7-2016

FROM *MILPAS* TO THE MARKET: A STUDY ON THE USE OF METAL SILOS FOR SAFER AND BETTER STORAGE OF GUATEMALAN MAIZE

José Rodrigo Mendoza
University of Nebraska-Lincoln, jrodrigomendoza@gmail.com

FROM *MILPAS* TO THE MARKET: A STUDY ON THE USE OF METAL SILOS FOR SAFER AND BETTER STORAGE OF GUATEMALAN MAIZE

by

José Rodrigo Mendoza

A THESIS

Presented to the Faculty of
The Graduate College at the University of Nebraska
In Partial Fulfillment of Requirements
For the Degree of Master of Science

Major: Food Science & Technology
Under the supervision of Professor Jayne Stratton

Lincoln, Nebraska
July, 2016

FROM *MILPAS* TO THE MARKET: A STUDY ON THE USE OF METAL SILOS FOR SAFER AND BETTER STORAGE OF GUATEMALAN MAIZE

José Rodrigo Mendoza, M.S.
University of Nebraska, 2016

Advisor: Jayne Stratton

This project aimed to implement the use of metal silos to improve quality and safety of maize consumed by inhabitants of the highlands of Guatemala. This manuscript includes a literature review of the maize production chain in Guatemala, a survey about agricultural practices used in the region of study, as well as a characterization of the analyzed maize regarding its mycoflora, nutritional composition, and insect infestation. To better understand the current situation regarding agricultural practices and maize consumption, a survey was carried out. Sample consisted of 280 families representing 14 rural communities distributed in the townships of Todos Santos and Chiantla, Huehuetenango, Guatemala. In addition, 25 farms from the same region were sampled for maize, which was evaluated for fungal count, fumonisin and aflatoxin, and insect analysis. Among surveyed farmers, 13 grew and harvested maize (denominated Chain 1, C1) while 12 had no land available to plant and consequently acquired maize by other means (denominated Chain 2, C2) such as local markets. Due to a clear diversity in the phenotype of the corn samples, proximate analyses of the various cultivars was conducted. Most Guatemalan farmers from the rural area have low income and large families, thus the economic aspect is a key factor for farmers desiring to improve in their lives. By the implementation of a metal silo, farmers who have the means to acquire this technology would improve their grain quality and safety and, with that, their livelihoods. A financial analysis was conducted to evaluate the economic feasibility of the farmers on purchasing the storage technology and either obtaining revenue (C1) or saving money (C2) in the process would be able to afford such a vessel.

Acknowledgements

Drs. Andréia Bianchini and Jayne Stratton. Thank you for letting me be part of your work team. Working for the Food Processing Center and doing applied research shaped me into a better professional. Thank you for your supervision, advice and friendship throughout my graduate studies.

Dr. Heather Hallen-Adams, thank you for sharing a portion of your vast knowledge, along with some laughs. I will always be thankful for your guidance through this project.

Dr. Fabio Mattos, for teaching me all the necessary tools for this important component of the project. Thank you.

Dr. Carlos Campabadal, thank you for all of your assistance and advice in these two years. I hope we can work together in the future.

My beloved friends. Your friendship was crucial for the conclusion of this project. Thank you for always believing in me Laura, Herbo, Lau, Martin, Shara, Carmen, Tico, Gabriela, Tiny, and many more.

My family. Adam, Anny, Enio, AnaGabbs, Karli, aunt Liz, Linda, and many others. Thank you for rooting for me.

There are no words to describe how thankful I am to you, mom and dad. Without your love and wisdom I would not be the person I am today. Thank you for teaching me about life. I love you.

TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

Scientific Manuscript One: Maize in Guatemala: seed development, production, limitations, and opportunities

Scientific Manuscript Two: Traditional maize post-harvest management practices amongst smallholder farmers in the western highlands of Guatemala

Scientific Manuscript Three: Assessment of smallholder farmer's maize in the western highlands of Guatemala: insect and mold infestation, mycotoxin contamination, and proximate analysis

Scientific Manuscript Four: Understanding the mycobiota of maize from the highlands of Guatemala, and implications to its quality and safety

Scientific Manuscript Five: Financial feasibility study of metal silo implementation for proper maize storage in the western highlands of Guatemala

MAIZE IN GUATEMALA: SEED DEVELOPMENT, PRODUCTION, LIMITATIONS, AND OPORTUNITIES *

R. Mendoza, J. Stratton, and A. Bianchini

Department of Food Science & Technology
University of Nebraska. Lincoln, NE, USA

Corresponding author:

Andréia Bianchini, Department of Food Science & Technology
The Food Processing Center, University of Nebraska-Lincoln
Phone: (402)472-3144, Fax: (402)472-1693
abianchini2@unl.edu

*Scientific manuscript formatted to meet guidelines for the Field Crops Research journal

ABSTRACT

Maize is a central component in Guatemala's culture. This is mainly attributable to the diet and traditions of the country's rural population. Throughout this tropical country there are several maize types, varying in shape, period of growth, resistance to adverse conditions, color, etc. and involved in different social roles. In order to reduce the food insecurity menacing the country, government development programs, including plant breeding institutions were formed. In this review, an overview of the maize seed breeding work of public and private institutions was included, with emphasis given to the former. Additionally, maize production zones in the country, as well as annual production, import and export patterns are discussed. Over the years, several government programs have been dismantled, but the Institute of Agricultural Sciences and Technology (ICTA), supported mainly by the International Maize and Wheat Improvement Center (CIMMYT), remains and has played a key role in attempting to improve the maize production systems in Guatemala. Furthermore, poor grain handling practices in the country are a setback resulting in mycotoxin contamination of staples including maize, affecting the overall health and life-span of the population. Improved maize varieties developed by ICTA may partially help to address this problem. In addition to governmental regulations, education for farmers is essential so they can be aware of the implications of possible improper practices performed and the health risk they present. Guatemalan government must encourage the economic growth of farmers located in the rural land of the country, specifically in the Highlands where most subsistence farming occurs. Such work would decrease food insecurity in the region, allowing farmers to enter the formal market with the ultimate goal of having safe, nutritious maize of high quality.

Keywords: Guatemala, maize, corn, maize production

INTRODUCTION

Guatemala is a country politically divided into 22 departments. In their World Economic Situation and Prospects (WESP) country classification report, the United Nations categorized Guatemala as a developing country, with a population of close to 15 million people and growing at a 1.86% rate per year (Index Mundi, 2015; United Nations, 2014). The majority of the population is distributed in the highland areas, many characterized by having an agrarian economy based on smallholder maize cropping systems (Valladares, 1989).

Due to historic, biological, economic, sociologic and strictly ideological factors, for Guatemalans maize is very important to their culture, so that the Mayan poem Popol Vuh depicts maize as the progenitor of humanity; the most perfect product of creation (Dowswell et al., 1996; Lima, 1988). Maize (*Zea mays*) kernels or caryopsis are formed on the female inflorescence fraction of the plant called the ear. Approximately 800 kernels are produced and removed from the cylinder of the ear, known as cob, by the process of shelling (White and Johnson, 2003). On average, each Guatemalan consumes 350 grams of maize on a daily basis (CIMMYT, 1981), registering one of the highest per capita consumption levels in the world (Sain and López-Pereira, 1999). Moreover, the United States Department of Agriculture has reported a consumption of 25,000 metric tons of maize in the year 2014 for Guatemala, and the amount has mostly showed an increase every year since 1960 (USDA, 2014). Noticeably, many Guatemalan dishes are based largely on this crop, which is a staple commodity for Guatemala's indigenous population (Argueta, 2013).

Maize is considered the vital input for crop production, being the highest source of employment in 2011 with over 50 million daily jobs (*jornales*), followed by beans with 15 million and sorghum with 1 million daily jobs (MAGA, 2013). Thus, proper handling, especially during storage, is fundamental. Fungal microflora is considered the major seed deterioration factor in storage (Saleem et al., 2012). It affects not only the seed viability, but also can lead to mycotoxins, which are secondary metabolites of filamentous fungi, hazardous to humans and animals (Wild and Gong, 2009). Fumonisin and aflatoxin are the

most prevalent fungal toxins found in Guatemalan maize destined for consumption (Torres et al., 2015; Torres, 2000). Even though contaminated lots of maize cannot be used for human consumption, due to food insecurity it is still consumed by some (Leslie and Logrieco, 2014), resulting in chronic and acute health implications. This is not only limited to maize. Several other commodities, including the two next most important staple foods for Guatemalans, beans and rice, have also been reported to be heavily contaminated by mycotoxins (de Campos and Olszyna-Marzys, 1979; Wiedenbörner, 2014)

THE NATIONAL MAIZE ECONOMY

Maize is still considered the staple food for most Guatemalans (Torres et al., 2015). According to the Ministry of Agriculture, Livestock and Food, for the 2015-2016 maize season, it was estimated that 876,000 hectares were planted to make approximately 40.9 million quintals (1 quintal = 100 lb) out of which 36.8 are white maize (MAGA, 2015a). For the country as a whole, Figure 1 shows fifty six years of maize production ranging from 1960 to approximately May of 2016. The national production for the last ten years averaged 1.91 million tons with a noticeable increase beginning 2006 (growth rate of 68%), and being rather stable until present day. Even with the larger presence of small-scale, subsistence-oriented maize production farms, approximately 45 % of the national maize area is due to farms larger than 7 hectares. In the commercial region of the Pacific Coast farms range between 7 and 45 hectares in extent (Dowswell et al., 1996).

Guatemala was considered self-sufficient in maize production in most years (Dowswell et al., 1996) up to late 1980s. Figure 2 includes both exports and imports of maize over a 56-year period from 1960 to 2016. Because of fluctuations in yields, mostly due to erratic climate, and pest and microbial occurrence, there is a clear continuous increase of imports starting approximately in the early 1990s. Such imports originated mostly (>90%) from the United States of America, followed by Argentina and Brazil (MAGA, 2015b) supplying close to a third of the national consumption in recent years (Van Etten and Fuentes, 2004). In contrast, exports have not changed positively over the past 20 years, except for an increase in 1996 (growth rate of 530%). Before that, exports

were small, and in some years non-existent. The countries importing maize from Guatemala include El Salvador (~92%) and Nicaragua (~7%) (MAGA, 2015a, 2015b). It can be seen that maize imports after 1990 had a marked rise, having severe repercussions in the agronomical sector in Guatemala, leading to a high unemployment rate in the rural area (Van Etten and Fuentes, 2004).

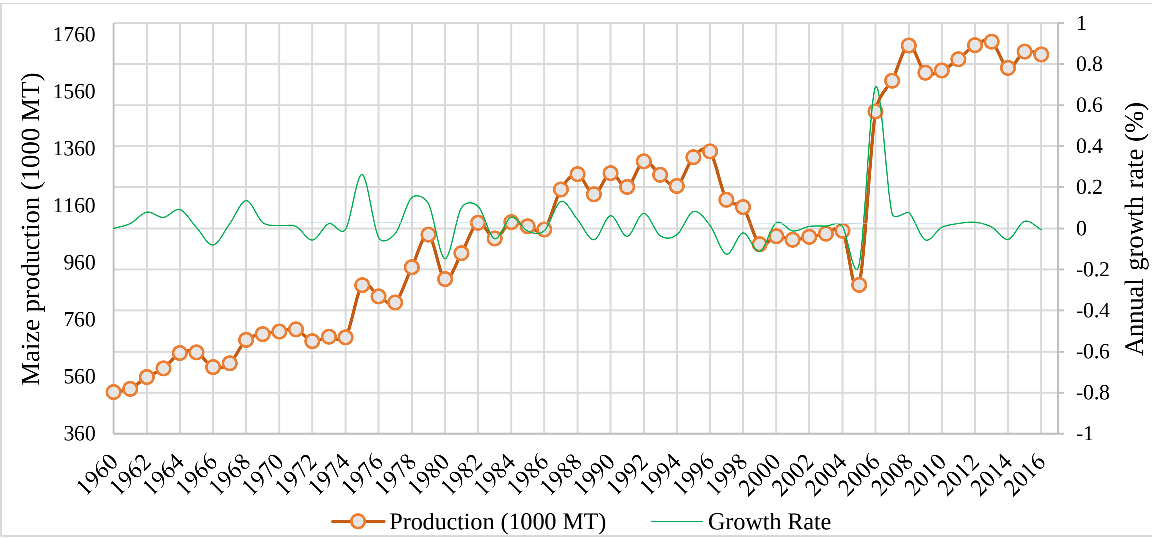


FIGURE 1. Guatemala maize production and annual growth rate. Years 1960 to 2016 (Index Mundi, 2016a). MT: metric tons

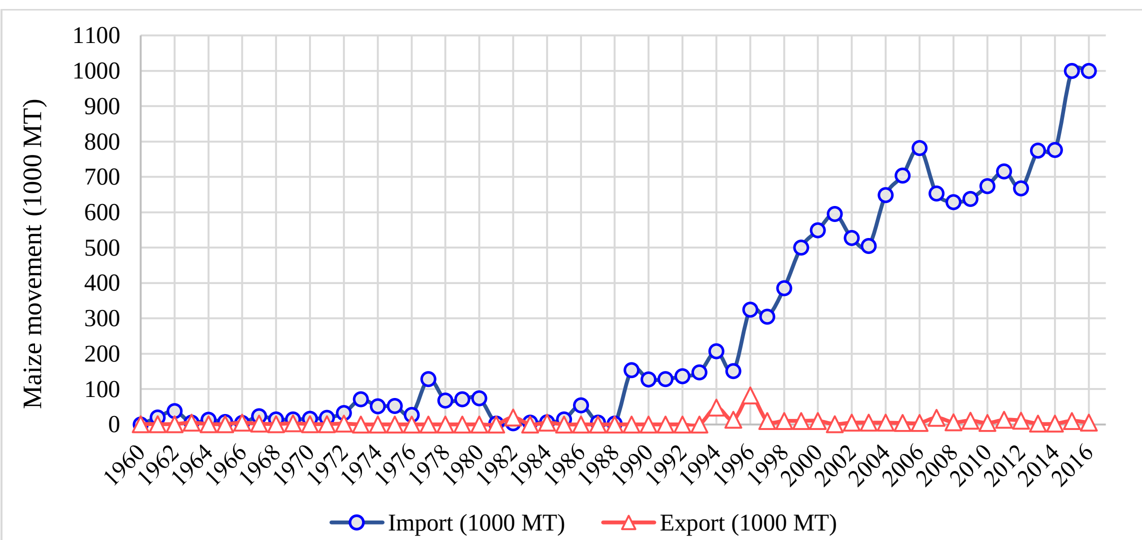


FIGURE 2. Guatemala maize import and export movements. Years 1960 to 2016 (Index Mundi, 2016b) MT: metric tons

After a sharp decline (Figure 1), maize production reached a new level in early 2000s to remain stable and augment in current times. The fact that the industry does not accept the national grain is a revealing symptom of the current problematic of the national production (Van Etten and Fuentes, 2004), especially due to high mycotoxin contamination, explained in further sections.

It is worth mentioning the National Institute of Agricultural Marketing (INDECA) as it had a direct impact on the maize's market price. INDECA was the state institution responsible for promoting and marketing services functions of agricultural production in the country, implementing marketing policies, price stabilization and supply of agricultural products to be determined by the Government through the Ministry of Agriculture, as it was dictated in the Legislative Decree 101-7: Law of the National Institute of Agricultural Marketing. Because of disruptions within the government, INDECA collapsed. An example of this occurred in 1976, when an earthquake took place in the country. Because the institute had grain reserves to maintain price stability, and the earthquake left many communities across the country in precarious conditions of food insecurity, the government decided to take such reserves, but failed to pay it back. Many stations from INDECA were left unused since 1983, year in which the government ordered the institute to forfeit its maize price regulator function (Sigüenza Ramírez, 2010).

It is important to include that maize consumption is affected by its accessibility, referring mainly to family economic limitations. The lack of economic means is reflected in the high rates of chronic malnutrition, because although sufficient production nationwide is obtained, consumers are not always able to purchase (MAGA, 2015a).

GUATEMALA'S AGRICULTURE AND MAIN MAIZE PRODUCTION AREAS

Agriculture is the pillar of the Guatemalan economy, where nearly half of the labor is engaged in agricultural activities for both domestic consumption and exports. This sector represents around 25 % of the GDP (Osorio, 2009). The Guatemalan

agriculture sector exports banana, cacao, coffee, sugar (crude and refined), and cardamom, among others (MAGA, 2015b). The major commercial agricultural area is found along the Pacific coast. This 21 to 37-mile wide strip is where most of the cattle, cotton, and sugarcane production occurs, along with basic food crops production (Dowswell et al., 1996).

Guatemala is located within tropical latitudes. Nonetheless, it does not have a solidly tropical climate (Osorio, 2009). Its elevation ranges from sea level to approximately 4000 masl (meters above sea level) resulting in a diversity of microclimates. Rainfall in the country ranges from 2000 millimeters per year in certain lowland areas to less than 500 millimeters per year in some semi-arid valleys. Marked rainy and dry seasons occur. The major crop production areas are shown in Figure 3, and in these areas maize is primarily produced in the West Central, East and the Southern Coastal regions of the country. In these areas, maize is grown under three major environmental classifications: Lowland tropical: Less than 1300 masl, where hybrids and varieties are planted in a 120-day growing season; Midaltitude areas: 1300-2000 masl; and Highland: above 2000 masl where the growing season is longer than 180 days. Of the aforementioned, the lowland setting accounts for more than 80 % of the total maize area in the country (Dowswell et al., 1996; Echeverría, 1990; INGUAT, 2016). López 2002 shows another way of fragmenting the maize production regions based on rainfall, shown in Table 1.

TABLE 1. Agroecological maize producing regions in Guatemala based on rainfall (López, 2002)

Region	**Area (ha)**	**%age (%)**	**Altitude (masl)**	**Pluviosity**
Tropic with favorable moisture	301000	43.0	0 – 1400	Relatively uniform rainfall
Tropic with limited moisture	175000	25.0	0 – 1400	Poor and erratic rainfall
Highlands (central and western)	224000	32.0	1400 – 3000	Relatively uniform rainfall

Based on this system, the largest production area occurs in altitudes ranging from 0 to 1400 masl, which comprises 68% of the production area of the country (approximately 476,000 ha). Within this area are the departments of San Marcos, Quetzaltenango, Retalhuleu, Escuintla, Santa Rosa, parts of Jutiapa, Alta Verapaz, Izabal, and the lower areas of Huehuetenango, Quiché and Petén. The area with a lower abundance of rainfall corresponds to the departments of Jutiapa, Chiquimula, Jalapa, Zacapa, El Progreso, Baja Verapaz, and some areas of drought in Quiché, Huehuetenango and Petén (López, 2002)

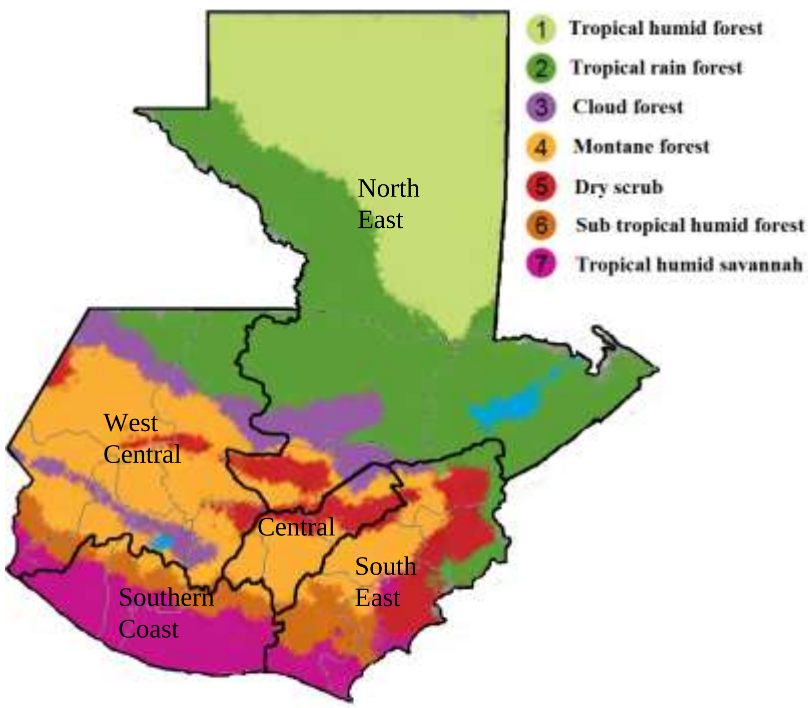


FIGURE 3. Maize production zones of Guatemala (Echeverría, 1990; INGUAT, 2016)

Food production in the central and western highlands is considered subsistence, where the majority of farmers produce food crops mainly for home consumption and are usually dependent upon off-farm sources of income (Isakson, 2009; Mendez et al., 2015). Farmers' landholdings are usually small (< 2 ha) and fragmented (CIMMYT, 1981; Dowswell et al., 1996). In lower elevations, where temperature and relative humidity can get very high (de Campos et al., 1980), agriculture is more commercially oriented, conformed mostly by smallholder farmers (Dowswell et al., 1996).

MAIZE SEED DEVELOPMENT AND PRODUCTION IN GUATEMALA

Maize is not only a major component in Guatemalan diets, but also a main constituent for livestock feeds, food production, and others. Therefore, its level of production has to be substantially raised to meet the demand for food, feed and industrial use. Such needs birthed a new priority in the country, the development of new varieties. For research purposes, different types of maize are referred to as “materials”. Developed materials can be "open pollinated" maize (OP), which allows the horticulturist to produce and save seed. This technique provides future crops with fresh and healthy seeds adapted to the local climate. The other type of maize material is denominated "hybrid", which is the result of crossed-pollination and consequently does not breed true (Ecology Action, 2010).

According to the number of parents that make the hybrids, these can be classified as single, double or triple, as shown in Figure 4 (López, 2002). Additionally, maize can also be classified based on its type. According to this classification, there are six general classes of maize: dent, flint, flour, sweet, pop and pod maize; all based on the kernel characteristics (Brown et al., 1985; White and Johnson, 2003). Unlike in developed countries where dent maize is preferred due to a higher yield, in Guatemala native flint varieties of maize are also grown, and in some instances favored. Dent maize has a vitreous endosperm on the side while the central core extending to the crown is soft and floury. Upon drying, the center part collapses to give a distinctive indentation. In contrast, flint maize has a thick vitreous endosperm surrounding a small granular center and is smooth and rounded with no denting (White and Johnson, 2003). This maize type may have an advantage where storage conditions are poor, as it may be more resistant to insect attack (Oregon State University, 2004).

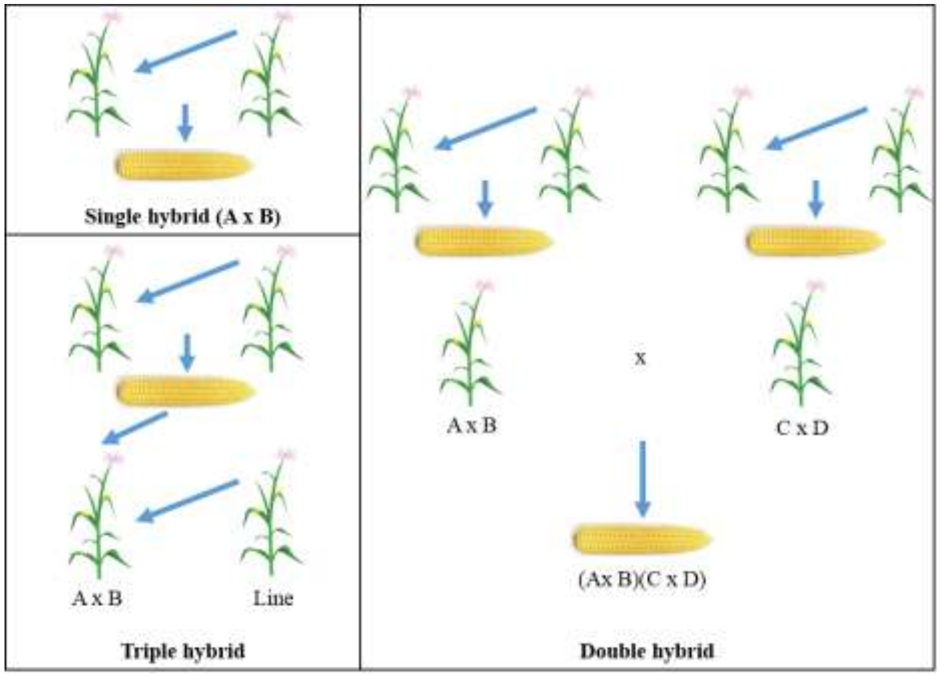


FIGURE 4. Types of hybrid corn, according to the number of parents (Biosciences for Farming in Africa, 2016; López, 2002)

Agricultural research in Guatemala started in the 1930s with the National Agricultural Chemical Institute (IQAN) mainly focusing on soil fertility, followed by the National Agricultural Institute (IAN) in 1944. Research in plant breeding consisted mainly in developing and evaluating lines (Osorio, 2009). During the 1950s, Guatemalan researchers participated in the collection of indigenous maize landraces as part of the US National Academy of Sciences-National Research Committee on Preservation of Indigenous Strains of Maize Project. In 1954, the Rockefeller Foundation initiated the Inter American Maize Improvement Program, aiming to test maize varieties which had already been exchanged between Colombia and Mexico. This provided germplasm, training, and technical assistance to maize researchers from the Guatemalan Ministry of Agriculture. Furthermore, Iowa State University established a tropical maize breeding station at Tiquisate on the Pacific coast in 1955. During this time, Guatemalan maize breeders released a number of improved OP varieties developed via selection. Some examples include two improved highland varieties, *Xela* and *San Marceño*, released in 1958. During the 1960s, seven lowland tropical varieties and one highland variety were developed, based mostly upon germplasm supplied by the Rockefeller Foundation maize

improvement programs working in Mexico, Colombia, and Central America (Baranski, 2014; Dowswell et al., 1996; Sigüenza Ramírez, 2010).

Later in 1970, the Agriculture Public Sector (SPA) was created in order to support agricultural and livestock production in the country. During the early 70s the Directorate General of Agricultural Services (DIGESA) was created within the Ministry of Agriculture, and it is the group in charge of coordinating activities of the SPA. In later years the SPA created over a dozen specialized units, out of which the Institute of Agricultural Sciences and Technology (ICTA) stands out for their work with maize. ICTA was founded in 1972 and its researchers' efforts were dedicated to improving several varieties of staple grain seeds (López, 2002), including rice, beans and maize, seeking better yields and higher nutritional content. The Rockefeller Foundation and United States Agency for International Development (USAID) financially supported the work done by ICTA. Over the years, the SPA was dismantled, however ICTA remained as a decentralized research entity under the overall responsibility of the Ministry of Agriculture (Dowswell et al., 1996; Sigüenza Ramírez, 2010).

ICTA was created as a decentralized state entity, autonomous, with legal personality, and with full capacity to acquire rights and obligations. Its organic law defines it as the public institution responsible for generating and promoting the use of agricultural science and technology (Sigüenza Ramírez, 2010), focused on generating, testing and transferring knowledge to small farmers (Echeverría, 1990). Much of the research work of ICTA focused on the area of breeding various crops, mainly to create resistance to pests and diseases (López, 2002; Osorio, 2009), improve adaptation to specific soils and climates, and to improve maize quality (Sigüenza Ramírez, 2010). Moreover, the International Maize and Wheat Improvement Center (CIMMYT) has played a key role in maize breeding research since 1973 providing maize germplasm (Figure 5); the reason why almost all, if not all, developed materials by ICTA has some CIMMYT germplasm within their composition (Morris and López-Pereira, 2000). During the 1970s, ICTA had approximately ten scientists involved in maize breeding, developing high-yielding OP varieties and hybrids. During the period of 1961-1991, ICTA released

39 improved maize varieties and hybrids for lowland areas and 6 improved varieties for the highlands of the country, out of which 35 contained CIMMYT germplasm in their background (Dowswell et al., 1996).

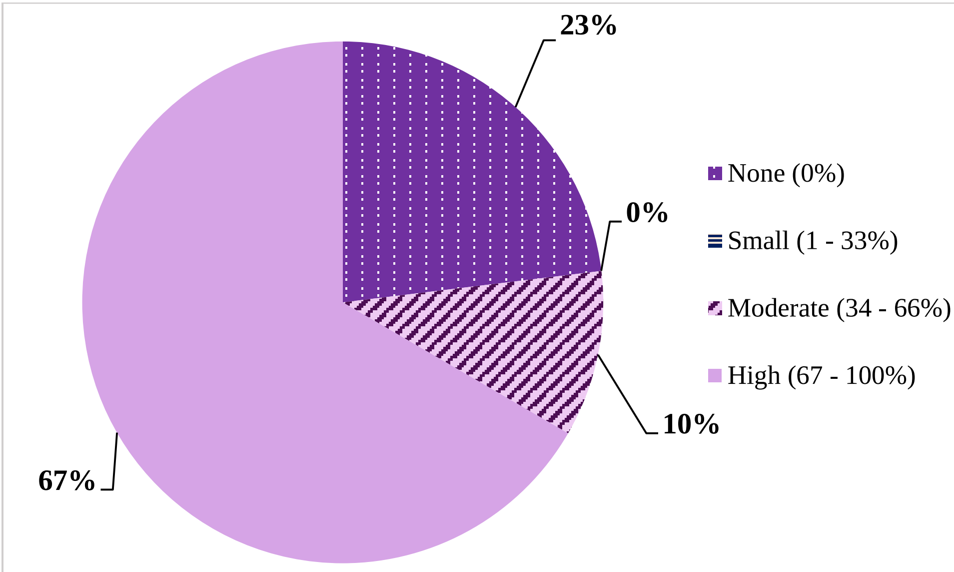


FIGURE 5. Approximate content of CIMMYT germplasm in maize materials generated by ICTA in Guatemala, 1966-1997. %ages in the graph are over the amount of materials developed. %ages in legend indicate how much of each germplasm composition belonged originally to CIMMYT (Morris and López-Pereira, 2000)

The maize research program has focused on two major environmental areas; Lowland and Highland. Accordingly, research priorities varied between regions. In most of the lowland agroclimatic region, where agriculture is commercially oriented, ICTA aimed to develop maize hybrids. In addition, ICTA’s maize research developed high-yielding OP materials with competitive yields that could be saved by the farm for subsequent harvests with little loss in yield potential. Within the highland zone, where most of Guatemala's subsistence farmers reside, ICTA scientists focused solely on OP materials that could be saved after every harvest without losing yield capacity (CIMMYT, 1981).

The seed industry policy in Guatemala was constructed upon a close partnership between the public and private sectors. In such partnership, the foundation seed of ICTA's maize varieties and hybrids was made available to private seed growers who produced the seed under the supervision of ICTA and Ministry of Agriculture seed specialists (Dowswell et al., 1996).

Private research in Guatemala has been encouraged by policies of ICTA and the government. ICTA may act as a maize provider to the private sector at a charge under a royalty system (Echeverría, 1990). These companies benefit from ICTA's research by using basic seed from this institution to create their own lines or by reproducing foundation seed from ICTA and selling it under their own labels (Echeverría, 1990). In the late 1980s, two private companies were involved in maize research. In 1990, these companies' research budgets were equivalent to nearly 50 % of ICTA's maize research budget (Dowswell et al., 1996). Between 1975 and 1990, the Guatemalan government invested Q1.4 million in maize research (Sigüenza Ramírez, 2010). During this time, the government dedicated the lowest share of public funds to agricultural research in the region (Mendez et al., 2015).

The commercial seed market is led by two private multinational establishments: Semillas Cristian-Burkard, recently acquired by Monsanto in 2008 (Klepek, 2012), and Seminal (Morris and López-Pereira, 2000). These are followed by Supersemilla and S. Ista. Altogether they hold 82 % share of the maize seed market (Morris and López-Pereira, 2000). The remaining fraction is distributed among ICTA (~8%) and other small seed firms (Echeverría, 1990). It is worth mentioning that approximately 40 % of the total volume of improved maize seed is based upon ICTA genotypes and the remaining fraction consists of proprietary hybrids by the private sector (Dowswell et al., 1996). As the private sector develops and serves the most favored areas (i.e. commercially-oriented) public research can concentrate on developing materials for less favored areas (i.e. highland region). Thus, to deliver improved seed to these area, in 1987 ICTA and DIGESA initiated an artisanal seed production program where ICTA had seed unit staff training local extension officers and artisan seed producers in seed production, providing

technical support, and supplying small amounts of foundation seed for seed reproduction (Dowswell et al., 1996).

