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KERNEL AND BULK DENSITY CHANGES DUE TO MOISTURE CONTENT, MECHANICAL DAMAGE, AND INSECT DAMAGE

A Thesis
Submitted to the Faculty
of
Purdue University
by
Danping Guo

In Partial Fulfillment of the
Requirements for the Degree
of
Master of Science

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To my mentor and parents. I couldn’t have done this without all of you. Thank you all for your supports along the way.

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ABSTRACT

Guo, Danping. M.S., Purdue University, May 2015. Kernel and Bulk Density Changes due to Moisture Content, Mechanical Damage, and Insect Damage. Major Professor: Klein Ileleji.

Corn (*Zea Mays*), is one of the major grain crops in the world and moisture content, mechanical damage and insect damage are three factors that affect its quality. The primary goal of this thesis was to investigate the effects of moisture content, mechanical damage and insect damage on kernel and bulk density of corn. The study was conducted using two corn hybrids, Pioneer 1352 and Pioneer 1221, that were grown on Purdue Agronomy Farm for Research and Education (ACRE), manually picked and shelled. In Objective 1, the effect of three different moisture conditioning processes (drying from harvest moisture, rewetting from 30% to 10%, and rewetting to 30% before drying from 30% to 10%) on kernel and bulk density was investigated. In Objective 2, the effect of mechanically damaged kernels at various damage percent levels created by blending undamaged whole kernels with damaged kernels was investigated. Objective 3 investigated the effect of insect damage, both artificially simulated internal damage by drilling a hole per kernel and actual insect damage by infesting with *Sitophilus zeamais* (maize weevil) was investigated.

For Objective 1, in all three moisture conditioning processes, both kernel and bulk density were found negatively and linearly correlated to moisture content. In general, Pioneer 1221 had a higher kernel and bulk density than Pioneer 1352 for all moisture conditions. A comparison of the last two conditioning processes (rewetting from 10% to 30% and drying from 30% to 10%) showed that neither kernel density nor bulk density was significantly different. Additionally, comparisons of the data from this research and the empirical models by Nelson (1980) and Brusewitz (1975) showed agreement at either the low moisture or high moisture of each hybrid.

As mechanical damage level increased, the kernel density changed positively, and the bulk density changed negatively. For the artificially induced insect damage by drilling a hole per kernel, kernel density increased and bulk density decreased as artificial damage level increased. However, for the actual insect infestation treatments, the kernel density at different life stages of *Sitophilus zeamais* decreased in the larva and pupa stages and this trend reversed at the adult stage. Comparisons of the data with the control (un-infested kernels) seem to indicate that the internal infestation was the cause of this decrease. Further work need to be conducted in order to better explain the results and verify whether kernel density can be used as a distinguishable indicator of internal insect infestation in corn kernels.

CHAPTER 1. INTRODUCTION

1.1 Thesis Organization

This thesis presents the study on the effect of moisture content, mechanical damage and insect damage on corn kernel and bulk density. The study involved conditioning batches of two corn hybrids to various levels of moisture content, mechanical damage and insect damage and measuring the kernel and bulk density of these batches. Statistical analyses were applied to the data to determine the relationship of the independent variables (moisture, mechanical damage and insect damage) and the dependent variables (kernel and bulk density).

In this chapter, the problem is outlined by giving an overview of the importance of corn quality in trade, storage and processing by discussing quality parameters, included in the USDA-FGIS (USDA Federal Grains Inspection Service, 1996) grading standards. These include test weight, damaged kernels and broken corn and foreign material. This chapter concludes with a statement of the specific objectives of this research.

1.2 The Importance of Corn Quality

Corn, also known as maize, or *Zea Mays*, is one of the major crops in the world. The importance of developing tests for measuring corn physical properties can be gained by considering the large quantity of corn produced worldwide every year. Annually, a total of around 700 million metric tons of this valuable crop are harvested worldwide (Tiller, 2007). During 2005, the world production of corn was 706 million metric tons (MMT), of which 40% (280 MMT) was produced by the United States (USDA, 2005). Corn is also known as the largest crop of the Americas. There is over 7,000 years of history of corn production in the United States, having been cultivated by the original people (native Americans) of the United States before the coming of settlers from Europe. During this period of time, important uses of corn have been developed such as livestock feed, human food, beverage and food ingredients, industrial bio-based products and fuel ethanol production. In 2014, 83.1 million acres of corn were harvested in United States (USDA-NASS, 2015).

Corn can be divided into three classes based on the color: yellow corn, white corn, and mixed corn. The United States Department of Agriculture (USDA) Federal Grain Inspection Service (FGIS) has developed grade standards that classify corn into one of five U.S. numerical grades or to U.S. Sample grade. The grade requirements are summarized in Table 1.1.

In addition to the U.S. numerical grades and U.S. Sample grade, there are also special grades. The special grades are defined to emphasize special qualities or conditions affecting its end-use value, and the special grades are added to the grade designation without affecting the numerical grade designation. There are also four special grades: flint, flint and dent, infested and waxy. And those four special grades are defined in Table 1.2.

Table 1.1. U.S. numerical grades and U.S. Sample grade of corn (after USDA-FGIS, 1996).

Grade	Minimum test weight per bushel (lb/bu)	Maximum Limits of -----		
		Damaged Kernels		Broken Corn and Foreign Material (percent)
		Heat Damaged Kernels (Percent)	Total (Percent)	
U.S. No. 1	56.0	0.1	3.0	2.0
U.S. No. 2	54.0	0.2	5.0	3.0
U.S. No. 3	52.0	0.5	7.0	4.0
U.S. No. 4	49.0	1.0	10.0	5.0
U.S. No. 5	46.0	3.0	15.0	7.0

U.S. Sample grade is corn that:

a) Does not meet the requirements for the grades U.S. Nos. 1, 2, 3, 4, or 5; or
b) Contains stones which have an aggregate weight in excess of 0.1 percent of the sample weight, 2 or more pieces of glass, 3 or more crotalaria seeds (Crotalaria spp.), 2 or more castor beans (Ricinus communis L.), 4 or more particles of an unknown foreign substance(s) or a commonly recognized harmful or toxic substance(s), 8 or more cockleburs (Xanthium spp.) or similar seeds singly or in combination, or animal filth in excess of 0.20 percent in 1,000 grams; or
c) Has a musty, sour, or commercially objectionable foreign odor; or
d) Is heating or otherwise of distinctly low quality.

Table 1.2. Four special grades of corn (after USDA-FGIS, 1996).

Special grades	Definitions
Flint corn	Corn that consists of 95 percent of more of flint corn.
Flint and dent corn	Corn that consists of a mixture of flint and dent corn containing more than 5.0 percent but less than 95 percent of flint corn.
Infested corn	Corn that is infested with live weevils or other insects that are injurious to stored grain.
Waxy corn	Corn that consists of 95 percent or more waxy corn.

In the subsequent paragraphs, some of grade parameters such as test weight, damaged kernels, broken corn and foreign materials shown in Table 1.1 and 1.2 are discussed with respect to how they affect corn quality and as they are related to this study.

Test weight of corn is expressed as the weight of corn kernels per bushel and is a density measurement. It is the weight of kernels occupied in a standard volume (often 1 pint). Test weight is one of the important grade measures of corn and consequently, affects its selling price. Test weight or density can be affected by moisture, mechanical and insect damage because all three parameters affect either the kernel volume or the bulk or kernel weight. Brusewitz (1975) showed that bulk and kernel density decreased for most grains including corn when moisture content was increased up to 30%. However, there is lack of work on the relative changes in corn kernel and bulk density during drying or rewetting, which was investigated in this study.

Damaged kernels are kernels and pieces of corn kernels damaged by ground, weather, disease, frost, germ, heat, insect, mold, sprouting or other material. Among the different types of damage, insect and mold damage are two that often happen during storage. Broken corn is defined by the USDA-FGIS grading standard as all matter that passes through a 12/64 round-hole sieve and over a 6/64 round-hole sieve. The foreign material is all matter other than corn that passes through the 6/64 round-hole sieve but remains on the 12/64 round-hole sieve. For this study, we investigated both mechanical damage (chipped or broken kernels) and insect damage caused by the maize weevil, *Sitophilus zemais* (Motschulsky), which is an internal feeder of corn kernels. Both types of damage potentially affect kernel and bulk density because they cause the reduction of kernel and bulk weight, and also bulk volume in the case of mechanical damage.

Mechanical damage primarily occurs during combine harvesting. During handling, mechanically damaged kernels dried using high-temperature dryers and cooled rapidly are susceptible to stress cracks, which could lead to kernel breakage. Kernels with high levels of mechanical damage are also more susceptible to spoilage in storage and also low yields of starch during processing.

Insect damage often happens in a grain bin during storage and is promoted by a warm and humid storage environment. Internal feeders like the maize weevils consume the interior of the kernels and contaminate kernels with excrement and body parts, all of which have a strong influence on the quality of the grain. Since internal feeders cannot be seen from outside during inspection, kernels that seem good from outside might still have significant problems. What’s more, insects produce heat and moisture due to their metabolic activities, which is favorable to microorganism growth and hotspot development in grain. It is estimated that the total economic losses in Canada could be millions of dollars annually in grains and oilseeds due to stored-product pests and microorganisms (White, 1993). Therefore, USDA-FGIS has made strict standards for classifying grain as infested with insects, as shown in Table 1.3.

Table 1.3. Federal Grain Inspection Service standards for grain graded as infested.

Grain 1000g Sample	Number and Type of Insects
Wheat, triticale, rye	2 or more live weevils 1 live weevil and 1 other live insect injurious to stored grain, or 2 other live insects injurious to stored grain
All other grain	2 or more live weevils 1 live weevil and five other live insects injurious to stored grain, or 10 other live insects injurious to stored grain

1.3 Objectives

The major goal of this research was to investigate how kernel density and bulk density change in corn kernels due to changes in moisture content, mechanical damage and internal insect damage. The findings of this study will provide some fundamental understanding of the relationships of these important parameters and could potentially be used to understand factors that affect corn quality or improve measures of corn quality. The specific objectives of this research were as follows:

1) Evaluate the effect of the following treatments on corn kernel density and bulk density:
    a. Drying corn kernels immediately after they are harvested from the field.
    b. Artificially rewetting naturally dried kernels from 10% to 30% moisture content in 2% point increments.
    c. Artificially rewetting naturally dried kernels to 30% moisture content, then drying from 30% to 10% in 2% point decrements.

2) Determine the effect of mechanical damage levels (broken and chipped kernels induced using a grain breakage tester) on corn kernel density and bulk density.

3) Determine the effect of internal insect infestation on corn kernel density and bulk density by means of:
    a. Drilling holes in kernels to simulate internal insect infestation damage by emerged *Sitophilus zeamais* (maize weevil) adults.
    b. Infestation of corn kernels with *Sitophilus zeamais* at different stages of their life cycle.

