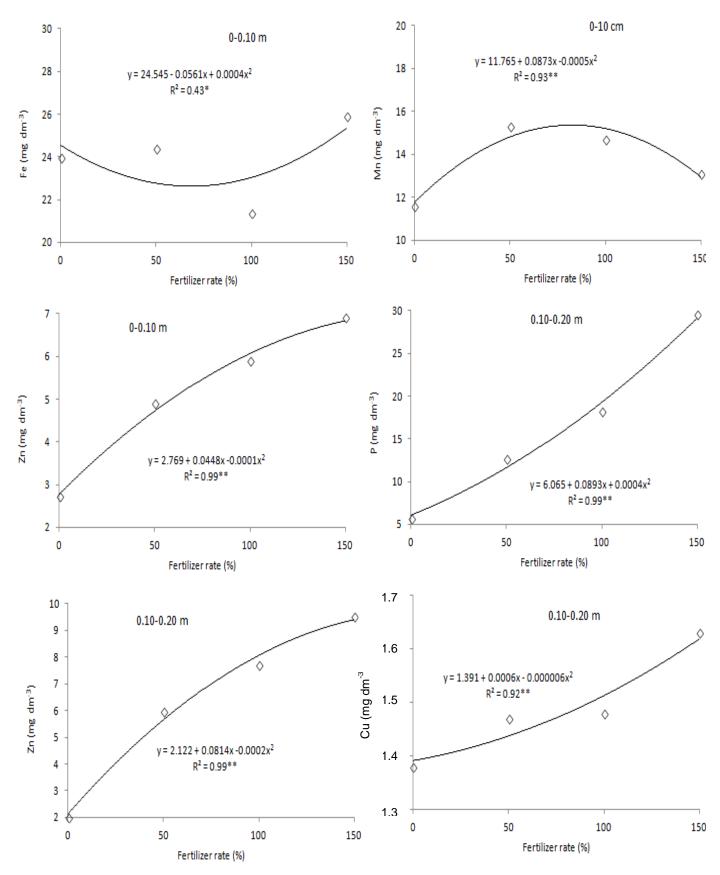


Figure 3. Chemical attributes of the soil (Fe, Mn and Zn in the depth 0 - 10 cm and P, Zn and Cu in the depth 10 - 20 cm) as a function of fertilization rates. Soil measured 24 months after first application of lime and phosphogypsum under the no-tillage system.

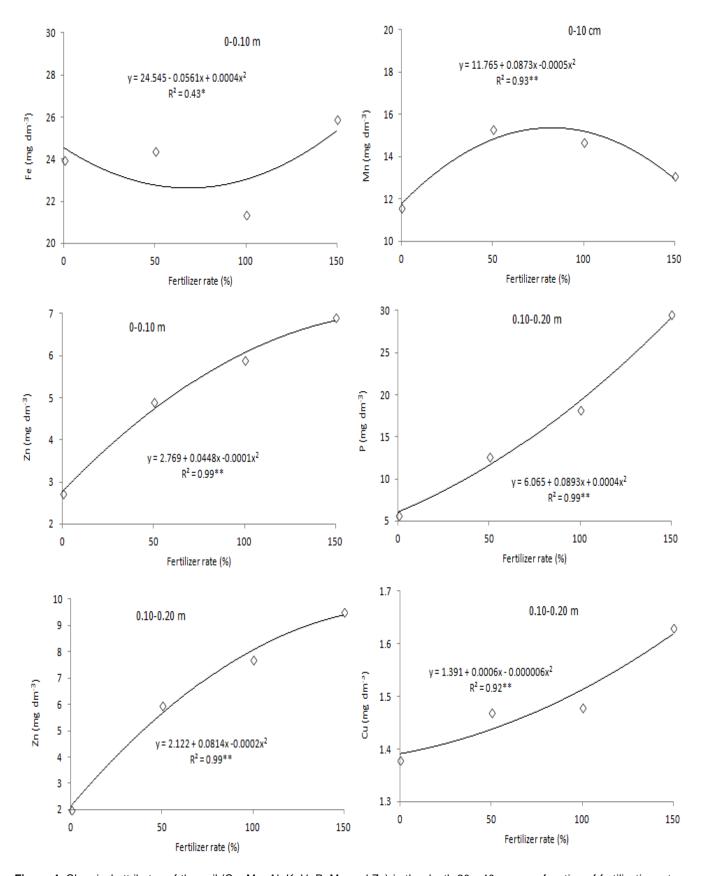


Figure 4. Chemical attributes of the soil (Ca, Mg, Al, K, V, P, Mn and Zn) in the depth 20 - 40 cm as a function of fertilization rates. Soil measured 24 months after first application of lime and phosphogypsum under the no-tillage system.

Table 4. Effects of the application of limestone (L) and/or phosphogypsum (PG), fertilization rates and growing season on the values of N, P, K, Ca, Mg and S in leaves sampled at the full flowering state (R2) in the soybean crop. Crop managed under the no-tillage system (NTS).

| | Growing season 2010/2011 | | | | | | | | |
|----------------------------|---------------------------|---------------------------|-----------------------------|---------------------------|---------------------------|---------------------------|--|--|--|
| Soil corrective | N | Р | K | Ca | Mg | S | | | |
| | | | g l | kg ⁻¹ | | | | | |
| Control | 39.8 ^a 1 | 2.42 ^a | 15.97 ^a | 12.88 ^{ab} | 3.18 ^a | 1.53 ^a | | | |
| Limestone | 37.1 ^a | 2.45 ^a | 16.01 ^a | 12.38 ^b | 2.56 ^b | 1.30 ^b | | | |
| L + PG | 37.5 ^a | 2.46 ^a | 15.63 ^a | 13.38 ^{ab} | 2.97 ^{ab} | 1.59 ^a | | | |
| Phosphogypsum | 37.3 ^a | 2.45 ^a | 16.59 ^a | 13.72 ^a | 3.09 ^a | 1.46 ^{ab} | | | |
| Fertilization rates | | | | | | | | | |
| 0 | 37.5 | 2.07 | 15.39 | 12.95 | 2.65 | 1.38 | | | |
| 0.5 | 37.2 | 2.53 | 16.11 | 12.13 | 2.84 | 1.43 | | | |
| 1.0 | 38.7 | 2.57 | 16.49 | 13.20 | 3.08 | 1.53 | | | |
| 1.5 | 38.2 | 2.61 | 16.20 | 14.08 | 3.23 | 1.54 | | | |
| Source of variation | | | F prol | oability | | | | | |
| Soil corrective (SC) | 0.5296 | 0.9732 | 0.8328 | 0.0221 | 0.0036 | 0.0057 | | | |
| Fertilization (F) | 0.8703 | < 0.001 | 0.7517 | 0.0009 | 0.0075 | 0.1898 | | | |
| SC × F | 0.6877 | 0.4265 | 0.9909 | 0.2578 | 0.6388 | 0.9456 | | | |
| Soil corrective | | | Growing seas | son 2011/2012 | | | | | |
| Control | 30.8 ^b | 3.12 ^a | 17.16 ^b | 10.39 ^b | 2.83 ^a | 1.64 ^b | | | |
| Limestone | 36.3 ^a | 3.18 ^a | 19.46 ^a | 10.25 ^b | 3.06 ^a | 1.40 ^c | | | |
| L + PG | 34.8 ^{ab} | 3.01 ^a | 16.69 ^b | 10.47 ^{ab} | 3.04 ^a | 1.61 ^{bc} | | | |
| Phosphogypsum | 33.2 ^{ab} | 3.21 ^a | 16.69 ^b | 11.52 ^a | 3.15 ^a | 1.88 ^a | | | |
| Fertilization rates | | | | | | | | | |
| 0 | 31.5 | 3.04 | 18.41 | 11.03 | 2.96 | 1.65 | | | |
| 0.5 | 34.9 | 3.27 | 16.93 | 9.97 | 2.89 | 1.67 | | | |
| 1.0 | 34.2 | 3.21 | 16.97 | 10.84 | 3.31 | 1.57 | | | |
| 1.5 | 34.7 | 3.01 | 17.69 | 10.80 | 2.92 | 1.64 | | | |
| Source of variation | | | Enrol | aahilitu | | | | | |
| Soil corrective (SC) | 0.0276 | 0.3699 | 0.0005 | oability 0.0118 | 0.4854 | <0.001 | | | |
| Fertilization (F) | 0.0270 | 0.0751 | 0.0003 | 0.0566 | 0.4654 | 0.6699 | | | |
| SC x F | 0.4622 | 0.0751 | 0.1291 | 0.0300 | 0.1094 | 0.4508 | | | |
| | | | | | | | | | |
| Soil corrective Control | 44.0 ^a | 2.99 ^a | 14.68 ^a | 9.47 ^a | 2.25 ^a | 2.33 ^a | | | |
| | 44.0 46.4 ^a | 2.99 3.06 ^a | 14.00 13.53 ^a | 9.47 9.30 ^a | 2.23 2.18 ^a | 2.33 2.43 ^a | | | |
| Limestone | 46.4 45.6 ^a | 3.06 3.13 ^a | 13.53 14.81 ^a | 9.30 9.96 ^a | 2.10 2.20 ^a | 2.43 2.44 ^a | | | |
| L + PG Phosphogypsum | 45.6 43.8 ^a | 3.13 3.09 ^a | 14.81 14.26 ^a | 9.96 9.90 ^a | 2.20 2.08 ^a | 2.44 2.48 ^a | | | |
| Fertilization rates | | | - | | | | | | |
| 0 | 41.4 | 2.60 | 14.20 | 8.93 | 2.01 | 2.22 | | | |
| 0.5 | 45.6 | 3.05 | 14.25 | 9.25 | 2.24 | 2.43 | | | |
| 1.0 | 45.9 | 3.29 | 14.23 | 10.17 | 2.23 | 2.43 | | | |
| 1.5 | 46.9 | 3.29 | 14.36 | 10.17 | 2.23 | 2.52 | | | |
| Source of variation | | | | oability | | | | | |
| Soil corrective (SC) | 0.1572 | 0.1295 | 0.0812 | 0.1192 | 0.4359 | 0.3306 | | | |
| Fertilization (F) | 0.1372 | <0.001 | 0.9603 | 0.0001 | 0.4339 | 0.0022 | | | |
| SC x F | 0.8952 | 0.3019 | 0.5854 | 0.8080 | 0.1200 | 0.6687 | | | |