The most important developed maize product between 2000 to 2010 was the OP material ICTA B-7, released in 2003, which had a large-scale dissemination in 2010 to 2013, and today has a high demand among producers on Eastern Guatemala due to its tolerance to drought. These were distributed during the years 2012-2014; in 2014 mainly supported by the HarvestPlus America Latina y Caribe program. This program is part of the Consortium of International Agricultural Research Centers (CGIAR) on Agriculture for Nutrition and Health, involved in agricultural development and seeking to provide health and nutritional benefits to the poorest populations (HarvestPlus, 2016; Raymundo, 2016)

Since mid-2012 the ICTA research program was assigned five professional researchers and had a five-year maize program plan, and a participatory plant breeding (PPB) approach was incorporated, emphasizing to the highlands of Guatemala. With the support of CIMMYT, in recent years ICTA has focused on maize hybrids to obtain materials resistant to tarspot, having similar material characteristics but higher productivity than the hybrid ICTA HB-83, to recover the genetics of their main hybrids (ICTA HB-83 and ICTA MayaQPM), and to obtain bio-fortified materials, mainly with a high Zinc (Zn) content, or high quality protein and high Zn. As a result of this work, ICTA currently has developed four high-yielding hybrids, namely two with high performance (one thought as a substitute for ICTA HB-83), and two tolerant to tarspot, which have already passed the validation stage (Raymundo, 2016).

ICTA is currently in the process of refining the procedure of increasing seed production. Additionally two hybrid materials high in Zn have been identified, which will begin the process of validation in the second half of 2016. In the case of OP varieties, ICTA has focused on the genetic improvement of traditional materials for the tropics with varieties ICTA B-1, ICTA B-5, ICTA B-7 and ICTA Maquina 7422. Highland materials include the previously mentioned San Marceño, the ICTA Compusto Blanco, ICTA C-

301 and ICTA Don Marshal. Also, synthetic materials of white and yellow grain have been developed, mainly with better performance than the local counterparts. Furthermore, varieties with high quality protein and high Zn, and PPB were developed, focused on improving native varieties based on the interests of farmers in the communities. The goal of the latter process is to obtain improved local varieties with a wide range of adaptation. As a result, ICTA has developed a maize material that is spreading among farmers in the tropics, with characteristics of good performance and high quality protein, named ICTA B-9^{ACP}. Furthermore, two additional materials of high quality protein and high Zn will be in the validation phase in 2016, out of which at least one is expected to start being distributed in 2017. Moreover, from traditional varieties, ICTA has also developed two improved varieties for the tropics, and two improved varieties for the plateau. Currently, there are 8 new materials of the plateau, also PPB oriented (Raymundo, 2016). Table 2 includes some developed OP and hybrid material by ICTA. Although ICTA has developed materials for mid- and high-altitude regions where most small-scale farmers are situated, most of the released OP varieties and all the hybrids are being used in the lowlands (Echeverría, 1990).

ICTA was one of the few institutions that survived closures near the 90s, however with a profile greatly diminished since. Until 1985, the public program had released an increasing amount of material each year, but began to decline afterwards (Sain and López-Pereira, 1999). In 1998 a process of structural adjustment to modernize the institute took place, changing to a research approach as user demand occurred. During the period from 1995 until 2011 they had a shoestring budget, funded mainly by CIMMYT with approximately $5,000 to $10,000 per year, and few professional investigators assigned.

Due to this continuous lack of funding, from 2007 to 2011 the program had only one professional researcher (Raymundo, 2016; Sain and López-Pereira, 1999; Sigüenza Ramírez, 2010). On average, from 2000 to 2011, ICTA had a government budget of Q10 M per year (~$1.3 M/year). The government budget for ICTA in 2012 was Q27 M (~$3.4M), Q32 M (~$4 M) in 2013, 2014 and 2015; and in 2016 it was Q35 M (~$4.4M)

(Raymundo, 2016). Other services provided by ICTA include technical recommendations for harvesting grains, providing detailed guides to aide farmers lacking proper grain-handling understanding (Instituto de Ciencia y Tecnologia Agricolas (ICTA), 2014).

MAIZE PRODUCTION LIMITATIONS

Once harvested, the fate of maize quality is dictated based on the farms' post-harvest handling practices. In the commercial sector (i.e. lowlands of the country), grain storage occurs in silos. For the smallholder sector, there is a variety of lower-cost storage structures. Small farmers consider storage of corn on the cob appropriate, thus both cob and kernel storage take place. In the highlands of Guatemala where cob storage is observed, it is performed with or without husk, in *trojas*, *mancuernas*, *tapancos*, *galeras*, *cajones* or other rustic storage vessels (Asociación Nacional del Cafe (ANACAFE), 2004; Food & Feed Grain Institute, 1984; Hirst Sole, 1994).

If the corn husk is intact, it encloses and protects the grain from insect attack. It also limits the exchange of moisture between the grain and the environment, reducing the probabilities of increased humidity which would otherwise jeopardize the safety of maize. Conversely, if the maize initially had 14 % moisture content or above, this enclosed condition slows the drying rate, therefore promoting microbial growth. In addition, insect damage provides an environment that encourages microbial infection (Ariño et al., 2009). *Sitotroga cerealella* (angoumois grain moth), *Rhyzopertha dominica* (grain borer), and the weevils *Sitophilus zeamais* and *S. oryzeae* are common insect pests encountered in Guatemalan maize fields and storage (Hirst Sole, 1994; López, 2002).

TABLE 2. Examples of maize materials developed by ICTA (López, 2002)

Zones	**Material**	**Commercial name**	**Characteristics**	**Yield* (qq/ma)**
Low tropical area	Open Pollinated (OP)	ICTA B-1	White. Dent texture. Resistant to wind currents. May be harvested between 90-120 days.	60-90
		ICTA La Maquina 7422	White. Semi-crystalline texture. Resistant to wind currents. May be harvested between 90-120 days. Tolerant to heavy rainfall.	60
		ICTA B-5	White. Crystalline texture. May be harvested between 80-95 days (precocious). Tolerant to areas with poor rainfall.	40
		ICTA A-6	Yellow. Semi-crystalline texture. May be harvested in approximately 90 days (precocious). Tolerant to areas with poor/erratic rainfall.	60
	Hybrids	ICTA HB-83	White, double hybrid. Semi-dent texture. Resistant to wind currents. Harvested at approximately 120 days.	70-100
		ICTA HB-83 improved	White, double hybrid. Semi-dent texture. Resistant to wind currents. Harvested at approximately 120 days.	78-100
		ICTA HB-Proticta	White, hybrid. Semi-crystalline texture. Higher tryptophan and lysine content. Harvested at approximately 120 days.	70-100
		HA-46	Yellow, hybrid. Semi-dent texture. Resistant to wind currents. Harvested at approximately 115 days.	65-90
		HA-48	Yellow, hybrid. Semi-dent texture. Resistant to wind currents. Harvested at approximately 115 days.	70-90
Highland area	Open Polinated (OP)	ICTA Compuesto bl.	White. Dent texture. Resistant to wind currents. Harvested at approximately 225 days. Altitude: 2100-2400 masl.	70-80
		ICTA San Marceño	Yellow. Dent texture. Harvested at approximately 210 days. Altitude: 2200-2400 masl.	70-80
		ICTA San Marceño impr.	Yellow. Dent texture. Resistant to wind currents and foliar disease. Harvested at approximately 210 days. Altitude: 1800-2000 masl.	84
		ICTA Chivarreto	Yellow. Semi-crystalline texture. Harvested at approximately 210 days. Altitude: 2500-2700 masl.	60-70
		ICTA Toto amarillo	Yellow. Semi-crystalline texture. Harvested at approximately 240 days. Altitude: 2200-2400 masl.	70-80
		Guateian Xela	Yellow. Dent texture. Recommended for the valley of Quetzaltenango specifically. Harvested at approximately 210 days.	60-70
		ICTA V-301	White. Dent texture. Resistant to wind currents. Harvested at approximately 190 days. Altitude: 1500-1900 masl.	60-70
		ICTA V-302	Yellow. Dent texture. Harvested at approximately 190 days. Altitude: 1500-1900 masl.	60-70
		ICTA V-304	Yellow. Dent texture. Harvested at approximately 210 days. Altitude: 1900-2100 masl.	60-70
		ICTA V-305	Yellow. Dent texture. Harvested at approximately 210 days. Altitude: 1900-2100 masl.	60-70
		ICTA Don Marshall	White and yellow. Dent texture. Harvested between 90-120 days. Altitude: 1400-2100 masl.	60-70

*depending on irrigation. 1ma = 0.7ha

Grain storage in silos provides a set of advantages such as semi-hermetic storage, in small quantities and almost independent of external environmental conditions. Being a closed container, the silo allows effective fumigation of the grain to control insects (Hirst Sole, 1994; Tefera et al., 2011). Previous findings from this country reveal that low-income farmers harvest their maize at high moisture levels, particularly when precipitation is excessive, so further drying is always necessary (Food & Feed Grain Institute, 1984; López, 2002), although this rarely, if ever, occurs. Sun-drying, a common practice in the rural areas, is a cost-efficient drying method for grains. However it is performed in uncontrolled environment for longer periods, promoting contamination by pests and fungi (Ashiq, 2015; Semple et al., 1991). Because of the high investment required for mechanical dryers, inexpensive options have been proposed for developing countries. Low-cost convective dryers such as the STR (from Vietnamese. S = sấy khô (*drying*) T = xe thớt (*flat-bed*) R = rẻ (*low cost*)) can be a viable alternative in small farming regions where electricity can be made available. Solar drying, which harnesses the radiative energy from the sun for drying applications, is a common process in many countries, particularly where environmental temperature reaches 30°C or higher (Chua and Chou, 2003). The latter however may encounter problems in the highlands as precipitation and clouds cover the area often, slowing the drying process via this low-cost approach. Furthermore, because some fungi take advantage of wounds on the grain surface to contaminate it, it has been suggested that if farms would consider shelling maize, such activity should be performed with dried maize, and at the last possible moment to reduce environmental exposure, thus decreasing the chances of fungal infection (Torres et al., 2015).

This maize safety issue is aggravated with mid-high temperature and moisture environments, thus farms in the lowland region (de Campos and Olszyna-Marzys, 1979; Food & Feed Grain Institute, 1984) are at high risk of insect damage and mycotoxin exposure. This is also of concern as this region is that commercializes the larger portion of maize nation-wide, possibly disseminating contaminated maize throughout the country. Previous studies have revealed the presence of mycotoxins or mycotoxigenic fungi in Guatemala. Martinez et al isolated, among other fungi, the mycotoxingenic

Penicillium sp., *Fusarium monoliforme,* and *Aspergillus flavus* from maize samples in domestic warehouses, local markets and farms throughout the country (Martinez et al., 1970). Maize from the Pacific coast of Guatemala stored for six months showed alarming aflatoxin contamination levels ranging from 28 to 240 ppb (de Campos and Olszyna-Marzys, 1979; de Campos et al., 1980). More recently, Torres et al performed a nation-wide mycotoxin screening revealing aflatoxin levels ranging from 1.6 to 2655ppb, whereas fumonisin contamination ranged from 0.01 to 14.6ppm (Torres et al., 2015). Some of the quantities mentioned exceed the FDA human exposure limits of 20ppb for aflatoxin and ~3ppm for fumonisin, respectively (NGFA U.S, 2011). Such significant level of contamination affects the Guatemalan population, especially those individuals having a monotonous grain-based diet who consume high amounts of maize on a daily basis. Some reported effects include the effect of fumonisin on the inhibition of ceramide synthase causing depletion of sphingolipids and accumulation of bioactive intermediates, interfering with the function of some membrane proteins, including the folate-binding protein (Marasas et al., 2004; Riley et al., 2015). Other effects include caused by this toxin include liver and esophageal cancer, and stunting (Lee and Ryu, 2015; Rocha et al., 2009). Likewise, continuous intake of aflatoxin may result in impaired child development, immune system suppression, liver cancer and even death (Cotty and Jaime-Garcia, 2007; Perrone et al., 2007)

GAPS IN THE CURRENT KNOWLEDGE AND FUTURE OPORTUNITIES FOR THE GUATEMALAN MAIZE PRODUCTION CHAIN

Regulations towards mycotoxin exposure limits must be addressed in the Guatemala. Currently there are no official limits in the country. As a result, marketing activities rely on international entities guidelines such as CODEX.   Currently, only guide values are established until regulation will be approved. In contrast, other regions in Latin America have advanced further in this regard. Specifically South America, where MERCOSUR, a trading block consisting of Argentina, Brazil, Paraguay and Uruguay, have harmonized mycotoxin regulations in place (FAO, 2004).   In addition to promoting the development of regulations, education should be provided to the poor sector from

rural areas, given that they do not necessarily market maize but do produce it for self-consumption. With education, farmers will be aware of the risks incurred in carrying out possible inefficient practices in the maize productive chain. In this regard, it is imperative to perform studies of current grain handling practices, including storage, microbial and pest infestation, among others. Taking advantage of the emerging technologies for mycotoxin screening with lower detection methods, more studies should be done in the country in order to assess the degree of risk the population is exposed to.

Several storage methods alternatives have been proposed in Latin America. The most popular one, the metal silo, is considered a key post-harvest technology in the fight against famine. This semi hermetically sealed cylindrical structure reduces pest and fungal infestation of its contents, thus increasing food quality, safety and security (Manuel et al., 2007; Tefera et al., 2011; Yusuf and He, 2011). Nonetheless, these technology comes with a cost that most smallholder farmers cannot necessarily afford. Non-governmental organizations and credit unions can aid the situation by providing loans so that the people of rural area can achieve financial and food security. On this subject, it is important to analyze the impact of the introduction of this technology on the income of Guatemalan farmers to assess its feasibility.

Regarding the maize plant breeding program in the country, from 2013 to date, seventeen young researchers (the majority under 30 years) have joined ICTA. The institute's forecast is that an additional group of nine young researchers will be joining the work-force in 2017 (Raymundo, 2016). This new talent will hopefully generate more maize material, specifically for those in need (i.e. highland farms) reducing the country's food insecurity.   Additionally, maize genetic diversity is rich in Guatemala, and is part of the inhabitants' cultural heritage (FAO and IPGRI, 2002; Van Etten, 2006). Provided that some developed materials have the intended impact, accompanied with proper grain handling practices (Food & Feed Grain Institute, 1984), farmers of low socioeconomic level could produce a surplus, allowing them enter into the formal market; that would in turn help meet the needs of every household, by the consumption and sale of safe, nutritious grain of high quality.

The implementation of a novel governmental approach to battle food insecurity is needed, restructuring weakened structures that had success in the past. Since the most limiting aspect for the success of crop programs in Guatemala is lack of funding, proper channeling must be performed in order to provide the necessary infrastructure to carry out maize-related activities. Additionally, with government support, prevention of pre-harvest contamination, and the detoxification of maize already contaminated, may open new export opportunities for Guatemalan farmers.

REFERENCES

Argueta, A., 2013. Moon Guatemala. Avalon Travel.

Ariño, A., Herrera, M., Juan, T., Estopañan, G., Carramiñana, J.J., Rota, C., Herrera, A., 2009. Influence of agricultural practices on the contamination of maize by fumonisin mycotoxins. J. Food Prot. 72, 898–902.

Ashiq, S., 2015. Natural occurrence of mycotoxins in food and feed: Pakistan perspective. Compr. Rev. Food Sci. Food Saf. 14, 159–175. doi:10.1111/1541-4337.12122

Asociación Nacional del Cafe (ANACAFE), 2004. Cultivo del Maíz. Programa de diversificacion de ingresos en la empresa cafetalera [WWW Document]. URL https://www.anacafe.org/glifos/index.php?title=Cultivo_de_maiz

Baranski, M., 2014. The wide adaptation of wheat: Expanding the Rockefeller Foundation's international agricultural research program.

Biosciences for Farming in Africa, 2016. What is a maize F1 hybrid [WWW Document]. URL http://b4fa.org/bioscience-in-brief/plantbreeding/intraspecific-f1-hybrids/what-is-a-maize-f1-hybrid/ (accessed 1.1.16).

Brown, W.L., Zuber, M.S., Darrah, L.L., Glover, D.V., 1985. Origin, adaptation, and types of corn. Natl. Corn Handb. 1–6.

Chua, K.J., Chou, S.K., 2003. Low-cost drying methods for developing countries. Trends

Food Sci. Technol. 14, 519–528. doi:10.1016/j.tifs.2003.07.003

CIMMYT, 1981. Investigacion y produccion de maiz en Guatemala. Guatemala.

Cotty, P.J., Jaime-Garcia, R., 2007. Influences of climate on aflatoxin producing fungi and aflatoxin contamination. Int. J. Food Microbiol. 119, 109–115. doi:10.1016/j.ijfoodmicro.2007.07.060

de Campos, M., Olszyna-Marzys, A.E., 1979. Aflatoxin contamination in grains and grain products during the dry season in Guatemala. Bull Env. Contam Toxicol. 22, 350–356.

de Campos, M., Santos, J.C., Olszyna-Marzys, A.E., 1980. Aflatoxin contamination in grains from the Pacific coast in Guatemala and the effect of storage upon contamination. Bull. Environ. Contam. Toxicol. 24, 789–795. doi:10.1007/BF01608190

Dowswell, C.R., Paliwal, R.L., Cantrell, R.P., 1996. Maize in the third world, 1st ed. WestviewPress, Boulder, Colorado.

Echeverría, R.G., 1990. Public and Private Investments in Maize Research in Mexico and Guatemala (No. 90/03). Mexico.

Ecology Action, 2010. Principio 7: Semillas de Polinización Abierta [WWW Document]. URL http://www.cultivebiointensivamente.org/Self_Teaching_8.html (accessed 6.6.16).

FAO, 2004. Worldwide regulations for mycotoxins in food and feed in 2003, 3. Mycotoxin regulations in 2003 and current developments. Rome.

FAO, IPGRI, 2002. The role of women in the conservation of the genetic resources of maize, 1st ed. Rome.

Food & Feed Grain Institute, 1984. Post-production systems for basic grains in the western highlands of Guatemala: a description and evaluation. Manhat.

HarvestPlus, 2016. HarvestPlus - Quienes somos [WWW Document]. URL http://lac.harvestplus.org/quienes-somos/

Hirst Sole, L.A., 1994. Algunos aspectos físicos y de procesamiento del maíz en grano y mazorca, con y sin tusa, almacenado en dos tipos de estructuras, con y sin tratamiento químico. Universidad del Valle de Guatemala. Ingeniería en Ciencias de Alimentos.

Index Mundi, 2016a. Guatemala Corn Production by Year [WWW Document]. URL http://www.indexmundi.com/agriculture/?country=gt&commodity=corn&graph=production (accessed 6.6.16).

Index Mundi, 2016b. Guatemala Corn Imports Annual Growth Rate [WWW Document]. URL http://www.indexmundi.com/agriculture/?country=gt&commodity=corn&graph=imports-growth-rate (accessed 6.6.16).

Index Mundi, 2015. Guatemala Population [WWW Document]. URL http://www.indexmundi.com/guatemala/population.html (accessed 1.1.16).

INGUAT, 2016. Guatemala Biomes [WWW Document]. Birding. URL http://birding.visitguatemala.com/biomes (accessed 6.6.16).

Instituto de Ciencia y Tecnologia Agricolas (ICTA), 2014. Recomendaciones técnicas para el cultivo de maiz para zonas de produccion comprendidas entre 0 a 1400 msnm.

Isakson, S.R., 2009. No hay ganancia en la milpa: the agrarian question, food sovereignty, and the on-farm conservation of agrobiodiversity in the Guatemalan highlands. J. Peasant Stud. 36, 725–759. doi:10.1080/03066150903353876

Klepek, J., 2012. Against the grain: knowledge alliances and resistance to agricultural biotechnology in Guatemala. Can. J. Dev. Stud. Can. d'études du développement 33, 310–325. doi:10.1080/02255189.2012.719824

Lee, H.J., Ryu, D., 2015. Advances in Mycotoxin Research: Public Health Perspectives. J. Food Sci. 80, T2970–T2983. doi:10.1111/1750-3841.13156

Leslie, J.F., Logrieco, A.F., 2014. Mycotoxin Reduction in Grain Chains, 1st ed. John Wiley & Sons, Inc., Manhattan, Kansas.

Lima, F.R., 1988. La Cultura Del Maiz En Guatemala, 1st ed. Ministerio de Cultura y Deportes, Guatemala.

López, M.F., 2002. Una guía para su manejo agronómico del maíz. Guatemala.

MAGA, 2015a. Situación del maíz blanco. Guatemala.

MAGA, 2015b. El Agro en Cifras 2014. doi:10.1017/CBO9781107415324.004

MAGA, 2013. El agro en cifras 2013, Direccion de Planeamiento. Guatemala.

Manuel, I., Abel, R., Muñoz, G., Ramírez, B., Hilario, V.J., Bellon, S.M., Mexicana,

R.F., Rosas, I.M., Muñoz, A.G., Valverde, B.R., Salgado, J.H.H., 2007. Physical and physiological seed quality of maize landraces stored in metalic silo and with traditional methods in Oaxaca, Mexico. Rev. Fitotec. Mex. 30, 69–78.

Marasas, W.F.O., Riley, R.T., Hendricks, K. a, Stevens, V.L., Sadler, T.W., Gelineau-van Waes, J., Missmer, S. a, Cabrera, J., Torres, O., Gelderblom, W.C. a, Allegood, J., Martínez, C., Maddox, J., Miller, J.D., Starr, L., Sullards, M.C., Roman, A.V., Voss, K. a, Wang, E., Merrill, A.H., 2004. Fumonisins disrupt sphingolipid metabolism, folate transport, and neural tube development in embryo culture and in vivo: a potential risk factor for human neural tube defects among populations consuming fumonisin-contaminated maize. Am. Soc. Nutr. Sci. 134, 711–716.

Martinez, E., Gomez, R., Bressani, R., 1970. Prevalencia de hongos en granos de maiz (Zea mays, L.) de Guatemala. Turrialba 20, 311–319.

Mendez, W., Galluzzi, G., Say, E., 2015. The importance of ininternational exchanges of plant genetic resources for national crop improvement in Guatemala (No. 154). Denmark.

Morris, M.L., López-Pereira, M.A., 2000. Impactos del mejoramiento de maíz en América Latina 1966-1997. Mexico.

NGFA U.S, 2011. FDA Mycotoxin Regulatory Guidance.

Oregon State University, 2004. Maize Taxonomy, Morphology and Reproduction [WWW Document]. CSS 330. World Food Crop. URL http://oregonstate.edu/instruct/css/330/six/

Osorio, L.R.M., 2009. Plant breeding programs in Guatemala [WWW Document]. Food Agric. Organ. United Nations. URL http://www.fao.org/in-action/plant-breeding/our-partners/americas/guatemala/en/ (accessed 1.6.16).

Perrone, G., Susca, A., Cozzi, G., Ehrlich, K., Varga, J., Frisvad, J.C., Meijer, M., Noonim, P., Mahakarnchanakul, W., Samson, R.A., 2007. Biodiversity of Aspergillus species in some important agricultural products. Stud. Mycol. 59, 53–66.

Raymundo, E.R., 2016. Interview: ICTA maize research work, period 2000-2016.

Riley, R.T., Torres, O., Matute, J., Gregory, S.G., Ashley-Koch, A.E., Showker, J.L., Mitchell, T., Voss, K.A., Maddox, J.R., Gelineau-van Waes, J.B., 2015. Evidence

for fumonisin inhibition of ceramide synthase in humans consuming maize-based foods and living in high exposure communities in Guatemala. Mol. Nutr. Food Res. 1, 2209–2224. doi:10.1002/mnfr.201500499

Rocha, L.O., Nakai, V.K., Braghini, R., Reis, T.A., Kobashigawa, E., Corrêa, B., 2009. Mycoflora and Co-occurrence of fumonisins and aflatoxins in freshly harvested corn in different regions of Brazil. Int. J. Mol. Sci. 10, 5090–5103. doi:10.3390/ijms10115090

Sain, G., López-Pereira, M.A., 1999. Maize Production and Agricultural Policies in Central America and Mexico (No. 99-02), CIMMYT Economics Working Paper. Mexico, D.F.

Saleem, M.J., Bajwa, R., Hannan, A., Qaiser, T.A., 2012. Maize seed storage mycoflora in Pakistan and its chemical control. Pakistan J. Bot. 44, 807–812.

Semple, R.L., Frio, A.S., Hicks, P.A., Lozare, J.V., 1991. Control of aflatoxin in maize, in: Mycotoxin Prevention and Control in Foodgrains. Bankok, Thailand, pp. 166–172.

Sigüenza Ramírez, P., 2010. Nuestro maíz, nuestro futuro : estudios para la reactivación de la producción nacional de maíz en Guatemala. Instituto de Estudios Agrarios y Rurales. -IDEARCoordinación de ONG y Cooperativas -CONGCOOP-, Guatemala.

Tefera, T., Kanampiu, F., De Groote, H., Hellin, J., Mugo, S., Kimenju, S., Beyene, Y., Boddupalli, P.M., Shiferaw, B., Banziger, M., 2011. The metal silo: An effective grain storage technology for reducing post-harvest insect and pathogen losses in maize while improving smallholder farmers' food security in developing countries. Crop Prot. 30, 240–245. doi:10.1016/j.cropro.2010.11.015

Torres, O., Matute, J., Gelineau-van Waes, J., Maddox, J.R., Gregory, S.G., Ashley-Koch, A.E., Showker, J.L., Voss, K.A., Riley, R.T., 2015. Human health implications from co-exposure to aflatoxins and fumonisins in maize-based foods in Latin America: Guatemala as a case study. World Mycotoxin J. 8, 143–159. doi:10.3920/WMJ2014.1736

Torres, O.R., 2000. Evaluacion de metodos para decontaminar maiz con micotoxinas a nivel del hogar en una comunidad indigena del altiplano de Guatemala. Guatemala.

United Nations, 2014. Country classification. Data sources, country classifications and

aggregation methodology.

USDA, 2014. Guatemala Corn Domestic Consumption by Year [WWW Document]. URL http://www.indexmundi.com/agriculture/?country=gt&commodity=corn&graph=domestic-consumption

Valladares, L., 1989. Culto al maíz en Guatemala; y, Psicología del indígena coloteco, 1st ed. Guatemala.

Van Etten, J., 2006. Molding maize: the shaping of a crop diversity landscape in the western highlands of Guatemala. J. Hist. Geogr. 32, 689–711. doi:10.1016/j.jhg.2005.12.002

Van Etten, J., Fuentes, R.M., 2004. La Crisis Del Maíz En Guatemala: Las importaciones de maiz y la agricultura familiar. Anu. Estud. Centroam. 30, 51–66.

White, P., Johnson, L., 2003. Corn: chemistry and technology, Second. ed. American Association of Cereal Chemists, Inc., St. Paul, MN.

Wiedenbörner, M., 2014. Mycotoxins in Foodstuffs, 2nd ed.

Wild, C.P., Gong, Y.Y., 2009. Mycotoxins and human disease: A largely ignored global health issue. Carcinogenesis 31, 71–82. doi:10.1093/carcin/bgp264

Yusuf, B.L., He, Y., 2011. Design , development and techniques for controlling grains post-harvest losses with metal silo for small and medium scale farmers. African J. Biotechnol. 10, 14552–14561. doi:10.5897/AJB11.1845

TRADITIONAL MAIZE POST-HARVEST MANAGEMENT PRACTICES AMONGST SMALLHOLDER FARMERS IN THE WESTERN HIGHLANDS OF GUATEMALA*

R. Mendoza[1], L. Sabillón[1], W. Martinez[3], C. Campabadal[2], H. Hallen-Adams[1], and A. Bianchini[1]

[1] Department of Food Science & Technology, University of Nebraska. Lincoln, NE, USA

[3] Grain Science & Industry, IGP, Kansas State University. Manhattan, KS, USA

[3] Development Organization SHARE Guatemala. Guatemala, GTM.

Corresponding author:

Andréia Bianchini, Department of Food Science & Technology

The Food Processing Center, University of Nebraska-Lincoln

Phone: (402)472-3144, Fax: (402)472-1693

abianchini2@unl.edu

*Scientific manuscript formatted to meet guidelines for the Journal of Storage Products Research

ABSTRACT

Much of the maize that is produced in the Guatemalan Highlands is planted, harvested and handled via subsistence-oriented agricultural practices, which are strongly connected to Mayan heritage. This post-harvest assessment study was done to characterize the current post-harvest practices used by farmers in the region of Huehuetenango, Guatemala, in order to identify the different grain handling practices in the region as well as possible factors contributing to post-harvest losses of maize. A total of 280 families representing 14 rural communities were surveyed through focus groups discussions and interviews. Survey revealed that most (88%) of interviewed farmers prefer to dry the maize cobs after harvest by laying them in stacks exposed to direct sunlight. After drying, harvested maize is stored until consumption along with purchased maize kernels from the market. Among storage practices 62% of surveyed families, mostly maize buyers rather than producers, store the maize as shelled kernels whilst 38% do so on cobs. When storing shelled maize, bags are the preferred containers among 81% of farmers, while only 14% use metal silos. Among farmers who stored maize on cobs, 74% use the *tapanco* (kitchen loft) as the preferred storage structure. Forty-one percent of farmers indicated storing the maize for at least 4 months. During the storage time, 61% of farmers perform grain quality checks once a week. Moreover, 65% perform pest control during storage; however, in most cases, the control is not preventive but corrective. For 49% of farmers, the main cause of maize loss between harvest and consumption is the mishandling of grain moisture, leading to insect infestation and fungal growth. With the data and analyzed information, it was possible to identify diverse maize harvesting, drying, storage and consumption practices within the studied communities. A better understanding of traditional post-harvest practices will help better design intervention steps to improve these practices and to increase food security and safety for smallholder farmers in the Guatemalan Highlands.

Keywords: Guatemala, maize, post-harvest, grain loss

INTRODUCTION

The majority of agriculture in the highlands of Guatemala is devoted to the culture of maize and beans. Wheat, squash and a few other vegetables are also grown on small farms (Williams and Menegazzo, 1988) mostly due to their attractive market value (Hamilton, 2005)(Reardon et al., 2009), but also for being less labor intensive (Immink and Alarcon, 1993). Of these, maize is considered a staple crop for Guatemala's population (Argueta, 2013), specifically for the poor sector (Van Etten and Fuentes, 2004), being the main food source for Guatemalans with an average annual production of 1.7 million metric tons (USDA, 2014). More than half of Guatemala's maize is consumed as tortillas, at approximately 170 kg per capita per year (Schmidt et al., 2012)(Kenneth F. Kiple, 2000). Besides this, several other Guatemalan dishes are based largely on maize, which also happens to be a very susceptible commodity to fungal contamination (Appell et al., 2009).

Fungi thrive in relatively low moisture environments compared with bacteria, which make grains, and more specifically maize, a perfect niche. This is further exacerbated when poor handling and storage conditions allow access to pests and/or promote moisture migration to the seed. Given that certain fungi can produce harmful compounds to humans and other animals, their presence becomes of concern. The toxicity of these compounds, known as mycotoxins, is dependent on several parameters such as dosage, chemical structure, length of exposure, and affected organism, among others (Cornell University, 2015)(Bryła et al., 2013). Depending upon the doses, the effects of food-borne mycotoxins can be acute, with symptoms of severe illness appearing rapidly. At lower doses fungal toxins show long term chronic effects on health, including the induction of cancer, and immune deficiency (Food and Agriculture Organization of the United Nations, 2016). Previous work in the lowlands of Guatemala indicate an incidence of mainly fumonisin but also aflatoxin, mycotoxins produced by fungi in the genera *Fusarium* and *Aspergillus*, respectively, acting synergistically or individually. Health problems attributed to these mycotoxicoses in the region include neural tube defects, stunting and hepatocellular carcinoma (Torres et al., 2015)(Torres et al., 2007).