1.4 Thesis Outline

The remainder of this thesis is divided into five chapters. In Chapter 2, literature pertaining to the relationship between density and moisture content, the effect of mechanical damage on quality of corn kernels, and how insect damage might affect kernel and bulk density and methods of internal insect detection in grains are presented. The equipment, materials and experimental design are presented in Chapter 3. Chapter 4 describes the results and analyses of the experiments. In Chapter 5, conclusions and recommendations are made.

CHAPTER 2. REVIEW OF LITERATURE

2.1 Introduction

This chapter is a review of the literature. Section 2.2 introduces two important physical properties of grain, which are kernel density and bulk density. Also, moisture-dependent relationships of bulk and kernel density have been reviewed. In Section 2.3, the literature review of grain mechanical damage and its effect on grain quality is discussed. Section 2.4 introduces the detection methods of internal infestation in grains and the potential effect of insect damage on kernel and bulk density. Lastly, the maize weevil, *Sitophilus zeamais* (Motschulsky) and its life stages, as one of the most common internal feeders of corn kernels is discussed.

2.2 Moisture-dependent Relationships of Bulk and Kernel Density

Grain density is an important physical property that is used as an indicator of quality. The USDA-FGIS grading standard defines the test weight as the amount of weight in pounds (lb) contained in a given volume expressed in bushels (~1.25 ft^3). Test weight is also a measure of grain bulk density, which can be expressed as kg/m^3.

Typically good quality corn of standard trade moisture content (15.5% wet basis according to USDA-FGIS) is expected to have a good test weight. Typically, high test weight means the kernels have a higher percentage of hard endosperm. Also, it is one of the quality tests run on corn that is used to decide premiums or discounts received at sale. The official minimum allowable test weight in the United States for No. 1 yellow corn is 56 lb/bu and for No. 2 yellow corn is 54 lb/bu (USDA-GIPSA, 1996). Grain test weights usually vary depending on moisture and hybrid. Various physical factors can affect the grain test weight, such as hybrid, kernel maturity, presence of diseased and mold infested kernels and mechanical damage, but the primary one is the grain moisture content. Moisture content, the amount of water in the kernel can be expressed on a percent wet basis (w.b.) by subtracting the amount of kernel dry matter from the total wet mass and dividing by the total wet mass. Because the kernel dry matter is denser than water, the bulk density should increase with a decrease of kernel moisture. It is known that there is an inverse relationship between bulk density and moisture content in the range from 10 to 30 percent moisture. In addition, other factors like kernel size and shape, thickness of seed-coat also influence the test weight of grains (Seglar et al., 2011).

There are three kinds of density that relate to the density of a single particle, which could be used to express the kernel density. The first is the true density, which is the weight per unit volume of the solid particle that excludes any internal and external pores. The true density could be determined via chemical analysis (Wassgren, 2015). The second is the apparent (aka skeletal) density, which is the weight per unit volume of the solid particle, which includes internal pores but excludes external pores. It is usually measured using a gas pycnometer. The third is the envelope (aerodynamic) density, which is the weight per unit volume of the solid materials including both internal pores and external pores. These three kinds of particle density can be described using the following equations (Equation 2.1, 2.2 and 2.3):

$$\rho_{true} = \frac{m}{v_{solid}} \tag{2.1}$$

$$\rho_{apparent} = \frac{m}{v_{solid} + v_{internal\,pores}} \tag{2.2}$$

$$\rho_{envelope} = \frac{m}{v_{solid} + v_{internal\,pores} + v_{external\,pores}} \tag{2.3}$$

From these equations we can see that $\rho_{true} \geq \rho_{apparent} \geq \rho_{envelope}$ (Wassgren, 2015). The kernel density in this research represents the apparent density, which includes the internal pores. Chang (1988) reported that the kernel density for corn, wheat and sorghum at 11 to 13% moisture content varied between 1.258 and 1.396 g/cm^3. Like bulk density, the kernel density is highly dependent on moisture content. Therefore, the kernel density determination should be accompanied with moisture content determination.

Moisture content is also an important physical property of grain. Though moisture content is not a factor when grading grains, it does affect the grain grades by influencing density, and in making grain storage and subsequent handling and storage decisions such as the need to dry. Kernel density is another important parameter, which affects the kernel hardness, breakage susceptibility, milling, drying rate, and resistance to fungal development (Chang, 1988).

Previous research on the influence of moisture content on bulk density and kernel density have been conducted and relationships of moisture-dependent bulk and kernel density have been published (Chung and Converse, 1971; Hall, 1972; Gustafson and Hall, 1972; Hall and Hill, 1974). Miles (1937) investigated the relationship between the weight per measured bushel and moisture content and found that the association was negative in the range from 10 to 30 % moisture. Browne (1962) investigated the relationship between the moisture content and bulk density using rewetted wheat, barley, and oats, and found that the test weight decreased with increase in moisture content.

Brusewitz (1975) found that most types of grain he tested decreased in bulk density with increasing moisture content up to 30%. In his research, a one-pint Boerner test weight apparatus was used to measure bulk density following the procedure developed by Boerner (1922). The averaged data for bulk density were plotted as a second-degree polynomial equation as a function of moisture content (Equation 2.4).

$$\rho_b = 1.0863 - 2.971M + 4.81M^2 \tag{2.4}$$

$$r = 0.921$$

Nelson (1980) measured both bulk density and kernel density of 21 lots of shelled, yellow-dent field corn over moisture-content ranges from 10% to 35%. Averaged data over all lots were presented graphically, and kernel-density and bulk-density dependence on moisture content were described with third-order and fourth-order polynomial equations, respectively. In his experiment, moisture content was determined by drying 2-g ground samples for 3h at 130ºC. For bulk density measurements, a sample holder with a volume 116.485 cm^3 was used following consistent procedures in filling the coaxial sample holder (Nelson, 1978). Kernel density was calculated from kernel-volume measurements obtained with a Beckman model 930 air comparison pycnometer. Equations 2.5 and 2.6 were developed by Nelson (1980) for estimating kernel and bulk density of shelled corn as a function of moisture content over the range of 10% to 35%.

$$\rho_k = 1.2519 + 0.00714m - 0.0005971m^2 + 0.00001088m^3 \qquad (2.5)$$

$$r = 0.998$$

$$\rho_b = 0.6829 + 0.01422m - 0.0009843m^2 + 0.00001548m^3 \qquad (2.6)$$

$$r = 0.996$$

Grain kernels were rewetted by adding distilled water. Small differences in test weight were found between corn kernels dried for the first time and rewetted corn kernels. Subsequent drying and rewetting resulted in smaller differences (Brusewitz 1975). Chung and Converse (1971) found that there were small hysteresis differences in test weight during absorption and desorption in wheat. Brusewitz (1975) indicated that the differences were greater for corn than for wheat.

2.3 Mechanical Damage

Mechanical damage is an important factor to evaluate during harvesting, handling, and marketing of corn kernels. The use of the grain combine and field-shelling attachment for corn pickers has resulted in mechanical damage to corn kernels. Mechanical damage to kernels mostly occurs when the moisture content is relatively high. Because of the development of grain dryers, corn kernels can be harvested at high moisture levels, which makes kernels more susceptible to damage. Mechanical damage to corn kernels can increase rapidly when the moisture content is above 20% (Waelti et al., 1969).

Mechanical damage in corn kernels could affect both short-term and long-term storage. It is reported that machine-shelled corn, with 29% mechanical damage, could deteriorate two to three times faster than hand-shelled kernels without damage (Waelti et al., 1969). When there are mechanical damages, fungal invasion inside corn kernels become easier and insect infestation is greater. Kalbasi-Ashtari et al. (1979) found that mechanically shelled corn deteriorated 2 to 3 times more than hand shelled corn. Also, mechanical damage increases breakage susceptibility in kernels, which can affect subsequent handling. Saul and Steel (1996) reported that the energy costs needed for drying mechanically damaged kernels increased by six to seven times over the energy required to dry hand-shelled kernels without damage, because damaged kernels needed faster drying rates in order to prevent deterioration. What is more, mechanical damage could also result in lower oil recovery, poorer milling ability and greater nutrient loss compared to undamaged kernels (Freeman, 1970).

Mechanical damaged corn as expressed by broken corn and foreign material is determined by passing 250 g of kernels through a 4.76mm (12/64 in) round-hole sieve according to United States Department of Agriculture- Federal Grain Inspection Service (USDA 1999). This method, however, does not include all mechanically damaged kernels ranging from hairline cracks to severe damaged ones. Chowdhury and Buchele (1978a) developed a numerical damage index by using one of the biological properties of the grain, germination in this instance, for critical evaluation of mechanical damaged corn. This numerical damage index can be a more effective measure of mechanical damage and represents both quantity (percentage) and quality (severity) of the damaged kernels.

Many methods have been developed to evaluate mechanical damage. The most commonly used one is visual inspection. Kernels with any visual damage or cracks are picked from the sample to estimate the damage percentage (Koehler 1957). Germination test is another way to estimate kernel mechanical damage. Germination estimates mechanical damage by correlating the ability of grain kernels to emerge and develop a healthy seedling (Al-Mahasneh et al. 2001). However, this method reports not only mechanical damage, but also other types of kernel damages (Chowdhury and Buchele 1978b). Dielectric properties of damaged corn were successfully used to develop a damage level prediction sensor (Al-Mahasneh et al. 2001). Machine vision is also another way of measuring corn kernel mechanical damage. It determines the mechanical damage level by extracting the damaged area stained by green dye from kernel images, and calculating the percentage of total projected kernel surface area that stained green (Ng et al., 1998).

2.4 Detection of Internal Insect Infestation in Grain

Corn could be stored for several years after drying to a safe moisture. Under the proper environment, corn kernels can have little or even no detectable quality losses. However, if the environment is improper such as high relative humidity and temperature, it is easy to have spoilage. For short-term storage, the spoilage usually means the result of microorganisms including bacteria, yeast, and fungi. If the storage time is longer than six months, it is important to monitor for potential damage by insect pests. Stored products insect pests can be categorized as primary and secondary pests. Secondary pest are external feeders such as beetles (red flour beetle, confused flour beetle, saw-toothed grain beetle), which consume broken kernels, fines and flour fragments of grain, and it is difficult to chew through whole intact kernels. Thus, as long as kernels remain intact and not damaged, it is difficult for them to feed on. On the other hand, primary insect pests, also known as internal feeders are capable of attacking whole grain kernels and typically infest kernels by chewing into kernels and in some species, the life stages develop inside the kernel till the adult emerges out of the kernel. Insects such as the maize weevil, rice weevil, granary weevil and the lesser grain borer are internal grain feeders. Because of the acute damages caused by internal grain feeders, there are very strict export control restrictions in place should a lot of grain be found contaminated with them. For example, the Canada Grain Act implements a zero tolerance for stored-product insects in Grain (Canada Grain Act 1975). Also, as was discussed in the introduction and shown on Table 1.3, he USDA-FGIS has very strict and low tolerance levels for internal grain feeders.