 $^{^{1}}$ Means followed by the same letter in a column do not differ by the Tukey test at p < 0.05.

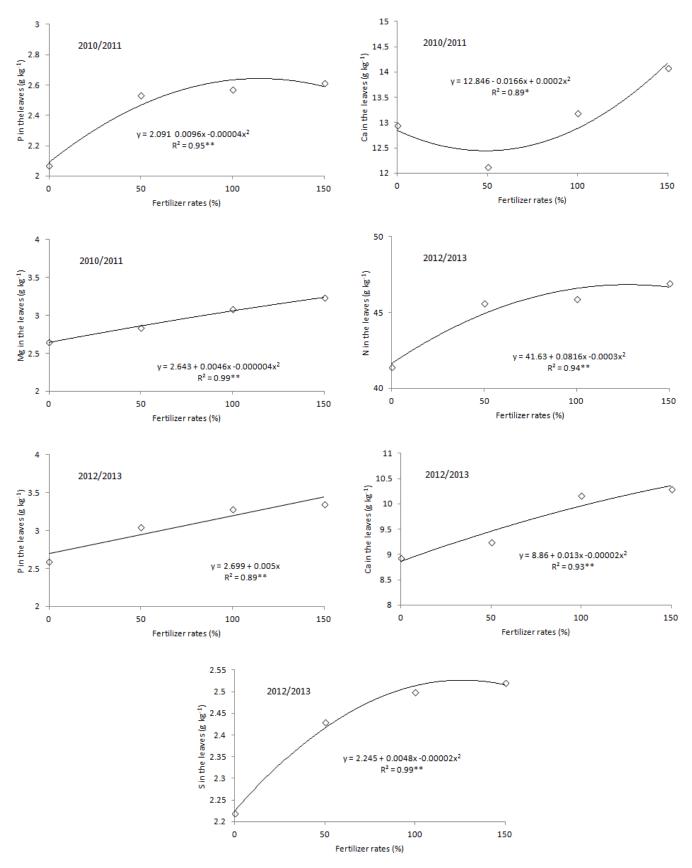


Figure 5. Concentration of nutrients (P, Ca, Mg in the growing season 2010/2011 and N, P, Ca and S in the growing season 2012/2013) in soybean leaves as a function of fertilization rates.

Table 5. Effects of application of lime (L) and/or phosphogypsum (PG), fertilization rates and growing season on the plant population (POP), number of pods plant (NPP), number of seeds pod (NSP), 100-seed weight (W100) and seed yield (Yield) of the soybean crop. Crop cultivated under the no-tillage system (NTS).

| | | | ing season 2010 | | |
|----------------------|----------------------|-----------------|-------------------|---------------------|---------------------|
| Soil corrective | POP | NPP | NSP | W100 | Yield |
| | nº × 1000 | nº | nº | g | kg ha ⁻¹ |
| Control | 180.6 ^a 1 | 71 ^a | 1.66 ^a | 14.20 ^{ab} | 2013 ^b |
| Limestone | 191.1 ^a | 71 ^a | 1.66 ^a | 14.49 ^a | 2494 ^a |
| L + PG | 203.5 ^a | 64 ^a | 1.78 ^a | 13.68 ^{ab} | 2534 ^a |
| Phosphogypsum | 195.7 ^a | 69 ^a | 1.61 ^a | 13.19 ^b | 2063 ^b |
| Fertilization rates | | | | | |
| 0 | 163.9 | 62 | 1.58 | 13.60 | 2042 |
| 0.5 | 181.0 | 67 | 1.76 | 13.98 | 2348 |
| 1.0 | 200.8 | 68 | 1.75 | 13.53 | 2358 |
| 1.5 | 225.1 | 77 | 1.61 | 13.46 | 2356 |
| Source of variation | | | F probability | | |
| Soil corrective (SC) | 0.1788 | 0.4581 | 0.0975 | 0.0094 | 0.0009 |
| Fertilization (F) | < 0.001 | 0.0242 | 0.0277 | 0.0785 | 0.0143 |
| SC x F | 0.6293 | 0.8306 | 0.1929 | 0.7521 | 0.1234 |
| Soil corrective | | | ing season 201 | 1/2012 | |
| Control | 278.1 ^a | 59 ^a | 1.54 ^a | 13.09 ^a | 1819 ^c |
| Limestone | 282.6 ^a | 64 ^a | 1.65 ^a | 13.16 ^a | 2538 ^a |
| L + PG | 282.8 ^a | 60 ^a | 1.63 ^a | 13.28 ^a | 2276 ^b |
| Phosphogypsum | 277.4 ^a | 57 ^a | 1.60 ^a | 12.59 ^a | 1874 ^c |
| Fertilization rates | | | | | |
| 0 | 278.3 | 54 | 1.46 | 12.96 | 1490 |
| 0.5 | 277.9 | 57 | 1.61 | 12.82 | 1899 |
| 1.0 | 282.5 | 61 | 1.66 | 13.16 | 2459 |
| 1.5 | 282.3 | 67 | 1.69 | 13.18 | 2659 |
| Source of variation | | | F probability | | |
| Soil corrective (SC) | 0.9122 | 0.5272 | 0.4898 | 0.2131 | < 0.001 |
| Fertilization (F) | 0.9423 | 0.0358 | 0.0239 | 0.6842 | < 0.001 |
| SC x F | 0.8982 | 0.2657 | 0.7106 | 0.2680 | 0.1293 |
| Soil corrective | | | ing season 2012 | 2/2013 | |
| Control | 393.3ª | 27 ^a | 1.90 ^a | 17.00 ^a | 2820 ^b |
| Limestone | 394.9 ^a | 30 ^a | 1.99 ^a | 17.58 ^a | 3151 ^a |
| L + PG | 400.1 ^a | 31 ^a | 2.01 ^a | 17.25 ^a | 3248 ^a |
| Phosphogypsum | 408.8 ^a | 27 ^a | 1.93 ^a | 17.29 ^a | 3148 ^{ab} |
| Fertilization rates | | | | | |
| 0 | 389.0 | 22 | 1.90 | 16.05 | 2674 |
| 0.5 | 404.0 | 28 | 2.00 | 17.27 | 3083 |
| 1.0 | 406.6 | 30 | 2.00 | 17.68 | 3228 |
| 1.5 | 397.5 | 34 | 1.93 | 18.12 | 3382 |
| Source of variation | | | F probability | | |
| Soil corrective (SC) | 0.0842 | 0.0159 | 0.4232 | 0.2216 | 0.0001 |
| Fertilization (F) | 0.1035 | < 0.001 | 0.9890 | < 0.001 | < 0.001 |
| SC x F | 0.2740 | 0.9129 | 0.6809 | 0.1584 | 0.0907 |

 $^{^{1}}$ Means followed by the same letter in a column do not differ by the Tukey test at p < 0.05.