Previous findings by CIMMYT revealed that for a significant part of the western highlands of Guatemala food production is subsistence; agricultural assets are generally very small and rural properties are highly fragmented (CIMMYT, 1981). It is in such rural regions where people have limited economic resources that a larger maize consumption is more noticeable (Torres et al., 2007), therefore even low levels of mycotoxin contamination could pose a substantial health risk to this population. Moreover, the lack of financial support results in limited technical knowledge and tools (Immink and Alarcon, 1993) for appropriate grain handling practices, among which proper storage and drying equipment stand out. Many of the farmers in the highlands of Guatemala use rudimentary and empirical techniques where little technology is involved. Consequently the present work aims to evaluate such conditions to better understand their potential role and impact on maize quality and safety.

This study took place in Huehuetenango, Guatemala. This department lies in the northwestern corner of Guatemala. Geographically, it is bounded to the north and west by Mexico, to the east by the department of Quiché and to the southeast by the department of Totonicapán. This region is largely mountainous with a total area of approximately 7500 square kilometers (~2900 mi^2) (Baepler, 2016). More specifically, the townships of Chiantla and Todos Santos Cuchumatán, of the Huehuetenango department were subject to investigation.

Understanding the different traditional maize-handling practices performed in the Highlands of Guatemala will help elucidate their potential influence on the class of maize produced, sold or consumed, as well as its safety and shelf-life. Additionally, data would guide the choice of better intervention steps, if necessary, to decrease smallholder farmers' maize spoilage and post-harvest losses, and ultimately increase the food security and safety of the region.

MATERIALS AND METHODS

Sampling method. Households located in communities or settlements known as landscapes, villages, towns, cities, etc. from Todos Santos and Chiantla were randomly selected. The sample size (n = 267 households) obtained from community conglomerates was determined using the following equation (Shein-Chung Chow, Hansheng Wang, 2007):

$$n = \frac{\left(Z_{\alpha/2}\right)^2 * p(1-p)}{d^2}$$

Where,

- $Z_{\alpha/2}$: 1.962, confidence level at 95%. Two-tail test.
- p: Ratio, 0.50. The variance of the indicators measured as a proportion reaches a maximum point as they approach 0.50, ensuring an adequate sample size.
- q: 1 – p, probability of failure. Complement of the event.
- d: accuracy or acceptable error limit. In this case 6% (0.06).

Possible losses of study subjects for various reasons (data loss, abandonment, no answer) was also taken into account with a sample increase. The adjusted sample size (n_{adj}=272) was determined as follows (Mariela Borda Pérez, Rafael Tuesca Molina, 2013):

$$n_{adj} = n\left(\frac{1}{1-R}\right)$$

Where,

- R: Proportion of expected losses, 2% (0.02) is expected.

A sample of 280 households was obtained, distributed in Chiantla (35.7%) and Todos Santos (64.3%). Although Todos Santos’ population and terrain are the smaller of the two, the selection of the sample is proportionately greater due to its variations in altitude.

Community selection. The communities were selected based on their altitude and maize production chain (producers or purchasers). Additionally two communities were included as a control group. Communities were divided in three groups depending upon the altitude: type C: altitude from sea level until 1,500 masl (meters above sea level), type B: between 1500 and 2700 masl, and type A: above 2700 masl.

Farmers having land available to plant and harvest maize (producers) were designated “Chain 1” farmers; while farmers who didn’t have land and thus rely on purchasing maize were identified as “Chain 2” farmers. With 20 families per community, 14 communities were covered in this study: 8 from Todos Santos, 4 from Chiantla and 2 as control group (1 for each township).

Surveying process. Two hundred and eighty families from the 14 communities of Todos Santos and Chiantla, townships of Huehuetenango in Guatemala, were surveyed between May and August 2014. The survey consisted of 80 questions in order to get acquainted with household composition, practices related to agriculture and grain handling, community organization, level of technical education, hygiene and health. Only results related to maize agriculture, harvest, grain handling and storage are included in the present article. Unless otherwise noted, farmers’ answers to questions were referred to the 2013-2014 harvest season.

Before the actual interviewing of the different households, the interviewers selected for the survey were properly trained. At the end of such trainings, interviewers reported having knowledge and understanding of the study objectives, mastering the survey instrument (i.e. ballots) to be used, having an impartial interview technique, knowing the areas the study comprised, logistics and contact with community, route plan, among others. In addition interviewers spent a day of work in the field to validate their skills and mastery over the instruments.

This validation was performed with people in the community of Taluca from the township of Chiantla. This community was not selected to participate in the study,

however it showed similar characteristics to those that were. Several consultations between post-harvest scientists and SHARE, the NGO providing field personnel, resulted in the refined survey instrument and procedures to be followed.

RESULTS AND DISCUSSION

Farmers and land tenure. Almost the entire sample (99.6%) mentioned having land available for agriculture. However 90.0% of respondents own the land while 10.0% rent or borrow. Of this, 82% of land owners and 15.7% of renters/borrowers expressed using the land specifically for planting maize. Much of the decision to plant or buy maize to meet household demands was associated with maize availability, the economic capacity for hiring labor and buying fertilizer, as well as the support from government/social programs (Washington Office on Latin America (WOLA), 2013).

Even though land is available, at times it is not necessarily enough and consequently farmers rent or borrow additional land. Table 1 provides a breakdown of land tenure and usage based on land size. Overall it is a subsistence agriculture (CIMMYT, 1981) with no abundant revenues, thus neither food security nor capital growth are maintained.

TABLE 1. Land tenure and usage according to size in Todos Santos and Chiantla, Huehuetenango, Guatemala

Land size (cuerda*)	Percentage of land		
	Used for planting (%)	Owned (%)	Rented (%)
1 - 3	27.1	29.6	30.6
4 - 8	34.5	34.0	50.0
> 9	38.4	36.4	19.4

*1 cuerda = 20*20 m

Even though the main purpose of farming in this region is home consumption, data indicates that there is not enough land available to reach the household demand. This could be related to weather conditions that affect production, as well as the average size

of families. Of the surveyed households, 62.1% were comprised of 6 or more members. This affects directly the food availability of every household, as their low economic means may prevent all members having access to nutritious food, and therefore they may not have a balanced diet.

Moreover, farmers have shown a positive attitude towards planting higher value crops for profit as they are aware of the opportunity to use their land for both economic and cultural heritage benefits (Hamilton, 2005). From an economic point of view, the proportion of land dedicated to maize agriculture may suffer a decrease in the future. Farmers from other regions of the Highlands of Guatemala are starting to be open to the idea of including non-conventional commodities such as mini-squashes and berries in their land. These products can result in higher profit compared to traditional commodities (Hamilton, 2005).

Maize planting and harvest. Although most (80.4%) farmers produce maize, as this traditional agricultural practice is an important component of their identity (Hamilton, 2005)(Van Etten and Fuentes, 2004), all of the surveyed farmers buy additional maize for home consumption confirming an insufficient production for the annual household demand.

Regarding the seed usage, 95% of the farmers reported using native (*criolla*) seeds. The most outstanding varieties used by the farms in the region included: annual white maize, short white maize, white maize, *San Lorenzo* yellow, dog's teeth, native yellow, pinto maize, *Salqueño* maize, black maize, *Sarquilito* and native *Chucuy* maize. These are the common names used by the farmers to describe their heirloom seeds which have been used for years, and have been selected for their best features (i.e. kernel uniformity in size, large cob size, resistance to pest damage/mold, etc.). More information on selection criteria will be presented.

A majority (66.8%) of farmers believe *criolla* seeds (mostly flint varieties) have higher yields than commercial (dent) varieties, while 38.4% find *criolla* seeds superior in

pest and disease resistance. This latter advantage is likely due to the maize composition. Unlike their dented counterparts, flint corn has shown to be more impervious to insect damage as it possesses a hard outer layer to protect the soft endosperm (Suleiman et al., 2015). Additionally, inhabitants of the region prefer some native varieties while preparing specific food products (Van Etten, 2006). For instance, farmers in the community of San Antonio Las Nubes mentioned that *Diente de Perro* (dog's teeth) is used for tortillas while *Salpor* is used for baking bread.

Regarding the availability of materials and tools to plant maize, nearly all of the farmers (99.5%) reported having tools (hoe, shovel, etc.) to work the land during planting and harvest. Also, 94.0% have access to fertilizers, 11.0% use improved seeds, and 61.5% use native seeds. Only 3.3% reported access to irrigation equipment. It is important to mention that in the region of study, located in the north-western region of Guatemala, there are generally two alternate planting seasons, based on elevation. The "January cycle", which is from January to October, usually is performed in the elevated areas or higher plateau at approximately 2600 masl. This planting is done leveraging the naturally occurring moisture in this region. The "May cycle" from May to December, in the lower regions, is dependent upon the rainy season. Accordingly, for those places where rain is not frequent, yields can be compromised due to poor plant development if not enough water is available in the growing stage of the plant life cycle. Conversely, those regions with excessive rainfall lead to high moisture levels in the field causing ear rot, premature sprouting, and mold growth. Furthermore, once harvested maize from these high moisture level areas may take longer to dry exposing the maize to the environment for a longer period, thus making it more vulnerable to pests and fungi.

Traditional knowledge in maize handling. A large portion of the sample (92.9%) reported having a minimum of five years of experience managing their land; most have done it throughout their lives. The maize is destined for self-consumption (98.9%) and only a small fraction is sold to neighbors or local markets.

A practice called *dobla* (to fold the stem of the maize plant to interrupt the transport of water and nutrients, accelerating drying) is done when the grain is fully formed and is no longer milky, which happens around 85 to 90 days after planting (Instituto de Ciencia y Tecnologia Agricolas (ICTA), 2014). This is practiced by 33% of the interviewees. The majority of farmers follow very specific conditions such as a period of prolonged rain to establish the harvesting time. For those who perform *dobla*, some will ascertain the proper time to perform it based on the color of the tassel (10.4%) or the leaves (26.4%), and a smaller fraction rely on the nail test (4.4%). This last method consists of evaluating the grain hardness by puncturing a kernel with a fingernail.

Regarding the *tapisca* (harvest), farmers follow specific practices or combination of practices inherited through generations. Similarly as for *dobla*, some farmers proceed to harvest based on the color of the tassel (12.0%) or leaves (27.2%); the nail test (9.2%); mouth test (2.2%) which consists of evaluating the maize hardness by biting a kernel; between 25-30 days (Instituto de Ciencia y Tecnologia Agricolas (ICTA), 2014) after *dobla* (42.9%); based on rain pattern (14.3%); and some proceed to harvest specifically after every December 1st (14.3%). In some cases, farmers will notice that the evaluated cob is *sazón* (ready), and proceed to collect the entire maize harvest at once rather than let it partially rot or germinate on the fields. In some cases, varying from farm to farm, maize growers have set dates for harvest based on relevant calendar dates (either Catholic or civil calendar). As an example, some farmers in Todos Santos prefer to perform their harvest after the *All Saints' Day*, celebrated on November 1st.

Seventy six percent of the respondents perform harvest entirely by hand while others use tools such as machetes or knifes to facilitate the task. Some farmers in San Antonio las Nubes mentioned that the harvest is performed during a full moon, as it results in much harder grain and is more resistant to pest attack (Bravo Martinez, 2009) during storage. Field observations revealed that most farmers didn't perform tillage on their farms. Even though no-till has advantages such as erosion control (Chulze et al., 2000), it can also significantly increase the relative frequency of mycotoxigenic and spoilage fungi present in the field, thus increasing the chances of contamination.

Maize damage. Upon harvest, 98.4% of farmers said they see some type of damage either on the maize plant or on the cob; in most cases both kinds of damage were observed. Figure 1 shows the different sources of damage identified by farmers. It is known that weather during the growing and harvesting season influences maize damage to some extent due to contamination with fungi (Cotty and Jaime-Garcia, 2007), which was the most frequently reported type of damage (~68%). It is important to note that farmers only reported fungal damage when it was evident (e.g. pink slurry, white mold), but even when not noticeable, fungal damage may have already occurred while they take this maize as visually safe (Martinez et al., 1970).

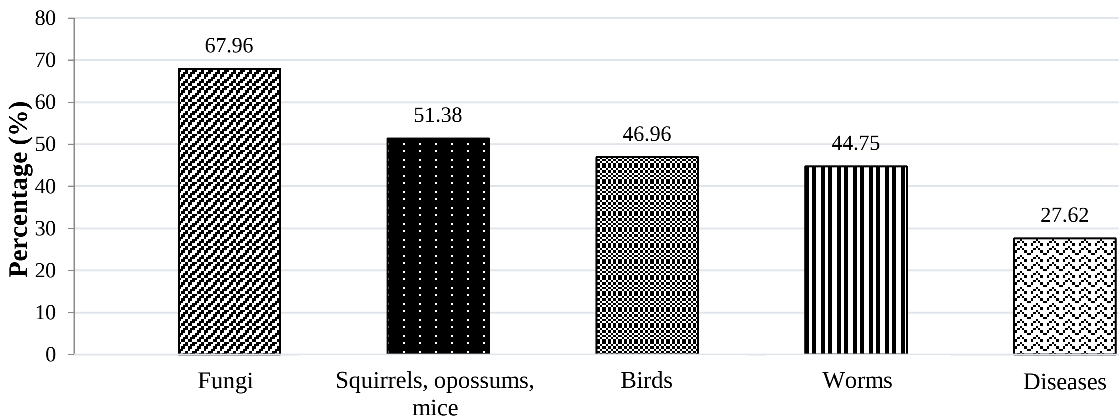


FIGURE 1. Farmers' observations regarding maize damage during harvesting period.

Other sources of damage reported were animal damage (namely squirrels, opossums and mice) followed by bird damage, worms and diseases. All of these ultimately result in entry points for fungi, explaining the reported dominance of fungal damage in the region.

Selection and drying practices. Selection is performed either for storage and consumption purposes, or for later use as seeds for the next production cycle. Interviews revealed that this is performed based on the color of the cob (77.17%), size of the kernel (52.17%), proper amount of kernels per cob (48.37%), kernels without visible damage or

stains (31.52%) and/or absence of fungi. Other less frequent but important features mentioned were cob weight and the combination of weight and color. For some farmers seed selection takes place in the field during harvest (37.36%) while others do it prior to drying (37.91%) or when the maize is being stored (28.02%.). *Mulco* maize are terms used to refer to maize damaged by fungi, insects or rodents. The fate of this maize will be discussed in later sections.

The majority of interviewed farmers (93.5%) who produce maize reported performing some type of drying practice before storage. Out of that fraction, 3.5% dry the maize in the whole plant (*milpa*), before cutting the cob. The rest remove the cob from the plant and then proceed to dry it. In this group, 88.4% of farmers indicated sun-drying the ears before storing, while 10.5% said they place the ears directly in the *tapanco*. However, farmers said they can combine different techniques (i.e. sun-drying and drying in *tapanco*). Alternatively, some farmers dry the maize after the shelling process. Once shelled, 10.0% of the interviewees place the maize on nylon sheets followed by sun-drying. Among all farmers the most common practice is to dry the ears after being harvested. Some farmers in San José las Flores reported to have *tapancos* above their living quarters with tin roofs. They use this as a space for drying maize over a period of approximately one month (November to December), and then place it in wooden boxes located inside the house, in silos or bags. This practice is due to the rainy conditions around harvesting in this area, which makes it impossible to do sun-drying. Although it is considered a promising post-harvest storage technology to maintain maize quality, there is a potential issue for farmers who store their maize in metal silos. Conditions such as temperature fluctuations and improper drying prior to storage (i.e. maize with unsafe moisture levels), common in this tropical country, could promote condensation on the storage vessel inner walls, rewetting the grain in specific areas and thus creating hot spots for fungi and an accompanying intensification in mycotoxin occurrence.

Storage and grain usage practices. Decision regarding grain readiness for storage is based on traditional practices that include tactile or finger-nail test (32%),

mouth test (16.9%), and a combination of sound and visual observation (45.4%). A small number of producers (~3%) have a surplus in their production out of which 1.7% sell maize in the local market or to families in the community. This small proportion is partially due to lack of economic means to have more land available to plant maize. Additionally, smallholder farmers do not have technical knowledge or assistance to meet the rapidly changing and stringent food quality standards (Hamilton, 2005) for commerce. It was observed that when maize was usually sold it was usually on market days: Wednesdays, Thursdays, and Sundays.

It was found that 74% of the farmers prefer to use the *tapanco* as storage for ears. This practice allows farmers to protect it from the elements. If the maize's husk is intact, it encloses the kernels and protects them against insects, but it also limits the exchange of moisture between the kernels and the environment (Sole, 1994). It was observed that some families rely on the greenhouse effect of the *tapanco* letting the heat of the roof finish the drying process. This is of concern as corn that is potentially not properly dried is stored, in some instances for years, with no adequate aeration or moisture control. This can lead to mold growth and potentially be a risk factor to future harvests placed in this space, resulting in the same issue as previously described for silo usage.

To a lesser degree, maize is stored in *mancuerna* (23%) which consists of partially husking the ears and using the remaining husks to hang them from the main beams in the porch area. This practice is associated with the cobs selected to be used as seeds for the next crop cycle. For some farmers the practice of *mancuerna* is more dependent on the variety, as field observations revealed that the *salpor* and other native varieties are preferably dried via this method. The remaining farmers (3%) utilize *troja* as a mean of storage. This is a self-standing box structure built from scrap wood and wire. This last storage method is becoming less popular compared to the others previously mentioned; tendency indicated that it may disappear in the future. Some of the storage methods described are pictured in Figure 2.

FIGURE 2. Different storage methods found in Chiantla and Todos Santos. a) Tapanco, b) Cajón (wooden box), c) Metal silo d) Mancuerna

Farmers that buy maize, or shell it upon drying, prefer (81%) to store it in bags woven on tubular fabric made from polypropylene. This practice is more widespread in the township of Todos Santos (64%) than in Chiantla (46%). Among farmers that buy maize the practice of using the original package (bag) as a storage container is popular due to its convenience.

Out of the 14% of respondents who use silos, 79% indicated using the *pastilla* (pill) of a phosphine or phosphamine salt for pest control. Field observations, however,

revealed that the method of implementation is not precise as farmers add either insufficient or excess amounts of it, being unaware of possible biological (i.e. prevailing pests) or chemical contamination of their crops.

Regarding the length of storage, 41% of respondents indicated storing maize for 4 months or more (Figure 3), followed by 30% who store their corn for 1 month, 19% for 3 months and 10% for 2 months.   According to visits during the study it was observed that some families may buy maize that is sufficient for only a week or one month since their income does not allow them to purchase large quantities of grain. This explains why almost one third of respondents store their maize for less than one month.

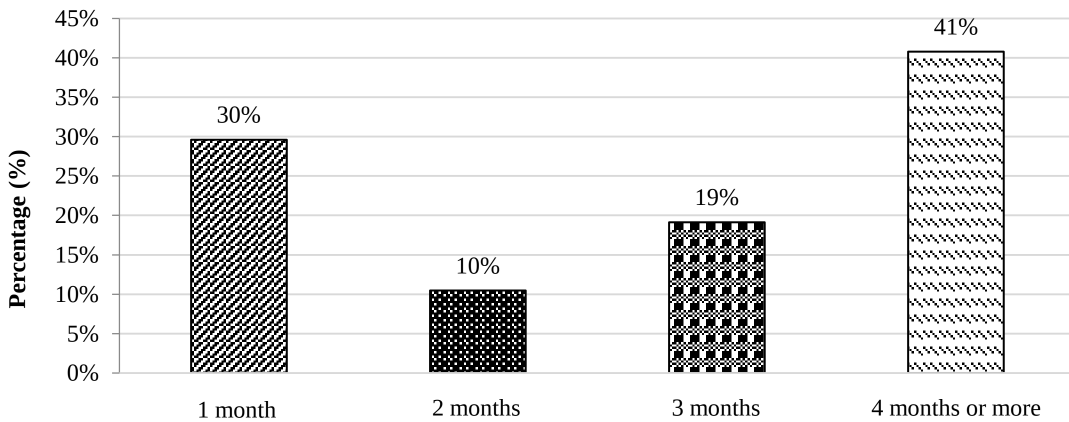


FIGURE 3. Maize storage period according to farmers. Graph reflects pattern observed by farmers throughout several seasons and it does not necessarily reflect the last season

Storage management. In both Chiantla and Todos Santos townships, the practice of cleaning the storage site was evident (98%). Of this, 98% indicated that they clean the storage location before placing the freshly harvested or purchased maize. Eight percent clean the storage location each month and 2% every two months. The practice of cleaning the storage site consists of sweeping the *tapanco* or cleaning the wooden box or silo using a broom and/or cloth. Ninety percent of these farmers perform quality checks before storage and among those, 61.4% check it once a week, 14.1% twice a month, and 5.2% once per month.

Sixty-five percent of farmers perform pest control for grain storage, focusing on rodent, moth and weevil control mostly in the *tapanco*. From these, 53% do it when rodents are observed, 10% do so at the time of storage, and 22% when they observe damage in the kernels, presence of moths, weevils or other insect damage. In most cases, field observations revealed that such control is not preventive but rather corrective. During field visits, farmers mentioned that smoke generated from the kitchen during food preparation helps with pest control for grain placed in the *tapanco*, as this storage assembly is usually above the stove area.

Post-harvest maize losses. According to farmers, losses after harvest are caused by rodents (18%), rot (32%), due to grain and environmental moisture (12%), fungal damage (5%), birds (5%) and insects (9%). As mentioned before, damage caused by pests contributes to the damage caused by fungi as wounds allow the latter to proliferate (Cotty and Jaime-Garcia, 2007). The rotting of ears and kernels is the most commonly identified reason for loss by farmers. It is important to mention that rot damage, moisture or presence of fungi are related thus overall 49% of farmers reported losses due to excessive humidity or mishandling of moisture in the grain. In many cases, they first consume the purchased maize leaving the maize they harvested for the end of the season which may aggravate the issue. Table 2 shows post-harvest losses reported by chain and township. A total production of 2237 quintals of maize was reported among surveyed farmers, and from this, 146.7 quintals (6.6%) were lost during storage. Among farmers that purchase maize (chain 2), a total of 5,229 qq were acquired during the period covered by the survey (harvest 2013-2014). From the total, a loss of 82 qq was reported representing a 1.5% loss.

TABLE 2. Perceived post-harvest losses in the region of study by chain and township

Township	Chain	Produced or Purchased maize (qq*)	Reported loss (qq)	Percentage of loss (%)
Todos Santos	1	1,197	82.5	6.9
	2	3,282	48.4	1.4
Chiantla	1	1,040	64.2	6.2
	2	2,247	33.1	1.4

*1 quintal = 100 lb

Additionally, it is imperative to mention that the reported loss must be understood from the point of view that "damaged" maize may be defined differently among farmers of the Highlands and from a commercially acceptable point of view. In practice, they discard very little maize even when it is damaged. Much maize that is elsewhere considered as damaged is still used in these regions for human or animal consumption, so one can expect that losses reported here may be underestimated.

Overall, this information shows how deficient current grain handling practices are in the region, since maize buyers (i.e. chain 2) had a considerably lower percentage of losses compared to maize producers: ~1% and ~6% respectively.

Damaged maize usage. Seventy percent of the interviewed households indicated that maize that shows any sign of damage is given to animals. Interestingly, although maize is considered damaged, 20% of the families would still consume it. Damaged maize can be either used for human consumption by mixing (“diluting”) with sound maize, for animal consumption or eventually discarded; a decision which usually lies with women as field observations and previous research (Hamilton, 2005) in the Highlands of Guatemala revealed. The practice of mixing *mulco* with healthy maize kernels impinges directly on food safety, as the *mulco* portion has more evident pest damage and is likely to be heavily infested by microorganisms, and possibly by several toxigenic species. This practice is more frequent in Todos Santos than in Chiantla since the waste of maize in Todos Santos is not considered socially acceptable. Culturally it is said that "People who waste or throw the maize kernels may develop rash on their skin" which would be "punishment from God" for the sin of wasting maize. Overall, 83% of farmers buy maize to replace what has gone bad. However, in Todos Santos, one quarter of all respondents indicated not replacing maize even when damage is evident. In Chiantla, 94% of farmers reported having replaced the damaged maize.

Maize and food security. According to the survey results, compared to previous seasons, maize production in 2014 fell by 20-60% due to a prolonged drought in the area under study, putting at risk the food availability for farmers in 2015. During this study

67.4% of farmers reported having harvested less than the previous year. Table 3 summarizes the yield of 2014-2015 harvest. During 2014 the prolonged drought began in July 18th and ended on August 14th. This drought affected the proper development of plants and consequently the maize yield.

TABLE 3. Maize production in Chiantla and Todos Santos, season 2014-2015

Harvest range (qq)	Percentage of farmers (%)
5 - 10	71.2
15 - 20	15.8
>25	13.0

For Guatemalan families maize is a staple food. Seventy-four percent of respondents indicated that they need more than 5 lb of maize per day to feed their family. However in some cases the need was between 12 and 15 pounds. When townships were compared, the study found that in Chiantla 86% of the families need more than 5 lb per day; while in Todos Santos this pattern occurs for 66% of respondents. Because of their dependence on maize, the region's food insecurity is intensified during erratic weather patterns, such as the irregular rainfall and extremely long heat wave observed during the 2014 season.

Maize availability. Table 4 reveals estimated maize consumption quantities for the region of study, where 52.9% of respondents consume less than 600 g/person/day (=1.32 lb/person/day) with an overall mean of 388 g/person/day (=0.85 lb/person/day) for this group. The remaining respondents consume more than 600 g/person/day, with some consuming as much as ~3000g/person/day. Eighty-four percent of respondents indicated having maize in storage at the time of data collection; quantities shown in Figure 4. Basing family maize requirements on a consumption of approximately 317.5-453.6 g/person/day (=0.7-1.0 lb/person/day) (CIMMYT, 1981)(Bressani, 1990), the majority of farmers would not have enough maize to support their families beyond 2 months, coinciding with previous findings (López, 2002). Twenty-five percent of respondents reported having more than 500 lb of maize in storage, however after asking if the current

stored maize would be sufficient to meet the food needs of the family until the next harvest, more than half of this fraction (67%) indicated that it was not enough, showing signs of food insecurity.

TABLE 4. Estimated average maize daily consumption in farms from Chiantla and Todos Santos, Huehuetenango, Guatemala.

Maize consumption (g/person/day)	Group average consumption ±SD (g/person/day)	Percentage (%)
<600	387.6 ±127.6	52.9
600-1000	757.4 ±118.7	35.4

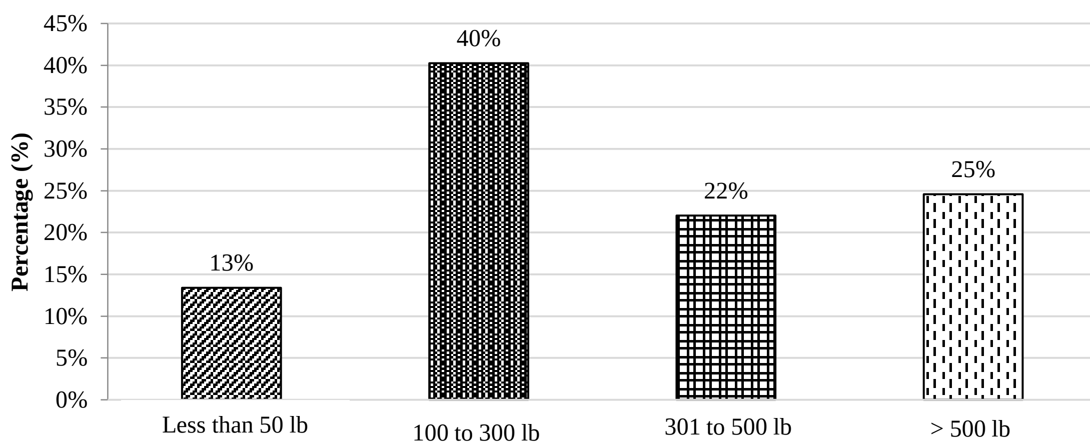


FIGURE 4. Available maize in storage among farmers surveyed (Season 2014-2015).

Data collected in the present study showed that farmers of the highlands of Guatemala are in need of improving their agricultural practices in conjunction with proper grain handling after harvest in their farms. Most improvements could be applied in the areas of drying and storage of grain. Aggravating this scenario, a reduced income and consequently a monotonous diet increases the risk of mycotoxicosis (acute and chronic) for the inhabitants of the region as the maize is clearly handled in such ways that threaten its quality and safety.

Acknowledgements. Authors wish to acknowledge SHARE Guatemala for the field work in Huehuetenango. Research done at the Food Innovation Center's laboratory is supported by USAID through the Feed the Future Innovation lab for the Reduction of Post-Harvest Loss, from Feed the Future, the U.S. Government's Global Hunger and Food Security Initiative. Global Hunger and Food Security Research Strategy: Climate Resilience, Nutrition, and Policy (RFA-AID-OAA-L-14-00002).

Conflict of interest. None.

REFERENCES

Appell, M., Kendra, D., Trucksess, M., 2009. Mycotoxin prevention and control in agriculture, 1st ed. American Chemical Society.

Argueta, A., 2013. Moon Guatemala. Avalon Travel.

Baepler, D.H., 2016. The Avifauna of the Soloma Region in Huehuetenango, Guatemala. Cooper Ornithol. Soc. 64, 140–153.

Bravo Martinez, J., 2009. Metal silos and food security. Lessons Learned from a Successful Central American Post-harvest program. Quito, Ecuador.

Bressani, R., 1990. Chemistry, technology and nutritive value of maize tortillas. Food Rev. Int. 6, 225–264.

Bryła, M., Roszko, M., Szymczyk, K., Jędrzejczak, R., Obiedziński, M.W., Sękul, J., 2013. Fumonisins in plant-origin food and fodder-a review. Food Addit. Contam. 30, 1626–1640. doi:10.1080/19440049.2013.809624

Chulze, S.N., Ramirez, M.L., Torres, A., Leslie, J.F., 2000. Genetic variation in Fusarium section Liseola from no-till maize in Argentina. Appl. Environ. Microbiol. 66, 5312–5315. doi:10.1128/AEM.66.12.5312-5315.2000

CIMMYT, 1981. Investigacion y produccion de maiz en Guatemala. Guatemala.

Cornell University, 2015. Aflatoxins: Occurrence and Health Risks [WWW Document]. URL http://poisonousplants.ansci.cornell.edu/toxicagents/aflatoxin/aflatoxin.html (accessed 10.10.15).

Cotty, P.J., Jaime-Garcia, R., 2007. Influences of climate on aflatoxin producing fungi

and aflatoxin contamination. Int. J. Food Microbiol. 119, 109–115. doi:10.1016/j.ijfoodmicro.2007.07.060

Food and Agriculture Organization of the United Nations, 2016. What are mycotoxins? [WWW Document]. Mycotoxins in Grains. URL http://www.fao.org/wairdocs/x5008e/x5008e01.htm (accessed 1.4.16).

Hamilton, S., 2005. Maya Farmers and Export Agriculture in Highland Guatemala: Implications for Development and Labor Relations. Lat. Am. Perspect. 32, 33–58. doi:10.1177/0094582X05279503

Immink, M.D.C., Alarcon, J.A., 1993. Household Income, Food Availability, and Commercial Crop Production by Smallholder Farmers in the Western Highlands of Guatemala. Econ. Dev. Cult. Change 41, 319–342.

Instituto de Ciencia y Tecnologia Agricolas (ICTA), 2014. Recomendaciones técnicas para el cultivo de maiz para zonas de produccion comprendidas entre 0 a 1400 msnm.

Kenneth F. Kiple, K.C.O., 2000. The Cambridge World History of Food, 1st ed. Cambridge University Press.

López, M.F., 2002. Una guía para su manejo agronómico del maíz. Guatemala.

Mariela Borda Pérez, Rafael Tuesca Molina, E.N.L., 2013. Métodos cuantitiativos. Herramientas para la investigación en salud, 4th ed. Barranquilla, Colombia.

Martinez, E., Gomez, R., Bressani, R., 1970. Prevalencia de hongos en granos de maiz (Zea mays, L.) de Guatemala. Turrialba 20, 311–319.

Reardon, T., Barrett, C.B., Berdegué, J.A., Swinnen, J.F.M., 2009. Agrifood Industry Transformation and Small Farmers in Developing Countries. World Dev. 37, 1717–1727. doi:10.1016/j.worlddev.2008.08.023

Schmidt, A., Eitzinger, A., Sonder, K., Sain, G., 2012. Tortillas on the Roaster (ToR). Central American Maize-Bean Systems and the Changing Climate.