Infestation of kernels by internal feeders causes huge losses. Also, it is very difficult to detect internal infestation, especially at early stages of infestation. Thus, it is very important to find a way to detect the internal infestation in grain kernels. While mass loss from insects feeding on the germ and endosperm is typically incurred, there have been limited studies carried out to understand the relationship between kernel density and the growth stage of the insect inside the kernel. Understanding the relationship between the kernel density and internal insect infestation might be important in developing new methods to detect internal insect infestation in grains.

Monitoring of insect infestation is a fundamental part of managing stored grain. Various techniques have been used for detecting hidden insects in whole kernels. Infestation of grains can be detected by staining secretions (egg plugs) or body fluids of insects (hemolymph) and entry holes (Frankenfeld, 1948). Also flotation methods have been developed to detect internal infestation by using suitable salt solutions with whole grains (White, 1957) or a mixture of alcohol solution and light mineral oil with grounded grains (AOAC, 1997). Howe and Oxley (1944) proposed the use of carbon dioxide (CO_2) produced by insect respiration in food grains and grain products as an indicator of internal insect infestation. Uric-acid measurement has been applied to hidden insect infestation since 1950s (Subrahmanyan et al., 1955; Venkatrao et al., 1957).

One effective imaging technique used in detecting internal insect infestation in grain uses X-rays. The technique has been extensively applied in detecting internal damage in food grains and in investigating the growth and development of insects (Shah and Khan, 2014) and it has become an official method for detecting hidden infestations in the United States (AACC, 1995). The X-ray method was first used by Milner et al. (1950b) in detecting hidden insects in grains. The X-ray method is reliable, and accurate in detecting internal grain feeders. It is reported that hidden insects at different life stages could be identified by the soft X-ray with greater than 96% accuracy (Karunakaran et al., 2003). According to Pearson et al. (2003), at least four life stages of insect could be classified by X-ray techniques by measuring the occupied area by the insect. Additionally, the X-ray is a non-destructive and direct method to detect insect infestations (Milner et al., 1952; Stermer, 1972). The X-ray method needs expensive machine to generate X-rays, and also an experienced personnel is required to operate the machine and interpret the radiographs. The type of grain, the degree of penetration, and the contrast required determines a required exposure time and voltage of X-rays (Rajendran, 1999). Typically higher moisture of grains would need a higher voltage for the penetration of X-rays (Semple, 1992), and the denser the matter, the greater the X-ray absorption. The time for completing an analysis is about 2.5h, which is longer than other methods like cracking and flotation methods that typically takes less than 1.5h (Brader et al., 2002). However, Haff and Slaughter (2004) proposed the use of real-time digital imaging rather than X-ray film for discriminating infested kernels, which has the possibility to shorten the X-ray procedure significantly.

Other imaging techniques such as near-infrared reflectance spectroscopy (NIRS) can also detect hidden infestations. The NIR spectroscopy has evolved as a fast, reliable and accurate method for grain analysis. The NIR technology is based on the absorption of electromagnetic wavelengths. The NIR method can be used to detect external and internal insect infestation in wheat (Ridgway & Chambers, 1996). Perez-Mendoza et al. (2003) determined that NIR is a rapid method and there is no need to prepare sample before experiment. However, NIR could not detect low levels of infestation in bulk samples. Also, it cannot differentiate between live and dead insects (Dowell et al., 1999).

Acoustic techniques are another effective technique. Brain (1924) suggested that hidden infestation in food products could be detected by amplifying the sounds of feeding and movements of the insect larvae inside the kernel. Acoustical detection methods can detect both internal and external insect infestation by amplifying and filtering sounds of their movements and feeding. Adams et al. (1953) suggested that the acoustical method could be a method to detect hidden infestation " without sampling or removing the grain from the bins in much the same manner as permanent thermocouple systems are now used for checking the heating of grain in storage". Thus, acoustical systems can be a quick and easy method that has the potential to detect hidden insects automatically and has an advantage over carbon dioxide and X-ray methods. The first studies using acoustic techniques to detect insect activities inside single kernels of grain used microphones and phonograph cartridges (Adams et al. 1953, Bailey and McGabe, 1965). In more recent acoustical measurement system for insects, a high frequency detector (40 kHz) was used to study insect feeding activities of cowpea weevils *Callosobruchus maculatus* (F.) inside cowpeas (Shade et al. 1990). Although these sensors could lower the background sound levels, they are still limited by the requirement of having the sensors be in contact with infested grains. Table 2.1 shows the advantages and disadvantages of various detection techniques (Rajendran, 2005).

Table 2.1. Advantages and disadvantages of various insect detection methods.

Test Method	Pros	Cons
Flotation method	Simple, quick, and requires minimum laboratory facilities.	Cannot tell species and insect stage, not suitable for all kinds of grains.
Staining techniques	Simple, and needs very little training.	Can result in false positives, destructive, and not suitable for all kinds of grains and all insect stages.
Acoustic	Automated, computer-based, continuous, quick and easy, grain samples do not need to be removed, and nondestructive.	Cannot detect dead insects and early stage of infestations like larvae.
X-ray method	Can detect both dead and live insects, able to tell the species and insect stages, highly accurate, and nondestructive.	High initial costs, ongoing costs and chemicals to develop X-ray film; high labor fee.
Near infrared spectroscopy (NIRS)	Rapid, and no sample preparation.	Not sensitive to low levels of infestation, affected by grain moisture content, complex and frequent equipment calibration.
CO_2 analysis	Rapid, low equipment cost, and low labor fee cost.	Not all the CO_2 measured is produced by insects, some can be attributed to the grain sample; not quantitative.

2.5 Maize Weevil, *Sitophilus zeamais* (Motschulsky) and Its Life Stage

The maize weevil, *Sitophilus zeamais* Motschulsky, is a species of beetle in the family Curculionidae. Typically it is a major pest of stored corn throughout the corn-growing regions of the world (Throne, 1986). It is 1/8 – 3/16 inch long, and varies from dull red-brown to nearly black and usually marked on the back with four light reddish or yellowish spots (Figure 2.1). The maize weevil is one of the major internal feeders on cereal grains. The life stages of maize weevil include egg, larva, pupa and adult. Before maize weevils become adults, all the other life stages (larva and pupa) feed inside the kernel until it emerges out as an adult. Therefore, fumigation is the only way to kill the immature stages within the kernels.

Figure 2.1. Maize weevil.

Adults of maize weevil could live 5-8 months, and each female could lay around 300 eggs in their whole life. The minimum life cycle from egg to adult is about 30 days, and the minimum temperature for development is above 12.8^{o}C (55ºF). An index of environmental suitability indicated that the optimal environment for maize weevil populations' growth on corn is 30ºC and 75% RH (Throne, 1994).

The female maize weevil deposit their eggs in holes bored into the grain kernel. After depositing her egg in this cavity, the female seals the opening with a gelatinous secretion. The eggs then hatch into the larval stage. By feeding inside the kernels, the larva grows into a pupa. The eggs and immature stages of maize weevil hide within the kernels of corn, which are invisible to the naked eyes. Ordinary physical inspection methods sometimes cannot detect internal infestations. Thus, kernels that look uninfested might actually be internally infested. Figure 2.2 shows the timeline of the development of the different life stages of the maize weevil.

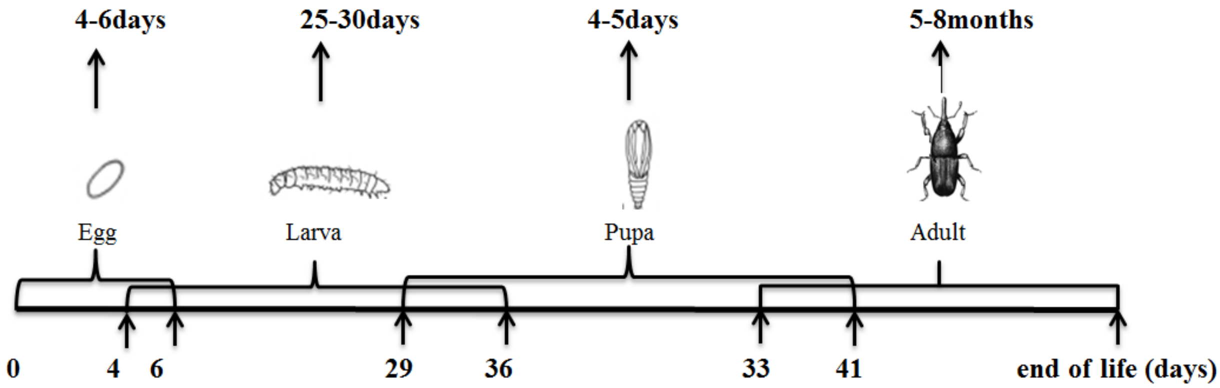


Figure 2.2. Timeline of the developmental stages of the maize weevil.

CHAPTER 3. MATERIALS AND METHODS

In this chapter, the materials used for the experiments and the test procedures used are introduced, including all equipment that were used in this research. The details of the samples are discussed in more detail in Section 3.1. In Section 3.2, corn sample treatments and methodology are described, including experimental design and all procedures used for corn sample treatments. Equipment used were also described in this section. Finally, Section 3.3 describes the statistical methods used to analyze the data.

3.1 Corn Sample Used for this Study

Two hybrids of manually shelled yellow dent corn were used in this research. They were Pioneer 1352 and Pioneer 1221. All of them were hand-picked from Agronomy Center for Research and Education (ACRE) of Purdue University located on state road (SR) 52 west during the fall of 2014. The corn husks were manually removed in the field after picking and ears were brought back to the lab. When the samples from the field arrived at the laboratory, they were hand-shelled with a Decker hand corn sheller (shown in Figure

3.1). The kernels were placed in trays on the laboratory bench after shelling, and allowed to dry naturally in the lab at about 24ºC room temperature. After drying to around 10% moisture content, the kernels were sealed in Ziploc® bags and stored in a walk-in cooler (5ºC) until needed. All kernels used in this research were damage free. Moisture contents are reported on a wet basis (w.b.). Details of two samples are given in Table 3.1.

Table 3.1. Details of corn hybrids used.

Hybrid	**Initial MC**	**Final MC**	**Date collected**	**Source**
Pioneer 1352	31.5%	11.8%	09/25/2014	From ACRE
Pioneer 1221	17.5%	8.1%	10/23/2014	From ACRE

Figure 3.1. Decker hand corn sheller.