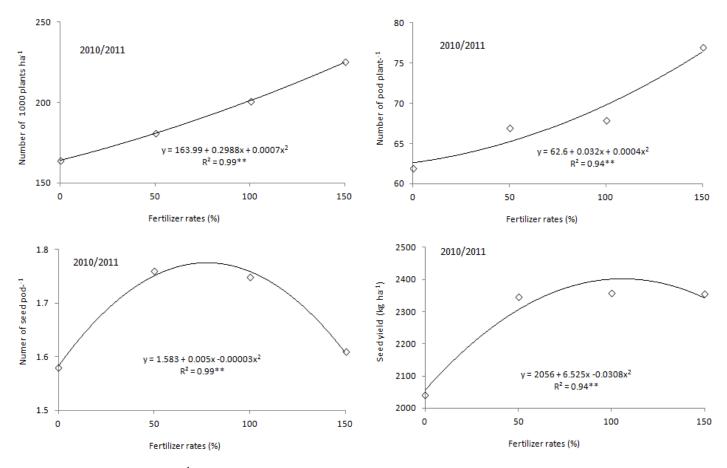


Figure 6. Number of plants ha⁻¹, yield components and seed yield of soybean in the growing season 2010/2011 as a function of fertilization rates.

Table 6. Efficiency of PK fertilizer use in the soybean crop cultivated under the NTS as a function of lime (L) and phosphogypsum (PG) application.

| Variable | No corrective | Limestone | L + FG | Phosphogypsum |
|---|----------------------|--------------------|--------------------|--------------------|
| SSASY - A ¹ | 6652 | 8183 | 8058 | 7085 |
| K ₂ O-P ₂ O ₅ applied by fertilizer - B ² | 360 | 360 | 360 | 360 |
| Fertilizer efficiency use - A/B | 18.48 B ³ | 22.73 ^A | 22.38 ^A | 19.68 ^B |

 1 SSASY - Sum of average of the soybean seed yield from application of rates 0, 50, 100 and 150% of the recommended fertilization in three growing seasons (2010/2011, 2011/2012 and 2012/2013). Average of the amount of P_2O_5 and K_2O applied via fertilizer at doses 0, 50, 100 and 150% of the recommended fertilization in three growing seasons (2010/2011/2011/2012 and 2012/2013). Thus, it was applied for each nutrient 0, 40, 80 and 120 kg ha⁻¹, totaling 240 kg ha⁻¹ of each nutrient, that is, applied 480 kg $K_2O + P_2O_5$. The average was 120 kg ha⁻¹ (480/4 fertilizer rates) for each year time 3 (growing season) = 360. 3 Means followed by the same letter in the row do not differ by LSD test at p < 0.05.

phosphogypsum than without liming. The fertilization rates had significant quadratic effects on the plant population, the number of pods plant⁻¹, the number of seeds pod⁻¹ and the seed yield (Figure 6).

During the second growing season (2011/2012), the lime treatment significantly affected soybean seed yield (Table 5). Lime applied alone provided the highest yield and this value differed from those produced by the other

treatments. The fertilization rates had significant quadratic effects on the number of pods plant⁻¹, number of seeds pod⁻¹ and seed yield (Figure 7). During the third growing season (2012/2013), significant treatment effects were found only for seed yield. The application of lime with or without phosphogypsum resulted in higher seed yields, which differed from the values found for the control. The fertilization rates had significant quadratic

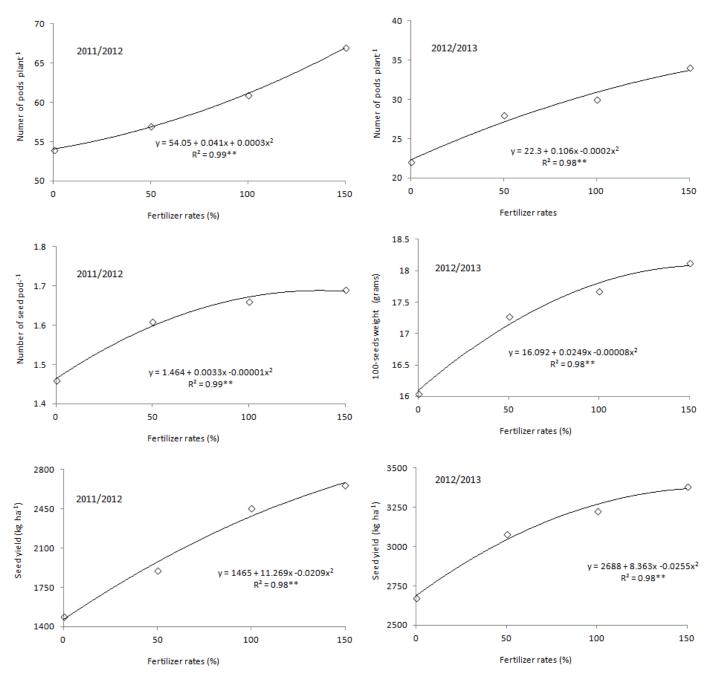


Figure 7. Yield components and seed yield of soybean in the growing season 2011/2012 and 2012/2013 as a function of fertilization rates.

effects on the number of pods plant⁻¹, 100-seed weight and seed yield.

Efficiency of fertilizer use

The treatments with lime, with or without phosphogypsum, allowed a higher efficiency of fertilizer use. This value differed from those found for the control and for phosphogypsum alone (Table 8).

DISCUSSION

Soil attributes

The lime treatment produced the greatest changes in soil properties, resulting in increased pH levels, an increased availability of Ca and Mg and a decreased content of Al and H + Al, with increases in CEC and base saturation at a depth of 0 to 0.10 m 24 months after the first application. However, the effects on soil attributes were

more pronounced in the surface layer, with small changes in the 0.10 to 0.20 m layer (only Mg, Al and Fe) and no change in the 0.20 to 0.40 m layer. Results from the field have shown that the movement of the limestone when applied on the soil surface is very low (Gascho and Parker, 2001; Conyers et al., 2003; Tang et al., 2003; Caires et al., 2008; Soratto and Crusciol, 2008a, b; Churka Blum et al., 2013). Our results imply that the reaction of the non-incorporated lime with the soil was delayed; only supplying acidity correction in the first few cm of the soil.

The application of phosphogypsum, which is not a soil corrective, did not result in significant alterations in the pH of the soil or in other attributes. If applied in large quantities, this soil conditioner can cause the leaching of bases (Ca, Mg and K) to deeper layers (Ramos et al., 2006; Caires et al., 2008). In our experiment, however, higher levels of these bases were not observed in the 0.20 to 0.40 m layer.

The pH level decreased linearly with increases in the amount of fertilizer applied. An explanation of this result is that nitrogen fertilizers were applied when common bean was cultivated during the off season between the soybean growing seasons. These fertilizers were applied in the same correspondent plots of soybean. In other words, the common bean plots which received a rate of 150% of fertilizer were the same with the ones which received 150% of fertilizer on soybean. The nitrogenous fertilizers ammonium sulfate and urea contain N-NH₄⁺ and N-NH₃⁻, respectively. In the soil, these ions rapidly oxidize to nitrate, releasing H⁺ and producing significant decreases in pH (Fageria et al., 2011; Nascente et al., 2012).

The content of Ca and Mg increased up to a certain level of fertilization and then began to decrease. This finding can be explained by competition with K for soil adsorption sites. The increased application of KCI increased the amount of K in the soil (Figure 2). According to Liebhardt (1981), the application of K to soils directly affects Ca and Mg, as they compete for the same adsorption sites in the soil. Thus, it is probable that greater amounts of K caused the release of Ca and Mg from soil colloids and possibly the leaching of these nutrients to deeper layers. The observed levels of these nutrients in the 0.20 to 0.40 m layer (Figure 4) are consistent with this suggestion.

Phosphorus increased significantly with increasing fertilization rates. This result can be explained by an increase of P in amounts larger than that exported by the seeds (Figures 9 and 10). Cu, Zn and Mn increased with increasing levels of fertilizer. These findings may reflect the decreased pH in those treatments that increased the availability of cationic micronutrients (Fageria et al., 2011). Micronutrients are influenced strongly by pH, and decreases in pH are accompanied by a greater availability of these nutrients (Fageria et al., 2002). Note that the levels of Fe were affected little by fertilization

rates. A possible reason for this outcome is that Oxisols typically contain large amounts of Fe, and agricultural management alters the balance of this nutrient in the soil only in rare cases (Fageria et al., 2002).

At a depth of 0.20 to 0.40 m, due to the probable leaching of Ca and Mg, increased levels of fertilization produced higher levels of these nutrients. Potassium, which is more readily leachable, also showed increases with increasing rates of fertilization. However, these increases were much lower than those obtained for Ca and Mg. These results indicate that the majority of K was absorbed by soil colloids in the superficial layers. Most likely, the addition of phosphate fertilizers at the 0.10 to 0.20 m depth produced increased levels of P at depth as fertilization rates increased. With rising levels of bases at depth (Ca, Mg and K), increases in base saturation were expected, and these expected increases were observed. The micronutrients Zn and Mn also showed significant increases with increasing levels of fertilization. This result may reflect the lower pH normally found in the deeper soil layers (Table 2).