Shein-Chung Chow, Hansheng Wang, J.S., 2007. Sample Size Calculations in Clinical Research, 2nd ed. CRC Press.

Sole, L.A.H., 1994. Algunos aspectos físicos y de procesamiento del maiz en grano y mazorca, con y sin tusa, almacenado en dos tipos de estructuras, con y sin tratamiento químico. Universidad del Valle de Guatemala.

Suleiman, R., Williams, D., Nissen, A., Bern, C.J., Rosentrater, K.A., 2015. Is flint corn naturally resistant to Sitophilus zeamais infestation? J. Stored Prod. Res. 60, 19–24. doi:10.1016/j.jspr.2014.10.007

Torres, O. a, Palencia, E., Lopez de Pratdesaba, L., Grajeda, R., Fuentes, M., Speer, M.C., Merrill, A.H., O'Donnell, K., Bacon, C.W., Glenn, A.E., Riley, R.T., 2007. Estimated fumonisin exposure in Guatemala is greatest in consumers of lowland maize. J. Nutr. 137, 2723–2729. doi:137/12/2723 [pii]

Torres, O., Matute, J., Gelineau-van Waes, J., Maddox, J.R., Gregory, S.G., Ashley-Koch, A.E., Showker, J.L., Voss, K.A., Riley, R.T., 2015. Human health implications from co-exposure to aflatoxins and fumonisins in maize-based foods in Latin America: Guatemala as a case study. World Mycotoxin J. 8, 143–159. doi:10.3920/WMJ2014.1736

USDA, 2014. Guatemala Corn Domestic Consumption by Year [WWW Document]. URL http://www.indexmundi.com/agriculture/?country=gt&commodity=corn&graph=domestic-consumption

Van Etten, J., 2006. Molding maize: the shaping of a crop diversity landscape in the western highlands of Guatemala. J. Hist. Geogr. 32, 689–711. doi:10.1016/j.jhg.2005.12.002

Van Etten, J., Fuentes, R.M., 2004. La Crisis Del Maíz En Guatemala: Las importaciones de maiz y la agricultura familiar. Anu. Estud. Centroam. 30, 51–66.

Washington Office on Latin America (WOLA), 2013. Government Investment in Family Agriculture New Opportunities in Mexico and Central America.

Williams, W., Menegazzo, G., 1988. Apple culture in the highlands of Guatemal. Acta-Horticulturae 232, 57–66.

ASSESMENT OF SMALLHOLDER FARMERS' MAIZE IN THE WESTERN HIGHLANDS OF GUATEMALA: INSECT AND MOLD INFESTATION, MYCOTOXIN CONTAMINATION, AND PROXIMATE ANALYSIS *

R. Mendoza[1], A. Rodas[4], A. Oliva[4], L. Sabillón[1], A. Colmenares[4], J. Clarke[1, 2], H. Hallen-Adams[1], C. Campabadal[3], and A. Bianchini[1]

[1] Department of Food Science & Technology, University of Nebraska. Lincoln, NE, USA

[2] Department of Statistics, University of Nebraska. Lincoln, NE, USA

[3] Grain Science & Industry, IGP, Kansas State University. Manhattan, KS, USA

[4] Department of Food Science & Engineering, Universidad Del Valle. Guatemala, GTM.

Corresponding author:

Andréia Bianchini, Department of Food Science & Technology

The Food Processing Center, University of Nebraska-Lincoln

Phone: (402)472-3144, Fax: (402)472-1693

abianchini2@unl.edu

*Scientific manuscript formatted to meet guidelines for the Journal of Food Protection

ABSTRACT

Maize, a staple in many developing countries, is prone to pest attack and fungal infestation when inadequate practices occur during production and storage. More specifically, in Guatemala, mycotoxin contamination of agricultural products is a result of a combination of environmental conditions, tradition, and poverty, among other factors. In order to assess the current conditions, mold and insect count as well as fumonisin and aflatoxin exposure from farms (n=25) in two townships of the Huehuetenango department, Chiantla and Todos Santos, were determined. Moreover, native and commercial maize varieties were arranged in 13 distinctive groups and a proximate analysis was carried out in order to see if there was a variety group with a nutritional potential advantage. Total fungal count in maize samples ranged from 3.6 to 6.83 log CFU/g and differences among farms of different altitudes were not significant at $p<0.05$. Farms where maize is not produced but bought are at higher risk of fumonisin contamination, whereas local producers are affected by aflatoxins. Overall, aflatoxin had an incidence of 100% ranging from 1.0 to 85.3 ppb, while fumonisin was in 52% of the evaluated farms ranging from 0.4 to 31.0 ppm. The means+SD of all detectable cases of mycotoxin contamination were above the Provisional Maximum Tolerable Daily Intake (PMTDI) for fumonisin as well as the recommended maximum ingestion levels of aflatoxin. Daily intake values ranged from 0.01 to 0.85 μg/kg bw/day for aflatoxin, and 2.91 to 310.0 μg/kg bw/day for fumonisin. The entomological analysis revealed an overall incidence of *Ephestia kuehniella* (flour moth) of 32%, *Sitophilus zeamais* (maize weevil) of 16% and *Tribolium sp*. (flour beetle) of 8% for the analyzed farms. In the case of maize buyers, only 5% of the analyzed samples from the highest altitude (>2700 masl) in the study had flour moths, however producers from the same altitude had no insect presence. In addition, maize producers from the lowest altitude (sea level-1500 masl) had the greatest amount of insects. Proximate analysis revealed a significant higher protein content for white (8.7%), yellow (9.1%) and white-yellow hybrid (8.9%) flint varieties from maize producers as well as for yellow dent (9.2%) and flint (8.9%) varieties from maize buyers. Farmers of the highlands of Guatemala need to improve their agricultural practices in order to have safe and nutritious maize and maize-based products.

Keywords: Guatemala, maize, corn, mycotoxins, insects, proximate analysis

INTRODUCTION

Maize (*Zea mays*) is one of the main cereals, widely consumed throughout the world in many forms as result of different processes (*7*). In developing countries such as Guatemala where maize is a staple, due to sub-optimal post-harvest management techniques, a large portion of the annual yield is lost during storage. This is exacerbated in rural areas of the country where considerably higher amounts of maize are consumed on a daily basis (*7*, *30*, *60*). Such losses have been attributed partly to microbial action during storage (*3*).

Microorganisms associated with grains are notorious for causing severe health hazards such as farmer's lung, aspergillosis, and mostly important, production of mycotoxins. Also, tropical climate with high temperature and relative humidity along with poor storage conditions adversely affect the preservation of harvest (*3*). In many cases subsistence farmers may not have no alternative but to consume a certain amount of damaged harvest (*5*), and as a result the food consumed is consistently contaminated with numerous microorganisms (*33*). Of these, fungi stand out due to some being mycotoxin-producers hence compromising not only the quality but also the safety of the food/feed products as well as market potential.

It has been established (*6*, *33*, *43*) that a combination of 70% relative humidity, and maize moisture equal to or below 14% are fair conditions to maintain grain quality over time, assuming restricted access to pests and prompt treatment with pesticides and fungicides when necessary (*53*). In rural areas, however, inhabitants that harvest maize for self-consumption do not necessarily have the technical knowledge or tools to perform preventive or corrective measurements to maintain the maize in safe conditions but are rather guided by their empirical experience. When storage is not properly controlled, many pests attack the grain and moisture can then accumulate from their activities providing ideal conditions for fungal activity (*11*).

Fungi found in grain can be classified into two groups, field fungi and storage fungi (*46*). Some examples of the former include *Alternaria* and *Fusarium* spp., growing

in the developing kernels when the moisture content is high (20-40%), but usually not in stored grain. These fungi gradually die during storage, so that old grain generally has low counts of viable field fungi. Low numbers of field fungi can also reflect arid growing conditions or high drying temperatures in maize (*56*). As the name implies, storage fungi invade grains or seeds during storage. These fungi are usually not present to any serious extent before harvest. Under improper storage conditions they can increase rapidly leading to significant problems, influenced mainly by moisture content, temperature, condition of the grain going into storage, the length of storage time and pest activity. The most common storage fungi include *Aspergillus* and *Penicillium* species (*58*).

Mycotoxins are low molecular weight secondary metabolites of filamentous fungi which represent a risk to both human and animal health (*51*). In humans, mycotoxins can cause liver necrosis, reduced growth, esophageal cancer, and depressed immune system response, among other symptoms (*12*, *29*) depending on the mycotoxin, dosage and frequency of exposure. Such toxins are produced by saprophytic fungi during storage or by endophytic fungi during plant growth (*26*). The mycotoxigenic fungi that infect maize can produce a wide range of mycotoxins, of which the carcinogenic aflatoxins and fumonisins are the most commonly detected and of the greatest public health importance (*33*). Also, feeding livestock contaminated grain has been shown to decrease animal productivity (*22*), with an according reduction in the food supply. Moreover, maize that appears healthy may also be contaminated if pre- and post-harvest conditions favored fungal growth (*37*).

Aflatoxin contamination is greater in maize that has been produced under stress conditions. Thus, drought, heat, insect damage, and fertilizer stress are all conducive to high levels of aflatoxins (*13*, *29*). *Aspergillus flavus* is the most common species associated with aflatoxin contamination of agricultural crops, being highly stable in soil and on the plant (*13, 47*). *A. flavus* produces aflatoxin B1 and aflatoxin B2. *A. parasiticus* produces these toxins as well as aflatoxin G1 and aflatoxin G2 (*33*). Additionally, for

lactating mammals, dietary aflatoxin is metabolized to another form of the toxin, aflatoxin-M, which is secreted in the milk (*37*).

It is estimated (*9*) that more than half of the maize and maize-based products consumed around the world are contaminated with fumonisins. These mycotoxins are produced by *Fusarium* species such as, *F. verticillioides*, *F. proliferatum* and others (*9*, *38*, *60*), and are usually found in regions where climate is warm and wet (*9*, *50*). Both of these endophytic species produce fumonisin B1, fumonisin B2 and fumonisin B3, and along with other several *Fusaria* that co-occur in maize, can also synthesize a wide range of additional toxins, including moniliformin, trichothecenes such as deoxynivalenol and nivalenol, zearalenone, fusaproliferin and beauvericin (*33*).

After harvest, maize is still a living organism, thus it continues to respire and generate heat, carbon dioxide and water. At this point, the viability of potential attacking organisms depends on nutrient availability, temperature, kernel composition, relative humidity and oxygen content of the gases surrounding the maize (*24*). In Guatemala environmental conditions provide an ideal scenario for pest invasion. In the northern part of the country as well as on the Pacific and Atlantic coasts the climate is hot and humid; in the interior of the northern part it is hot and dry and in the highlands, it is cold or temperate and humid (*10*).

Insects destroy at least 5% of harvested and stored grain worldwide. They can also affect the maize respiration rate proportional to their damage (*24*) and cause a carbohydrate depletion (*3*, *31*), which ultimately reduces maize quality. Insects can also decrease seed germination. Moreover, insect presence can result in excreta production, body parts, unpleasant odors and flavor, and unwanted microflora (*24*). Regarding the latter, insect pests carry pathogenic fungi and expose or predispose plants to disease development (*23*) compromising the safety of the commodity.

In order to better understand the current situation regarding maize quality and safety in the Highlands of Guatemala, a study comprising 25 farms of different altitudes

from Todos Santos and Chiantla located in the department of Huehuetenango was conducted. In this study maize was evaluated for insect infestation, fungal load, and mycotoxin content, specifically fumonisin and aflatoxin. Additionally, different maize varieties were identified, and to further characterize the staple of the region proximate analyses were also conducted.

MATERIALS AND METHODS

Studied area. In general for Guatemala, maize agriculture is focused in the highlands and parts of the south-western and north-eastern coast, between altitudes of 0 to approximately 3000 masl (meters above sea level) (*53*). Thus the region of study was categorized into three altitudes: Type C altitude (temperate to warm) from sea level until 1500 masl, type B altitude (mild) between 1500 and 2700 masl, and type A altitude (cold) above 2700 masl.

Maize sampling. Twenty five farms from 9 communities distributed in Todos Santos (n=6) and Chiantla (n=3), townships of Huehuetenango, were selected for this phase of the study. Communities in Chiantla were San José Las Flores, Cumbre La Botija and San Antonio Las Nubes. In the Todos Santos region communities included were Tres cruces, Tuiboch, Rio Ocho, Chichim, Chemal II, and Chicoy.

Samples were collected at different time points, including harvest, and days 0, 30, 60 and 90 of storage from the 2014-2015 harvesting season. Farms where maize was planted and harvested were identified as “chain 1” farms. Conversely, farms where there was insufficient land to plant maize and thus farmers had to rely on purchasing grains from markets or similar were designated “chain 2” farms.

Environmental readings. Relative humidity (RH) of the storage area was monitored for 16 farms from different altitudes every 60 minutes with a Temperature/Relative Humidity data logger. HOBO-ONSET UX100 data loggers were used for traditional storage inside the households. 5 Sony CR2032 HRB-TEMP-1 data

loggers were used for traditional storage exposed to the environment. Approximately 4000 readings were obtained per farm. Out of the 25 farms included in the study, 9 did not have a logger placed at their location and therefore their RH data was assigned based on data from a farm that was at an equivalent altitude and similar physical location (i.e. department, township and community).

Moisture determination. Moisture was measured immediately after sampling at each farm utilizing a John Deere Grain Moisture Tester (model SW08120, US), used according to manufacturer's instructions.

Entomological analysis. Maize (kernels, cob) samples from each time point were checked visually to establish insect incidence. For those cases where insects belonged to the order *Coleoptera* (i.e. beetles), isolates collected directly from the sample and placed in a container with ethanol (Fisher Scientific, USA) 80% v/v. When insects were moths, samples were placed in a refrigerator for 15 minutes to inactivate the insects. After refrigeration insects were placed in a plastic container. An individual count was performed followed by photographing and identification under stereoscope. Samples were kept under observation for 15 days at room temperature in a container that allowed the entry of oxygen to the sample. After this period, another check was conducted to evaluate any internal infestation. Total incidence per altitude/chain was reported.

Mold count. Maize samples from each time point were aseptically transferred to sterile containers, soaked with 0.1% peptone water (DIFCO, USA) and they were left to soak for 30 minutes. Sample was then blended (high-grind) for a minimum of 3 minutes or until the sample was properly ground. Samples were serially diluted and plated on Dichloran Rose Bengal Chloramphenicol –DRBC– (DIFCO, USA) agar followed by a 5 day incubation period at 25°C. Results were reported as logarithm of colony-forming units per gram of maize.

Mycotoxin analysis. Analysis was performed on maize samples collected throughout the storage period as well as during harvest. Total aflatoxin (B1, B2, G1 and

G2) and fumonisin (B1, B2 and B3) were measured using an Agravision® Agrastrip lateral-quantifiable ELISA test (Romer Labs, Missouri). Briefly, maize samples were milled so that 75% would pass through a 20-mesh screen and a 10 g sub-sample was mixed with 70% methanol (Merck, USA) solution. After 1 min of vigorous shaking, sample was left to sediment for approximately 2.2 minutes. Prior to the reading, 50 µL of the supernatant was mixed with 950 µL of dilution buffer for fumonisin or 1000 µL of dilution buffer for aflatoxin analysis. The range of detection for aflatoxin was 0 to 100 ppb with a detection limit (LOD) of 3.6 ppb and a limit of quantitation (LOQ) of 5.0 ppb. The range of detection for fumonisin was 0 to 5 ppm with a LOD of 0.3 ppm and a LOQ of 0.4 ppm. Readings below the LOD were taken as zero. Results between LOD and LOQ were considered as LOQ/2 (*44*). For readings above the maximum limit, the extracts were diluted until a measurement within the range of detection was obtained and the amount was reported after applying the proper dilution factor.

Proximate analysis. Raw samples of native maize varieties were assayed for proximate composition. Moisture content was determined as the weight loss after being exposed in a convection oven (AOAC 931.04). Protein content was determined by the Dumas method (AOAC 992.23). Ash content was determined as the residue remaining after incinerating the sample in a muffle furnace (AOAC 923.03). The lipidic fraction was determined using the Soxhlet solvent extraction method (AOAC 920.39). Carbohydrate content was determined by difference (*2*, *19*).

Data analysis. R version 3.2.3 was used to perform the statistical analysis. The three different altitudes were compared using an ANOVA test with the objective of evaluating any significant differences among the means of relative humidity. After confirming that the ANOVA was significant (data not shown, $p<2e^{-16}$), pairwise comparisons using t tests with pooled SD were done. Bonferroni was used as p value adjustment method to evaluate significant differences. Instead of using all of the RH values (n>4000), 6 representative values were used to summarize the distribution: mean, 25 percentile, median, min, max and 50 percentile.

Maize moisture content (field values) was evaluated using Wilcoxon rank sum test pairwise comparisons. Analysis was done between altitudes within chain (1 or 2), and each altitude across chains (1 and 2). Proximates for all maize varieties were evaluated using a pairwise comparisons ANOVA to see any significant difference; p-values shown as a matrix. Finally a Recursive Partitioning Model (RPM) was used for mycotoxin analysis as it did not follow a normal distribution.

RESULTS AND DISCUSSION

Controlling maize post-harvest conditions helps maintain its quality or minimize any deteriorative changes (*24*). Given that moisture is a key parameter that will define the overall grain quality throughout storage until consumption, moisture measurements were taken in the field maize at harvest and during storage at different altitudes of Chiantla and Todos Santos, Huehuetenango, Guatemala; data shown in Figure 1.

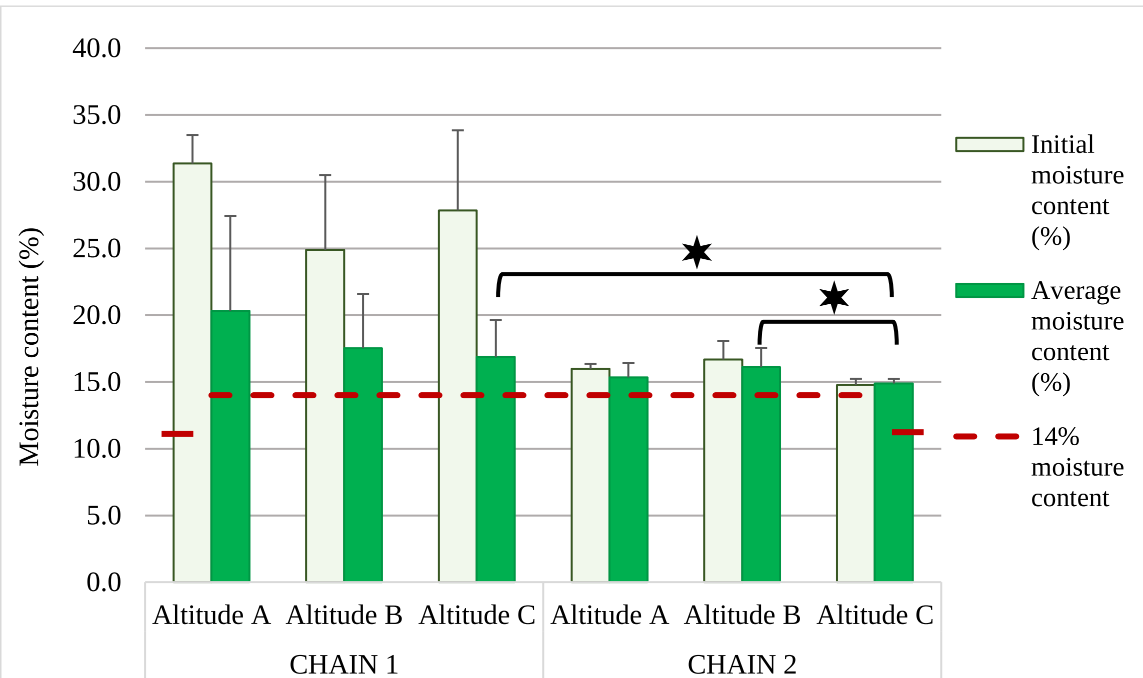


FIGURE 1. Average moisture content of maize from Chiantla and Todos Santos, 2014-2015 maize season. Bars on the left (□) indicate initial moisture at harvest (Chain 1) or storage at day zero (Chain 2) and bars on the right (■) indicate an average measurement of all readings during storage. ✶ denotes significant difference among maize moisture data.

For chain 1 farms (i.e. maize producers) maize is naturally high in moisture after harvest, at approximately 30%. Therefore some farmers from the region proceed to naturally dry it via diverse methods followed by storage in a variety of structures depending on the farm and maize variety. After removing harvest data due to traditional drying processes, within chain 1 all pairwise comparisons of average storage moisture between altitude levels were not significant (smallest *p*-value was >0.30). Similarly, within chain 2 (i.e. maize buyers) most pairwise comparisons of average storage moisture between altitude levels were not significant except for B and C ($p<0.029$). From the Wilcoxon rank sum test, chain 2-altitude B farms being at higher risk of presenting spoilage and safety issues. All farms located in the 3 different altitudes across chains surpassed the safe limit of ≤14% moisture (*43*), with that being more evident for chain 1 farms. This finding suggests that, under these conditions, grain would be prone to degradation, decreasing not only its nutritional value but also its safety for consumers as the maize would be highly predisposed to mold development and thus mycotoxin contamination. Comparing each altitude across chains, the only significant difference observed was for altitude C, with $p<0.037$ from Wilcoxon rank sum test. Based on these findings, it can be said that altitude does not play a significant role in maize moisture during storage in the traditional storage types (e.g. bags, *tapanco*, *troja*) for chain 1 farms.

Another factor affecting grain quality involves pests that can infest maize both in the field and in storage. As rodents and birds are comparatively large in size and can move rapidly from one place to another, it is difficult to quantify their presence. However, insect occurrence can be quantified and was consequently evaluated. Three insects were identified in the region of study: flour moth (*Ephestia kuehniella*), flour beetle (*Tribolium sp.*) and maize weevil (*Sitophilus zeamais*) (Figure 2). Insect incidence can affect not only the nutritional value of the maize and its quality, but also its safety as infestation creates entry points for microorganisms (*13*, *14*) and can act as vectors.

Maize weevil, commonly encountered in warm regions (*24*), can infest stored maize kernels or corn cobs before harvest. When the husks are removed, or following

bird damage on the ears in the field, weevils can feed on the grain and spawn inside of the kernels (*20*) which classifies them as a *primary insect*. In contrast, the secondary insects, flour beetle and flour moth, develop outside of the grain. The latter is destructive of the maize plant and grain, both in field and storage, and can cause an unpleasant flavor when kernels are consumed (*24*). Additionally, it has been reported that the European corn borer moth's (*Ostrinia nubilalis*) larvae feeding on the plant may transport *Fusarium* spores from the leaf surface into the plant interior tissues (*9*). Similarly, other moths, including the moth detected in the studied region, could also increase fungal contamination of grain. Many insects carry aflatoxin-producing fungi (*13*). Some secondary insects also feed specifically on moldy kernel surfaces, such that their presence is attributed to poor storage conditions.

Moreover, both abiotic (temperature, humidity) and biotic (host, vegetative biodiversity) stresses significantly impact the insects and their population dynamics. Specifically, at low temperature, a high mortality has been observed as well as an altered developmental rate (*27*). From this, a low incidence for altitude A was expected. Based on results, observed (Figure 2), this trend appears to be truth as only 5% of the maize samples (1/20) for altitude A chain 2 had flour moths, while its chain 1 counterpart had no insect presence (0/12).

In addition, altitude C (lowest altitude) of chain 1 had the greatest amount of insects with about 20% (5/24) incidence of flour moth, 8.3% (2/24) of flour beetle and 8.3% (2/24) of maize weevil. Interestingly, altitude C of chain 2 had no insect presence (0/9). Since chain 2 represents a commercial chain, this may indicate a fair initial quality regarding insects of purchased maize, or that storage conditions were properly secluded from the environment. In general, insect attack occurred predominantly for chain 1 farms, where the maize was exposed to the elements (field, drying). Figure 2 shows the incidence of insects (presence/absence of insects), however it must be mentioned that the highest counts of insects were also found in chain 1. For instance, flour moth had a maximum count of 1 for chain 2 altitude A, while chain 1 altitude B had a maximum count of 143 (data not shown), per occurrence.

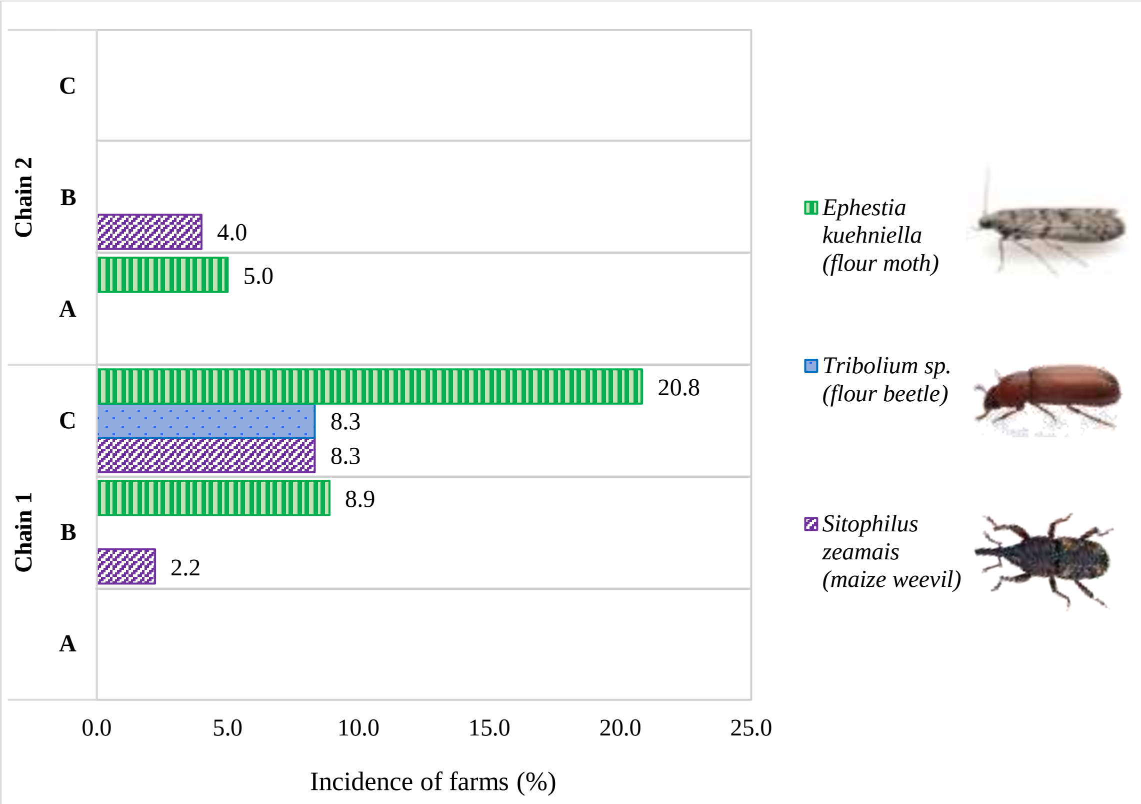


FIGURE 2. Incidence of insect infestation on maize from farms in Chiantla and Todos Santos, Huehuetenango, Guatemala. Maize season 2014-2015. Each group of samples per altitude was taken as 100%. Images credits: (*1*, *4*, *49*)

Under high humidity, grain moisture content equilibrates with the environment leading to microbial growth and contamination. Equilibrium moisture content and environmental temperature dictate the extent of such contamination (*13*). Mold count was performed in all collected samples to evaluate the fungal load, and with this a possible indication of mycotoxin contamination. Figure 3 reflects the influence of the high moisture content findings (see Figure 1) since both chain 1 and chain 2 farms have an approximate average fungal load of 5 log CFU/g at harvest and throughout storage. During the counting process, the fungal community observed was quite diverse between time points so further analysis is necessary to evaluate a potential population change throughout the studied period (manuscript in preparation).

Regarding mycotoxin contamination, the target fungal toxins evaluated in this study were aflatoxin and fumonisin as previous work (*6*, *18*, *37*, *53*, *59*, *60*) dealing with maize has reported these two toxins consistently. Environment has an impact on mycotoxin contamination partly by direct effects on causative fungi; as it changes, so do the complex communities of mycotoxin-producing fungi (*13*). The three different altitudes' relative humidity measurements were compared using an ANOVA test with the objective of evaluating any significant differences among the means of the environmental data. After confirming that the ANOVA was significant at $p<2e^{-16}$ (data not shown), pairwise comparisons using *t* tests with pooled SD were done. Bonferroni was used as *p value* adjustment method to evaluate significant differences. Relative humidity between altitudes A (62.0%), B (71.1%) and C (68.8%) was significantly different at $p<0.05$ (data not shown).

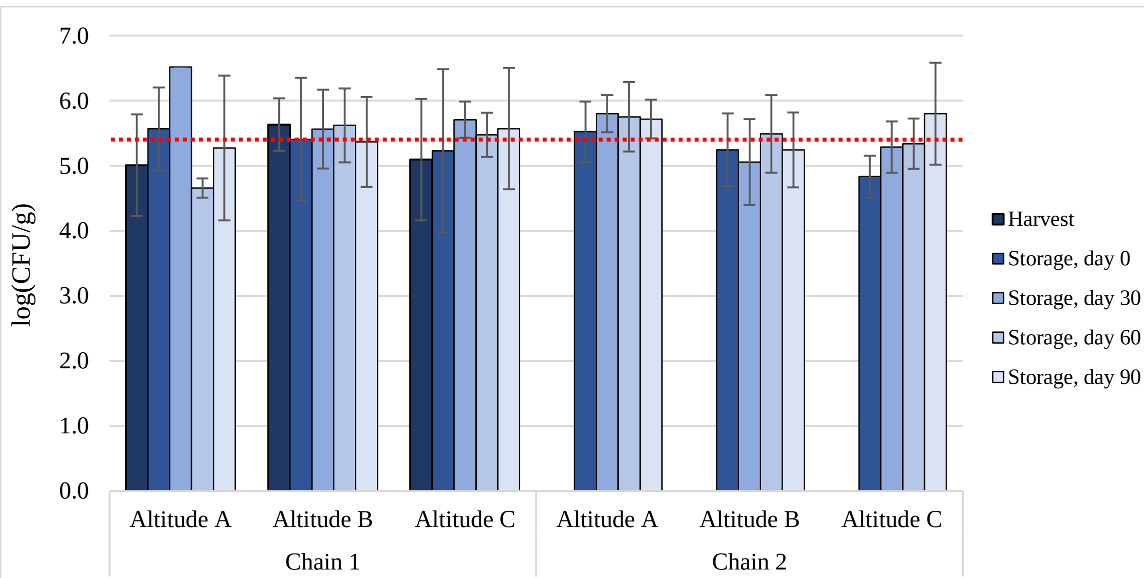


FIGURE 3. Average mold infestation in maize from Chiantla and Todos Santos, 2014-2015 maize season. Data shown as logarithmic CFU per gram of maize. Dotted line represents an average across chains and altitudes.

It can be seen in Figure 4 that for chain 1 farmers, field conditions allowed a considerable development of aflatoxin-producing fungi as the toxin was detected upon harvest for 84% of chain 1 farms ranging from 1.8 to 17.9 ppb (individual farm-data not shown). Although *Fusarium* species are predominantly considered as field fungi, it has

been reported that fumonisin production can also occur post-harvest when storage conditions are permissive (*11*). Data presented here supports this because even though no fumonisin was detected at harvest for any chain 1 farm, the toxin was detected during storage at a range from 1 to 4.7 ppm. Conversely, for buyers (chain 2) both mycotoxins were found at all sampling points with aflatoxins ranging from 2 to 85 ppb, and fumonisins ranging from 0.4 to 31 ppm. In 2005 Torres et al (*60*) reported similar values for fumonisin from maize bought in Huehuetenango local markets. Inhabitants of this region purchase their maize from departments in the lowland region (<600masl) of the country or Mexico (*21*, *60*), where warmer climate (correlated with low altitude), leads to more severe problems with fumonisin (*9*).