3.2 Corn Treatments and Methodology

In this section, the treatments for each objective are introduced. In Section 3.2.1, three treatments are described to evaluate the effect of moisture content on kernel and bulk density. The methods to determine the effect of mechanical damage levels on kernel and bulk density are discussed in Section 3.2.2. Finally, an overview of how the effect of insect infestation on kernel and bulk density were evaluated are given.

3.2.1 Moisture Content Conditioning and Methods

In objective 1, corn kernels were conditioned to various moisture levels by drying or rewetting while the kernel and bulk density were measured at these moisture contents. In these treatments, the dependent variables were kernel and bulk density and the independent variable was moisture content.

Three different moisture conditioning treatments were performed as shown in Figure 3.2. In the first treatment, corn kernels were dried naturally in the lab from the field-harvested moisture level (31.5% for Pioneer 1352 and 17.5% for Pioneer 1221) down to around 10% moisture. About 5000g kernels of each hybrid were used in this treatment. During this drying process, moisture content was first measured every day using a John Deere moisture meter (Model 38900, Deere & Company Moline, Illinois, U.S.) (Figure 3.3). The goal was to measure both bulk and kernel density daily for every 2 percent or more point decrease in moisture content during the natural drying process. Thus, the John Deere moisture meter was used as a quick tool to determine moisture of the corn during drying. If there was a 2% difference in moisture compared with the reading of last day, then the kernel and bulk density were measured, and also moisture content was determined using the more accurate air oven method. As would be expected, the drying rate was not constant and the moisture content first decreased rapidly when it was relatively high and more slowly as the moisture decreased. Therefore, the moisture loss rate was not constant throughout conditioning by drying. Moisture content was accurately determined with triplicate 15g samples by means of the whole corn kernel air-oven method by drying samples for 72h at 103 ºC (ASAE, 2012). The air oven used is shown in Figure 3.4 (Model 21-250, Gilson Company Inc., Lewis Center, Ohio, U.S.). Upon completion of drying, both hybrids were divided into two equal halves with a Boerner Sample Divider and labelled as S_1 and S_2 (for both hybrids). Dried samples were sealed in Ziploc® bags and placed in a walk-in cooler at 5°C. Table 3.2 shows the information for corn samples before and after the drying process.

Table 3.2. The mass and moisture content of samples before and after the first treatment.

Hybrid	MC % at Harvest**	MC% after drying	Initial weight (g)*	Final Weight S_1 (g)	Final Weight S_2 (g)
Pioneer 1352	31.5	11.8	5000	1803	1805
Pioneer 1221	17.5	8.1	5000	2130	2128

*All weights in this table were rounded to the whole number
**MC are reported on a wet basis

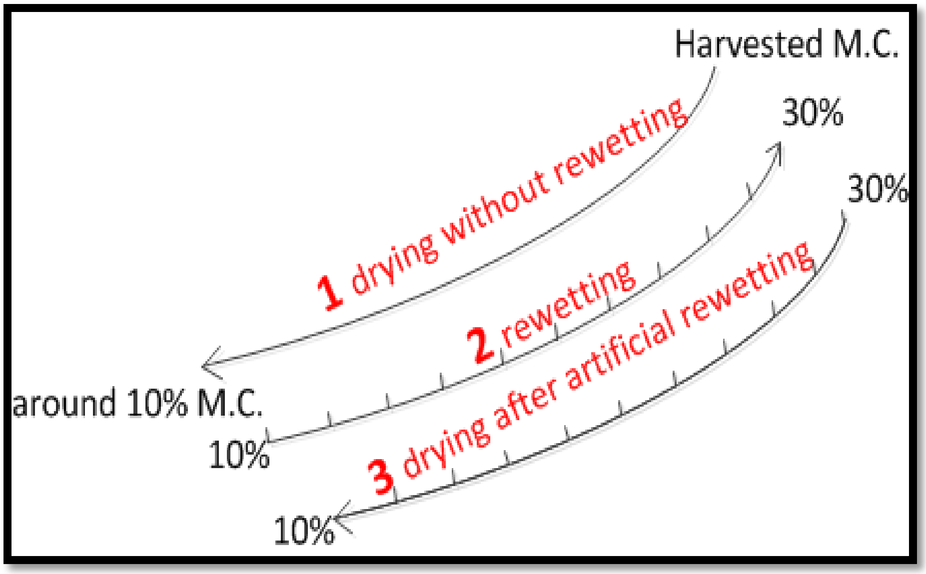


Figure 3.2. Three conditioning treatments.

Figure 3.3. John Deere Model 38900 moisture meter.

Figure 3.4. Air Oven.

In the second treatment, corn kernels were conditioned by rewetting kernels with a predetermined quantity of distilled water in order to increase moisture from 10% to 30% moisture content in 2 percent points increments. For each hybrid, Sample S_1 (shown in Table 3.2) was used in this treatment. When samples were taken out of the walk-cooler for moisture conditioning, they were left to warm up to the room temperature by leaving on the bench still sealed air-tight in the Ziploc® bag.

For rewetting, the mass of water needed to achieve the target moisture level was determined by Equation 3.1.

$$\mathrm{M_{initial}} \times (1 - \mathrm{M.C._{initial}}) = (\mathrm{M_{initial}} + \mathrm{M_{water}}) \times (1 - \mathrm{M.C._{target}}) \quad (3.1)$$

Where:

$\mathrm{M_{initial}}$ is weight of sample before rewetting

$\mathrm{M.C._{initial}}$ is the moisture content before rewetting

$\mathrm{M_{water}}$ is the water needed for rewetting to the target moisture level

$\mathrm{M.C._{target}}$ is the target moisture content after rewetting

The mass of distilled water needed to achieve the target moisture level was measured using an electronic balance in a beaker. About 0.5g extra distilled water was added to compensate for the loss of water adhering to the wall of the beaker. To ensure uniform rewetting, the sample was placed in a plastic container (Figure 3.5) that was rotated on a tumbler (Figure 3.6) for 4 h. The rewetted kernels were sealed in the Ziploc® bag again and placed to equilibrate in the walk-in cooler for 24 h prior to testing. Both hybrids, S_1 sample lost moisture to below 10% during storage. Therefore they were first conditioned to 10% moisture to begin the study and then conditioned to higher moisture contents in 2 percent points increment up to 30%. At each moisture level, the moisture content, kernel and bulk density, after each moisture increment were measured (Table 3.3).

Figure 3.5. Container used for rewetting.

Figure 3.6. Tumbler device for rotating container to ensure thorough mixing.

Table 3.3. Moisture content levels during rewetting and tests conducted at each moisture level.

Rewetting Process	Moisture content: 10% → 12% → 14% →16% → 18% → 20%→ 22% → 24% → 26% → 28% → 30%
Tests	Kernel density, bulk density and moisture content

In the third treatment, corn kernels were rewetted to 30% first, then dried from 30% to 10% in 2 percent point decrements. The samples used in this treatment were the S_2 sample that had been stored in the walk-in cooler after the first treatment. Corn kernels were rewetted directly to 30% moisture. The rewetting procedure was the same as that used in the second treatment. Because a larger quantity of distilled water was added for conditioning kernels to 30%, the container was rotated on the tumbler overnight. After rewetting, samples were sealed in a Ziploc® bag and placed in the walk-in cooler to equilibrate for 24 h.

Before starting the drying process, samples were left sealed in the Ziploc® bag to equilibrate to the room temperature. Then samples were left in thin layers on a tray to dry at room temperature from 30% to 10% moisture level in 2 percent point decrements. During the drying process, both the John Deere moisture meter and weight loss measurements were used to check when drying had been completed. Equation 3.2 was used to determine the sample weight that should have been achieved for a given target moisture.

Table 3.4. Moisture content levels during drying and tests conducted at each moisture level.

Rewetting Process	Moisture content: 30% → 28% → 26% →24% → 22% → 20%→ 18% → 16% → 14% → 12% → 10%
Tests	Kernel density, bulk density and moisture content

$$\mathrm{M}_{\mathrm{initial}} \times (1 - \mathrm{M.C.}_{\mathrm{initial}}) = \mathrm{M}_{\mathrm{final}} \times (1 - \mathrm{M.C.}_{\mathrm{target}}) \qquad (3.2)$$

Where:

$\mathrm{M}_{\mathrm{initial}}$ is the mass at the previous moisture level before rewetting

$\mathrm{M.C.}_{\mathrm{initial}}$ is the moisture content at the previous moisture level before rewetting

$\mathrm{M}_{\mathrm{final}}$ is the mass after rewetting

$\mathrm{M.C.}_{\mathrm{target}}$ is the target moisture content after rewetting

3.2.2 Mechanical Damage Levels and Methods

In this treatment, the dependent variables were kernel and bulk density, and the independent variable was the mechanical damage level. The objective of this investigation was to determine the effect of mechanical damage on kernel and bulk density.

For each hybrid, about 14500g of kernels were prepared. Prior to the experiments, corn kernels had been stored sealed in Ziploc® bags and stored in the walk- in cooler. In this treatment, corn kernels were conditioned to 15% moisture content using the same procedures as previously described 3.2.1. Details of sample moisture contents are shown in Table 3.5.

Table 3.5. Details of sample moisture content used for the mechanical damage study.

Pioneer 1352		Pioneer 1221	
Initial Moisture Content[a] (% w.b.)	Moisture Content (% w.b.) after Rewetting[b]	Initial Moisture Content[a] (% w.b.)	Moisture Content (% w.b.) after Rewetting[b]
8.7	15.1	7.4	15.0

[a] Average initial moisture content of samples after storage in the cooler

[b] Average moisture content after rewetting to 15% moisture content

To create mechanical damage on kernels, the Grain Breakage Tester (Serial C011P, Grain Research Laboratory, Minneapolis, MN, U.S.) was used. Samples of 15% moisture corn were passed through the instrument. Sound kernels were mixed with damaged kernels to create five different levels of mechanical damage. For each damage level, 1000g total weight of corn kernels were prepared. The weights of damaged kernels and sound kernels at different damage levels are shown below (Table 3.6).

Samples at different mechanical damaged levels shown in Table 3.6 would need 1950g damaged kernels and 3050g sound kernels. In order to prepare enough samples, 2500g kernels were sent to the Grain Breakage Tester and 3500g sound kernels were prepared for subsequent mixing. After mixing the mechanically damaged kernels with undamaged kernels, the kernel density, bulk density and moisture content using the air-oven at all damage levels were measured.

Table 3.6. The weight of damaged kernels and sound kernels needed at various mechanical damage levels.