Nutrient content in soybean leaves

The decrease in Ca, Mg and S content in the treatment with lime can be explained by the dilution effect because the Ca and Mg content in the soil increased, resulting in better plant development and higher seed yields (Table 5). Caires et al. (2001) have reported that in soybean crops, liming allowed the absorption of nutrients to increase, resulting in higher seed yield. In contrast, increased levels of N and K, the nutrients accumulated in greater amounts in the plant, should be related to adequate conditions for the development of the root system, with positive effects on biological nitrogen fixation and the absorption of K. According to Rosolem and Marcello (1998), Costa and Rosolem (2007) and Veronese et al. (2012), liming can increase the growth of soybean root in terms of length and surface area. The increase in nutrient content in the soybean leaves with the increasing levels of P and K resulting from fertilization was expected given the original low soil fertility (Table 1). This increase was associated with an increase in the levels of these nutrients in the soil (Figures 3 and 4) and the decrease in soil acidity caused by liming, increasing the availability of the nutrients to the plants (Fageria et al. 2011). According to Gonçalves Junior et al. (2010) as the soil from the Cerrado Region normally has low fertility, P and K application to the soybean crop provided better plant nutrition and resulted in higher grain yield than no fertilization.

Yield components and seed yield

Liming increased soybean seed yield in the three

seasons. This result may be a consequence of the changes in the soil properties caused by the application of the limestone to the soil, such as increased pH, increased availability of Ca and Mg, base saturation and CEC. Likewise, Caires et al. (2001) have reported significant increases in the seed yield of soybean due to the use of surface liming without incorporation in the NTS. The authors attributed this result to the greater availability of Mg.

On the other hand, the application of phosphogypsum did not increase the crop seed yield over the 3 years of cultivation. Similar results have been reported in other studies in areas of annual crops when the soil was under the NTS (Caires et al., 1998, 2003). The lack of a soybean to the application response by phosphogypsum may have occurred because application of this soil conditioner does not provide acidity correction in the topsoil and because the growth of sovbean roots in the absence of soil water deficit is not influenced by decreased Al saturation in the subsoil (Caires et al., 2006a). Indeed, it appears that the values of Al in the deep layers of the soil were low (less than 0.3) mmol^c dm⁻³) and were affected little by the application of phosphogypsum, showing no differences from the results of the other treatments.

Increased fertilization rates produced increases in the plant population, yield components and seed yield to a certain point. Thus, it can be inferred that it is likely that other nutrients may have limited the seed yield after fertilization with only P and K.

Efficiency of fertilizer use

For every kilogram of fertilizer applied, 22.73 kg of soybean seeds were obtained from the lime application and 22.38 kg from the application of lime with phosphogypsum. These values differed from the control (18.48 kg of soybean seeds) and from the application of phosphogypsum alone (19.68 kg of soybean seeds). The results show that the lime effectively increased the soybean seed yield and improved the efficiency of fertilizer use. In contrast, the application of phosphogypsum produced no significant increase in seed yield.

Conclusion

The use of lime alone or in combination with phosphogypsum improved soil chemical characteristics, the nutritional status of the soybean plants and yield. In contrast, the application of phosphogypsum alone did not produce significant alterations in the soil or affect soybean seed yield. The use on soybean of increasing application rates of fertilizer containing P and K (simple superphosphate, triple superphosphate and potassium

chloride) during three growing seasons produced significant increases in the seed yield of the crop. Accordingly, the application of lime increased fertilization efficiency, enabling a greater production of soybean seeds per kg of fertilizer applied. Based on these results, it is clear that, particularly in areas of the Cerrado, the surface application of lime without incorporation under the NTS is a better practice to increase fertilizer efficiency than phosphogypsum application. The application of phosphogypsum alone was not effective under the conditions tested in the experiment, which included soil acidity in the surface layer.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Self-build silos for storage of cereals in African rural villages

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Prototypes of silos for the storage of cereals were designed and built at the Department of Agricultural, Food and Forestry Systems of University of Firenze. The silos were planned specifically to be used in Tanzania, by individual farmers operating in the inland areas of the country. Nevertheless, with a capacity of 1 m³ and 2 m³, they can be considered suitable for several African areas. Except for some details that require local blacksmith workshops, the users can self-build the silos. The possibility to use materials and equipment normally available on site is included among the criteria to take into account during the design phase. For example, corrugated galvanized iron, employed by the local population as cover for houses, can be considered a suitable material, as well as raw-earth, traditionally used in African rural areas. To demonstrate the reliability of the design and the functionality of the adopted solutions, unskilled staff of the Department built the silo in the workshop, using only simple tools available almost anywhere. The paper gives suitable information to replicate the building of this kind of silos, where unskilled labor is available at the family and/or farm level.

Key words: Silos, self-building, raw-earth, rural villages, maize storage, Tanzania.

INTRODUCTION

Especially for the development of the agricultural sector, two main categories of interventions can be schematized in developing countries in support of the economy and social progress (Dixon et al., 2001; Barker, 2007; Hoffmann, 2011).

The first category includes interventions that can transfer on-site facilities and organization having a markedly industrial nature, similar to those present in the countries of origin. In these cases, to be effective and

long lasting, the actions generally need to be set in an industrialized context, capable of providing the new body the contribution of complementary tools, continuity of energy supply and qualified technical and administrative workers, essential for operation. When these conditions do not occur, the intervention is likely to result in an ephemeral attempt, as not infrequently it has happened in the past. These interventions are characterized by a significant financial commitment and by limited needs

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related to the number and duration of the presence of qualified people provided by donors. Anyhow, this type of intervention has a low impact on the social-economic local environment, at least in the short and medium term,

since it involves only a small number of highly qualified local technicians (HSI, 2011).

The second category comprises the interventions that at first analyze local conditions, in particular taking into consideration the socio-economic situation and the traditional production means and techniques actually available on site. As a result, the introduction of few elements of scientific progress improves the agricultural and livestock productions thank to adoption intervention strategies by local people, which do not seriously change traditional methods. This type of action is characterized by a limited financial commitment and, mainly in the initial stages, by a need for the presence of technical personnel on site. The social impact of these actions is generally very high since, in addition to technical personnel for the control and dissemination of knowledge, numerous local agricultural operators are directly involved. These interventions can result in extensive agro-livestock chains able to develop the socioeconomic conditions of relatively large areas, in gradual but sustained and solid way (Kwa, 2001; Smalley, 2013).

The Department of Agricultural, Food and Forestry Systems of University of Firenze (GESAAF) has operated in various ways in the context of both types of interventions above described, also carrying out trainings in Italy and locally with the implementation of on-site pilot interventions.

The solution proposed in this work can be considered an intervention of the second type. Silos have been planned for the specific needs of a typical family in the area of Itigi, Singida Region, Tanzania that is a typical rural village of Africa, where GESAAF has been called to solve problems related to grains storage. Usually, farmers in this area owns an average of 1 to 2 ha of farmland, which has a typical yield of about 1.000 kg/ha of maize or other grains (FAO, 2013).

In this area, the rainy season starts in early November and ends in late March so the sowing is done in late October and the harvesting in early April (FAO, 2013). After about two weeks of outdoor drying, the maize reaches a water content of around 12 to 13%, and then it can be stored with a water content below 14%, level considered safe for maize storage (Golob, 2009; Wambugu et al., 2009; Yakubu et al., 2010). The crop must be able to be stored both for the next sowing, which occurs 6 months after harvest, and for food use until the next harvest, which occurs after 11 to 12 months.

Currently a significant portion of the grain stored is lost because of poor storage conditions (Proctor, 1994; Coulter and Schneider, 2004; Golob et al., 2009; Yakubu et al., 2010). The stored material can undergo direct attack by animals (insects, small mammals and birds), while the increase of moisture and temperature, in particular in the course of the rainy season, can cause the onset of

mildew and microorganisms, and the germination of the seeds. GESAAF has designed containers able to reduce losses of food. Besides, the storage of maize, the designed silos can store other kind of cereals (wheat, barley) and other seeds (e.g. beans).

The total size of the silos depends on the presence of walls and empty spaces, planned to increase its insulation from the heat. Cereal grain can generate heat, but this process is generally negligible for small amounts, and in any case cannot be avoided unless ventilation is used. In this case, however, other, more severe, drawbacks have to be highlighted such as the increase of humidity (Wambugu et al., 2009).