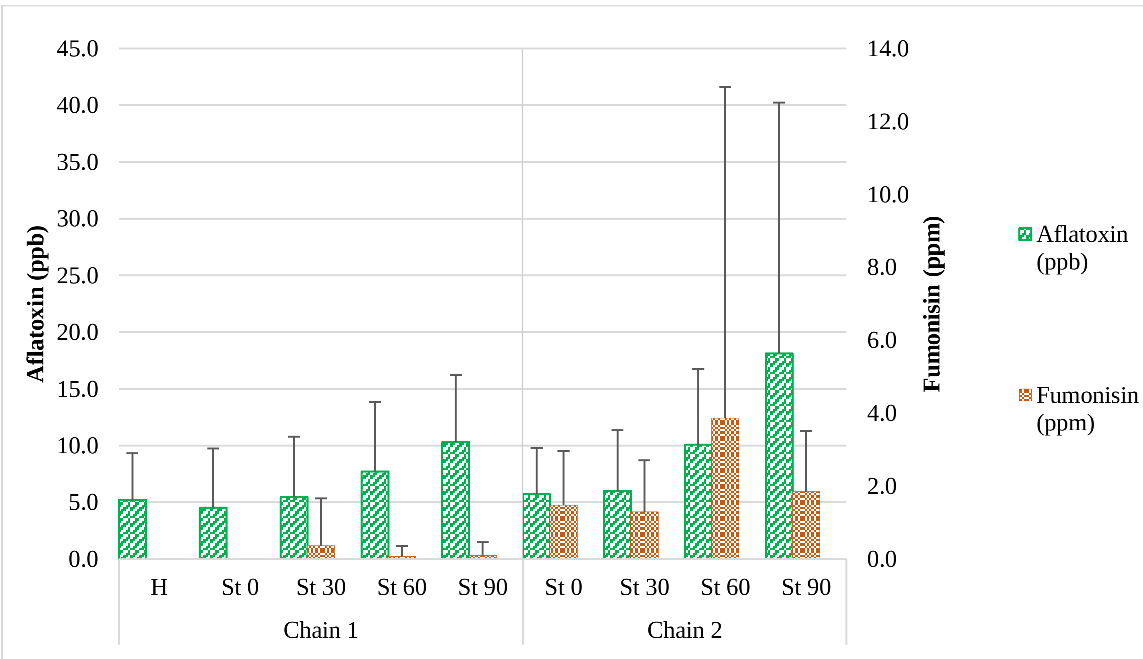


FIGURE 4. Overall mycotoxin levels during harvest and different storage days in Chiantla and Todos Santos, 2014-2015 maize season. H = Harvest, St = Storage at the designated day.

Overall, fumonisin was detected in 52% of the farms on purchased maize whereas aflatoxin was in 100% of the samples. Deviations that lead to the lack of normality of the mycotoxin data were most likely due to sampling. Samples were gathered randomly from the stored grain, and hot spots are a well-recognized issue associated with mycotoxin surveying (*32*). On account of this, a Recursive Partitioning Model (RPM) was used for

data analysis. RPM revealed that farmers who buy maize (chain 2) are at higher risk of fumonisin contamination. Aflatoxin, present for both producers and buyers, is influenced here possibly by the relative humidity (specifically >74.5%) of the storage location.

Furthermore, it can be seen in Figures 5 and 6 that all farmers where either fumonisin or aflatoxin was present had mean+SD daily ingestion levels above the recommended levels established either by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) or other studies (*34*, *65*). An average weight of 70 kg men and 60 kg for women (*54*) was used for these calculations. Also, it is estimated (*7*) that in Guatemala the consumption of maize is approximately 600 g and 400 g per day for men and women, respectively, which was used for calculation. Nevertheless this amount is usually higher. A previous study in this region (see Chapter 2) revealed that more than 40% of farms from Chiantla and Todos Santos consume more than 600 g of maize per person on a daily basis, indicating that inhabitants of this region may be at an even greater risk than shown in Figures 5 and 6. Additionally, co-occurrence of mycotoxins may exacerbate the harmful effects (*9*, *61*) of mycotoxin exposure.

Although there is no official Provisional Maximum Tolerable Daily Intake (PMTDI) for aflatoxin (*62*), previous studies (*34*, *65*) have recognized recommended levels. For those readings not considerably higher than the suggested level (0.001 μg Aflatoxin/kg bw*day) it must be remarked that the evaluated families are of scarce economic means and do not have a diverse diet, making them rely mostly on contaminated maize consumption and thus resulting in a continuous consumption (*64*) of the secondary metabolites. This is in addition to their culture which condemns the waste of grain (see Chapter 2), regardless of its condition, increasing the use of contaminated grain. The highest values for daily ingestion found for aflatoxin, the most potent carcinogenic mycotoxin known to date (*47*), are 280 times the recommended level of 0.001μg / kg bw / day (*34*) for men and 150 times for women. Regarding women specifically, aflatoxin is known to cross the placenta (*64*), thus affecting embryos and so the problem transcends generations. And when born, infants will likely be exposed to Aflatoxin M1 via breast-milk (*40*, *55*) as a direct result of the ingestion of contaminated maize.

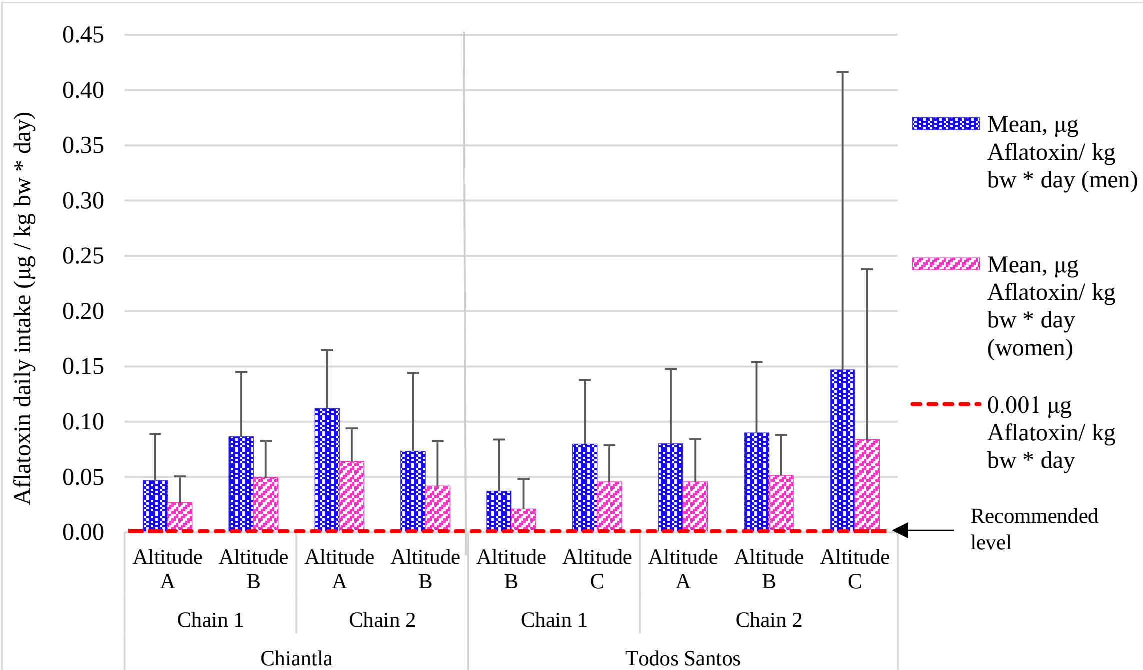


FIGURE 5. Aflatoxin average daily ingestion in Chiantla and Todos Santos, 2014-2015 maize season. Dotted line represents the recommended level of maximum exposure of aflatoxin for humans.

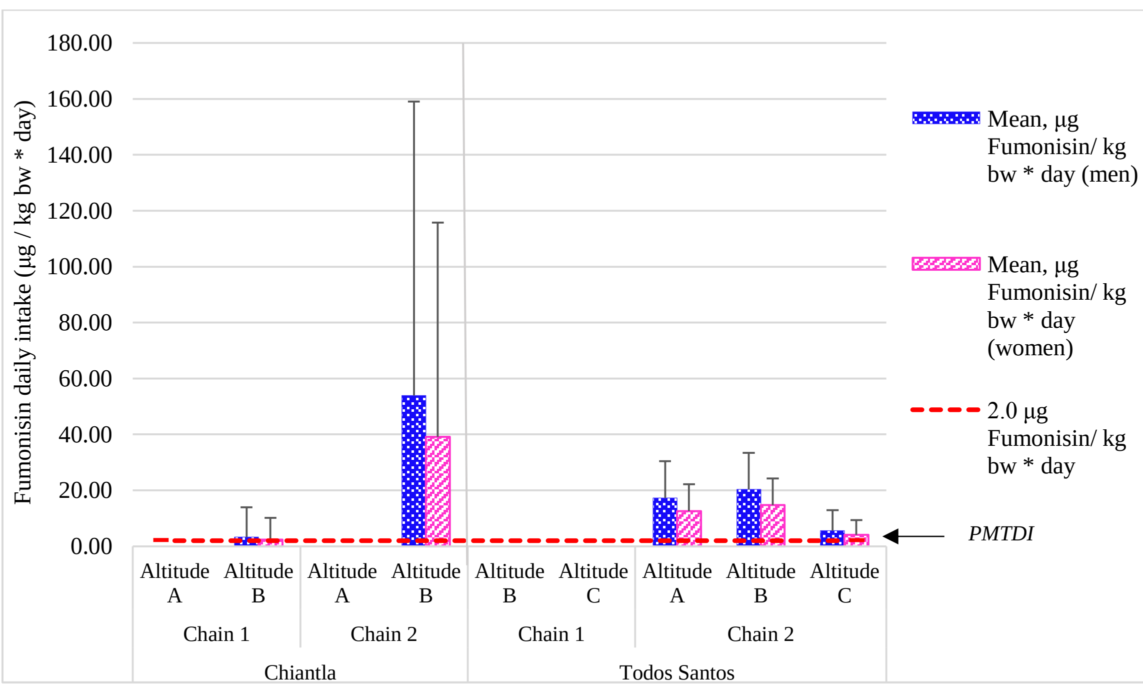


FIGURE 6. Fumonisin average daily ingestion in Chiantla and Todos Santos, 2014-2015 maize season. Dotted line represents the Provisional Maximum Tolerable Daily Intake (PMTDI) of fumonisin for humans.

Likewise, for those consuming fumonisin-contaminated maize, the highest calculated daily ingestion values are around 52 times the PMTDI of 2.00 µg / kg bw / day (*63*) for men and 38 times for women. Torres et al (*60*) found fumonisin was prevalent in the lowland region of Guatemala, and to a lesser degree in the highlands. Our data supports those findings, and indicates not only a low incidence of fumonisin, but also an apparent dominance for aflatoxin-producing fungi in the highlands of the country. As an example, the highest reported reading for fumonisin of 31 ppm was a chain 2 farm (i.e. maize buyers) from Chiantla.

Maize tortillas, the most widely consumed product of the country (*53*), involve heating the ground nixtamalized (lime-treated) maize. Such thermal treatment would degrade bacteria and fungi but some mycotoxins, including aflatoxin, have been shown to be resistant (*26*, *37*) and therefore would still be present after food is cooked. Specifically for fumonisin, alkaline cooking releases matrix-associated fumonisins, forming hydrolyzed forms of fumonisins solubilized into the *nejayote*, being thus a potential method for reducing this mycotoxin content from the diet (*15*) provided the *nejayote* is discarded. Both mycotoxins were detected in 52% of the evaluated samples hence synergistic effects (*57*) between fumonisin and aflatoxin may well further increase the health hazards for habitants of the region.

Malnutrition in Guatemala, the highest in Latin America and among the highest in the world, is mostly protein-caloric (*10*, *20*). This increases the risk of death and impairs cognitive development in children, affecting their future productivity and incomes (*32*, *36*). The World Bank estimated (*20*, *25*) that only one-third of surveyed infants from Guatemala received an adequate intake of protein, while 12% did not even reach 50% of the recommended requirement. The quality of diets in low-income populations in developing countries is compromised by limited access to high-cost, nutrient-rich foods, especially animal source foods, and relies instead mostly on grains like maize. Moreover, it has been reported that there is a correlation between stunting in infants with exposure to aflatoxin and fumonisin (*22*, *28*). Of the latter, there is additionally a correlation with the inhibition of ceramide synthase disrupting sphingolipid formation promoting

conditions such as Alzheimers (*48*), and neural tube defects (NTD) (*32*, *39*, *52*). Regarding NTD, in Guatemala the prevalence of this condition is approximately of 2.34 per 1000 live births, representing the most common congenital irregularity (*35*).

Maize kernels consist essentially of the embryo and endosperm, embedded in the pericarp, which is part of the ovary. Generally, the grain contains approximately 75% carbohydrate, 10% protein, 5% of lipids and 10% water (*20*). In rural areas of Guatemala maize tortillas, the most popular maize product in the country, account for up to 64-80% of the total caloric intake and 70-75% of the protein intake (*8*). Even though previous studies have shown that there is no correlation between mycotoxin production and protein, oil, starch and total fiber (*46*) in grains, fungi growing on stored grains can reduce the amount of carbohydrate, protein and total oil content. It can also increase the moisture content and free fatty acids, enhancing biochemical reactions (*3*) that lead to degrading the final quality of the commodity prior to consumption. Field observations during sampling supported this as it was seen that the households had a high consumption of poor quality maize which may well be translated into low daily protein ingestion.

There are several native and commercial maize varieties grown in Guatemala. Depending on the environmental, cultural, and genetic parameters they can vary in color, quantity, weight, and nutrient composition (*45*). Several are these were evaluated (see Figure 7) in this study representing the maize consumed in the area of interest.

From all samples collected, based on visual characteristics of the maize, 13 groups were defined. For each group, because variety names are generally related to biological characteristics (*17*)(Chapter 1, 2), to simplify this evaluation, samples were grouped based on a combination of phenotypic traits, namely shape (flint or dent) and color (red, black, yellow, white). A proximate analysis was carried with the purpose of better understanding the nutritional composition of the varieties. Table 1 includes the results obtained for the proximate analysis for every maize variety found in the townships of Chiantla and Todos Santos.

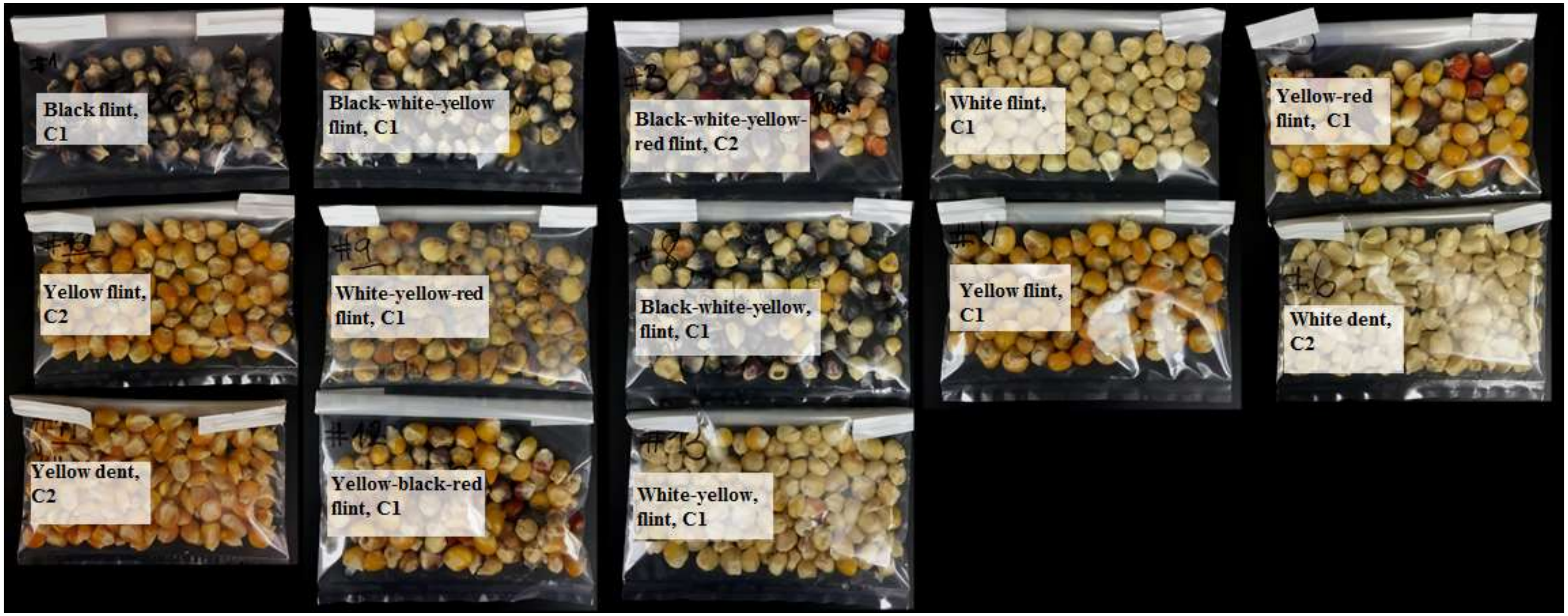


FIGURE 7. Guatemalan native (*criollo*) and commercial maize varieties consumed by inhabitants of Chiantla and Todos Santos, Huehuetenango. These varieties represent the farms included in this study and it does not mean that these are the only ones in the region of study.

TABLE 1. Proximate analysis (wt%, dry basis) for native and commercial maize found in Chiantla and Todos Santos, Huehuetenango, Guatemala. Unless otherwise noted varieties belong to Chain 1 (i.e. maize producers). Data showed is the mean of three replications ± standard deviation

Maize variety	Shape	Ash			Protein			Lipids			Carbohydrates		
Black	Flint	1.21	±	0.03	7.01	±	0.08	2.57	±	0.13	88.13	±	0.22
White	Flint	1.16	±	0.09	8.71	±	0.40	3.25	±	0.16	85.76	±	0.62
White C2	Dent	1.06	±	0.05	7.19	±	0.16	3.57	±	0.15	87.02	±	0.34
Yellow	Flint	1.23	±	0.03	9.13	±	0.36	4.36	±	0.20	84.16	±	0.58
Yellow C2	Flint	1.12	±	0.10	8.88	±	0.16	3.73	±	0.14	85.12	±	0.38
Yellow C2	Dent	1.15	±	0.03	9.21	±	0.39	3.22	±	0.25	85.26	±	0.62
Yellow Red	Flint	1.07	±	0.04	8.07	±	0.21	3.25	±	0.28	86.47	±	0.53
White Yellow	Flint	1.24	±	0.05	8.94	±	0.15	3.49	±	0.26	85.23	±	0.44
Yellow Black Red	Flint	1.30	±	0.03	8.11	±	0.62	3.39	±	0.08	86.12	±	0.69
White Yellow Red	Flint	1.28	±	0.09	5.23	±	0.23	1.77	±	0.12	90.90	±	0.40
White Black Yellow	Flint	1.43	±	0.09	7.30	±	0.34	3.54	±	0.19	86.65	±	0.60
Black White Yellow	Flint	1.26	±	0.12	6.98	±	0.32	2.95	±	0.24	87.69	±	0.66
Black White Yellow Red	Flint	1.21	±	0.11	7.36	±	0.39	3.09	±	0.26	87.25	±	0.73

Ash, protein, lipid and carbohydrate fractions differed significantly between varieties. In general, it can be seen in Table 1 that all maize varieties had a proximate composition where, based on percentage weight, it was higher in carbohydrate, followed by protein, lipid, moisture and ash. The higher proportion of carbohydrates is due to the starch content of the grain, which is not considered to contribute greatly to the nutritional profile but is more a functional attribute. For those varieties with a higher carbohydrate content, it was observed that the lipid and, in higher degree, the protein content were reduced, while ash and moisture remain rather constant. No specific difference was found due to the shape of the kernel, however only two dent varieties were included, compared with eleven flint varieties. Figure 8 shows the *p*-values when the proximate composition of the different maize groups were compared using pairwise comparisons ANOVA. It can be seen that the White-Yellow-Red flint variety from chain 1 showed the most significant difference among all 13 groups evaluated followed by Black flint from chain 1and Yellow dent from chain 2. More importantly, a higher protein content may be indicative of a more nutritious variety. In that regard, the proximate analysis revealed a significant higher protein content for white (8.7%), yellow (9.1%) and white-yellow (8.9%) flint varieties from maize producers (chain 1) as well as for yellow dent (9.2%) and flint (8.9%) varieties from maize buyers (chain 2). Additional analysis for protein quality as well as mineral analysis would be needed to further support these findings and definitively describe these varieties as higher in nutritional value. Nonetheless, it must be remarked that other protein sources (i.e. animal protein) are necessary for a balanced protein intake as the overall protein quality of maize is insufficient (*45*).

In conclusion, farmers of the highlands of Guatemala need to improve their agricultural practices in order to have safe and wholesome maize and maize-based products. The use of fertilizers in the field is recommended. Plants that have nutrient availability during their growth stage are generally stronger, healthier and better able to compensate for pest/fungal damage than those under nutritional deficiencies (i.e. stress) (*42*).

	Black	Black White Yellow	Black White Yellow Red	White	White Black Yellow	White Yellow	White Yellow Red	White, dent, C2	Yellow	Yellow Black Red	Yellow Red	Yellow, C2	Yellow, dent, C2	Yellow, dent, C2	Yellow, C2	Yellow Red	Yellow Black Red	Yellow	White, dent, C2	White Yellow Red	White Yellow	White Black Yellow	White	Black White Yellow Red	Black White Yellow	Black
													Lipid	Ash												
Black		0.52	0.12	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.02	1.00	1.00	0.90	0.82	0.96	0.95	0.03	1.00	0.00	1.00	1.00	0.66	
Black White Yellow	1.00		1.00	0.79	0.05	0.09	0.00	0.03	0.00	0.29	0.79	0.00	0.89	0.91	0.59	0.04	1.00	1.00	0.06	0.00	1.00	0.37	0.59	0.87		1.00
Black White Yellow Red	0.98	0.96		1.00	0.29	0.43	0.00	0.19	0.00	0.81	1.00	0.02	1.00	1.00	1.00	0.72	0.96	1.00	0.80	0.01	1.00	0.01	1.00		1.00	0.74
White	0.00	0.00	0.00		0.86	0.95	0.00	0.74	0.00	1.00	1.00	0.20	1.00	1.00	1.00	0.94	0.76	0.93	0.97	0.04	0.99	0.00		0.09	0.01	0.00
White Black Yellow	1.00	0.99	1.00	0.00		1.00	0.00	1.00	0.00	1.00	0.86	0.99	0.75	0.01	0.00	0.00	0.24	0.11	0.00	0.00	0.05		0.71	0.97	0.53	0.10
White Yellow	0.00	0.00	0.00	1.00	0.00		0.00	1.00	0.00	1.00	0.95	0.95	0.88	1.00	0.99	0.30	1.00	1.00	0.38	0.00		0.12	0.99	0.01	0.00	0.00
White Yellow Red	0.00	0.00	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.04	0.59	0.00	0.00	0.49		0.00	0.00	0.00	0.00	0.00	0.00
White, dent, C2	1.00	1.00	1.00	0.00	1.00	0.00	0.00		0.00	0.99	0.75	1.00	0.61	0.74	0.97	1.00	0.10	0.21		0.00	0.02	1.00	0.24	1.00	0.94	0.42
Yellow	0.00	0.00	0.00	0.92	0.00	1.00	0.00	0.00		0.00	0.00	0.03	0.00	1.00	0.93	0.16	1.00		0.00	0.00	0.47	0.00	0.05	0.00	0.00	0.00
Yellow Black Red	0.02	0.01	0.24	0.57	0.16	0.15	0.00	0.07	0.03		1.00	0.66	1.00	0.98	0.76	0.07		0.01	0.72	0.00	0.72	0.99	1.00	0.40	0.07	0.01
Yellow Red	0.02	0.02	0.32	0.47	0.22	0.11	0.00	0.10	0.02	1.00		0.20	1.00	0.65	0.94		1.00	0.00	0.99	0.00	0.27	1.00	0.91	0.85	0.29	0.04
Yellow, C2	0.00	0.00	0.00	1.00	0.00	1.00	0.00	0.00	1.00	0.23	0.17		0.13	1.00		0.17	0.56	0.63	0.01	0.00	1.00	0.07	0.96	0.00	0.00	0.00
Yellow, dent, C2	0.00	0.00	0.00	0.78	0.00	1.00	0.00	0.00	1.00	0.01	0.01	0.98			1.00	0.30	0.76	0.43	0.02	0.00	1.00	0.14	0.99	0.01	0.00	0.00
Protein																										Carbohydrate

FIGURE 8. *p*-values for native (*criollo*) and commercial maize proximate analysis corresponding to protein, lipid, ash and carbohydrate content. Highlighted cells indicate a significant ($p \leq 0.05$) difference. Unless otherwise noted, maize varieties belong to Chain 1 (i.e. maize producers) and are flint-shaped.

Proper drying to moisture content below 14%, along with broken kernel and debris removal (*23*) prior to storage could reduce the opportunity for fungal infestation; while controlled storage units (e.g. metal silos) would prevent pest interaction with the crop. To better control insect infestations, a plausible solution for this problem could be the use of pesticides. However in Guatemala, as in other third world countries, the excessive use of pesticides is alarming. Because of pesticide residues, farmers with limited knowledge of pest control are at danger of pesticide poisoning (*24, 41*), thus proper dosage must be used. Additionally, the seed should be protected by pesticide application or other means prior to planting (*20*), and aeration should be used during grain storage (*16*), although these alternatives may not be attainable in the region of study due to the lack of financial means. Moreover, any delay in the harvest increases the possibility of post-harvest damage due to insect infestation and fungi decreasing grain quality, consequently farmers should perform the harvesting process in a timely manner.

REFERENCES

1. Andermatt Biocontrol AG. 2016. Ephestia kuehniella.
2. AOAC International. 2005. Official Methods of Analysis of AOAC international, 18th ed. Maryland.
3. Bhattacharya, K., and S. Raha. 2003. Deteriorative changes of maize, groundnut and soybean seeds by fungi in storage. *Mycopathologia* 155:135–141.
4. BioNET-EAFRINET. 2011. Sitophilus zeamais Motschulsky, 1855 - Maize Weevil.
5. Boxall, R. A. 2002. Damage and loss caused by the Larger Grain Borer Prostephanus truncatus. *Integr. Pest Manag. Rev.* 7:105–121.
6. Bravo Martinez, J. 2009. Metal silos and food security. Lessons Learned from a Successful Central American Post-harvest program. Quito, Ecuador.
7. Bressani, R. 1990. Chemistry, technology and nutritive value of maize tortillas. *Food Rev. Int.* 6:225–264.
8. Bressani, R., G. Arroyave, and N. Scrimshaw. 1953. The nutritive value of central

american corns. I. Nitrogen, ether extract, crude fiber, and minerals of twenty-four varieties in Guatemala. *J. Food Sci.* 18:261–267.

9. Bryła, M., M. Roszko, K. Szymczyk, R. Jędrzejczak, M. W. Obiedziński, and J. Sękul. 2013. Fumonisins in plant-origin food and fodder-a review. *Food Addit. Contam.* 30:1626–1640.
10. Campos, M. de, and A. E. Olszyna-Marzys. 1979. Mycotoxin contamination in grains and grain products during the dry season in Guatemala. *Bull. Environ. Contam. Toxicol.* 22:350–356.
11. Chulze, S. N. 2010. Strategies to reduce mycotoxin levels in maize during storage: a review. *Food Addit. Contam. Part A. Chem. Anal. Control. Expo. Risk Assess.* 27:651–7.
12. Cornell University. 2015. Aflatoxins: Occurrence and Health Risks.
13. Cotty, P. J., and R. Jaime-Garcia. 2007. Influences of climate on aflatoxin producing fungi and aflatoxin contamination. *Int. J. Food Microbiol.* 119:109–115.
14. de Campos, M., J. C. Santos, and A. E. Olszyna-Marzys. 1980. Aflatoxin contamination in grains from the Pacific coast in Guatemala and the effect of storage upon contamination. *Bull. Environ. Contam. Toxicol.* 24:789–795.
15. De Girolamo, A., V. M. T. Lattanzio, R. Schena, A. Visconti, and M. Pascale. 2016. Effect of alkaline cooking of maize on the content of fumonisins B1 and B2 and their hydrolysed forms. *Food Chem.* 192:1083–1089.
16. Delcour, J., and C. Hoseney. 2013. Principles of cereal science and technology, 3rd ed. American Association of Cereal Chemists, Inc.
17. Etten, J. van. 2006. Changes in farmers' knowledge of maize diversity in highland Guatemala, 1927/37-2004. *J. Ethnobiol. Ethnomed.* 2.
18. Etten, J. Van, and R. M. Fuentes. 2004. La Crisis Del Maíz En Guatemala: Las importaciones de maiz y la agricultura familiar. *Anu. Estud. Centroam.* 30:51–66.
19. Food and Agriculture Organization of the United Nations. 2003. Food energy – methods of analysis and conversion 93.
20. Fuentes, R. M. 2002. El cultivo del maiz en Guatemala. Una guía para su manejo agronómico.
21. Galemba, R. 2012. Corn is food, not contraband. *Am. Ethnol.* 39:716–734.

22. Gallant, A. Mycotoxins Linked to Malnutrition in Developing Countries. *Glob. Food Saf. Resour.*

23. Guo, B. Z., N. W. Widstrom, T. E. Cleveland, and R. E. Lynch. 2000. Control of preharvest aflatoxin contamination in corn: fungus-plant-insect interactions and control strategies. *Recent Res. Dev. Agric. Food Chem.*

24. Hirst Sole, L. A. 1994. Algunos aspectos físicos y de procesamiento del maíz en grano y mazorca, con y sin tusa, almacenado en dos tipos de estructuras, con y sin tratamiento químico. Universidad del Valle de Guatemala. Ingeniería en Ciencias de Alimentos.

25. Jones, K. M., M. Ramirez-Zea, C. Zuleta, and L. H. Allen. 2007. Prevalent vitamin B-12 deficiency in twelve-month-old Guatemalan infants is predicted by maternal B-12 deficiency and infant diet. *J. Nutr.* 137:1307–13.

26. Kaushik, G. 2015. Effect of Processing on Mycotoxin Content in Grains. *Mycotoxins Fruits Veg.* 55:1672–1683.

27. Khaliq, A., M. Javed, M. Sagheer, M. Sohail, M. Sohail, and M. Sagheer. 2014. Environmental effects on insects and their population dynamics. *J. Entomol. Zool. Stud. JEZS* 1:1–7.

28. Khlangwiset, P., G. S. Shephard, and F. Wu. 2011. Aflatoxins and growth impairment: A review. *Crit. Rev. Toxicol.* 41:740–755.

29. Koenning, S., and G. Payne. 2000. Mycotoxins in Corn.

30. Krause, V. M., K. L. Tucker, H. V. Kuhnlein, C. Y. Lopez-Palacios, M. Ruz, and N. W. Solomons. 1992. Rural- urban variation in limed maize use and tortilla consumption by women in Guatemala. *Ecol. Food Nutr.* 28:279–288.

31. Lacey, J., A. Hamer, and N. Magan. 1994. Respiration and losses in stored wheat under different environmental conditions. *Proceeding 6th Int. Work. Conf. Stored-Product Prot.* 2:100.

32. Lee, H. J., and D. Ryu. 2015. Advances in Mycotoxin Research: Public Health Perspectives. *J. Food Sci.* 80:T2970–T2983.

33. Leslie, J. F., and A. F. Logrieco. 2014. Mycotoxin Reduction in Grain Chains, 1st ed. John Wiley & Sons, Inc., Manhattan, Kansas.

34. Magrine, I. C. O., S. S. C. Ferrari, G. F. Souza, L. Minamihara, C. Kemmelmeier,

E. Bando, and M. Machinski. 2011. Intake of aflatoxins through the consumption of peanut products in Brazil. *Food Addit. Contam. Part B* 4:99–105.

35. Manucci, G., and E. von Quednow. 2014. Como Lo Hago Yo: Anomalías del Tubo Neural en Guatemala — Mielomeningocele Unidad de Espina Bífida e Hidrocefalia. *Surg Neurol Int.* 5:S12–S22.