Mechanical Damage Level	Total weights (g)	Mechanical Damaged kernels (g)	Sound kernels (g)
0%	1000	0	1000
15%	1000	150	850
30%	1000	300	700
50%	1000	500	500
100%	1000	1000	0

3.2.3 Insect Damage Levels and Methods

For this Objective, two investigations were conducted. The first artificial insect damage was created in the kernels by drilling a hole in the kernel to simulate the internal infestation of kernels by a primary insect pest such as the maize weevil. In this case, the dependent variables were kernel and bulk density, and the independent variable was the artificially damaged kernel level. The second investigation involved infesting corn kernels with unsexed adult maize weevils with the goal of achieving insect damaged kernels. In this case the dependent variables were kernel and bulk density, and the independent variable was the life stage of the maize weevil. For each hybrid, a total of 6000g sample at 15% moisture content (details provided in Section 3.2.2) was used.

For the artificially damaged kernels used to simulate internal insect infestation, a Black & Decker electric drill (model GC1801, Black & Decker Corporation, Towson, Maryland, USA) with a 1/16 inch diameter drill bit was used to drill a single hole in the kernel endosperm. Five different levels of artificially damaged corn kernels were created by mixing sound kernels with damaged kernels. For each damage level, 1000g total weight of corn kernels were prepared for the tests. The weights of damaged kernels and sound kernels at different damage levels are shown below (Table 3.7). For each level, the kernel density, bulk density and moisture content were measured.

Table 3.7. The weight of damaged kernels and sound kernels needed at various artificial damage levels.

Artificial Damage Level	Total weights (g)	Artificial Damaged kernels (g)	Sound kernels (g)
0%	1000	0	1000
15%	1000	150	850
30%	1000	300	700
50%	1000	500	500
100%	1000	1000	0

For actual insect damaged kernels, about 2500g of corn kernels were prepared for each hybrid. First, the kernel density was tested at the initial moisture content 15%. This was the initial kernel density of corn kernels without maize weevil infestation. Then for each hybrid, the corn kernels were divided into four lots with a Boerner sample divider (Seedburo Equipment Co., Des Plaines, IL, USA) (Figure 3.7) giving four lots of about 600g. For each hybrid, four 1-quart glass jars were prepared and numbered as J_1, J_2, J_3 and J_4 (Figure 3.7). Then 600 mixed-age, mixed-sex maize weevils were prepared. A counter and a vacuum device shown in Figure 3.8 were used to collect and count the number of maize weevils placed in each jar. For sample J_1, J_2 and J_3, 600g of corn kernels and 200 maize weevils were placed in each jar. For J_4, only the 600g of corn kernels were added without maize weevils, which was used as the control jar. Jars were sealed with filter paper and wire mesh (shown in Figure 3.8) and stored in a temperature-controlled chamber (Figure 3.10). The chamber was set at 30ºC temperature. In order to increase the relative humidity in the environmental chamber, a beaker (1420ml) of water was placed in the chamber. A temp/RH HOBO sensor and data logger (model U10-003, Onset Corp, Bourne, MA, USA) (Figure 3.10) was placed in the chamber to collect data on the chamber's temperature and relative humidity. The average temperature and RH% were 30 °C and 30%, respectively. After four days, jars labeled J_1, J_2 and J_3 were taken out of the chamber and sieved with a No. 6 U.S. standard testing sieve to remove all maize weevils out of the kernels. Kernels were placed back in the original jars and the jars were placed back into the chamber. It was assumed that 4 days was enough time for eggs to be laid by the adults on corn kernels.

Figure 3.7. Boerner Sample Divider.

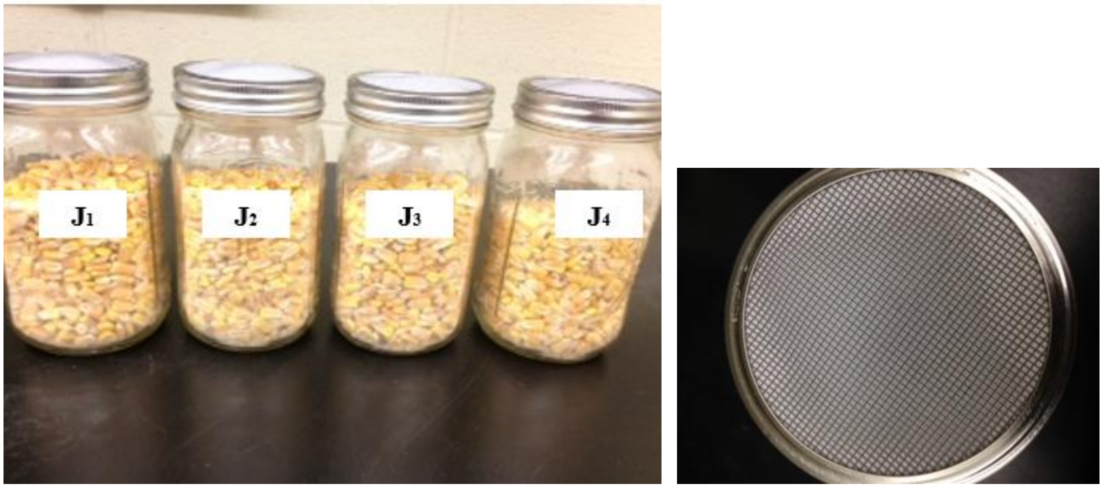
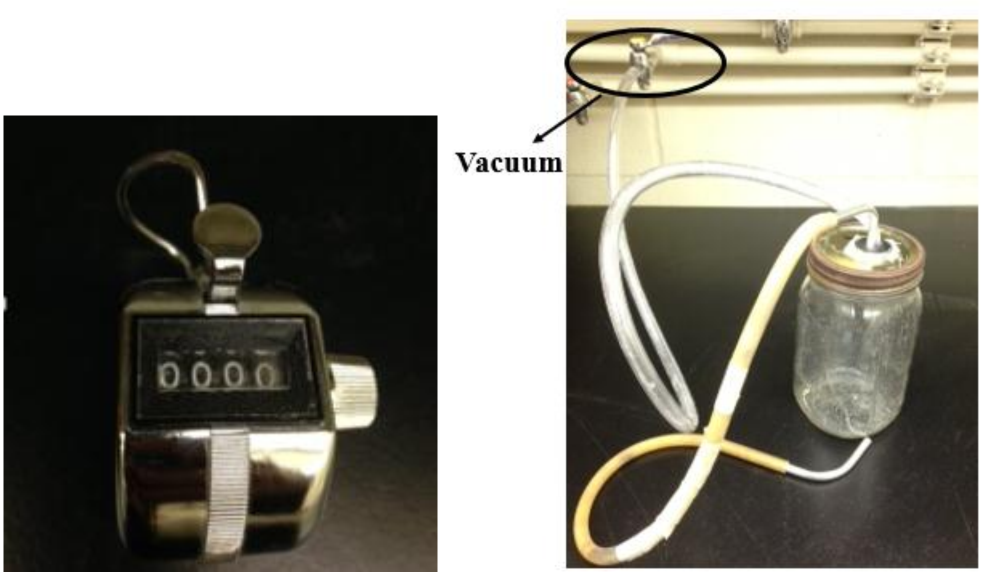


Figure 3.8. Samples in the glass jars and the jar cap.



(a) (b)

Figure 3.9. Counter Figure (a) and vacuum device (b).

(a) (b)

Figure 3.10. Temperature controlled chamber (a) and HOBO data logger (b).

Based on the life cycle of the maize weevil, different stages of weevils occur after different time periods. Therefore, corn kernels were sampled for kernel density measurements at the time periods when the egg, larva, pupa and adult would have developed, which correspond to day 4, 20, 35 and 50, of incubation respectively (Table 3.7). In Figure 3.11, test 1 at day 0 was the initial kernel density of corn kernels at 15% M.C. without maize weevil infestation.

Table 3.7. Specific dates corn kernels were sampled for kernel density tests.

Life stage	Tests	Date
Initial	1	0
Egg	2	4th
Larva	3	20th
Pupae	4	35th
Adult	5	50th

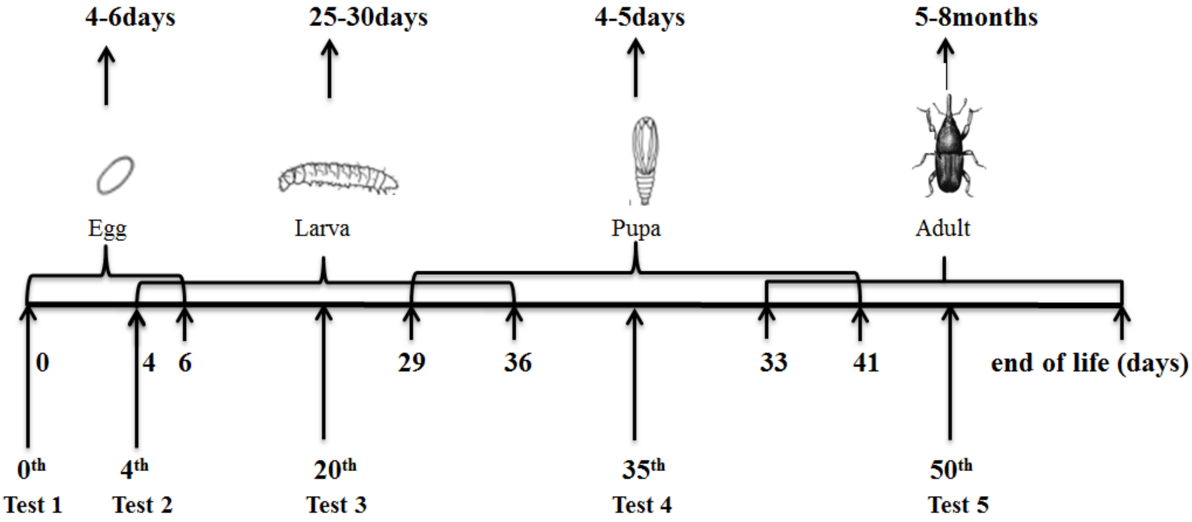


Figure 3.11.Timeline of the developmental stages of the maize weevil and specific sampling dates for kernel density tests.

3.2.4 Kernel and Bulk Density Measurement Methods

Kernel density was measured by using a multipycnometer (Quantachrome Instruments, Boynton Beach, FL, USA) with nitrogen gas (Figure 3.12). The gas is viewed as an ideal gas and thus the ideal gas law $PV = nRT$ can be applied. The multipycnometer determines the apparent density, which is the particle density including internal pores but excludes the external pores. This technique employs Archimedes principle and determines the particle volume by measuring the pressure difference when a known quantity of nitrogen is flowing from a precisely known reference volume (V_R) to the sample cell, which contains the solid material. The multipycnometer used was calibrated with a large sphere and large cell before every test. The equation used to determine the powder (kernel) volume are given in Equation 3.3 and Equation 3.4 (Quantachrome Instruments Manual, 2009). For each sample, three sub-samples were taken from the original samples using a Boerner sample divider, and subsamples were run three times. Thus there were nine replicates for one sample and the average represented the kernel density of this sample.

$$V_P = V_C - V_R[(\frac{p_1}{p_2}) - 1] \tag{3.3}$$

$$\rho = \frac{m}{V_P} \tag{3.4}$$

Where:
V_P = Volume of powder (kernels) (cm^3).
V_C = Volume of sample cell (cm^3).
V_R = Reference volume (cm^3).