MATERIALS AND METHODS

Two silos were designed, one of 1 m³ and another of 2 m³, constructed in corrugated galvanized iron and insulated with earthen walls, with materials and equipment existing in Itigi, Tanzania. The same equipment and materials are available in many other rural areas in African Countries. The container has been structurally designed according to the relevant Eurocodes (EN 1991-4, 2006; EN 1993-1-3, 2006; EN 1993-4-1, 2007). The drawing of the content must be made on a daily basis without the outside agents penetrate inside (Petrovskij, 1990; Proctor, 1994; Udoh et al., 2000; Coulter and Schneider, 2004), so silos should be impenetrable, airtight and able to maintain a temperature as low and constant as possible. It has to be accessible for cleaning and filling on a yearly basis.

For the construction of the silo the following materials are necessary: corrugated galvanized steel sheet, galvanized smooth sheet, angle section bars 30×30×3 mm, rivets 4 mm nominal diameter, strips of waxed fabric, e.g. truck tarpaulin, which can be formed both from organic or synthetic material, sturdy fabric, sturdy thread to sew the fabric, twine, bitumen, galvanized network, 20×20 mm mesh, clayey soil, coarse gravel, leafy branches, aluminum thin sheets, if available.

The tools needed for the construction are: hand shears (necessary to cut sheets), electric or hand driller and 4 mm diameter drill bits, hand riveter, welder, a small metal container to heat the bitumen, a brush to spread the bitumen, sturdy needle to sew the fabric. The silo consists of the following elements shown in Figure 1.

- 1. Bottom sheet: round edges allow adapting the container inside the perimeter wall (Figure 1:1).
- 2. Sidewall of the container cylinder: this part consists of three sheets joined together by rivets; the joints are sealed with outside applied bitumen (Figure 1:2).
- 3. Angle section bar frame: this structure has the task to strengthen the upper closure of the container, to allow a person to rest over in order to enter into the container (Figure 1:3).
- 4. Upper closure: the hole allows a person to enter into the container; round edges allow to adapt the container inside the perimeter wall and to eliminate the trouble of alignment with the bottom (Figure 1:4).
- 5. Lid: it must be affixed and sealed after filling; yearly it must be removed and repositioned (Figure 1:5).
- 6. Drawing device (Figure 2): it consists of an air lock chamber closed by a sluice valve; the length of the device is due to the need to reach the outside of the external walls (Figure 1:6).
- 7. Bag: to complete the emptying (Figure 1:7); the rear and the lateral parts of the bag are raised by means of the twines according to the scheme in Figure 3.

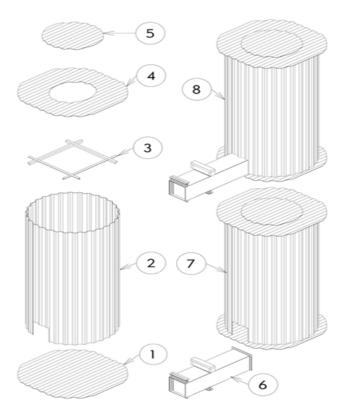


Figure 1. The silo and its components.

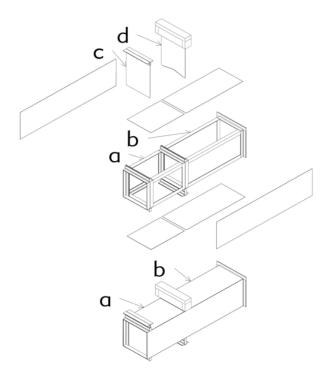


Figure 2. The drawing device and its components: a) drawing chamber b) extension outside of the wall to allow the actuation of the device d; c) outside guillotine for grains drawing; d) inside guillotine (valve) for closure during the drawing.

8. The container assembled is shown in Figure 1: 8.

The main container must be built with corrugated galvanized steel sheet, a thin sheet (0,4 mm) which is normally used by the local population as cover for houses. Also galvanized smooth sheets thicker (1-2 mm) are locally available, but because they are more expensive, they can be used only in small quantities for special applications. Due to the low thickness the sheets are not easily weldable. Therefore, the sheets should be joined each other and to the section bars by means of rivets.

For the construction of the container it is necessary to use a sturdy fabric, which can be formed either from vegetable or synthetic material. To seal the container, hot bitumen of the type used for asphalting roads is used. The bitumen is always coated on strips of waxed fabric, e.g., truck tarpaulin, which are arranged outside of the container, so that the bitumen is never in contact with the edible content.

The construction of raw earth brick walls ensures the thermal insulation of the container. In order to insulate it from the rainwater, from the moisture of the soil and from insects and other animals that burrow in the soil, a frustum of a pyramid constructed of a coarse gravel base supports the whole construction.

To avoid direct sunlight, a thick layer of leafy branches must cover the vertical walls and the roof of the construction. All these materials are available on site. The use of aluminum thin sheets to increase the thermal insulation from radiation heat transfer would be helpful. However, it is not certain that this material is available on site.

A test has been prepared in order to assess the ease of construction of the silo and to verify the functionality of the design solutions adopted. The workshop of the department was specifically arranged to create the same atmospheric conditions and the typical working situations of the Itigi village in Tanzania. In particular, the temperature and the humidity of the environment and the lack of resources and materials were taken into account. In this background, two unskilled workers built a prototype of the smaller silo using only the tools and the materials envisaged in the plan.

RESULTS AND DISCUSSION

The assembling of the silo, except the earth walls, required two days of work, during which several issues related with the construction and the use of materials were highlighted. Main considerations explaining some of the measures and decisions taken to improve the design of silo after the results stressed in the test are reported.

To ensure sealing of the container to external agents during the extraction of the grains, the silo is equipped with a slide valve. As illustrated in Figure 2, the ensiled material is present in part b, which is in direct communication with the interior of the container. Opening the guillotine d, the material enters the part a, which is closed by the guillotine c. Closing the guillotine, the gain to drawing chamber can be accessed and the contents can be drawn without external agents enter in b. The part b must be sufficiently long to allow the operation of the guillotine d, when the silo is inside the thermal insulation walls.

The above-said behavior happens until the ensiled material has reached the angle of repose, which is about 35° for maize and wheat (EN 1991-4, 2006). To allow the complete emptying of the silo, it is necessary that the grain is contained within a fabric bag which, reached the second phase (Figure 3), can be operated by means of

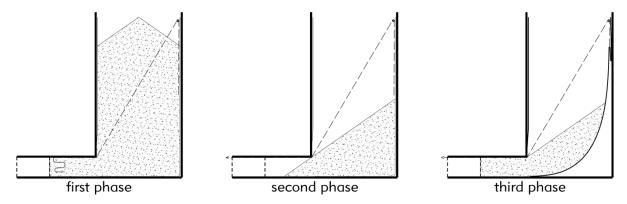


Figure 3. Phases of the container emptying.

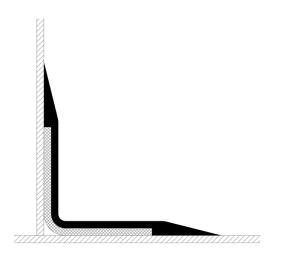


Figure 4. Joints ribbon-bitumen sealing.



Figure 5. The prototype under construction.

small ropes protruding from the outside of the mouth, so as to convey the content to the drawing device.

L-shaped elements, fastened with rivets to the two parts, made the joint between the vertical and horizontal

sheets. By cutting and bending little pieces of sheet, it is also possible to create L-shaped elements. The cylindrical sidewall sustains the pressure of the content, so it slightly stresses the joint between the bottom sheet and the sidewall.

The joint between the vertical and horizontal sheets and between the lid and the upper closure must be sealed affixing a ribbon of waxed fabric on which the bitumen is spread, according to the scheme shown in Figure 4.

In this way, it is possible to seal the spaces corresponding to the corrugations of the sheets. Furthermore, in comparison to the direct sealing of the sheets, this constitutes a safer, less sensitive to the deformations of the support sheets joint. It is also less prone to contamination of the contents.

A prototype of the inside metal container has been made in the workshops of GESAAF by two unskilled people (Figure 5).

The silo should be placed on a foundation consisting of a loose coarse gravel embankment, resting on the ground level. In this way, it is insulated from moisture and meteoric water. Furthermore, it is hindered the access to the internal cavities by soil-borne insects and mammals. On the top of the foundation, a sheet of material impervious to moisture and animals should be placed.

In order to store up the grains in the most appropriate manner, it is very important to minimize the increase in their temperature caused by external factors. For this purpose, the container must be thermally insulated from the external environment. To obtain this goal in the designed silos, it was decided to use the raw earth for the external structure. The raw material is widely available on the same building site and most people may carry out its processing, as it requires no special skill or complex equipment.

Raw earth buildings, if properly executed, have considerable strength and, if properly maintained, can have a virtually unlimited life (Barbari et al., 2014a,b). Maintenance consists essentially in repair by plastering parts that might be damaged by weathering, performed using the same clay mortar. Raw earth has a high

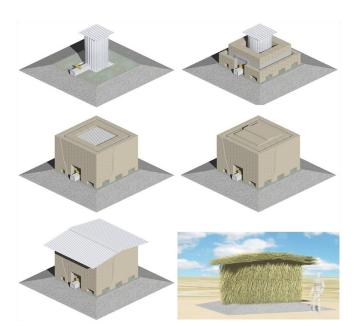


Figure 6. Phases of the silo completion (type A).