36. Marini, A., and M. Gragnolati. 2003. Malnutrition and Poverty in Guatemala. *World Bank* 1–54.

37. Martinez, E., R. Gomez, and R. Bressani. 1970. Prevalencia de hongos en granos de maiz (Zea mays, L.) de Guatemala. *Turrialba* 20:311–319.

38. Meredith, A. A., R. Olga, S. Saenzde, and T. Ronald. 1999. Fumonisin B1 and Hydrolyzed Fumonisin B1 (AP1) in Tortillas and Nixtamalized Corn (Zea mays L .) from Two Different Geographic Locations in Guatemala. *J. Food Prot.* 62:1218–1222.

39. Missmer, S. A., L. Suarez, M. Felkner, E. Wang, A. H. Merrill, K. J. Rothman, and K. A. Hendricks. 2006. Exposure to fumonisins and the occurence of neutral tube defects along the Texas-Mexico border. *Environ. Health Perspect.* 114:237–241.

40. Mohammadi, H. 2001. A Review of Aflatoxin M 1 , Milk and Milk products. *Aflatoxins - Biochem. Mol. Biol.* 1:397– 414.

41. Morales, H., and I. Perfecto. 2000. Traditional Knowledge and Pest Management in the Guatemalan Highlands. *Agric. Human Values* 17:49–63.

42. Morales, H., I. Perfecto, and B. Ferguson. 2001. Traditional fertilization and its effect on corn insect populations in the Guatemalan highlands. *Agric. Ecosyst. Environ.* 84:145–155.

43. Nash, M. J. 1985. Crop conservation and storageSecond. Pergamon Press, England.

44. Nielsen, L. K., D. J. Cook, S. G. Edwards, and R. V. Ray. 2014. The prevalence and impact of Fusarium head blight pathogens and mycotoxins on malting barley quality in UK. *Int. J. Food Microbiol.* Elsevier B.V. 179:38–49.

45. Nuss, E. T., and S. A. Tanumihardjo. 2010. Maize: A Paramount Staple Crop in the Context of Global Nutrition. *Compr. Rev. Food Sci. Food Saf.* 9:417–436.

46. Ono, E. Y. S., L. Biazon, M. Da Silva, É. Vizoni, Y. Sugiura, Y. Ueno, and E. Y.

Hirooka. 2006. Fumonisins in corn: Correlation with Fusarium sp. count, damaged kernels, protein and lipid content. *Brazilian Arch. Biol. Technol.* 49:63–71.

47. Perrone, G., A. Susca, G. Cozzi, K. Ehrlich, J. Varga, J. C. Frisvad, M. Meijer, P. Noonim, W. Mahakarnchanakul, and R. A. Samson. 2007. Biodiversity of Aspergillus species in some important agricultural products. *Stud. Mycol.* 59:53–66.
48. Pralhada Rao, R., N. Vaidyanathan, M. Rengasamy, A. Mammen Oommen, N. Somaiya, and M. R. Jagannath. 2013. Sphingolipid metabolic pathway: an overview of major roles played in human diseases. *J. Lipids* 2013:178910.
49. Rentokil. 2016. Common Stored Product Insect Species.
50. Rheeder, J. P., W. F. O. Marasas, and H. F. Vismer. 2002. Production of fumonisin analogs by Fusarium species. *Appl. Environ. Microbiol.* 68:2101–2105.
51. Richard, E., N. Heutte, V. Bouchart, and D. Garon. 2009. Evaluation of fungal contamination and mycotoxin production in maize silage. *Anim. Feed Sci. Technol.* 148:309–320.
52. Riley, R. T., O. Torres, J. Matute, S. G. Gregory, A. E. Ashley-Koch, J. L. Showker, T. Mitchell, K. A. Voss, J. R. Maddox, and J. B. Gelineau-van Waes. 2015. Evidence for fumonisin inhibition of ceramide synthase in humans consuming maize-based foods and living in high exposure communities in Guatemala. *Mol. Nutr. Food Res.* 1:2209–2224.
53. Roberto, M., and F. López. 2002. Una guía para su manejo agronómico del maíz. Guatemala.
54. Russell, M. 1976. Parent-child and sibling-sibling correlations of height and weight in a rural population of preschool children. *Hum. Biol.* 48:501–515.
55. Sadeghi, N., M. R. Oveisi, B. Jannat, M. Hajimahmoodi, H. Bonyani, and F. Jannat. 2009. Incidence of aflatoxin M1 in human breast milk in Tehran, Iran. *Food Control* 20:75–78.
56. Sauer, D. B., C. L. Storey, O. Ecker, and D. W. Fulk. 1982. Fungi in U.S. export wheat and corn. *Postharvest Pathol. Mycotoxins* 72:1449–1452.
57. Streit, E., K. Naehrer, I. Rodrigues, and G. Schatzmayr. 2013. Mycotoxin occurrence in feed and feed raw materials worldwide: long-term analysis with

special focus on Europe and Asia. *J. Sci. Food Agric.* 93:2892–9.

58. Sweets, L. 1996. Stored Grain Fungi. *Agric. Electron. Bull. Board*.
59. Torres, O., J. Matute, J. Gelineau-van Waes, J. R. Maddox, S. G. Gregory, A. E. Ashley-Koch, J. L. Showker, K. A. Voss, and R. T. Riley. 2015. Human health implications from co-exposure to aflatoxins and fumonisins in maize-based foods in Latin America: Guatemala as a case study. *World Mycotoxin J.* 8:143–159.
60. Torres, O. a, E. Palencia, L. Lopez de Pratdesaba, R. Grajeda, M. Fuentes, M. C. Speer, A. H. Merrill, K. O'Donnell, C. W. Bacon, A. E. Glenn, and R. T. Riley. 2007. Estimated fumonisin exposure in Guatemala is greatest in consumers of lowland maize. *J. Nutr.* 137:2723–2729.
61. Waes, J. G. van. Mycotoxins and birth defects. *Creight. Univ.*
62. WHO. 2015. Evaluations of the Joint FAO/WHO Expert Committee on Food Additives (JECFA). Aflatoxin. 5639.
63. WHO. 2015. Evaluations of the Joint FAO/WHO Expert Committee on Food Additives (JECFA). Fumonisin. 2–3.
64. Wild, C. P., and Y. Y. Gong. 2009. Mycotoxins and human disease: A largely ignored global health issue. *Carcinogenesis* 31:71–82.
65. Yazdanpanah, H., A. Zarghi, A. R. Shafaati, S. M. Foroutan, F. Aboul-Fathi, A. Khoddam, F. Nazari, and F. Shaki. 2013. Analysis of Aflatoxin B1 in Iranian Foods Using HPLC and a Monolithic Column and Estimation of its Dietary Intake. *Iran. J. Pharm. Res.* 12:81–87.

UNDERSTANDING THE MYCOBIOTA OF MAIZE FROM THE HIGHLANDS OF GUATEMALA, AND IMPLICATIONS TO ITS QUALITY AND SAFETY *

R. Mendoza, H. Hallen-Adams, J. Stratton and A. Bianchini

Department of Food Science & Technology
University of Nebraska.
Lincoln, NE, USA

Corresponding author:

Andréia Bianchini, Department of Food Science & Technology
The Food Processing Center, University of Nebraska-Lincoln
Phone: (402)472-3144, Fax: (402)472-1693
abianchini2@unl.edu

*Scientific manuscript formatted to meet guidelines for the Journal of Crop Protection

ABSTRACT

Maize is considered a staple crop in Guatemala, having a major impact in the rural regions where it is consumed in high amounts. Given that traditional pre- and post-harvest practices lead to exposure to the environmental surroundings where pests and microorganisms may be present, maize quality and safety can be compromised severely. In order to assess the potential degree of risk, maize from six farms from Huehuetenango, Guatemala were evaluated based on their mycobiota. DNA was extracted from the maize samples, and ITS1F-TW13 amplicons were subjected to Illumina sequencing. This survey allowed the identification of 52 fungal taxa in the 90-day maize storage period. For the farms where the maize moisture content exceeded 20%, a high yeast content was observed which can reflect spoilage during storage. Findings showed a significant amount of *Fusarium* and *Aspergillus*, mycotoxin-producing molds, which could lead to mycotoxin contamination. This indicates a potential for compromising the health of the inhabitants of the region where maize represents a significant portion of the diet. Moreover, fungal endophytes *Stenocarpella maydis* and *Acremonium sp*. were also found in significant amounts across farms, indicating damage of the maize plant. Insect damage is another indicator of risk as it may result not only in entry points for fungi but insects can also act as vectors for such microorganisms. The fungi *Ophiostoma ips* and *Hannaella zeae* which have been related to insect infestation were found, and it is likely that these organisms are related to incidence of *Ephestia kuehniella* (flour moth), *Sitophilus zeamais* (maize weevil) and *Tribolium sp.* (flour beetle) present in the analyzed farms. Results from this study can help better understand the current health-risk scenario in the Highlands of Guatemala incurred by poor grain handling practices.

Keywords: Guatemala, maize, corn, fungal population

INTRODUCTION

Mycotoxicosis cases have increased during the past two decades in Latin America and worldwide, and the number of patients at risk has risen dramatically, specifically for those that are immunocompromised such as pregnant women, organ transplant recipients, HIV positive individuals (Romanelli et al., 2014; Sifuentes-Osornio et al., 2012), or people suffering from certain medical conditions such as hepatitis (Kew, 2003) where exposure to specific mycotoxins may have a synergistic effect. In tropical developing countries, such as Guatemala, environmental conditions coupled with poor grain handling practices are conducive to microbial growth, exposing inhabitants to staples that are often contaminated (Cotty and Jaime-Garcia, 2007). Moreover, a considerably larger maize consumption than average occurs in rural communities of Guatemala (Torres et al., 2007), where people have limited economic resources. Consequently even small levels of mycotoxin contamination could pose a health risk to this population. Mycotoxin effects are dose-dependent, producing a variety of symptoms in the consumer. Aflatoxins and fumonisins are recurrently implicated in mycotoxin contamination of maize. Health effects of aflatoxin poisoning, synthesized by some Aspergilli, include liver necrosis and tumors, reduced growth, and depressed immune response (Cornell University, 2015; Perrone et al., 2007; Wild and Gong, 2009). Fumonisin, a mycotoxin produced by *Fusarium moniliforme* and others, is correlated with esophageal cancer, stunting and other symptoms (Bryla et al., 2013).

Fungi are frequently encountered in agricultural products at different stages including pre-harvest, harvest, processing and handling (Perrone et al., 2007), thus there is a risk of microbial contamination in every step of the maize production chain. The contamination may lead to spoilage, possible development of mycotoxins and decrease in yield. Factors promoting mold proliferation and mycotoxin development include oxygen availability, heat, rain or insect damage (Richard et al., 2007). Among the changes due to spoilage, which would include those caused by *Aspergillus* species, are sensorial (discoloration), nutritional and qualitative (rot, off odors) damage to different commodities (Perrone et al., 2007). Yeasts colonize maize with high levels of moisture

(Glewen et al., 2013), and are often associated with quality issues. Overall, fungi can be considered food safety, quality, economic and security issue that affect communities, especially those heavily reliant on a single staple food.

Morphology (i.e. spores, hyphae, etc.) of cultures grown on defined media have been used for traditional fungal identification. However a significant proportion of microorganisms, including fungi, cannot be cultivated in axenic conditions. Moreover, such conditions of analysis are laborious and entail isolation and purification of each microbial species prior to their identification (Richard et al., 2009). In addition, molds may not always produce spores in culture and thus are not distinguishable by classic mycological methods (Romanelli et al., 2014).

Molecular-based approaches are becoming more commonly used for fungal identification because they allow for a more rapid and objective identification, and provide insight into microbial occurrence, relative abundance and microbial niches (Romanelli et al., 2014; Tedersoo et al., 2010). Nuclear ribosomal genes and markers, particularly the small subunit (SSU) are widely used for barcoding prokaryotes because of the simplicity of amplification, the occurrence of universal primer locations, and the alignability across phyla and domains. However, for fungi SSU is highly conserved thus providing little resolution to species level, sometimes confounding even different fungal genera. Consequently, the internal transcribed spacer (ITS) region has been used for fungal identification (Nilsson et al., 2006; Tedersoo et al., 2010). This is a variable region located between conserved genes encoding the 18S, 5.8S and 28S ribosomal subunits (Romanelli et al., 2014).

The aim of this research was to investigate the mycoflora diversity of maize from the Western-highlands of Guatemala during a 3-month period, from harvest through storage, in order to assess the safety and quality of this regional staple commodity utilizing a DNA-based identification approach. This would represent the first study describing the mycobiota of maize in this part of Guatemala.

MATERIALS AND METHODS

Sample selection. Hand-shelled maize samples from the 2014-2015 harvest season from six farms distributed in Todos Santos (n=3) and Chiantla (n=3), townships of Huehuetenango, Guatemala, were analyzed in this study. The farms were distributed among three different altitudes: Type C altitude from sea level to 1500 meters above sea level (masl), type B altitude between 1500 and 2700 masl, and type A altitude above 2700 masl. Sampling time points included harvest, as well as days 0, 30, 60 and 90 of storage. Farms where maize was grown were identified as "Chain 1" farms. Farms with insufficient land to plant maize where farmers had to rely on purchasing maize for consumption were designated "Chain 2" farms.

Preparation of maize samples. Prior to analysis, all samples were kept frozen at -20°C. In order to conduct a complete fungal profiling, both symptomless and visually contaminated kernels were included in the analysis. Samples were homogenized and a subsample of 45g was used for subsequent steps.

Preparation of template DNA. DNA extraction from maize was performed according to the procedure established by the European commission's Community Reference Laboratory for GM Food and Feed (CRL-GMFF). Forty-five grams of maize kernels were aseptically weighed and ground (Mr. Coffee grinder IDS77). Two grams of ground corn was transferred into a 50 mL centrifuge tube and 10 mL of extraction buffer (Directorate General-Joint Research Centre of the European Commission, 2007) pre-warmed to 65°C was added. After a 10 min incubation, supernatant was transferred to a clean 15 mL tube and an equal volume of chloroform:isoamyl alcohol (24:1, Sigma Cat. No. C0549-1PT) was added. The tube was slowly inverted 20 times for extraction, and was then centrifuged (Jouan BR4i) at 7000 x *g* for 10 min. The resulting upper aqueous phase was transferred to another 15 mL tube, and 10 μL of RNAse A (10mg/mL, Qiagen No. 19-101) were added followed by a 30 min incubation at 37°C. Approximately 10% of the extract volume of warm (55°C) 10% CTAB Buffer (Sigma Cat. No. H-6269) was added, mixed and followed by an addition of an equal volume of chloroform:isoamyl

alcohol. Sample was once again centrifuged at 7000 x *g* for 10 min and the upper aqueous phase was transferred to a 50 mL tube.

Three volumes of precipitation buffer (Directorate General-Joint Research Centre of the European Commission, 2007) were added, followed by a 10min waiting period. The sample was then centrifuged at 7000 x *g* for 15 min at room temperature. The liquid layer was discarded and the resulting pellet was washed two times with 4 mL 70% ethanol (Merck Cat. No. 1009831000), and centrifuged at 7000 x *g* for 5 min at room temperature. The ethanol was carefully removed by decanting to prevent the detachment of the washed pellet. After alcohol evaporation at room temperature, the DNA pellet was dissolved in 200 μL of Tris-Base and EDTA buffer (Directorate General-Joint Research Centre of the European Commission, 2007), and left overnight at 4°C. DNA sample was incubated at 65°C for 15 min followed by a spin at the maximum speed in bench top microfuge (Beckman Coulter microfuge 16) of 16160 x *g* for 5 min. To further clean the DNA template, the sample was placed into Pall Nanosep MF 0.2 μm column (Pall Corporation P/N ODM02C33) and spun at 14462 x *g* until all sample had passed through. The sample was then placed into a Pall Nanosep 30K omega column (Pall Corporation P/N OD030C33) and the purified DNA was collected.

Polymerase chain reaction (PCR). After DNA was extracted and purified, primers ITS1-F:TW13 (Anderson and Cairney, 2004; Bokulich and Mills, 2013; Bruns, 2002) (IDT, Integrated DNA Technology) along with a Taq PCR master mix kit (Qiagen 201443) were used to carry out the reaction using a thermal cycler (BIORAD T100). PCR reactions were performed in a volume of 50 μL per reaction, including the DNA template. Conditions included an initial denaturation at 94°C for 4 min, 40 amplification cycles at 94°C for 1 min, annealing at 50°C for 1 min, extension at 72°C for 3 min and final extension of 10 min at 72°C. At least every set of 10 DNA samples were run with 1 negative (no DNA) control. After the reaction was completed, a 10 μL aliquot of each amplicon was run on a 0.7% agarose gel and visualized by ethidium bromide (BIORAD, molecular imager GelDoc XR+) to confirm amplification.

Nucleotide sequencing and analysis. PCR amplicons were sequenced by MrDNA (Shallowater, TX) using barcoded and adaptor-modified ITS1F:ITS2 primers for Illumina Myseq sequencing. Nested PCR was performed since fungal DNA abundance was not known and it might have been in relatively low proportions. Artifacts such as possible chimeric sequences were removed by MrDNA. Results were expressed as proportion of identified taxa per time-point (harvest, days 0, 30, 60 and 90 of storage). Organisms receiving unclear identification from MrDNA (i.e. top BLAST hit was "Environmental sample") were run on BLAST (National Center for Biotechnology Information, NCBI). Samples which ITS1 sequence could not distinguish to species were clustered at genus level when necessary. In those cases, fungal identification was made based on maximum identities (>98%). Differences in the fungal community were reflected in the relative abundance at each time-point.

RESULTS AND DISCUSSION

This study had the objective of providing an understanding of the current food security situation in the Highlands of Guatemala by means of identifying several fungi present at two different stages of maize production; harvest and throughout storage. In order to evaluate any possible influence of the altitude, farms from three different altitudes were selected to participate in this study. As part of analysis, fungi were categorized as biologically significant, i.e. reported from maize or the maize environment (22 taxa, Table 1) or incidental (not likely to be primary or secondary colonizers of maize; 30 additional taxa). It can be seen in Figure 1 that fifty-two fungal taxa in total were identified throughout the farms. Different species were distributed in relation to the geographical areas, with environmental conditions (e.g. moisture level, relative humidity) varying accordingly (see Chapter 3).

In instances where ribosomal targets are not sufficiently sensitive for discrimination of closely related species, additional loci can be sequenced (Romanelli et al., 2014). The ITS region used in this research was perhaps not the best approach to identify species of some genera including *Penicillium, Fusarium, and Alternaria*, thus

such taxa were reported only at the genus level. Where species-level identification is critical for cases of this nature, a highly specific primer can be selected to amplify a region specific for the species of interest. However this would likely require culturing, as such regions occur in single copy genes. Nonetheless the technique used here allowed for a broader understanding of the population by using a less specific primer.

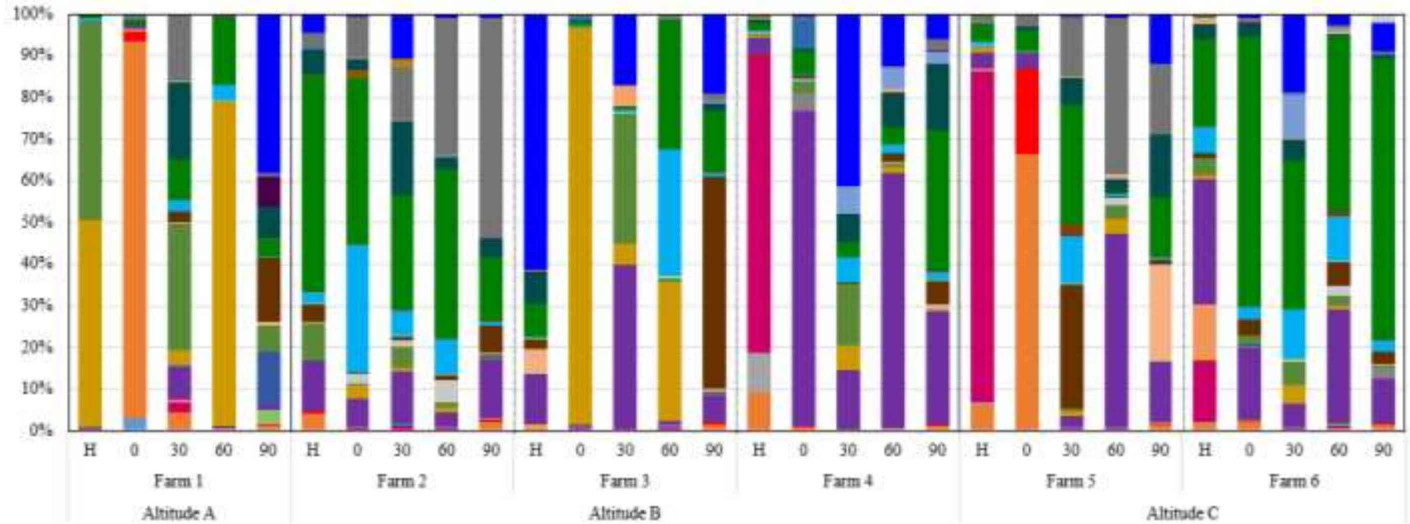
TABLE 1. Fungal taxa with a documented association with maize, isolated from maize from Chiantla and Todos Santos, Huehuetenango, Guatemala. Growing season 2014-2015.

Taxa	Isolated from	Reference
Candida sake	Maize	(Michele A Mansfield, 2005)
Candida sp.	Maize	(Lumi-Abe et al., 2015)
Candida quercitrusa	Maize	(Nguyen et al., 2007; Su-lin L. Leong, 2012; Xiao et al., 2014)
Cryptococcus sp.	(C. flavescens) Maize	(Kohl et al., 2015; Kurtzman, 1973; Michele A Mansfield, 2005)
Debaryomyces sp.	Maize (D. hansenii)	(Nout et al., 1997)
Hannaella zeae	Isolated from guts of corn rootworm	(Global Catalogue of Microorganisms (GMC), 2008)
Trichoderma hypocrea koningii	Affects maize growth in conjunction with F. solani	(C. B. MsAllister, I. García-Romera, A. Godeas, 1994)
Trichoderma hypocrea rufa	Isolated from corn infected with southern leaf blight	(C. T. Hou, A. Ciegler, 1972)
Acremonium sp.	Maize (A. zeae) (A. strictum)	(Poling et al., 2008; Tagne et al., 2002; Wicklow et al., 2005)
Aspergillus flavus	Maize	(de Lange et al., 2014; Payne et al., 2006; Torres et al., 2015)
Botrytis cinerea	Maize	(Shurtleff et al., 2016; ten Have et al., 2001)
Cladosporium sp.	Maize	(Lumi-Abe et al., 2015)
(Fusarium) Gibberella zeae	(Fusarium graminearum) Maize	(Broders et al., 2007; Reid et al., 1999)
(Fusarium) Gibberella intermedia	(Fusarium Proliferatum) Maize	(Logrieco et al., 1995; Scarpino et al., 2015)
(Fusarium) Gibberella moniliformis	(Fusarium verticillioides) Maize	(Jurgenson et al., 2002; Maschietto et al., 2015)
Monographella sp.	Maize	(Müller, E.; Samuels, 1984)
Mucor fragilis	Maize and rice	(Lumi-Abe et al., 2015)
Nigrospora sp.	Maize	(Blaney et al., 1986; D. T. Wicklow, C .W. Hesseltine, O. L. Shotwell, 2014; Standen, 1941)
Penicillium sp.	Maize	(Mansfield et al., 2008; Marin et al., 1998)
Phoma sp.	Isolated from maize roots.	(Orole and Adejumo, 2009; Remesova et al., 2007; Stephen Peterson, Cletus Kurtzman, 2015)
Stenocarpella macrospora	Maize	(European and Mediterranean Plan Protection Organization (EPPO), 2016)
Stenocarpella maydis	Maize	(European and Mediterranean Plan Protection Organization (EPPO), 2016; Wicklow et al., 2011)

Two organisms can be identified as partly dominant in the maize farms. The fungal endophytes in the genus *Fusarium (Gibberella)* ranging from 109 (0.4%) to 57478 (67.2%) sequence hits across farm time-points, as well as *Stenocarpella maydis* ranging from 13 (0.2%) to 55081 (61.3%) sequence hits across farm time-points. Regarding *Fusarium* spp., these pathogens have been reported to survive in the soil, in infected debris as well as inside of apparently healthy seeds (Morales-Rodriguez et al., 2007), affecting the plant throughout its development, including the edible parts. Previous reports of contaminated maize from the lowlands of Guatemala included the organisms *F. moniliforme* and *F. oxysporum* (Martinez et al., 1970). Additionally, *Gibberella moniliformis* (anamorph *Fusarium verticillioides*), which was detected in this study, produces the mycotoxin fumonisin (Jurgenson et al., 2002). This indicates that maize from the region if not maintained under conditions that would control mold growth and toxin production, could represent a food safety hazard for the population that would consume it.

Stenocarpella maydis is a pathogen of importance as it is the causative agent of dry-rot of maize ears. It has also been reported to be related with a neuromycotoxicosis in cattle grazing harvested maize fields in southern Africa and Argentina, activity which is also common in Guatemala (Wicklow et al., 2011). In the same genus, *S. macrospora* was detected in much less prevalence. This plant pathogen is commonly widespread in humid subtropical and tropical zones where plants exhibiting dry-ear rot and stalk rot may also display symptoms of leaf and husk striping (Wicklow et al., 2011) affecting alternate uses of additional maize parts (e.g. to make *chuchitos*), a practice commonly observed in the country.

Cladosporium as well as *Nigrospora*, found in Farms 1 and 3, have also been reported previously in Guatemala (Martinez et al., 1970). *Cladosporium* can produce undesirable effects on maize quality such as discoloration, reduced germination, mustiness, sour odors, chemical changes, and loss of weight (Katab, 2012). Similarly, *Nigrospora* affects the proper development of the maize plant, as it is frequently involved in ear-rots (Blaney et al., 1986).



Taxon	Hits	Taxon	Hits	Taxon	Hits	Taxon	Hits
Candida sake	3846	Candida sp.	206738	Candida quercitrusa	10691	(Kurtzmaniella) Candida ra lenensis	173507
(Kurtzmaniella) Candida zeylanoides	15	Cryptococcus flavescens	493	Cryptococcus rajasthanensis	127	Cryptococcus sp.	32
Debaryomyces sp.	35860	Hannaella zeae	67	Hyphopichia burtonii	12	Malassezia restricta	91
Malassezia sympodialis	13	Meyerozyma aff. caribbica	8434	Pichia sp.	112	Rhodotorula sp.	408
Torulaspora delbrueckii	482	Trichoderma hypocrea koningii	1307	Trichoderma hypocrea rufa	201	Acremonium sp.	233466
Aspergillus sp.	4970	Aspergillus aff. flavus	103863	Botrytis cinerea	5548	Cladosporium sp.	103794
Cordyceps confragosa	591	Diaporthe ampelina	20552	Diaporthe phaseolorum	5571	Diaporthe sp.	73
(Fusarium) Gibberella zeae	59989	(Fusarium) Gibberella aff. intermedia	1055	(Fusarium) Gibberella aff. moniliformis	49479	Fusarium polyphialidicum	2821
(Fusarium) Gibberella sp.	295328	Fusarium poae	1204	Lophiostoma cynaroidis	235	Monographella sp.	68178
Mucor fragilis	11	Nectria mauritiicola	4438	Nectria sp.	90	Neonectria sp.	11
Nigrospora sp.	15838	Ophiostoma ips	86	Penicillium sp.	4427	Phacidiopycnis washingtonensis	2799
Phoma sp.	87063	Plectosphaerella cucumerina	623	Rhinocladiella similis	30	Sphaeronaemella fimicola	63
Stenocarpella macrospora	11	Stenocarpella maydis	165076	Wallemia sp.	2128	Calocera cornea	205

FIGURE 1. Distribution of fungi in maize farms from Chiantla and Todos Santos, Huehuetenango, Guatemala. Maize season 2014-2015. The bar graph shows the relative abundance of the 52 taxa present in the farms.

Aspergillus was detected, with higher presence on farms 2 and 5 (Figure 1), increasing the chances of food spoilage (Perrone et al., 2007). In addition to DNA analysis, samples were also subjected to a plating regime. In this traditional method, *Aspergillus* presence was evident based on its distinctive morphology on the plates and characteristic conidiophores under the microscope (data not shown).

Another organism of health concern found in a higher proportion for Altitudes A and B consisted of the aflatoxin-producer *Aspergillus flavus*, previously reported in lowland areas of the country (Martinez et al., 1970; Torres et al., 2015). This organism is one of the most common species associated with agricultural products mainly due to its stability in soil (Perrone et al., 2007), being likely to affect future harvests when the land is not properly treated between seasons. Regarding this matter, field observations revealed that tilling practices were not common in the region of study. Although multiple benefits have been reported associated with not tilling, fungal biomass is enhanced in the topsoil under no-till conditions (Jansa et al., 2003) further aggravating the problem after each harvest.

The amount of aflatoxin-producing fungi associated with crops varies with climate. *A. flavus* is more common in warm areas, whereas it decreases in presence in colder areas (Cotty and Jaime-Garcia, 2007). This pattern, however, was not observed for the analyzed farms as the ones located in the coldest temperature had a higher presence of this fungi compared to some lower-altitude farms. This finding could be the result of erratic weather conditions, the maize handling process (e.g. kernel exposure during shelling) which may have promoted wounds, or because the farm did not have proper pest control, which may have allowed for entry points for this organism.

It can be observed that the occurrence of *Phoma* was higher in Todos Santos farms whereas *A. flavus* was more abundant in Chiantla. *Phoma* had a low presence in early stages of storage; it slowly colonized the maize, increasing in later storage time-points (Figure 1, farms 2 and 5).

Regarding *A. flavus*, solely based on the results shown here, maize production in Chiantla would be at higher risk of aflatoxin contamination, compared to maize from Todos Santos. Positively, some endophytes, such as the case of the detected *Phoma sp.,* have been found to be beneficial to the plant as a promoter of health, improving growth potential and acting as biological control agents against fungal (e.g. *Fusarium*) and bacterial diseases of plants (Orole and Adejumo, 2009).

Part of the fungal diversity detected included organisms not commonly found in the commodity of interest. Some examples include *Candida railenensis*, detected in several farm time-points, and having a particularly high presence on the harvest samples from farm 4 and 5. *C. railenensis* has been previously reported in oak trees, a cold-resistant tree found in the region of study (Terradas, 1999; Tzuk, 2011), hence their presence is comprehensible since all the farmers in the region perform traditional agricultural practices, such as exposing the maize to the environment for sun drying. Part of these traditional practices is the shelling of maize cobs by hand, an activity which may have contributed to the presence of *C. zeylanoides, Malassezia restricta,* and *Malassezia sympodialis,* frequently reported on skin (Dawson, 2007; Khosravi et al., 2013). More generally, *Candida sp*. was also detected among farms, and in considerably high abundance for farm 1 (altitude A) and farm 5 altitude C. In both instances, it occurred in time points near harvest, suggesting that the yeast outcompeted other fungi due to a high moisture content (Figure 1, Chapter 3), and may be associated with freshly harvested samples in the region. This finding indicates that insufficient drying upon harvest would result in spoilage during storage, diminishing the quality and shelf-life of the grain reserves.

Overall, results revealed a high presence of both field fungi (e.g. *Alternaria*, *Cladosporium*, and *Gibberella*) and storage fungi (e.g. *Aspergillus* and *Penicillium*), with some degree of succession of across farms, indicating an elevated moisture during harvesting and throughout storage, as well as possible pest damage generating wounds (Hirst Sole, 1994). Regardless of the cause, the presence of this variety of detrimental fungi reflects poor practices throughout the maize chain in this rural region of the country

which may compromise the quality and safety of the maize consumed. Furthermore, the association of maize with such spoilage organisms may further compromise the food security in the region. Fungi are ubiquitous, and controlling their presence presents a degree of difficulty, however, proper pre- and post-harvest practices may help attain conditions that delay fungal growth (see Chapters 3 and 5).

ACKNOWLEDGEMENTS

Authors wish to acknowledge the NGO SHARE Guatemala for the field work in Huehuetenango. Research done at the Food Innovation Center’s laboratory is supported by United States Department of Agriculture (USDA) Scientific Cooperation and Research Program (SCRP); CFDA number 10.961 .