P_1 = Pressure reading after pressurizing the reference volume.
P_2 = Pressure reading after including sample.

Figure 3.12. Multipycnometer with Nitrogen gas tank.

Bulk density was measured with a one-pint Boerner test weight apparatus (Figure 3.13) by filling the container following the standard test weight procedure (Boerner, 1922). The volume of the cylinder was one pint dry, which has a volume of 550.6 cubic centimeters for dry measure. The container weights were measured to the nearest 0.1g with an electronic balance. Corn was added to the funnel and the standard cylinder was placed coaxially under the funnel. After making sure the test weight apparatus was level by checking the bubble on the stand, the sliding valve on the funnel was open and the corn kernels fell into the standard cylinder. A v-shaped pan was placed at the base of apparatus to collect kernels that spilled over the side of the cup during filling. A wooden striker was used to level the top of the one pint container after the funnel was empty. Then the bulk density of corn kernels at each moisture content level were determined from the weight of the contents of the one-pint cylinder. Bulk density was calculated using the following Equation 3.5. Triplicate measurements were conducted, and the average and standard deviation of three measurements were reported.

$$\rho_{\text{bulk}} = \frac{\text{Sample Mass}}{\text{Volume}} \qquad (3.5)$$

Where:

Sample mass is the mass of corn kernels filling the standard cylinder and $v, volume$ is one pint (550.6 cm^3).

Figure 3.13. Boerner test weight apparatus.

3.3 Statistical Data Analysis

To evaluate the effect of moisture content, mechanical damage and insect damage on kernel and bulk density, respectively, data were subjected to one-way ANOVA ($\alpha=0.05$) using SAS (SAS, 1999). The PROC REG procedure was used for linear regression and the PROC GLM procedure was used for polynomial regression. The dependent variables were kernel density and bulk density, and the independent variables were moisture content, mechanical damage, and insect damage (including artificially simulated damage and insect damage by maize weevils), respectively.

In addition, for the two treatments in Objective 1, densities were compared to see if the patterns of kernel and bulk density changes with moisture during drying and rewetting were the same for corn kernels rewetted from 10% to 30% moisture in 2% increments, and dried from 30% to 10% moisture in 2% decrements and two-way ANOVA ($\alpha=0.05$) analysis using SAS (SAS, 1999) was conducted in this analysis. In Objective 3, a paired t-test was used to compare the kernel density of infested kernels with un-infested kernels. Graphs were plotted with MS Excel (Microsoft Office Excel 2003, Microsoft Corp., Seattle, Wash.).

CHAPTER 4. RESULTS AND CONSLUSIONS

This chapter presents results and statistical analyses of the study to determine the effect of moisture content, mechanical damage and insect damage on kernel and bulk density. This chapter consists of three sections. Section 4.1 presents results of the measurements made on corn kernels with three different moisture treatments: drying after harvest from the field, rewetting gradually for 10% to 30%, and drying gradually after artificially rewetting from 30% to 10%. Also, two treatments (rewetting and drying after artificial rewetting) were compared to see if they followed the same path. The second section (4.2) examines the effect of mechanical damage on bulk and kernel density. And the last section (4.3) describes the results of insect damage (artificially simulated insect damage and damage caused by maize weevil infestation) on bulk or kernel density. All sections include results of linear regression and also polynomial regression analyses. Equations and graphs are also used to gain insight into the relationship between moisture content and bulk or kernel density.

4.1 Effects of Moisture Contents on Bulk and Kernel Density

In all three treatments, kernel and bulk density were tested at various moisture contents. The test results are presented in Tables 4.1, 4.2 and 4.3. Table 4.1 shows the kernel and bulk density changes in corn kernels dried from harvest moisture. Table 4.2 shows kernel and bulk density changes in corn kernels rewetted from 10% to 30% moisture in 2% increments while Table 4.3 shows kernel and bulk density changes in corn kernels rewetted to 30% and then dried from 30% to 10% in 2% point decrements.

Table 4.1. Means of kernel density and bulk density of corn at various moisture contents dried from harvest moisture.

Hybrid	Moisture Content (% w.b.) [a]	Kernel Density (Kg/m^3) [b]	Bulk Density (Kg/m^3) [c]
Pioneer 1352	31.5	1230.6 (2.58)	673.0 (1.29)
	23.5	1240.6 (90.4)	698.3 (2.73)
	19.4	1241.9 (42.3)	731.4 (3.55)
	16.3	1251.4 (62.8)	759.3 (4.78)
	13.9	1251.7 (80.5)	758.1 (4.19)
	11.8	1252.0 (67.0)	757.3 (1.51)
Pioneer 1221	17.5	1275.1 (10.4)	784.1 (1.73)
	15.7	1280.1 (11.5)	790.9 (1.01)
	15.7	1280.3 (10.5)	791.7 (2.08)
	11.1	1295.3 (68.2)	795.6 (1.32)
	8.1	1298.1 (55.4)	801.0 (1.09)

[a] Average moisture content for 3 replicates (% wet basis) measured with an air oven
[b] Average kernel density for 9 replicates (standard deviation in parenthesis)
[c] Average bulk density for 3 replicates (standard deviation in parenthesis)

Table 4.2. Means of kernel density and bulk density of corn at various moisture contents during rewetting process from 10% to 30% in 2% point increments.

Hybrid	Moisture Content (% w.b.) [a]	Kernel Density (Kg/m^3) [b]	Bulk Density (Kg/m^3) [c]
Pioneer 1352	10.3	1254.8 (75.0)	767.1 (3.30)
	12.1	1253.2 (63.0)	763.7 (2.24)
	13.9	1253.0 (21.5)	754.9 (1.65)
	16.1	1248.3 (84.1)	753.0 (2.71)
	18.1	1246.7 (105.3)	749.3 (4.09)
	20.0	1247.4 (98.0)	731.1 (3.28)
	22.1	1243.9 (101.2)	724.0 (4.03)
	23.8	1242.6 (89.2)	710.4 (4.67)
	25.9	1237.0 (63.3)	689.8 (1.98
	28.1	1230.3 (64.1)	686.3 (3.20
	29.9	1229.8 (79.4)	677.1 (2.11)
Pioneer 1221	10.1	1295.1 (66.1)	797.1 (3.62)
	12.2	1293.5 (102.4)	796.3 (1.88)
	14.0	1293.7 (63.0)	788.5 (1.62)
	16.2	1289.4 (35.0)	783.2 (1.04)
	18.1	1280.1 (44.7)	778.4 (3.31)
	20.3	1279.2 (86.0)	776.7 (1.67)
	22.2	1277.2 (59.6)	762.0 (1.53)
	23.8	1269.9 (52.0)	743.4 (3.01)
	25.6	1262.7 (90.3)	733.6 (2.41)
	28.1	1262.1 (103.2)	727.9 (2.33)
	30.6	1260.6 (39.1)	719.4 (2.89)

[a] Average moisture content for 3 replicates (% wet basis) measured with an air oven
[b] Average kernel density for 9 replicates (standard deviation in parenthesis)
[c] Average bulk density for 3 replicates (standard deviation in parenthesis)

Table 4.3. Means of kernel density and bulk density of corn at various moisture contents during drying process from 30% to 10% in 2% point decrements after rewetting.

Hybrid	Moisture Content (% w.b.) [a]	Kernel Density (Kg/m^3) [b]	Bulk Density (Kg/m^3) [c]
Pioneer 1352	30.1	1237.1 (49.2)	679.2 (3.25)
	28.4	1240.9 (64.1)	691.8 (1.54)
	25.7	1243.1 (73.3)	704.1 (2.48)
	23.8	1244.3 (89.0)	711.0 (4.01)
	22.3	1247.2 (101.3)	726.7 (1.77)
	20.3	1250.7 (28.2)	734.9 (2.26)
	18.3	1251.6 (75.5)	742.3 (3.11)
	16.1	1252.0 (64.1)	750.2 (4.06)
	13.6	1253.4 (71.5)	758.7 (1.06)
	12.1	1257.3 (63.3)	767.5 (3.49)
	10.0	1257.5 (41.1)	773.3 (2.93)
Pioneer 1221	30.0	1264.2 (21.5)	733.2 (3.62)
	28.2	1269.3 (30.1)	742.8 (1.88)
	26.3	1271.2 (89.2)	749.7 (1.62)
	23.7	1275.5 (19.7)	751.2 (1.04)
	22.3	1276.2 (102.4)	755.4 (3.31)
	20.3	1282.0 (71.9)	762.1 (1.67)
	18.2	1286.7 (77.3)	767.8 (1.53)
	15.9	1291.7 (91.2)	777.9 (3.01)
	13.8	1292.4 (13.6)	782.3 (2.41)
	12.1	1295.3 (62.2)	788.1 (2.33)
	10.3	1295.9 (73.1)	791.0 (2.89)

[a] Average moisture content for 3 replicates (% wet basis) measured with air oven
[b] Average kernel density for 9 replicates (standard deviation in parenthesis)
[c] Average bulk density for 3 replicates (standard deviation in parenthesis)

From the results of the three treatments made on samples shown in Table 4.1, 4.2 and 4.3, we can see that both the kernel and bulk density decreased with increase in moisture content. Additionally, both hybrids behaved differently and followed the same trends for corn dried from harvest moisture and for rewetted and dried kernels. The kernel and bulk density for Pioneer 1221 was always higher than for Pioneer 1352 in all treatments. Correlations were determined using both linear and polynomial regression with SAS (SAS, 1999) for both hybrids.

For both hybrids, a scatter plot of the data were plotted and the trends were linear (for both kernel density and bulk density) (Figure 4.1 and Figure 4.2). One-way ANOVA was conducted on the data by using PROC REG procedure in SAS. The details are shown in Table 4.4 and Table 4.5. The linear equations for both kernel and bulk density are as follows:

Pioneer 1352:

$$\rho_{\mathrm{kernel}} = -1.16\mathrm{MC} + 1267.17 \tag{4.1}$$

$$\mathrm{R}^2 = 0.944$$

$$\rho_{\mathrm{bulk}} = -4.89\mathrm{MC} + 824.31 \tag{4.2}$$

$$\mathrm{R}^2 = 0.934$$

Pioneer 1221

$$\rho_{\mathrm{kernel}} = -2.59\mathrm{MC} + 1321.03 \tag{4.3}$$

$$\mathrm{R}^2 = 0.968$$

$$\rho_{\mathrm{bulk}} = -1.52\mathrm{MC} + 813.35 \tag{4.4}$$

$$\mathrm{R}^2 = 0.902$$

Where:

MC is moisture content (% w.b.).
ρ_{kernel} is the kernel density

ρ_{bulk} is the bulk density

Table 4.4. Linear regression analysis of kernel density and moisture content for corn dried from harvest moisture.