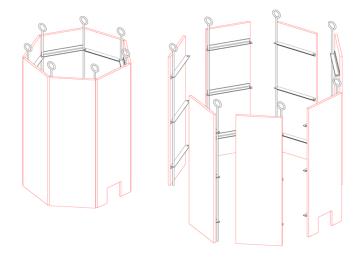


Figure 7. The dismantable internal formwork.

thermal inertia. Therefore, when a raw earth wall is heated on one face, it takes some time so that, while heating the mass of the wall, the temperature rises also on the opposite face. Since the temperature in the external environment has a sinusoidal trend in the 24 h, the temperature on the unheated face has not the time to reach the external value before night cooling, so it remains lower than outside. If the values of the thickness of the wall and of the day/night temperature swing are sufficiently high, the temperature rise of the inner face can be reduced to very small values or also to zero.

The daytime temperature rise of the outer face of the wall can be greatly limited if solar radiation does not

reach it directly. This can be achieved with an effective shading that can be realized thereby charging to the outer surfaces a sufficiently thick layer of plant material, such as grasses, straw, reeds, brushwood and similar as well as with suitably arranged trees.

The temperature of the outer face of the wall can be maintained at a value very similar to outside air temperature realizing a ventilated wall that is built outside a second wall to form a cavity in which air can circulates for the chimney effect. It is possible to place on the outer face of the internal wall a layer of reflective material. In this way, the heat radiation from the outer wall is rejected and the thermal insulation increased.

For these reasons, to achieve the thermal insulation a building made of considerable thick raw earth walls with a significant thermal inertia enclosed the container. An effective thermal lag in heat transfer is achieved (Doat et al., 1991; NZS, 1998; Morton, 2008; Minke, 2009). The roof covering must be removable to allow the annual cleaning and filling. The upper opening is closed with corrugated sheets, on which several layers of unbaked bricks are placed in order to obtain the same effect of thermal inertia characteristics of the vertical walls. On these bricks, galvanized corrugated sheets are placed in turn covered by a thick layer of plant material.

In applying the above principles to the designed silos, two construction methods are proposed, which use: a) unbaked clay bricks (adobe), b) rammed earth monolithic walls.

a) Unbaked clay bricks

Outside the first wall, a second wall is built, forming a cavity wall with natural air circulation. If available, on the outer surface of the first wall a reflective aluminum thin sheet should be applied. Furthermore, the vertical walls and the roof of the construction must be covered with a thick layer of leafy branches to avoid direct sunlight.

This solution requires, for the 1 m³ capacity silo, the construction of about 800 bricks, of various measures, which must be walled using as mortar the same mixture used for the bricks, but with a different degree of humidity. Finally, the exterior vertical surfaces of the building should be plastered (Figure 6).

b) Rammed earth monolithic walls

The rammed earth construction technique consists in laying raw earth in specially crafted formworks, and ramming it until the desired consistency is obtained. After removing the formwork, the result is a wall of considerable strength and durability.

In order to carry out the construction, two formworks are required: an internal formwork (Figure 7) and an external one (Figure 8). Both formworks must be able to be disassembled for their reuse. In order to favor carrying

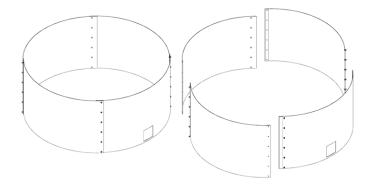


Figure 8. The dismantable external formwork.



Figure 9. First phases of the silo completion (type B).

and moving, it is necessary to contain the weights of formworks. Therefore, the external formwork has been designed to perform the casting of the wall in two phases, moving it up (Figures 9 and 10).

Conclusions

The proposed silos can be a suitable solution to store cereals and other seeds in African rural villages, thanks



Figure 10. Second phases of the silo completion (type B).

to the low cost of realization and to the simplicity of building, turning to unskilled labor of farmer's family.

The container and brick walls of adobe were made in the workshops of GESAAF using only materials and equipment actually available in the area to which the project is intended. Furthermore, the staff who implemented the construction did not have professional skills. This allowed us to verify the actual possibility of implementing the project on site.

Several improvements were introduced in the design after the test phase, related with the construction and the use of materials. In particular, a better system of unloading and a proper way to provide thermal insulation were developed.

Next step of the research will be the construction of the silo in an area of use in order to record and evaluate operating parameters.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Effect of stem density on growth, yield and quality of potato variety amethyst

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The study was carried out to determine the effect of stem density on growth, yield and quality of potato (Solanum tuberosum L.) variety amethyst in Zimbabwe. Three stem density treatments were used and these were initially derived from the number of sprouts or eyes per tuber: 2 stems/hill, 4 stems/hill and +6 stems/hill. Emergence, haulm growth, yield and quality characteristics of tubers were the main parameters measured. There was a significant difference in emergence among the 3 treatments (P<0.05). A mean of 30 plants/plot were recorded from the treatment with 6 stems/hill compared to 27.50 plants/plot from the treatment with 2 stems/hill at 12 day after planting. No statistical differences in tuber yield (P>0.05) among the three stem density levels was achieved. A high marketable yield was obtained at 2 stems/hill compared to 6 stems/hill. An average of 21.08 small tubers per hill that is, those considered unsellable were obtained from treatments derived from 6 stems per hill compared to 3.75 small tubers /plant from plants with 2 stems per hill. Seed producers whose main objective is to increase the multiplication rate, high stem densities per planting station have the potential of increasing plant populations resulting in many smaller tubers. For ware production, low stem densities promote greater proportion of medium to oversized tubers.

Key words: Potato, stem density, growth components, yield.

INTRODUCTION

The potato cultivar amethyst is arguably the most popular and widely grown potato variety in Zimbabwe (Manzira and Ackerman, 2011). A few comprehensive studies on stem population densities have been done on local potato cultivars. Regardless of the numerous studies on the potato worldwide, there is a gap in the area regarding interactions that contribute to the growth and subsequent yield of the crop (Masarirambi et al., 2012). In addition, no work has been reported on the response of amethyst to variation in stem. The growth and yield of potato is affected by such factors as stem density, seed tuber size,

planting depth, ridge volume and nitrogen supply (Barry et al., 1990; Masarirambi et al., 2012; Nielson et al., 1989; O'Beirne and Cassidy, 2006). The stem density of a potato crop is the number of stems per unit area and differs significantly from the plant population mostly given in production recommendations.

The potato plant consists of various stems and each stem forms roots, stolons and tubers and behaves like an individual plant. The density of the potato crop consists of two components. The first component is the number of plants generally referred to as plant density and the second is the number of stems per plant (Wiersema, 1987). The latter is more relevant in yield studies (Bussan et al., 2007). The number of eyes determines stem number per seed piece (Nielson et al., 1989) and similarly correlation with stem density in cultivars such as Nooksack was found in Europe. The number of eyes and stems produced per seed piece increased as cut seed piece increased (Nielson et al., 1989). Each stem from a single eye can be regarded as an independent production unit. Thus a sufficient number of strong stems should develop per seed tuber.

However, as much as higher stem density results in greater yield, it also affects the size of tubers. Growth is limited when competition among stems is high. At higher stem density, the tubers produced remained smaller than at lower stem density, while the percentage of large tubers decreased (Güllüoglu and Arıoglu, 2009). conducted Controlled environment studies characterize differences in canopy growth and dry matter production among single and multiple stemmed potatoes showed that vegetative and dry matter yield production were unaffected by stem density (Fleisher et al., 2011). As stem density increased, fewer tubers were produced and this was due to a reduced multiplication rate which was defined as number of tubers produced per seed tuber (Fleisher et al., 2011). The primary purpose of the current study was to study the influence of stem density on the growth, yield and quality of the potato variety amethyst.

MATERIALS AND METHODS

Description of the study site

The study was done at Africa University farm, Mutare, Zimbabwe. The area is under the Agro-ecological region 2 (18°53'70, 3"S: 32°36'27.9"E) at 1131 m above sea level. Average annual rainfall ranges from 750 to1200 mm.The average maximum temperature ranges from 18°C in July to 32°C in October. Very hot weather conditions prevail between September and December. High mean maximum temperatures in excess of 30°C are recorded in October, which is the hottest month of the year.