REFERENCES

Anderson, I.C., Cairney, J.W.G., 2004. Diversity and ecology of soil fungal communities: Increased understanding through the application of molecular techniques. Environ. Microbiol. 6, 769–779. doi:10.1111/j.1462-2920.2004.00675.x

Blaney, B., Ramsey, M., Tyler, A., 1986. Mycotoxins and toxigenic fungi in insect-damaged maize harvested during 1983 in far North Queensland. Aust. J. Agric. Res. 37, 235. doi:10.1071/AR9860235

Bokulich, N.A., Mills, D.A., 2013. Improved selection of internal transcribed spacer-specific primers enables quantitative, ultra-high-throughput profiling of fungal communities. Appl. Environ. Microbiol. 79, 2519–2526. doi:10.1128/AEM.03870-12

Broders, K.D., Lipps, P.E., Paul, P. a., Dorrance, a. E., 2007. Evaluation of *Fusarium graminearum* Associated with Corn and Soybean Seed and Seedling Disease in Ohio. Plant Dis. 91, 1155–1160. doi:10.1094/PDIS-91-9-1155

Bruns, T., 2002. Primer sequences [WWW Document]. Nucl. Prim. - 28s rRNA. URL https://nature.berkeley.edu/brunslab/tour/primers.html (accessed 5.4.16).

Bryla, M., Roszko, M., Szymczyk, K., Jedrzejczak, R., Obiedzinski, M., Sekul, J., 2013. Fumonisins in plant-origin food and fodder - a review. Food Addit. Contam. Part A. Chem. Anal. Control. Expo. Risk Assess. 30, 1626–1640.

C. B. MsAllister, I. García-Romera, A. Godeas, J.A.O., 1994. Interactions between Trichoderma koningii, Fusarium solani and Glomus mosseae: Effects on plant growth, arbuscular mycorrhizas and the saprophyte inoculants. Soil Biol. Biochem. 26, 1363–1367.

C. T. Hou, A. Ciegler, C.W.H., 1972. New Mycotoxin, Trichotoxin A, from Trichoderma viride Isolated from Southern Leaf Blight-Infected corn. Appl. Microbiol. 23, 183–185.

Cornell University, 2015. Aflatoxins: Occurrence and Health Risks [WWW Document]. URL http://poisonousplants.ansci.cornell.edu/toxicagents/aflatoxin/aflatoxin.html (accessed 10.10.15).

Cotty, P.J., Jaime-Garcia, R., 2007. Influences of climate on aflatoxin producing fungi

and aflatoxin contamination. Int. J. Food Microbiol. 119, 109–115. doi:10.1016/j.ijfoodmicro.2007.07.060

D. T. Wicklow, C .W. Hesseltine, O. L. Shotwell, G.L.A., 2014. Interference competition and aflatoxin levels in corn. Postharvest Pathol. Mycotoxins 70, 761–764.

Dawson, T.L., 2007. Malassezia globosa and restricta: Breakthrough Understanding of the Etiology and Treatment of Dandruff and Seborrheic Dermatitis through Whole-Genome Analysis. J. Investig. Dermatology Symp. Proc. 12, 15–19. doi:10.1038/sj.jidsymp.5650049

de Lange, E.S., Balmer, D., Mauch-Mani, B., Turlings, T.C.J., 2014. Insect and pathogen attack and resistance in maize and its wild ancestors, the teosintes. New Phytol. 204, 329–341. doi:10.1111/nph.13005

Directorate General-Joint Research Centre of the European Commission, 2007. Maize Seeds Sampling and DNA Extraction. Report on the Validation of a DNA Extraction Method from Maize Seeds and Grains.

European and Mediterranean Plan Protection Organization (EPPO), 2016. Stenocarpella macrospora and Stenocarpella maydis [WWW Document]. Data Sheets Quar. Pests. URL https://www.eppo.int/QUARANTINE/data_sheets/fungi/DIPDSP_ds.pdf (accessed 6.6.16).

Glewen, B.M., Kung, L., Shaver, R.D., Hoffman, P.C., 2013. Dealing with High Yeast Levels in High Moisture Corn and Corn Silage.

Global Catalogue of Microorganisms (GMC), 2008. Hannaella zeae [WWW Document]. HB 1209. URL http://gcm.wfcc.info/Strain_numberToInfoServlet?strain_number=CBS 11179

Hirst Sole, L.A., 1994. Algunos aspectos físicos y de procesamiento del maíz en grano y mazorca, con y sin tusa, almacenado en dos tipos de estructuras, con y sin tratamiento químico. Universidad del Valle de Guatemala. Ingeniería en Ciencias de Alimentos.

Jansa, J., Mozafar, A., Kuhn, G., Anken, T., Ruh, R., Sanders, I.R., Frossard, E., 2003. Soil tillage affects the community structure of mycorrhizal fungi inmaize roots. Ecol. Appl. 13, 1164–1176. doi:10.1890/1051-0761(2003)13[1164:STATCS]2.0.CO;2

Jurgenson, J.E., Zeller, K.A., Leslie, J.F., 2002. Expanded genetic map of Gibberella moniliformis (Fusarium verticillioides). Appl. Environ. Microbiol. 68, 1972–1979. doi:10.1128/AEM.68.4.1972-1979.2002

Katab, Y.N.T., 2012. Effect of certain fungi on maize plants with special reference to seed deterioration in field and storage and their control. Cairo University.

Kew, M.C., 2003. Synergistic interaction between aflatoxin B1 and hepatitis B virus in hepatocarcinogenesis. Liver Int. 23, 405–409. doi:10.1111/j.1478-3231.2003.00869.x

Khosravi, A.R., Shokri, H., Nikaein, D., Erfanmanesh, A., Fatahinia, M., Hlan, J.A., 2013. Evaluation of the pathogenicity of candida zeylanoides in BALB/c mice. Turkish J. Vet. Anim. Sci. 37, 408–413. doi:10.3906/vet-1105-40

Kim, J.J., Kim, S.H., Lee, S., Breuil, C., 2003. Distinguishing Ophiostoma ips and Ophiostoma montium, two bark beetle-associated sapstain fungi. FEMS Microbiol. Lett. 222, 187–192. doi:10.1016/S0378-1097(03)00304-5

Kohl, J., Lombaers, C., Moretti, A., Bandyopadhyay, R., Somma, S., Kastelein, P., 2015. Analysis of microbial taxonomical groups present in maize stalks suppressive to colonization by toxigenic Fusarium spp.: A strategy for the identification of potential antagonists. Biol. Control 83, 20–28. doi:10.1016/j.biocontrol.2014.12.007

Kurtzman, C.P., 1973. Formation of hyphae and chlamydospores by Cryptococcus laureni. Mycologia 65, 388–395.

Logrieco, A., Moretti, A., Ritienti, A., Bottalico, A., Corda, P., 1995. Occurence and Toxigenicity of F. proliferatum from Preharvest Maize Ear Rot and Associated Mycotoxins in Italy. Plant Dis.

Lumi-Abe, C.A., Faria, C.B., Fernandes De Castro, F., Regina De Souza, S., Cristina, F., Santos, D., Novais, C., Silva, D., Tessmann, D.J., Parra Barbosa-Tessmann, I., 2015. Fungi Isolated from Maize (Zea mays L.) Grains and Production of Associated Enzyme Activities. Int. J. Mol. Sci 16, 15328–15346. doi:10.3390/ijms160715328

Mansfield, M.A., Jones, A.D., Kuldau, G.A., 2008. Contamination of fresh and ensiled maize by multiple penicillium mycotoxins. Phytopathology 98, 330–336. doi:10.1094/PHYTO-98-3-0330

Marin, S., Sanchis, V., S??enz, R., Ramos, A.J., Vinas, I., Magan, N., 1998. Ecological

determinants for germination and growth of some Aspergillus and Penicillium spp. from maize grain. J. Appl. Microbiol. 84, 25–36. doi:10.1046/j.1365-2672.1997.00297.x

Martinez, E., Gomez, R., Bressani, R., 1970. Prevalencia de hongos en granos de maiz (Zea mays, L.) de Guatemala. Turrialba 20, 311–319.

Maschietto, V., Marocco, A., Malachova, A., Lanubile, A., 2015. Resistance to Fusarium verticillioides and fumonisin accumulation in maize inbred lines involves an earlier and enhanced expression of lipoxygenase (LOX) genes. J. Plant Physiol. 188, 9–18. doi:10.1016/j.jplph.2015.09.003

Michele A Mansfield, 2005. Fungi and Mycotoxins in Fresh and Ensiled Maize and the Affects of Agronomic Practices, Weather Conditions and Silage Characteristics on Toxin Contamination. Pennsylvania State University.

Morales-Rodriguez, I., Yañez-Morales, M. de J., Silva-Rojas, H. V., García-de-los-Santos, G., Guzmán-de-Peña, D.A., 2007. Biodiversity of Fusarium species in Mexico associated with ear rot in maize, and their identification using a phylogenetic approach. Mycopathologia 163, 31–39. doi:10.1007/s11046-006-0082-1

Müller, E.; Samuels, G.J., 1984. Monographella maydis sp. nov. and its connection to the tar-spot disease of Zea mays. Inst. Microbiol 40, 113–121.

Nguyen, N.H., Suh, S.-O., Blackwell, M., 2007. Five novel Candida species in insect-associated yeast clades isolated from Neuroptera and other insects. Mycologia 99, 842–858. doi:10.3852/mycologia.99.6.842

Nilsson, R.H., Ryberg, M., Kristiansson, E., Abarenkov, K., Larsson, K.H., Köljalg, U., 2006. Taxonomic reliability of DNA sequences in public sequences databases: A fungal perspective. PLoS One 1. doi:10.1371/journal.pone.0000059

Nout, M.J., Platis, C.E., Wicklow, D.T., 1997. Biodiversity of yeasts from Illinois maize. Can. J. Microbiol. 43, 362–367. doi:10.1139/m97-050

Orole, O.O., Adejumo, T.O., 2009. Activity of fungal endophytes against four maize wilt pathogens. African J. Microbiol. Res. 3, 969–973.

Payne, G. a, Nierman, W.C., Wortman, J.R., Pritchard, B.L., Brown, D., Dean, R. a, Bhatnagar, D., Cleveland, T.E., Machida, M., Yu, J., 2006. Whole genome

comparison of Aspergillus flavus and A-oryzae. Med. Mycol. 44, S9–S11. doi:Doi 10.1080/13693780600835716

Perrone, G., Susca, A., Cozzi, G., Ehrlich, K., Varga, J., Frisvad, J.C., Meijer, M., Noonim, P., Mahakarnchanakul, W., Samson, R.A., 2007. Biodiversity of Aspergillus species in some important agricultural products. Stud. Mycol. 59, 53–66.

Poling, S.M., Wicklow, D.T., Rogers, K.D., Gloer, J.B., 2008. Acremonium zeae, a protective endophyte of maize, produces dihydroresorcylide and 7-hydroxydihydroresorcylides. J. Agric. Food Chem. 56, 3006–3009. doi:10.1021/jf073274f

Reid, L.M., Nicol, R.W., Ouellet, T., Savard, M., Miller, J.D., Young, J.C., Stewart, D.W., Schaafsma, a W., 1999. Interaction of Fusarium graminearum and F. moniliforme in Maize Ears: Disease Progress, Fungal Biomass, and Mycotoxin Accumulation. Phytopathology 89, 1028–1037. doi:10.1094/PHYTO.1999.89.11.1028

Remesova, J., Kolarik, M., Prasil, K., 2007. Microfungi on the kernels of transgenic and non-transgenic maize damaged by the European corn borer. Czech Mycol. 59, 205–213.

Richard, E., Heutte, N., Bouchart, V., Garon, D., 2009. Evaluation of fungal contamination and mycotoxin production in maize silage. Anim. Feed Sci. Technol. 148, 309–320. doi:10.1016/j.anifeedsci.2008.02.004

Richard, E., Heutte, N., Sage, L., Pottier, D., Bouchart, V., Lebailly, P., Garon, D., 2007. Toxigenic fungi and mycotoxins in mature corn silage. Food Chem. Toxicol. 45, 2420–2425. doi:10.1016/j.fct.2007.06.018

Romanelli, A.M., Fu, J., Herrera, M.L., Wickes, B.L., 2014. A universal DNA extraction and PCR amplification method for fungal rDNA sequence-based identification. Mycoses 57, 612–622. doi:10.1111/myc.12208

Scarpino, V., Reyneri, A., Vanara, F., Scopel, C., Causin, R., Blandino, M., 2015. Relationship between European Corn Borer injury, Fusarium proliferatum and F. subglutinans infection and moniliformin contamination in maize. F. Crop. Res. 183, 69–78. doi:10.1016/j.fcr.2015.07.014

Shurtleff, M.C., Edwards, D.I., Noel, G.R., Pedersen, W.L., 2016. Diseases of Corn or Maize Diseases of Corn or Maize (Zea mays L .).

Sifuentes-Osornio, J., Corzo-Le??n, D.E., Ponce-De-Le??n, L.A., 2012. Epidemiology of invasive fungal infections in Latin America. Curr. Fungal Infect. Rep. 6, 23–34. doi:10.1007/s12281-011-0081-7

Standen, J.H., 1941. Variability and pathogenicity of Nigrospora oryzae (B . and Br .) Petch in maize. Iowa State College.

Stephen Peterson, Cletus Kurtzman, T.N., 2015. Protectice endophytes of maize that inhibit fungal pathogens and reduce mycotoxin contamination. doi:10.1002/pro.2705.Price

Su-lin L. Leong, 2012. Microbial populations during maize storage in Cameroon. African J. Biotechnol. 11, 8692–8697. doi:10.5897/AJB12.108

Tagne, A., Neergaard, E., Hansen, H.J., The, C., 2002. Studies of host-pathogen interaction between maize and Acremonium strictum from Cameroon. Eur. J. Plant Pathol. 108, 93–102. doi:10.1023/A:1015092030874

Tedersoo, L., Nilsson, R.H., Abarenkov, K., Jairus, T., Sadam, A., Saar, I., Bahram, M., Bechem, E., Chuyong, G., K??ljalg, U., 2010. 454 Pyrosequencing and Sanger sequencing of tropical mycorrhizal fungi provide similar results but reveal substantial methodological biases. New Phytol. 188, 291–301. doi:10.1111/j.1469-8137.2010.03373.x

ten Have, a, Breuil, W.O., Wubben, J.P., Visser, J., van Kan, J. a, 2001. Botrytis cinerea endopolygalacturonase genes are differentially expressed in various plant tissues. Fungal Genet. Biol. 33, 97–105. doi:10.1006/fgbi.2001.1269

Terradas, J., 1999. Holm Oak and Holm Oak Forests: An Introduction, in: Studies, E. (Ed.), Ecology of Mediterranean Evergreen Oak Forests. Springer-Verlag, pp. 3–14. doi:10.1007/978-3-642-58618-7_1

Torres, O. a, Palencia, E., Lopez de Pratdesaba, L., Grajeda, R., Fuentes, M., Speer, M.C., Merrill, A.H., O'Donnell, K., Bacon, C.W., Glenn, A.E., Riley, R.T., 2007. Estimated fumonisin exposure in Guatemala is greatest in consumers of lowland maize. J. Nutr. 137, 2723–2729. doi:137/12/2723 [pii]

Torres, O., Matute, J., Gelineau-van Waes, J., Maddox, J.R., Gregory, S.G., Ashley-

Koch, A.E., Showker, J.L., Voss, K.A., Riley, R.T., 2015. Human health implications from co-exposure to aflatoxins and fumonisins in maize-based foods in Latin America: Guatemala as a case study. World Mycotoxin J. 8, 143–159. doi:10.3920/WMJ2014.1736

Tzuk, A., 2011. Especies de árboles a sembrar en Guatemala por tipo de clima [WWW Document]. Reforestando Guatemaya. URL http://www.slideshare.net/AjTzuk/especies-de-rboles-segn-clima (accessed 1.1.16).

Wicklow, D.T., Rogers, K.D., Dowd, P.F., Gloer, J.B., 2011. Bioactive metabolites from Stenocarpella maydis, a stalk and ear rot pathogen of maize. Fungal Biol. 115, 133–142. doi:10.1016/j.funbio.2010.11.003

Wicklow, D.T., Roth, S., Deyrup, S.T., Gloer, J.B., 2005. A protective endophyte of maize: Acremonium zeae antibiotics inhibitory to Aspergillus flavus and Fusarium verticillioides. Mycol. Res. 109, 610–618. doi:10.1017/S0953756205002820

Wild, C.P., Gong, Y.Y., 2009. Mycotoxins and human disease: A largely ignored global health issue. Carcinogenesis 31, 71–82. doi:10.1093/carcin/bgp264

Xiao, M., Wang, H., Lu, J., Chen, S.C.A., Kong, F., Ma, X.J., Xu, Y.C., 2014. Three clustered cases of candidemia caused by Candida quercitrusa and mycological characteristics of this novel species. J. Clin. Microbiol. 52, 3044–3048. doi:10.1128/JCM.00246-14

FINANCIAL FEASIBILITY STUDY OF METAL SILO IMPLEMENTATION FOR PROPER MAIZE STORAGE IN THE WESTERN HIGHLANDS OF GUATEMALA

R. Mendoza[1], F. Mattos[2], C. Campabadal[3], H. Hallen-Adams[1], and A. Bianchini[1]

[1] Department of Food Science & Technology. University of Nebraska. Lincoln, NE, USA

[2] Department of Agricultural Economics. University of Nebraska. Lincoln, NE, USA

[3] Grain Science and Industry, Kansas State University. Manhattan, KS, USA

Corresponding author:

Andréia Bianchini, Department of Food Science & Technology

The Food Processing Center, University of Nebraska-Lincoln

Phone: (402)472-3144, Fax: (402)472-1693

abianchini2@unl.edu

ABSTRACT

Guatemala has encountered great challenges in terms of food security, influenced by the deficiency in traditional post-harvest practices, including storage structures, among others. This study aimed to evaluate the financial feasibility of introducing metal silos as part of an improved storage system for maize in the Western Highlands of Guatemala. White dent maize was used to assess different feasibility scenarios via simulations. Data gathered for the analysis included two small surveys as well as governmental data from previous maize seasons (2004-2014). Simulation output showed the feasibility of purchasing a silo in a period of one year in terms of relative expense. Comparisons were made between a specific scenario (where a farmer obtained a silo and followed a specific maize purchasing regime) and a baseline where there was no silo, and farmers had to buy maize on a monthly basis or every four months. One scenario was conducted describing maize producing farms (chain 1), and six scenarios were carried out for farms where maize was not planted but only purchased and consumed (chain 2). The best scenarios described the relative expense of chain 2 farmers who would purchase a silo and an entire year's maize supply at once. This scenario had a cumulative probability of 0.43 and 0.27 when compared to a baseline of buying maize on a 4-month period and 1 month purchase period, respectively. The remaining scenarios for chain 2 farmers showed lower probabilities (<0.2). The setting for chain 1 farms' probabilistic output indicated a cumulative probability of 0.13 of having less expenses overall when purchasing a silo. Probabilistic histograms revealed that farmers have a low likelihood of being debt-free after the first year of technology implementation and usage. However, if simulation conditions were to include a loan extension or the addition of off-farm money, it is likely that this technology will be feasible for farmers to obtain. The constant use of the silo in subsequent years would result in profit, which will ultimately aid to improve the quality of life of the inhabitants of the region.

Keywords: Guatemala, maize, corn, Monte Carlo

INTRODUCTION

Maize is a staple grain largely produced and consumed throughout Guatemala, especially in the rural areas in the form of tortillas (Torres et al. 2007; Etten and Fuentes 2004). This commodity is typically cultivated and handled through traditional agricultural practices. Because of this, substantial amounts are lost, aggravating starvation and malnutrition in the country (FAO 2009). Moreover, while storage is considered an important part of the farming process as it is necessary for guaranteeing the household food security, traditional storage practices in developing countries cannot assure the safety of the grains (Yusuf and He 2011).

It has been reported previously (Tefera et al. 2011) that damage caused by pests during storage is responsible for an estimated maize loss of up to 30%, particularly when no agrochemicals are used. This affects families' well-being both from a financial and a food availability perspective (Manuel et al. 2007). Besides causing quantitative losses, pests are commonly associated with mycotoxin contamination, compromising food and feed safety. Subsistence farmers may have no alternative but to consume a certain amount of damaged product (Boxall 2002). The consumption of contaminated grain, depending on the fungal toxin, leads to a diversity of symptoms from stunting to systemic cancer, and even death (Torres et al. 2007; Torres et al. 2015; Bravo 2009). Additionally, pests such as rats are able to transmit diseases such as poliomyelitis, leptosporiosis, teniasis, among others (Bravo 2009).

Several maize diseases caused by fungi prevail in warm, humid, tropical areas (Tefera et al. 2011), including those caused by mycotoxin-producing fungi. According to the Food and Agriculture Organization of the United Nations (FAO), at least 25% of the world's food crops are contaminated with mycotoxins (D.L. Park 1999), causing significant economic and trade problems at almost every stage of marketing between producers and consumers. When animal feed is contaminated, it can result in animal death, or lowered meat, egg and poultry productivity as well as decreased work performance (Dawson 1991).

Approximately 2% of Guatemalan farms are commercially driven, selling in local markets and some even in the international market. Some experience success, and several struggle to be profitable, complementing their income with off-farm activities. Many farms in Guatemala function merely at the subsistence level (Washington Office on Latin America (WOLA) 2013). The extant precarious conditions of maize production and storage in Guatemala continue to be an obstacle to obtaining a good quality product. It is common to observe that the best quality products are marketed outside of the country while the low grade food products remain in the region, affecting the health of the population (Dawson 1991; Etten and Fuentes 2004; Leslie and Logrieco 2014).

In order to avoid post-harvest losses from pests and microorganisms during storage, smallholder farmers are constrained to either barter (FAO and IPGRI 2002) or to sell their maize soon after harvest (Boxall 2002) when market prices are low, only to purchase it back a few months after at an expensive price, falling in a poverty trap. Even though traditional grain practices, including storage methods, require little investment, they lead to substantial losses over time, contributing to food insecurity. Therefore, promoting small scale improvements in agricultural practices is the key to achieve food security in developing countries (Tefera et al. 2011; Yusuf and He 2011).

There are several storage systems for small- and medium-size producers, such as plastic containers, plastic bags and metal silos. The farmers’ choice of the system will depend on availability, convenience, efficiency and cost-effectiveness (FAO 2014). A metal silo is considered a key post-harvest technology in the fight against hunger and ensuring food safety. It is a cylindrical structure constructed from a galvanized iron sheet and (semi) hermetically sealed, preventing the humidity exchange between the environment and the grain. The airtight seal reduces the oxygen content, killing over time any pests that may be present or preventing access from outside pests, thus extending the shelf life of its contents. Additionally, fungal development ceases when the oxygen level decreases to approximately 1%, and, a high content of carbon dioxide (product of grain

respiration) has been shown to reduce aflatoxin production of *Aspergillus flavus* (Manuel et al. 2007; Yusuf and He 2011; Tefera et al. 2011).

Metal silos provide more food security in quantitative and qualitative terms than traditional storage systems (Hermann 1991). They offer several attractive advantages, including maintaining a stable grain quality over time, with maize possibly being stored for up to three years without any problem. Other benefits include reduced use of insecticides, use of less storage space, none o low post-harvest losses, prevention of pest damage, and disposition of grain at different periods. This last advantage helps with the family income since any can be sold at premium prices for revenue when the market is favorable, empowering smallholder farmers (FAO 2014). With additional income, farmers could also gain access to more education, improving their managerial abilities in the farms to increase production, as these two have shown to be positively correlated (Kalaitzandonakes and Dunn 1995). When produced locally, silos provide an additional benefit to the community as it promotes job opportunities for local blacksmiths (MAGA 2008; Tefera et al. 2011).

To effectively store grains in a metal silo it is critical to maintain a low moisture content of the grain. Maize should be dried to less than 14% moisture content and stored under a relative humidity of 75-85% (Bravo 2009; Alberta Agriculture and Forestry 2010). Moreover, silos must be placed under a roof or cover to protect them from environmental conditions which may influence the grain quality over time. Sun drying of grain is the most common and inexpensive drying practice in developing countries, however may not provide the degree of dryness required for a safe storage (Yusuf and He 2011).

In Central America, the investment in a metal silo pays for itself when losses are prevented for two harvesting seasons, and by purchasing maize in low demand periods when its price is favorable (Bravo 2009). The majority of the staple grain producers in Central America are small to medium-sized family farms, and poverty is widespread among them (Bokusheva et al. 2012). Lack of adequate access to credit commonly

prohibits smallholder farmers from assuming the risk of investing in storage technologies. The investment in the maize sector should be a task of the government, due to the multiple benefits it can bring, not only to producers but also to consumers. Unfortunately in some instances such efforts are not fully implemented. (Etten and Fuentes 2004; Washington Office on Latin America (WOLA) 2013). As in much of Latin America, Guatemala is reducing state support for production, service, and credit activities in agriculture and small-scale commerce (Barham, Boucher and Carter 1996). Nevertheless, efforts have been made in Latin America in order to alleviate this problem. Since 1980, the Swiss Agency for Development and Cooperation (SDC) has sponsored a maize and beans post-harvest loss reduction program, disseminating metal silos in Honduras, Nicaragua, Guatemala and El Salvador (Swiss Agency for Development and Co-operation 2008b; Bokusheva et al. 2012). Studies conducted in these regions have revealed that metal silos used for storing maize increased from 5000 to 176,000 in the period of 1995 to 2007, preventing 13 to 15% of crop loss that would have otherwise occurred due to improper storage (Swiss Agency for Development and Co-operation 2008a).

In this study, a metal silo design was evaluated as means to improve maize quality and safety over time, thus leading to a positive impact in the health of the people living in the Huehuetenango department, in Guatemala. Since the storage technology represents an investment which farmers may or may not be able to afford, the feasibility of its acquisition was evaluated.

MATERIALS AND METHODS

Region of study. The feasibility analysis was performed considering agricultural practices and costs related to purchasing metal silos in Chianta and Todos Santos Cuchumatán, townships of Huehuetenango Guatemala. Farms involved in this study included those where maize was planted, harvested and consumed ("Chain 1" farms) and farms where land was not used or available to plant maize, so farmers purchased grain from the market ("Chain 2" farms).

Data collection. In order to gather essential information to perform the financial analysis such as maize yield patterns, purchase patterns, and price fluctuations in the region of study, two surveys were developed and distributed among twenty-three farms. The first survey included 55 questions regarding household composition, last season yield, daily maize consumption per household, list of places where maize was sold, costs involved in each maize harvest, maize storage facilities, and alternative used of maize (i.e. feed purposes). While the second survey consisted of 14 questions covering maize varieties, amount sold, previous year yields, household income, and drying practices. Information such as the influence of inflation on maize prices and additional maize prices covering several production seasons was obtained via Guatemalan government entities such as the National Institute of Statistics (INE), and the Ministry of Agriculture and Livestock (MAGA). Interest and loan information was gathered from SHARE Guatemala, a local NGO, as well as from banks and credit unions in the region of study. Only relevant survey results are shown as tables or figures while the remaining data was used as input for the simulation software and is not shown. Even though several maize varieties, including native or *criollo*, are grown and consumed in the region, the present financial analysis was performed exclusively for white dent variety. This variety was chosen since it is the one with the most market data availability and it currently dominates the commercial maize market throughout the country.

Data analysis. Monte Carlo Simulation Template version 1.2.0 (Vertex42 LLC 2014) was used for data analysis. Seven scenarios were made and simulations were run. Six scenarios included the chain 2 farms, and one scenario comprised the chain 1 farms. After comparing a scenario of a farmer having a silo, with the same conditions but without the storage technology, probabilistic histograms of relative expense were generated. These histograms revealed the cumulative probability of the feasibility of purchasing the metal silo in a period of one year. The scenarios were set up comparing the use of a silo versus the traditional conditions based on average maize purchasing patterns following a normal distribution and average monthly consumption. The annual production was also included in the model but as a probabilistic distribution, in this case as a triangular distribution (min, mode, max), due to insufficient local production data. Opportunity cost was also included taking into account an average interest rate from local banks. Since most farmers do not have the financial means for acquiring the storage technology, a loan with interest rate was included in the model. Additionally, it has been shown elsewhere that maize should be dried to a safe level of 14% or less prior to storage. Consequently, a drying service was included as an additional cost in the models. Simulation iterations (n) were set to 1000.

Metal silo. Based on field observations of existing metal silos in the region of study, coupled with farmers' and researchers' suggestions for design improvements, a refined design of a flat-bottom metal silo, sourced locally, was constructed. This metal silo of Gauge 26 (thickness), included lids with handles for easy access, as well as the option to add padlocks for security purposes. Moreover, to avoid the tilting of the silo, a shovel-like instrument was included, as well as a rubber stoppers in each lid to improve its hermetic conditions. This design had a cost of 100.00 USD (~785.00 GTQ). At the given dimensions (Figure 2) it has the capacity to store up to 10 quintals (1qq = 100lb) of maize. The price of the storage technology was included in the probabilistic model. As part of the study, 25 improved metal silos were distributed among farmers participating in the study.

RESULTS AND DISCUSSION

Field observations and surveys performed in Chiantla and Todos Santos regarding maize production indicated that the region followed the national maize production pattern (MAGA 2015). It was perceived that the main selling period for maize stored in traditional structures was directly after harvest, from November to January depending on the farm (data not shown). For those farms that can sell maize, this option may not be ideal since at this time prices are low, resulting in low revenue (Figure 1). For farms purchasing maize, they would prefer to do so at this time in order to spend the least amount of money. Unfortunately, traditional storage methods which would allow them to take advantage of the market as sellers or buyers, usually leads to losses during storage. If maize was stored in better storage systems, such as metal silos, it would allow chain 1 farmers to retain the harvested maize and sell it in later months, ideally right before the next harvest time, when the high demand results in favorable prices (e.g. Figure 1, month of July). Likewise, chain 2 farmers could buy more maize in times of low cost and store it for a long time without worrying about possible losses.

In this study, an improved silo based on the design of existing silos in the region was evaluated with regards to improving food safety and security in the Highlands of Guatemala. Field observations indicated that the existing silos, usually from the "Postcosecha" project through the SDC (MAGA 2008), presented denting on the bottom of the opposite section of the outlet. This was believed to be a result of tilting the storage structure in order to obtain the remaining portion of the grains. Additionally, silo users reported difficulty in opening both the inlet during the filling process, and outlet during the maize dispensing process. In order to further prolong the metal silo's life, an improved design (shown in Figure 2) was proposed. The improved design has an estimated life span of 20 years or more (Yusuf and He 2011; Bravo 2009) depending on the care of the owner.

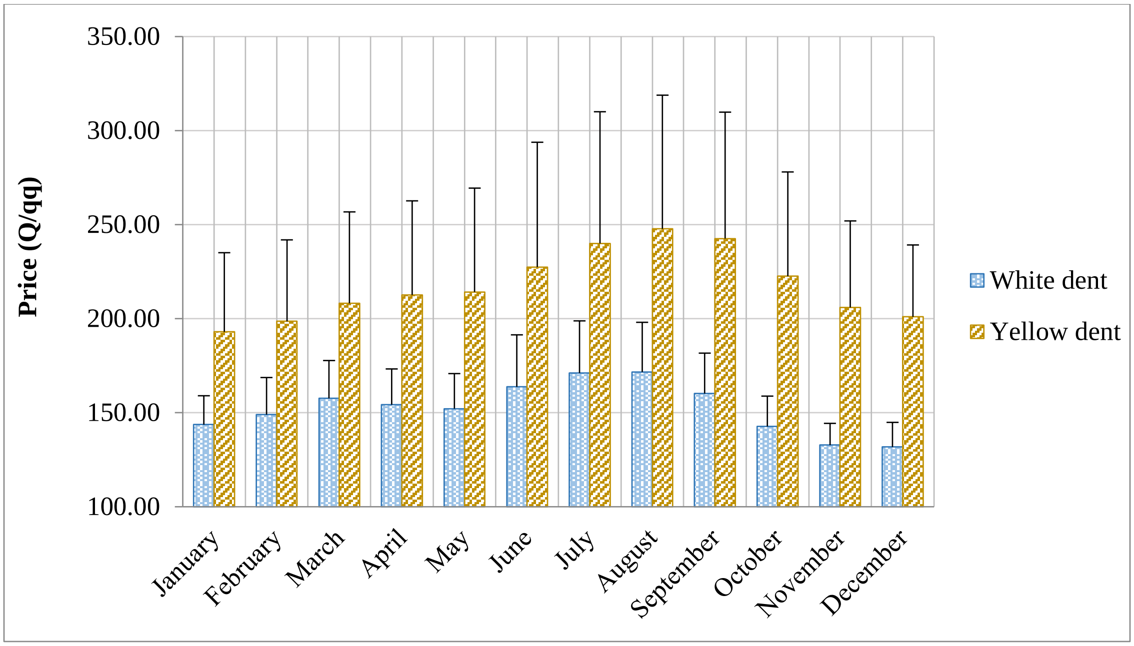


FIGURE 1. Maize average monthly prices adjusted to inflation. White dent, 2004 – 2013. Yellow dent, 2004 – 2012. Unadjusted prices obtained from the Ministry of Agriculture, Livestock and Food (MAGA 2015).