Dependent Variable: Kernel Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.944	Intercept MC	1267.17 -1.16	<.0001 0.001
Pioneer 1221	0.968	Intercept MC	1321.03 -2.59	<.0001 0.002

Table 4.5.Linear regression analysis of bulk density and moisture content for corn dried from harvest moisture.

Dependent Variable: Bulk Density				
Type	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.934	Intercept MC	824.31 -4.89	<.0001 0.002
Pioneer 1221	0.902	Intercept MC	813.35 -1.52	<.0001 0.014

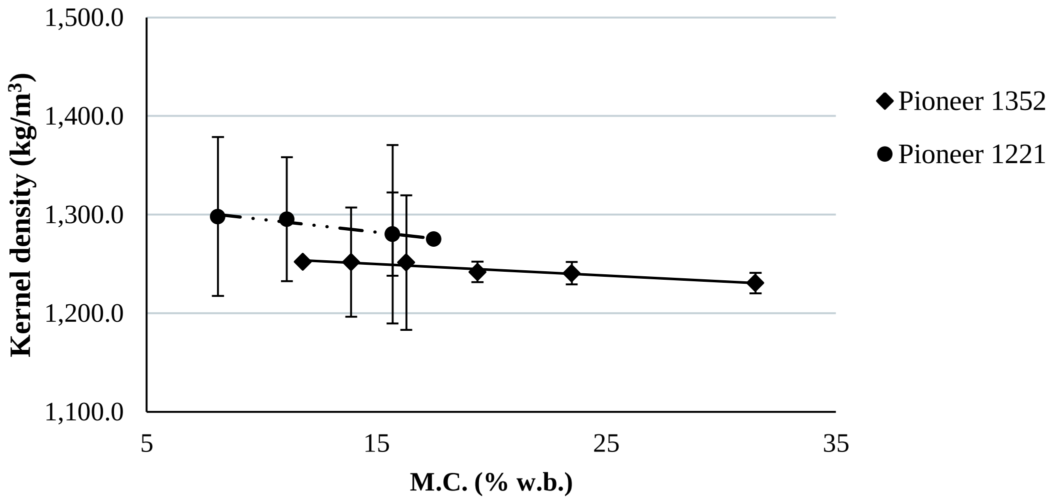


Figure 4.1. Kernel density changes for corn dried from harvest moisture.

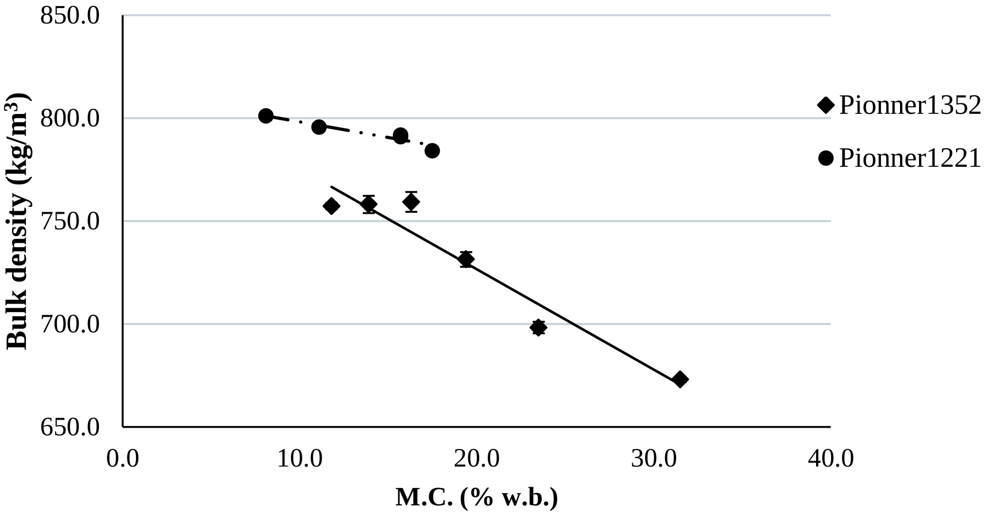


Figure 4.2. Bulk density changes for corn dried from harvest moisture.

For corn kernels rewetted from 10% to 30% in 2% point increments (Table 4.2), kernel density decreased by 25.0 kg/m^3 and the bulk density decreased by 90.0 kg/m^3 for Pioneer 1352. This was the same trend for Pioneer 1221; the kernel density decreased by 34.5 kg/m^3 while bulk density decreased by 77.7 kg/m^3. From the scatter plots, we can conclude that the relationships between kernel density and moisture content (Figure 4.3), and also bulk density and moisture content (Figure 4.4) are significantly linearly correlated with high R-squared. The statistics of one-way ANOVA using PROC REG procedure in SAS (SAS, 1999) is presented in Table 4.6 and Table 4.7. The linear equations for both kernel and bulk density are given below.

Pioneer 1352:

$$\rho_{\text{kernel}} = -1.29\text{MC} + 1270.03 \tag{4.5}$$

$$R^2 = 0.936$$

$$\rho_{\text{bulk}} = -4.90\text{MC} + 826.00 \tag{4.6}$$

$$R^2 = 0.961$$

Pioneer 1221

$$\rho_{\text{kernel}} = -1.96\text{MC} + 1317.83 \tag{4.7}$$

$$R^2 = 0.968$$

$$\rho_{\text{bulk}} = -4.17\text{MC} + 848.11 \tag{4.8}$$

$$R^2 = 0.946$$

Where:
MC = moisture content (% w.b.).
ρ_{kernel} = kernel density.
ρ_{bulk} = bulk density.

Table 4.6. Linear regression analysis of the kernel density and moisture content during rewetting process from 10% to 30% moisture in 2% point increments.

Dependent Variable: Kernel Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.936	Intercept MC	1270.03 -1.29	<.0001 < .0001
Pioneer 1221	0.968	Intercept MC	1317.83 -1.96	<.0001 < .0001

Table 4.7. Linear regression analysis of the bulk density and moisture content during rewetting process from 10% to 30% moisture in 2% point increments.

Dependent Variable: Bulk Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.961	Intercept MC	826.00 -4.90	<.0001 <.0001
Pioneer 1221	0.946	Intercept MC	848.11 -4.17	<.0001 <.0001

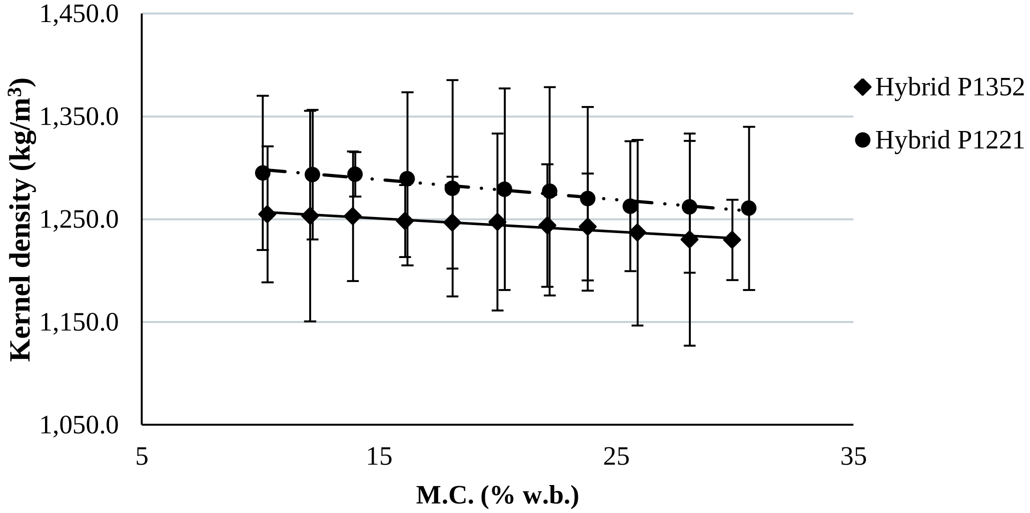


Figure 4.3.Kernel density as a function of moisture content during the rewetting process from 10% to 30% in 2% point increments.

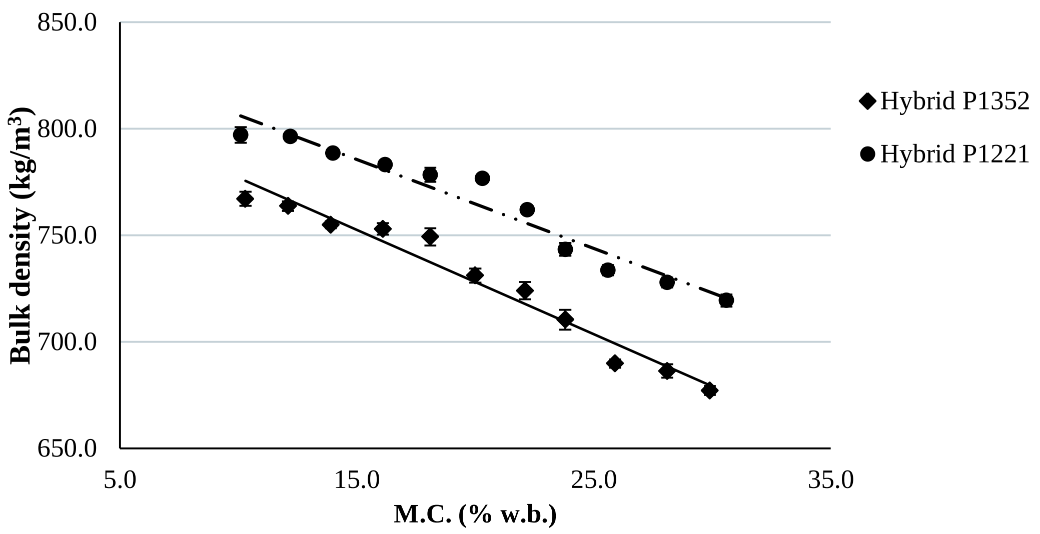


Figure 4.4. Bulk density as a function of moisture content during the rewetting process from 10% to 30% in 2% point increments.