Planting material

Amethyst is presently the most widely grown potato variety in Zimbabwe. It is a late maturing potato variety (17 to 19 weeks) and AA Class seed was procured from the Potato Seed Association of Zimbabwe. The seed was induced to sprout by dipping tubers in Gibberellic acid (32 ml of GA₃/100 L of water) for 1 min as per chemical label. The seed tubers were allowed to dry before being put in a room with subdued light until sprouting. Seed tubers were planted in furrows and ridged twice as is standard practice. The first ridging was when plants were about 20 cm in height and the second was at 35 cm.

Design of the experiment

The potato cultivar amethyst was evaluated for its response to

various stem density levels in an experiment laid out in a randomized complete block design (RCBD). Treatments were as follows: 2 sprout or stems/hill, 4 sprouts or stems/hill and ≥6sprouts or stems/hill. Stem density was maintained by pruning excess growth beyond that which was stipulated by the desired number of stems per treatment. Plots were represented by 4 × 4.0 m long rows, spaced at 0.90 m between rows and 0.30 m in the row. The harvested plot was 2 × 2 m.

General management and fertiliser applications

The experimental site was deep ploughed, disked and harrowed to ensure a fine tilth. Planting stations were made as per treatment. The planting depth was 10 cm. A soil analysis of the trial site gave the following information: pH (CaCl₂) 5.01, and a total content of the following elements: N 45.37 (ppm), P 0.797 (ppm), Mg 4.40 (meq%), Cl 7.1 (meq%), Zn 3.53 (ppm) and Cu 0.65 (ppm). Basal Compound C (6%N, $17\%P_2O_5$, 14% K_2O) was applied at the commercial rate of 1500 kg/ha prior to planting. Top dressing of ammonium nitrate (AN) was applied at 300 kg/ha three weeks after emergence. This was followed by split application sulfate of potassium (K₂SO₄) at 400 kg/ha at flowering and at two weeks after flowering. Basal fertilizer was applied in each planting station while top dressing was banded. The first ridging commenced after the top dressing with AN, when all plants had an average height of 20 cm. An allowable soil moisture depletion level of 40% was maintained throughout the growing period and soil moisture monitoring was done through the use of field tensiometers. Sprinkler irrigation was used for application of water as a supplement to the summer rain.

Data collection

The number of emerged stems was recorded from 8 to 12 days after planting. Leaf counts per plot were done from 30 to 60 days after emergence (DAE). A minimum leaf length of 0.5 cm was used to indicate leaf appearance. Final leaf counts included senesced and green leaf numbers (Fleisher et al., 2011). The length of the main stem per planting station of three randomly selected plants for each plot was recorded once every 10 days from 30 to 70 days after emergence. Relative growth rate in leaf number (RGRIn) was calculated from the formula (Balderrama and Chazdon, 2005):

$$RGRln = \frac{loge(ln60) - loge(ln30)}{60 - 30}$$

Where In60 is leaf number at 60 and In30 is leaf number at 30 DAE. Thus the parameter was calculated using data from 30 to 60 DAE. Tuber density was recorded at the time of harvesting. Four plants were selected per plot to measure the average number of tubers per plant. Tuber size distribution was also determined; samples were graded into small (25 to 37.5 mm in diameter) which made up non-marketable tubers (ware), medium (37.5 to 50 mm), large (50.00 to 56.25 mm) and oversized tubers (56.25 to 62.25 mm) which composed the marketable grades as by Masarirambi et al. (2012). Quality determinants such as number of green tubers, brown heart, tuber specific gravity and dry matter were obtained by randomly selecting 20 tubers from each treatment. In determining specific gravity, selected tubers from each treatment were first weighed in air and then re-weighed suspended in water. Specific gravity was then calculated using the following formula (Burton, 1989):

Specific gravity = Weight in air/[Weight in air - Weight in water]

Dry matter content was obtained using the formula which correlates specific gravity to dry matter by a correlation factor of 0.937% dry

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Table 1. Plant emergence counts at 7, 8, 9, 10, 11 and 12 Days after planting (DAP).

| Treatment | E at 7 DAP | E at 8 DAP | E at 9 DAP | E at 10 DAP | E at 11 DAP | E at 12 DAP |
|------------|--------------------|--------------------|---------------------|-------------|-------------|--------------------|
| 2 Sprouts | 7.25 ^a | 13.00 ^a | 16.50 ^a | 22.50 | 26.00 | 27.50 ^a |
| 4 Sprouts | 13.00 ^b | 14.50 ^a | 19.25 ^{ab} | 24.75 | 27.00 | 29.50 ^a |
| +6 Sprouts | 14.75 ^b | 19.75 ^b | 22.50 ^b | 26.50 | 28.50 | 30.00 ^b |
| LSD | 2.840 | 3.011 | 4.345 | 4.421 | 3.106 | 2.079 |
| P value | * | * | * | NS | NS | NS |
| CV | 14.1 | 11.0 | 12.9 | 10.4 | 6.6 | 4.1 |

^{*}Denotes significance at P<0.05; NS denote non-significance at P>0.05. E = emergence. The means not sharing a common letter in a column differ significantly at P<0.05.

Table 2. Leaf number counts (LN) recorded at 30, 40, 50 and 60 days after emergence (DAE).

| Treatment | LN at 30 DAE | LN at 40 DAE | LN at 50 DAE | LN at 60 DAE |
|--------------|--------------------|--------------|---------------------|---------------------|
| 2 Stems/hill | 22.37 ^a | 74.80 | 116.20 ^a | 126.89 ^b |
| 4 Stems/hill | 40.19 ^b | 71.40 | 112.70 ^a | 122.67 ^a |
| +6Stems/hill | 62.25 ^c | 73.70 | 100.90 ^b | 121.14 ^a |
| LSD | 3.730 | 14.48 | 10.08 | 2.924 |
| P value | * | NS | * | * |
| CV(%) | 12.6 | 10.8 | 8.3 | 3.2 |

^{*}Denotes significance at P<0.05; NS denotes non-significance at P>0.05. The means not sharing a common letter in a column differ significantly at 0.05 probability.

matter =(24.182±0.035)+(211.04±3.33) specific gravity-1.09)(Burton, 1989).

Data analysis

The data collected were analyzed using Genstat Discovery 3rd edition. The data were subjected to analysis of variance and mean were separated using Fisher's least significant difference (LSD) test at P=0.05.

RESULTS

Emergence of stems

Stem density significantly (P< 0.05) influenced the number of days taken for the crop to emerge at 7, 8 and 9 Days after planting (DAP) (Table 1). Plants from seed tubers with more than four sprouts (p <0.05) emerged earlier than plants from seed tubers with 2 sprouts. There was an increase in the days to emergence as the number of sprouts per planted tuber increased. However, this was not sustained at 10, 11 and 12 DAP.

Leaf counts

Table 2 shows significant variation in leaf numbers at 30, 50 and 60 Days after emergence (DAE). At 30 DAE the

leaf counts increased with increasing number of stems per hill. However at 50 and 60 DAE the reverse was observed. No differences were observed at 40 DAE. During the initial growth, leaf number appeared to be influenced by the number of stems that emerged but later it was greatest where the stems were fewer, perhaps showing the effects of reduced competition.

Relative growth rate in leaf emergence

Table 3 shows the relative growth rate (RGR) in leaf numbers (LN) from 30 to 60 DAE. There was a significant difference in the rate of leaf production at 30 to 40 DAE (P<0.05); 4 stemmed plants had relatively higher leaf production rate than 2 stemmed plants. There were no significant differences in RGR from 40 to 60 DAE among the 3 stem density treatments.

Main stem lengths

Table 4 shows the changes in stem length as affected by number of stems per station. The effect of stem density on the length of main stems showed some variations across the 3 stem density treatments. At 30 DAE the stem length increased with decreasing stem density. This trend was maintained at 40 and 60 DAE.

Table 3. Relative growth rate (Rgr) in leaf numbers (LN) from 30, 40, 50 and 60 days after emergence (DAE).

| Treatment | RgrLn 30-40DAE | RgrLn 40-50 DAE | RgrLn 50- 60 DAE |
|--------------|----------------------|-----------------|------------------|
| 2 Stems/hill | 0.0335 ^a | 0.1239 | 0.01239 |
| 4 Stems/hill | 0.0498 ^b | 0.1190 | 0.01190 |
| +6Stems/hill | 0.0440 ^{ab} | 0.1126 | 0.01126 |
| LSD | 0.01290 | 0.03616 | 0.003616 |
| P value | * | NS | NS |
| CV (%) | 42.6 | 40.8 | 42.6 |

^{*}Denote significance at P<0.05; NS denote non-significance at P>0.05. The means not sharing a common letter in a column differ significantly at 0.05 probability.