A challenge associated with the use of the metal silos is their relatively high cost, given that people in need of the storage technology are of scarce economic resources. Since traditional methods used in the region (e.g. *tapanco*, *mancuerna*, *cajón*) represent a sunk cost when compared to a metal silo, their cost was not included in the financial analysis. Additionally, when grain is being stored for longer periods than in the past, the opportunity cost of capital tied up in this grain must be computed (FAO 1994). To help in this process, banks and credit unions distributed in the region of study were asked about their savings account and interest rates (data not shown). As a result, an average interest rate of 0.5% was included in the analysis and associated with the amount of grain stored.

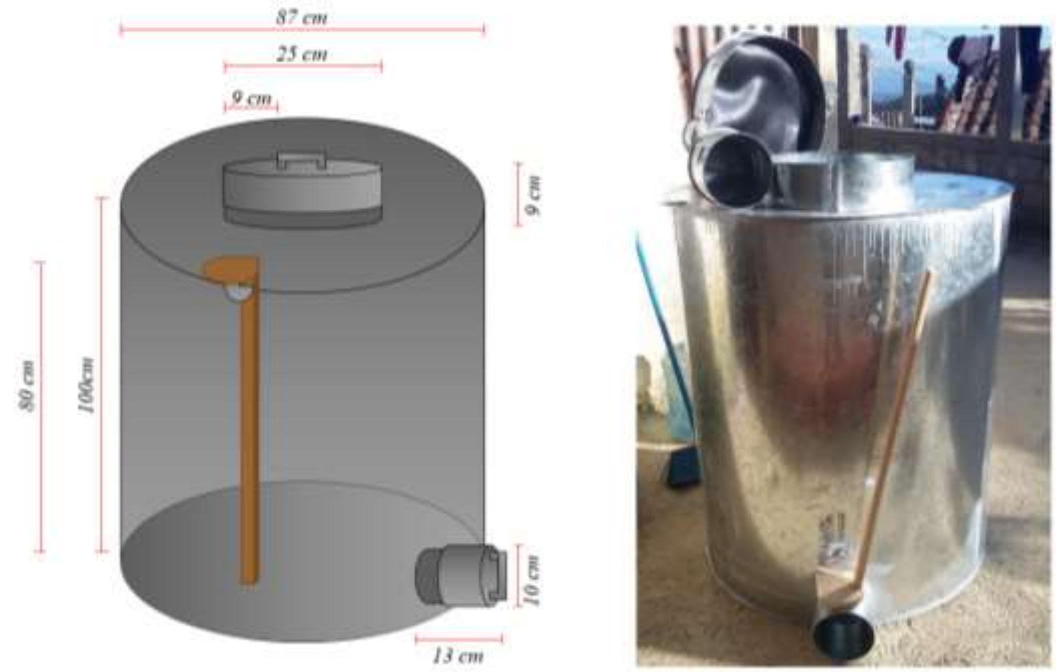


FIGURE 2. Improved flat-bottom silo design (left), and constructed prototype (right) of silo to be used by farms from Todos Santos and Chiantla, Huehuetenango, Guatemala.

Furthermore, since farms from Huehuetenango were not formally established, farmers didn’t necessarily have documentation systems including yields of previous seasons. Consequently, gathering of information was dependent on the farmers recalling such information. Due to the uncertainty of data on surveyed farms, a triangular distribution was used employing the collected records. The majority of farms that dealt with white maize showed a minimum yield of 3qq, a maximum of 12qq and a mode of 7qq. Field observations revealed that even though yellow maize is available in the market, farmers prefer the white variety due to its lower price (see Figure 1). If there is no white maize, they would then turn to the yellow maize variety but this rarely occurred in the region of study. Therefore, yellow maize as an output is not included in the present analysis.

Sun drying, although cost-effective, increases the exposure of grain to microbial contamination, as well as to pests such as rats and birds. If the process requires many

days, the potential for mold growth and toxin production is exacerbated. In order to prevent further contamination of the maize, it is recommended to rapidly dry the maize using mechanical dryers. The cost of the latter can be a limiting factor for the majority of the farms given their poverty levels. Therefore in this financial analysis, drying cost is calculated as a service fee of 50 GTQ (~6.35 USD) per quintal, as opposed to purchase and maintenance cost of the machine, resulting in a more feasible amount to pay.

Consumer Price Index (CPI) from the Bank of Guatemala was used to adjust maize price data from 2004 to 2015 for inflation (data not shown). After adjustments, it seems that prices are more influenced by supply and demand than by inflation. This data was included in the analysis to predict future values for subsequent years (Banco de Guatemala 2014).

Survey results revealed that close to 80% of farms in the region have an average monthly consumption of 2.7 quintals of maize per month (Table 1). Therefore, it was assumed in the simulation that farmers would consume 2.7 quintals every month. This amount was used for all of the models subsequently described.

TABLE 1. Farmers estimated monthly maize consumption in households from Chiantla and Todos Santos, Huehuetenango, Guatemala

Range (qq)	**Average (qq)**	**Percentage (%)**
< **1.5**	1.2	9.1
1.5 - 5.0	2.7	77.3
> **5**	6.0	13.6

Seven scenarios were proposed: six regarding chain 2 farms and one regarding chain 1 farms (see Table 2). In each case, comparisons were made between a specific scenario (where a farmer obtained a silo and followed a specific maize purchasing regime), and a baseline scenario where there was no silo, and farmers had to buy maize on a monthly basis or every four months.

TABLE 2. Simulation scenarios for financial feasibility analysis for silo purchase.

Scenario	Chain 2						7, Chain 1
	1	**2**	**3**	**4**	**5**	**6**	
Farmers with silo	Farmer buys maize in November (harvest), and stores corn for the whole year.	Farmer buys maize in November (harvest), and stores corn for the whole year.	Farmer purchases and stores corn twice in a year: November and April.	Farmer purchases and stores corn twice in a year: November and April.	Farmer purchases and stores corn three times in a year: November, March and July.	Farmer purchases and stores corn three times in a year: November, March and July.	Farmer harvests corn in November, stores it, and consumes it until it is gone. Then farmer purchases and stores the quantity of corn needed for the rest of the year.
Farmers without silo (baseline)	Farmer buys maize every 4 months	Farmer buys maize on a monthly basis	Farmer buys maize every 4 months	Farmer buys maize on a monthly basis	Farmer buys maize every 4 months	Farmer buys maize on a monthly basis	

The baseline for scenarios one, three and seven reflects those farmers without silo who buy maize every four months, and consequently having an approximate grain loss of 5% of the purchases amount every 2 months (i.e. end of 2nd and 4th, 6th and 8th, and 10th and 12th month of storage). The baseline for scenarios two, four and six is reflective of those farmers who buy maize on a monthly basis, thus no losses were included as those losses occur only approximately after 2 months of storage.

Scenarios one and two describe farmers having a silo where maize is purchased for the entire year in 1 month. Losses during storage are minimum to none while using metal silos along with proper timely drying (Bravo Martinez 2009; Tefera et al. 2011). Consequently for scenarios including silos no loss was deducted. Since the farmers in this case would purchase all maize in one month, November prices were used to reflect maize value at harvest time (see Figure 1), this being the least expensive. Results from these two scenarios can be seen in Figure 3. Probabilistic histograms showed a cumulative probability of having paid off the investment in a 12 month period of approximately 0.43 and 0.27 for farmers who would otherwise be purchasing maize every four months or every month, respectively, with the former results being more attractive. For every scenario outcome, the cumulative probability of reaching a relative expense of 0 or above was calculated. The lower this cumulative probability was, the higher were the risks associated with purchasing a silo, and higher was the likelihood of the farmer accumulating a debt by the end of the year. The result for the farmer would likely be the forfeiting of the metal silo as collateral. Additionally, as the relative expense associated

with having the silo increases, so does the risk. Moreover, although maize price fluctuation throughout the years was taken into account in the simulations (period 2004-2013), there is a possibility for unexpected events (e.g. climatic phenomena) that may further increase the risk.

Scenarios three and four included farmers with a silo purchasing maize two times per year. November (harvest) and April were used for the analysis. Results are shown in Figure 4. Probabilistic histograms exhibited a cumulative probability of approximately 0.16 for a 4-month purchase period and 0.036 for the 1-month purchase period, the former, again, being a more attractive financial option.

Scenarios five and six describe farmers with a silo purchasing maize three times per year. Months of November, March and July were used for this analysis (see Figure 5). In these simulations there was no significant probability of an overall lower expense on the storage investment in a 12 month period.

Finally, scenario seven includes those farms where maize is planted, harvested and consumed, as well as bought when yield did not meet the household monthly requirements. Harvest yield was assigned randomly based on previous year data (MAGA 2015), and it was linked to the average monthly consumption of 2.7 qq in order to maintain food security. Probabilistic histograms showed a cumulative probability of return on investment in a 12 month period of approximately 0.13.

The highest reported cumulative probability among farmers from chain 2 for a break-even point (or better) was close to 0.3 (second scenario). For chain 1 the cumulative probability for the breakeven point was 0.128. This however reflects the financial scene merely for a 12 month period, when a 12 month loan was used for acquiring the technology.

Once silos have been paid, the presented scenarios would show a more attractive relative expense, becoming mostly negative (data not shown). After paying off the silos,

chain 1 farmers would still have to pay for the drying service every season to maintain the safety of the harvested lot, but the silo would not generate further costs. This results in additional income for farmers of both chains as they would have less losses during storage, they would maintain grain integrity and safety for a longer period, and in the case of chain 1 farms any surplus could be sold for additional income. Any future income could be used to either invest in additional silos or be used to diversify the household diet. Chain 2 farms would have the benefit of buying larger amounts of maize when market prices are convenient instead of resorting to buy maize in months when prices are quite high.

Traditional storage practices in Guatemala cannot guarantee protection against pests, leading to grain damage, fungal growth, and mycotoxin contamination. A metal silo is a suitable storage structure for grains, provided a drying regime prior to storage. Although this key food safety factor cannot be measured financially, it does have weight in the farmer's decision to adopt this technology storage.

Considering the long life span of a metal silo, its durability, and reduction or prevention of grain post-harvest losses, ultimately this storage technology can result in savings over traditional methods, it would allow smallholder farmers from the Western Highlands of Guatemala to achieve an economically stable living, allowing them to consume safe products, to diversify their diet, and improve their overall livelihoods.

ACKNOWLEDGEMENTS

Authors wish to acknowledge the NGO SHARE Guatemala for the field work in Huehuetenango. Research done at the Food Innovation Center’s laboratory is supported by United States Department of Agriculture (USDA) Scientific Cooperation and Research Program (SCRP); CFDA number 10.961.

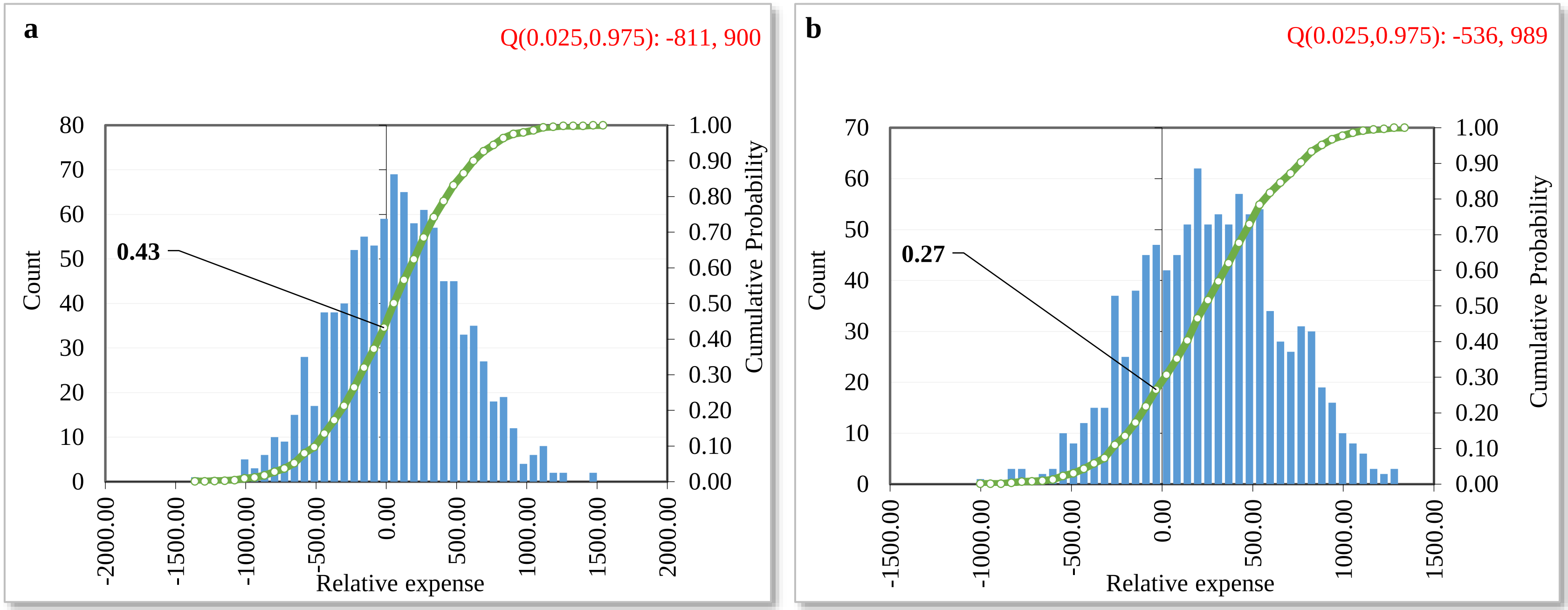


FIGURE 3. Probabilistic histogram of financial feasibility of silo purchase shown as relative expense. Chain 2 farmers purchased the silo, and buy all maize required to meet the household need for one year in November (harvest). a) Baseline scenario 1: Farmer without silo buys maize every 4 months. b) Baseline scenario 2: Farmer without silo buys maize on a monthly basis. Bar graph represent the probability of iterations of relative expense. Line graph represents the cumulative probability. Value shown in the chart associated with the relative expense of “zero” is the cumulative probability of paying the silo in a period of 12 months under the scenario considered.

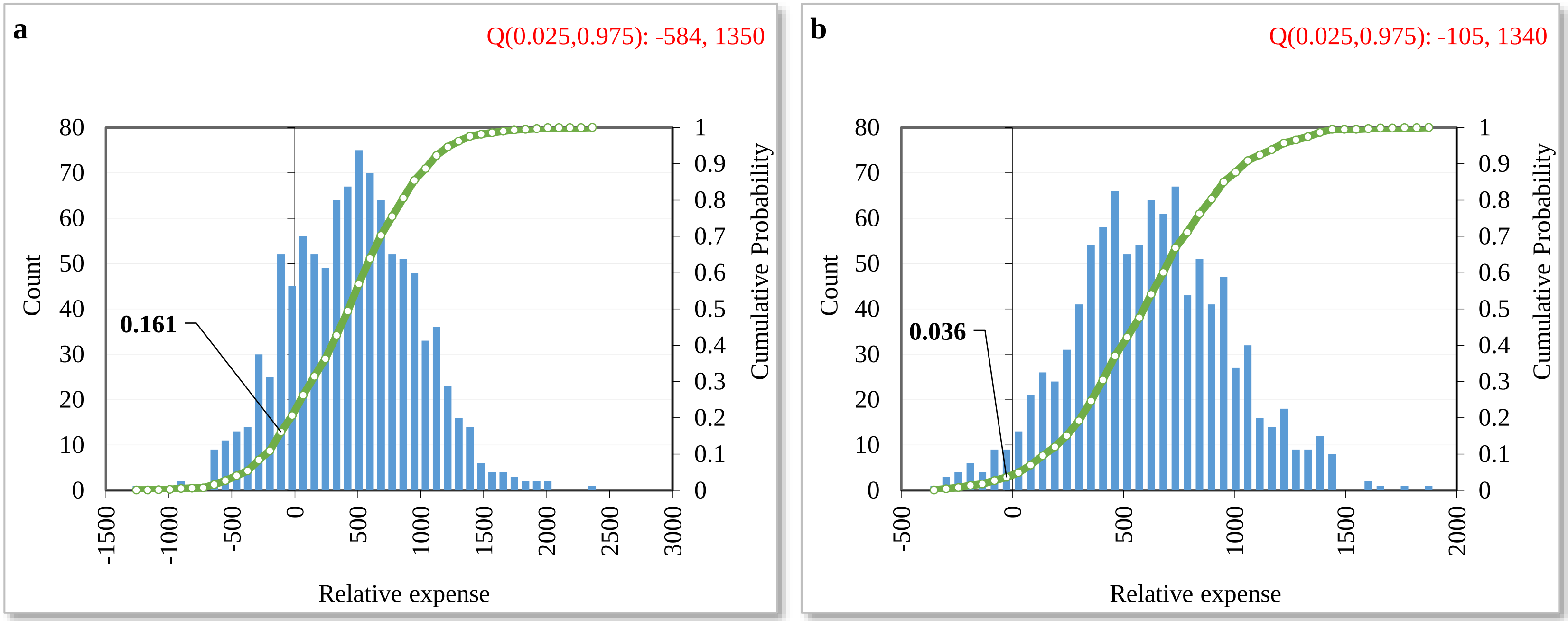


FIGURE 4. Probabilistic histogram of financial feasibility of silo purchase shown as relative expense. During November and April, Chain 2 farmers who purchased the silo, buy all maize required to meet the household need for one year. a) Baseline scenario 3: Farmer without silo buys maize every 4 months. b) Baseline scenario 4: Farmer without silo buys maize on a monthly basis. Bar graph represent the probability of iterations of relative expense. Line graph represents the cumulative probability. Value shown in the chart associated with the relative expense of “zero” is the cumulative probability of paying the silo in a period of 12 months under the scenario considered.

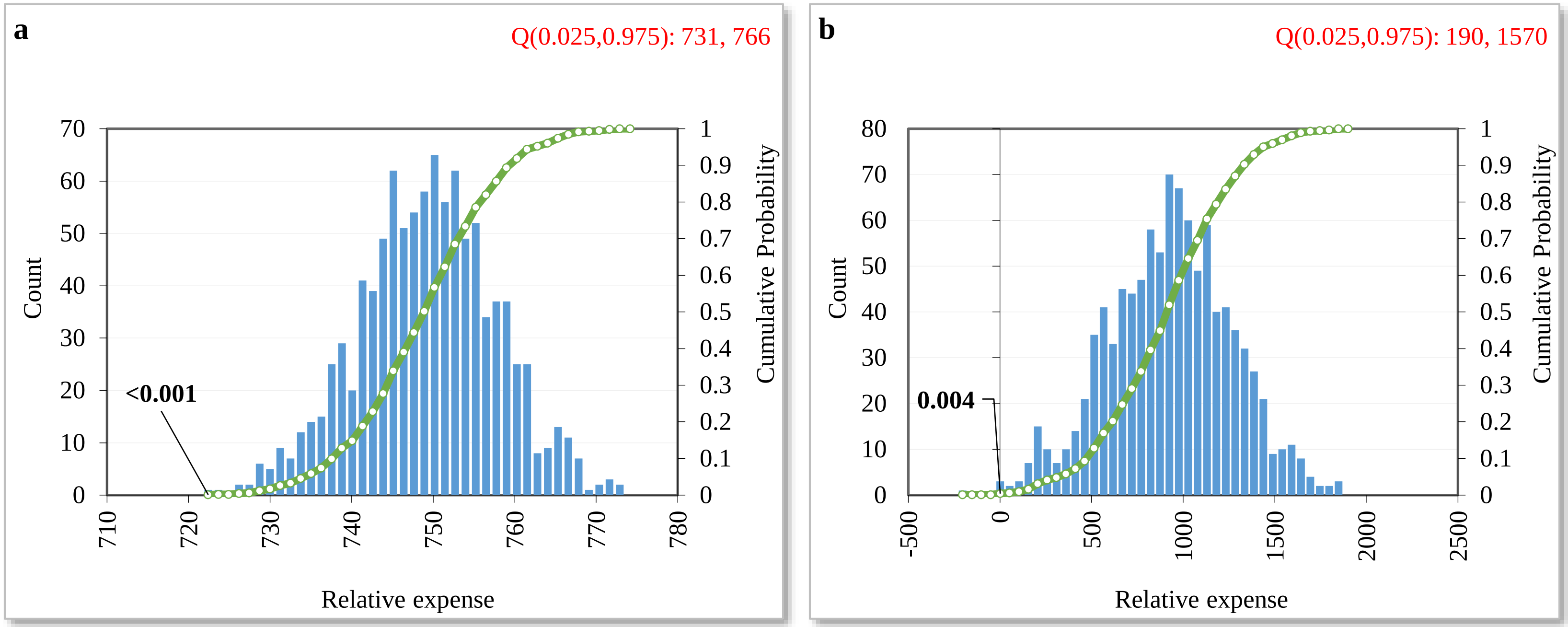


FIGURE 5. Probabilistic histogram of financial feasibility of silo purchase shown as relative expense. During November, March and July, Chain 2 farmers who purchased the silo, buy all maize required to meet the household need for one year. a) Baseline scenario 5: Farmer without silo buys maize every 4 months. b) Baseline scenario 6: Farmer without silo buys maize on a monthly basis. Bar graph represent the probability of iterations of relative expense. Line graph represents the cumulative probability. Value shown in the chart associated with the relative expense of “zero” is the cumulative probability of paying the silo in a period of 12 months under the scenario considered.

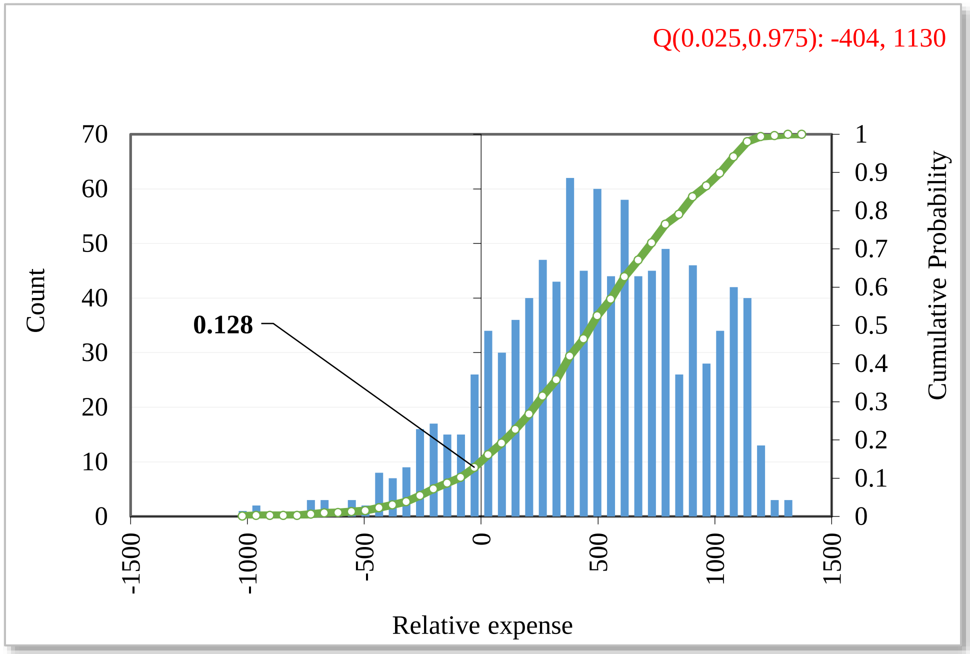


FIGURE 6. Probabilistic histogram of financial feasibility of silo purchase shown as relative expense. Scenario 7: Chain 1 farmers purchased a silo and buy maize once harvest is consumed. Baseline: Variable, dependent upon randomized harvest yield. Bar graph represent the probability of iterations of relative expense. Line graph represents the cumulative probability. Value shown in the chart associated with the relative expense of “zero” is the cumulative probability of paying the silo in a period of 12 months under the scenario considered.

REFERENCES

Alberta Agriculture and Forestry. 2010. “Cereal Grain Drying and Storage.” *Adapted from the Canada Grain Council’s Complete Guide to Wheat Management*. Available at: http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/crop1204 [Accessed January 7, 2016].

Banco de Guatemala. 2014. “Indice de precios al consumidor.” *Estadísticas económicas de Inflación*. Available at: http://www.banguat.gob.gt/inc/main.asp?id=1778&aud=1&lang=1 [Accessed April 6, 2016].

Barham, B.L., S. Boucher, and M.R. Carter. 1996. “Credit constraints, credit unions, and small-scale producers in Guatemala.” *World Development* 24(5):793–806.

Bokusheva, R., R. Finger, M. Fischler, R. Berlin, Y. Marín, F. Pérez, and F. Paiz. 2012. “Factors determining the adoption and impact of a postharvest storage technology.” *Food Security* 4:279–293.

Boxall, R.A. 2002. “Damage and loss caused by the Larger Grain Borer Prostephanus truncatus.” *Integrated Pest Management Reviews* 7(2):105–121.

Bravo Martinez, J. 2009. *Metal silos and food security. Lessons Learned from a Successful Central American Post-harvest program*. Quito, Ecuador.

Bravo, J. 2009. *Metal Silos and Food Security* 1st ed. Ibarra, Ecuador.

D.L. Park, H.N. and E.B. 1999. “Minimizing risks posed by mycotoxins utilizing the HACCP concept.” *FAO Corporate document repository*. Available at: http://www.fao.org/docrep/x2100t/x2100t08.htm [Accessed June 6, 2016].

Dawson, R.J. 1991. “Fungi and Mycotoxins in Stored Products.” In and J. I. P. B.R. Champ, E. Highley, A.D. Hocking, ed. *A global view of the mycotoxin problem*. Bangkok, Thailand, p. 50. Available at: http://aciar.gov.au/files/node/2152/fungi_and_mycotoxins_in_stored_products_part_1_34773.pdf.

Etten, J. Van, and R.M. Fuentes. 2004. “La Crisis Del Maíz En Guatemala: Las importaciones de maiz y la agricultura familiar.” *Anuario de Estudios Centroamericanos* 30(1-2):51–66. Available at:

http://revistas.ucr.ac.cr/index.php/anuario/article/view/1341.

FAO. 1994. “Annex 1 (Reference Chapter 1) - Application of cost-benefit analysis to storage projects.” In *Grain storage techniques - Evolution and trends in developing countries*. Available at: http://www.fao.org/docrep/t1838e/T1838E1o.htm.

FAO. 2014. *Manual técnico para la construcción y el uso de los silos metálicos familiares para almacenar cereales y leguminosas* 1st ed. A. A. Danilo Mejia, Matthew Howell, ed. Rome.

FAO. 2009. “Post-harvest losses aggravate hunger.” *News Article*. Available at: http://www.fao.org/news/story/en/item/36844/icode/ [Accessed June 6, 2016].

FAO, and IPGRI. 2002. *The role of women in the conservation of the genetic resources of maize* 1st ed. Rome. Available at: https://www.amherst.edu/media/view/92334/original/role+women+mais.pdf.

Hermann, H. 1991. “Seguridad alimentaria.: comparación de impactos socio-eoconomicas en la tenencia del silo métalico versu sistema tradicional de almacenamiento.” Available at: https://www.shareweb.ch/site/Agriculture-and-Food-Security/focusareas/Documents/phm_herrmann_seguridad_alimentaria.pdf.

Kalaitzandonakes, N.G., and E.G. Dunn. 1995. “Technical Efficiency , Managerial Ability and Farmer Education in Guatemalan Corn Production : A Latent Variable Analysis.” *Agricultural and Resource Economics Review* 24(1).

Leslie, J.F., and A.F. Logrieco. 2014. *Mycotoxin Reduction in Grain Chains* 1st ed. Manhattan, Kansas: John Wiley & Sons, Inc.

MAGA. 2015. “El Agro en Cifras 2014.”

MAGA. 2008. “Unidad especial de ejecucion del proyecto postcosecha MAGA.” Available at: https://www.donorplatform.org/index.php?option=com_cobalt&task=files.download&tmpl=component&id=1345&fid=15&fidx=0&rid=1709&return=aHR0cHM6Ly93d3cuZG9ub3JwbGF0Zm9ybS5vcmcvY29iYWx0P3N0YXJ0PTY2MA%3D%3D.

Manuel, I., R. Abel, G. Muñoz, B. Ramírez, V.J. Hilario, S.M. Bellon, R.F. Mexicana, I.M. Rosas, A.G. Muñoz, B.R. Valverde, and J.H.H. Salgado. 2007. “Physical and physiological seed quality of maize landraces stored in metalic silo and with traditional methods in Oaxaca, Mexico.” *Revista Fitotecnia Mexicana* 30:69–78.

Swiss Agency for Development and Co-operation. 2008a. “Central America : fighting poverty with silos and job creation.” *Latin Brief*:1–4. Available at: https://www.shareweb.ch/site/Agriculture-and-Food-Security/focusareas/Documents/phm_sdc_latin_brief_silos_central_america_e.pdf [Accessed January 1, 2016].

Swiss Agency for Development and Co-operation. 2008b. *Manual for Manufacturing Metal Silos for Grain Storage* 2nd ed. Postharvest National Programmes in Honduras, Guatemala, Nicaragua and El Salvador. Available at: https://www.shareweb.ch/site/Agriculture-and-Food-Security/focusareas/Documents/phm_postcosecha_manual_silos_e.pdf.

Tefera, T., F. Kanampiu, H. De Groote, J. Hellin, S. Mugo, S. Kimenju, Y. Beyene, P.M. Boddupalli, B. Shiferaw, and M. Banziger. 2011. “The metal silo: An effective grain storage technology for reducing post-harvest insect and pathogen losses in maize while improving smallholder farmers’ food security in developing countries.” *Crop Protection* 30(3):240–245. Available at: http://www.sciencedirect.com/science/article/pii/S0261219410003376.

Torres, O. a, E. Palencia, L. Lopez de Pratdesaba, R. Grajeda, M. Fuentes, M.C. Speer, A.H. Merrill, K. O’Donnell, C.W. Bacon, A.E. Glenn, and R.T. Riley. 2007. “Estimated fumonisin exposure in Guatemala is greatest in consumers of lowland maize.” *The Journal of nutrition* 137(12):2723–2729.

Torres, O., J. Matute, J. Gelineau-van Waes, J.R. Maddox, S.G. Gregory, A.E. Ashley-Koch, J.L. Showker, K.A. Voss, and R.T. Riley. 2015. “Human health implications from co-exposure to aflatoxins and fumonisins in maize-based foods in Latin America: Guatemala as a case study.” *World Mycotoxin Journal* 8(2):143–159. Available at: http://www.scopus.com/inward/record.url?eid=2-s2.0-84928343699&partnerID=tZOtx3y1\nhttp://wageningenacademic.metapress.com/index/D52007701363785T.pdf\nhttp://wageningenacademic.metapress.com/openurl.asp?genre=article&id=doi:10.3920/WMJ2014.1736.

Washington Office on Latin America (WOLA). 2013. “Government Investment in Family Agriculture New Opportunities in Mexico and Central America.” Available at: http://www.wola.org/files/Government_Investment_in_Family Agriculture.pdf.

Yusuf, B.L., and Y. He. 2011. “Design , development and techniques for controlling grains post-harvest losses with metal silo for small and medium scale farmers.” *African Journal of Biotechnology* 10(65):14552–14561.