Table 4. Stem length (cm) measured at 30, 40, 50, 60 and 70 days after emergence (DAE).

| Treatment | STL at 30DAE | STL at 40DAE | STL at 50DAE | STL at 60DAE | STL at 70DAE |
|--------------|---------------------|----------------------|--------------|--------------------|--------------------|
| 2 stems/hill | 30.88 ^b | 34.93 ^b | 41.37 | 48.96 ^b | 63.28 ^a |
| 4 stems/hill | 28.25 ^{ab} | 34.00b ^{ab} | 40.05 | 47.02 ^a | 64.51 ^a |
| +6stems/hill | 26.94 ^a | 33.04 ^a | 40.94 | 46.49 ^a | 60.80 ^a |
| LSD | 2.746 | 1.340 | 1.766 | 1.425 | 4.20 |
| P value | * | * | NS | * | NS |
| CV% | 13.4 | 1.34 | 1.768 | 1.425 | 4.2 |

STL = stem length.*Denote significance at P<0.05; NS denote non-significance at P>0.05. The means not sharing a common letter in a column differ significantly at 0.05 probability, STL= stem length.

Table 5. Tuber yield in kg/ha, tuber density /plant and marketable yield at harvest in t/ha.

| Treatment | Mean tuber yield (Kg/ha) | Tuber density (counts/plant) | S1 (non-Marketable) | S2-3-4 (Marketable) |
|--------------|--------------------------|------------------------------|---------------------|---------------------|
| 2 stems/hill | 18972 | 7.58 ^a | 3.75 ^a | 11.83 ^b |
| 4 stem/hill | 23327 | 17.50 ^b | 16.17 ^b | 9.33 ^a |
| +6 stem/hill | 28885 | 24.75 ^c | 21.08 ^c | 11.67 ^b |
| LSD | 10863.7 | 4.697 | 2.332 | 2.009 |
| P value | 0.162 | 0.001 | < 0.001 | 0.028 |

^{*}Denote significance at P<0.05; NS denote non-significance at P>0.05. The means not sharing a common letter in a column differ significantly at 0.05 probability; SI = tuber diameter of 25 to 37.5 mm; S2= tuber diameter of 37.5 to 50 mm; S3= tuber diameter of 50.00 mm to 56.25 and S4= tuber diameter of 56.25 to 62.25 mm).

Yield and tuber density counts per plant

Table 5 shows mean yield, number of tubers per plant, non-marketable and marketable yield. Although the lower stem density tended to yield less, this was not significant. Tuber density increased with increase in number of stems/hill (Table 5).

Marketable yield and non-marketable yield

Tuber size distribution which affects the volume of economic yield suitable for ware consumption was classified into marketable and non-marketable yield (Table 5) all measured in number of tubers per plant. The S1 size (tuber diameter of 25 to 37.5 mm) represented

non marketable output which were tubers regarded too small for ware consumption or commercial processing while S2 (tuber diameter of 37.5 to 50 mm), S3 (tuber diameter of 50.00 to 56.25 mm) and S4 (S4= tuber diameter of 56.25 to 62.25 mm) represented the marketable yield suitable for both ware and commercial processing. Non-marketable yield increased with increase in stems/hill while marketable yield was least at 4 stems/hill and larger at 2 and >6 stems per hill.

Tuber greening, specific gravity and dry matter content

Table 6 shows the response of tuber specific gravity and the subsequent dry matter content to stem density. The

Table 6. Number of green tubers (GT/plant), Specific Gravity (Sp gravity) and per cent dry matter content of tubers (%DM content) at harvest.

| Treatment | G T/plant | Sp Gravity | %DM Content |
|--------------|-------------------|----------------------|--------------------|
| 2 Stems/hill | 0.25 ^a | 1.06930 ^a | 18.02 ^a |
| 4 stem/hill | 1.75 ^b | 1.08368 ^b | 21.01 ^b |
| +6 stem/hill | 4.25 ^c | 1.07257 ^a | 18.70 ^a |
| LSD | 0.999 | 0.005195 | 1.079 |
| P value | * | * | * |
| CV% | 27.7 | 1.606 | 0.350 |

^{*}Denote significance at P<0.05; NS denote non-significance at P>0.05. The means not sharing a common letter in a column differ significantly at 0.05 probability.

number of green tubers increased with increasing stem density. Specific gravity was highest at 4 stems and lower at 2 and >6 stems and this was mirrored by percentage dry content (Table 6).

DISCUSSION

Emergence counts

There are a number of factors that could explain the differences in crop emergence rate shown in Table 1. The size of seed tuber at planting, planting depth, method of planting, physiological condition of seed tuber material, edaphic and environmental factor are some of the causes of uneven crop emergence. Seed tubers with multiple sprouts can have a significant competitive edge over tubers with a fewer eyes or less sprouts as they can produce emergence of several strong seedlings per square inch (Cortbaoui, 1988). Nielson et al. (1989) established that some cultivars such as Nooksack with few spaced eyes or high percentage of blind tubers had a negative correlation of stem number to yield suggesting that uneven emergence and or missing plants help to explain the performance of stem density with respect to rate of emergence.

Leaf counts

At low stem densities production of basal lateral branches was more vigorous as the crop intended to increase its ability to intercept light and photosynthetic active radiation (PAR). Our findings agree with the findings of Fleisher et al. (2011) and Engels et al. (1993) who observed a similar trend in leaf production amongst various stem densities. As a result of increased interstem competition amongst high stem densities the allocation of accumulated assimilates to production of photosynthetic apparatus was reduced (Moorby, 1967).

Relative growth rate in leaves

The initial differences in RGR in LN shown in Table 3 can

be attributed to the difference brought about by stem numbers soon after crop establishment. Plants derived from high stem densities had a relatively high RGR as the effects of leaf shading and competition for light interception might have been minimal during the early growth phase. Masarirambi et al. (2012) suggested that differences caused by uneven sprout numbers at emergence had an effect on the haulm growth as those plants emanating from high sprout densities had a "rich" background of nutrients stored in the parent tuber. The lack of significant difference in RGR in LN from 40 to 60 DAE may be attributed to the increase in the rate of lateral brunching among low density plants which nullified the initial differences in leaf production among the 3 densities. This phenomenon was also reported by Fleisher et al. (2011) and Masarirambi et al. (2012), in which they showed that secondary stems and lateral branching increased with a decline in stem population.

Main stem lengths

The increase in stem length with decreasing stem density as shown in Table 4 may be attributed to less inter stem competition for nutrients. Our findings confirm work by Moorby (1967) and Fleisher et al. (2011) who observed an increase in stem length at low stem density which they attributed to low inter-plant completion for both nutrients and photosynthetic active radiation.

Yield

The observations validated the findings by Hammes (1985) in which stem populations had no significant effect on tuber yield despite that more tubers were produced at high stem densities. Bussan et al. (2007) found that yield increased with an increase in stem density per hill. Our findings show that the number of tubers/plant, increased with increasing stem density as shown in Table 5. This was largely due to the fact that each stem is an independent production unit. Hence an increase in their number will consequently translate to more tubers per station. These finding are in agreement with observation

by Wiersema (1987) who showed an increase in the number of tubers as stem density increased.

The non-marketable yield increased with increasing stem density. This was attributed to an increase in competition for water, nutrients and sunlight during tuber bulking and confirms work by Rex (1990) and Wiersema (1987) who reported a reduction in total marketability of tubers as stem populations increased. Marketable yield was best at the lowest and highest stem densities. We, however, expected that the proportion of marketable tubers would fall as stem density increased. Our findings which were contrary perhaps indicate that our upper cut off point for stem density was still in the optimal range for the variety we used.

Tuber greening, specific gravity and dry matter content

While other studies illustrated the significance of manipulating cultural practices such as planting depth, hilling time and interval, ridge geometry, planting method as the major factors affecting tuber greening (Bohl and Love, 2005; Mburu, 1984; Ravishankar et al., 2007; Svensson, 1962), our work showed that stem density may also be added to this myriad of factors. That the number of green tubers increased with increasing stem density was probably because the chances of tubers being exposed to the sun were greater as stems competed for space (Kouwenhoven et al., 2003; Pavek and Thornton, 2009). The specific gravity of tubers was highest at the lowest and highest stem density and highest at the intermediate stem density. Perhaps, this shows that our stem densities were within a range where trends were not firm. The same trend as with specific gravity was evident with dry matter since the dry matter was estimated from specific gravity.

Conclusions

The study showed that differences in sprout numbers per tuber had an effect on the rate of emergence. Plants established from 6 sprouts/tuber emerged earlier followed by 4 sprouts/tuber and lastly 2 sprouts/tuber. The effect of leaf number (LN) among the three subsequent densities showed a significant difference at 30 days after emergence (DAE). Plants derived from 2 stem/hill had longer main stems followed by 4 and 6 stem treatments respectively. Stem density had no effect on the total tuber yield. Tuber density increased with an increase in stem density. Marketable yield declined with an increase in stem density.

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Conflict of Interests

The author(s) have not declared any conflict of interests.

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