The third treatment was drying corn kernels from 30% to 10% in 2% point decrements (Table 4.3.). For corn kernels rewetted to 30% and dried from 30% to 10% in 2% point decrements, the kernel density increased by 20.4 kg/m^3 and the bulk density decreased by 94.0kg/m^3 for Pioneer 1352. Likewise, the same trend occurred for Pioneer 1221; the kernel density decreased by 31.5 kg/m^3 while bulk density decreased by 57.8 kg/m^3. From the scatter plots, the relationships between kernel density and moisture content (Figure 4.5), and also bulk density and moisture content are linear (Figure 4.6). One-way ANOVA using PROC REG procedure in SAS (SAS, 1999) shows significant linear correlation (Table 4.8 and Table 4.9). The linear equations for both kernel and bulk density are given below (Equation 4.9 - 4.12).

Table 4.8. Linear regression analysis of the kernel density and moisture content during rewetting from 10% to 30% moisture in 2% point increments.

Dependent Variable: Kernel Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.966	Intercept MC	1268.33 -0.98	<.0001 < .0001
Pioneer 1221	0.968	Intercept MC	1315.59 -1.68	<.0001 < .0001

Table 4.9.Linear regression analysis of the bulk density and moisture content during rewetting from 10% to 30% moisture in 2% point increments.

Dependent Variable: Bulk Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.987	Intercept MC	824.08 -4.65	<.0001 <.0001
Pioneer 1221	0.988	Intercept MC	821.63 -2.88	<.0001 <.0001

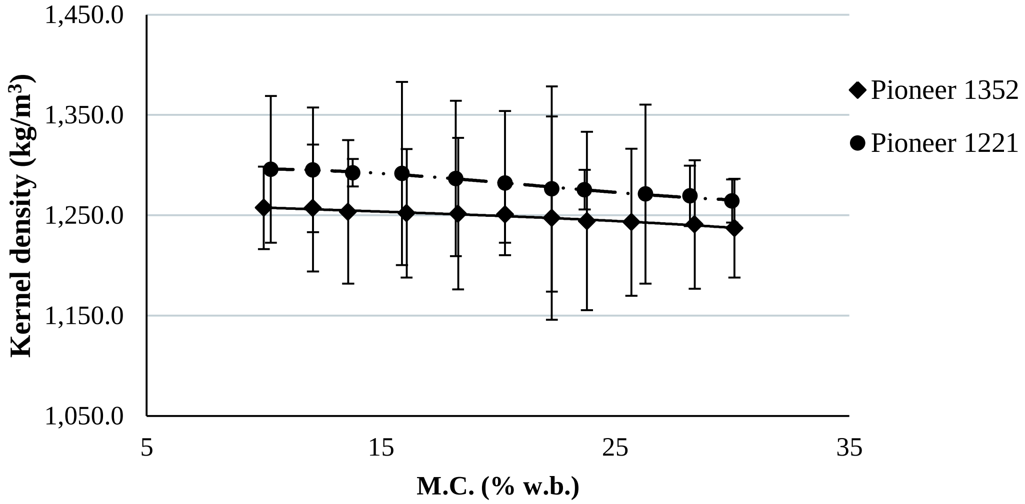


Figure 4.5. Kernel density as a function of moisture content during the drying process from 30% to 10% in 2% point decrements.

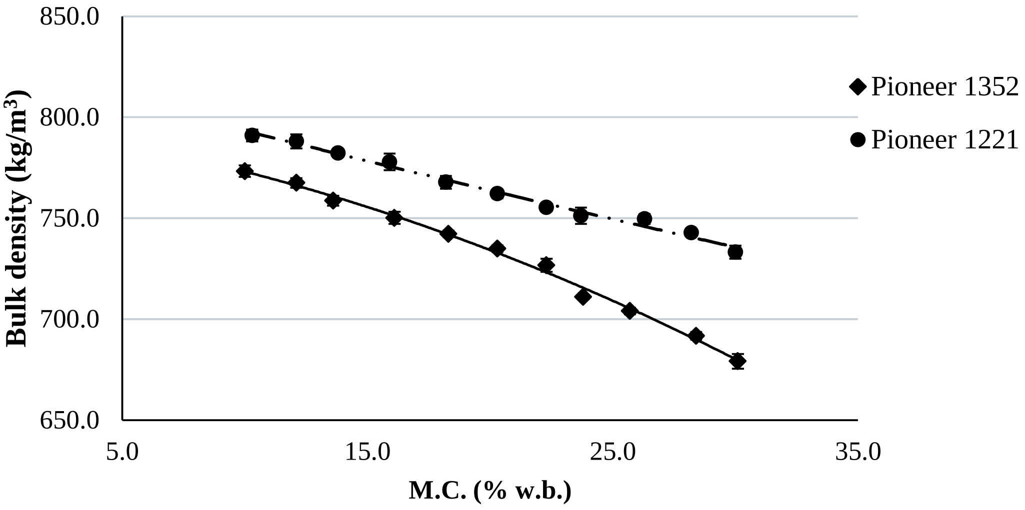


Figure 4.6.Bulk density as a function of moisture content during the drying process from 30% to 10% in 2% point decrements.

Pioneer 1352:

$$\rho_{\text{kernel}} = -0.98\text{MC} + 1268.33 \tag{4.9}$$

$$R^2 = 0.966$$

$$\rho_{\text{bulk}} = -4.65\text{MC} + 824.08 \tag{4.10}$$

$$R^2 = 0.987$$

Pioneer 1221

$$\rho_{\text{kernel}} = -1.68\text{MC} + 1315.59 \tag{4.11}$$

$$R^2 = 0.968$$

$$\rho_{\text{bulk}} = -2.88\text{MC} + 821.63 \tag{4.12}$$

$$R^2 = 0.988$$

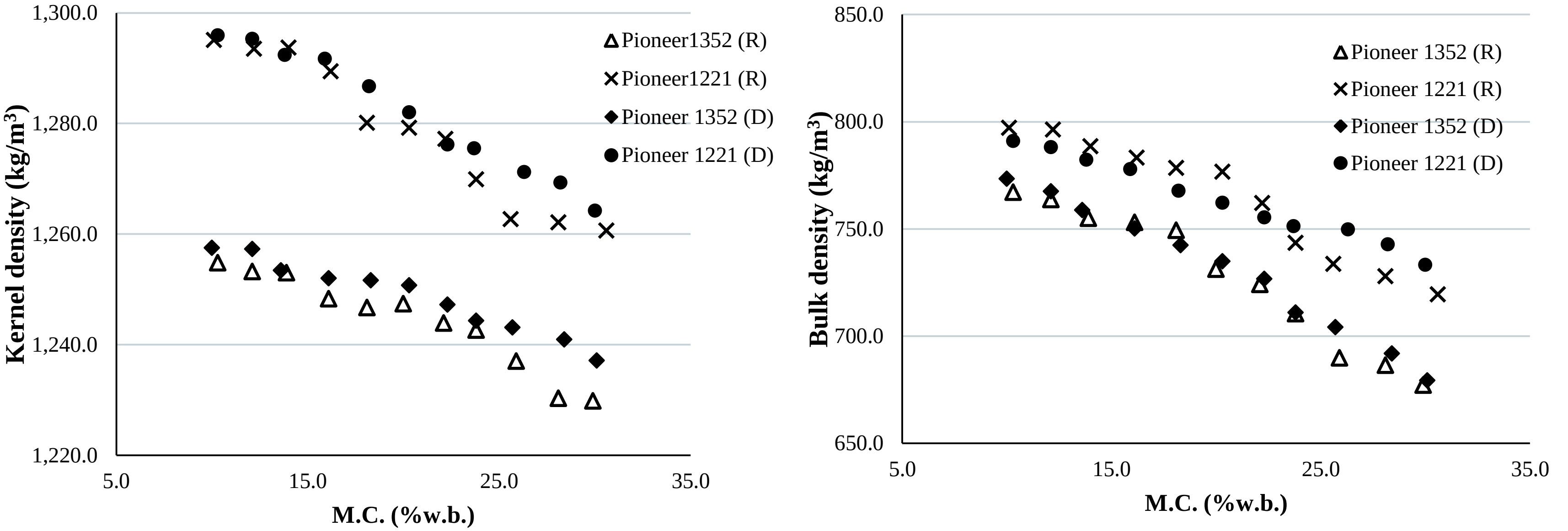
Where:

MC is moisture content (% w.b.).

ρ_{kernel} is the kernel density.

ρ_{bulk} is the bulk density.

Previous research on the relationship between moisture content and density of grain have been conducted on rewetting and drying cycles. Also, during grain storage in a bin, rewetting or drying of the bulk might happen due to weather changes. In Objective 1, the second treatment (rewetting) and the third treatment (drying after artificial rewetting) were compared (Figure 4.7), to see if the relationship between moisture content with kernel density, or bulk density, would be different for the drying or rewetting treatments. Two-way ANOVA was conducted by SAS to show if there was significant different in those two treatments at 0.05 α level. The results show that neither kernel density nor bulk density would be significantly different in rewetting and drying treatments. Thus, rewetting and drying would not affect the kernel and bulk density of corn kernels. Additionally, the curves for rewetting and drying intersected and crossed over at some point for both hybrids.



(a) (b)

Figure 4.7. Comparison of (a) kernel density and (b) bulk density.

(R) - Rewetting from 10% to 30% moisture in 2% point increments.
(D) - Drying from 30% to 10% moisture in 2% point decrements after artificial rewetting to 30%.

Research on moisture-dependent kernel and bulk density relationships was conducted by Nelson (1980). Twenty-one lots of hybrid, yellow-dent corn were used. Two empirical equations on the relationship of kernel and bulk density with moisture were developed by Nelson (Equation 3.18 and Equation 3.19). The equations developed by Nelson (1980) were plotted and compared with both the corn rewetting and drying treatments in this research (Figure 4.8 - 4.11).

$$\rho_k = 1.2519 + 0.00714m - 0.0005971m^2 + 0.00001088m^3 \tag{4.13}$$

$$r = 0.998$$

$$\rho_b = 0.6829 + 0.01422m - 0.0009843m^2 + 0.00001548m^3 \tag{4.14}$$

$$r = 0.996$$

Where:
ρ_k is kernel density.
ρ_b is bulk density.
m is moisture content.

Brusewitz (1975) also conducted kernel and bulk density tests at various moisture contents with corn kernels. In his research, one pint Boerner test weight apparatus was used for bulk density determination and helium-air pycnometer was used for kernel density test. Two empirical equations on the relationship of kernel and bulk density with moisture were given by Brusewitz (Equation 3.20 and Equation 3.21). The equations given by Brusewitz (1975) were plotted and compared with both the corn rewetting and drying treatments in this research (Figure 4.8 - 4.11).

$$\rho_k = 1.352 - 0.367m \tag{4.15}$$

$$r = 0.697$$

$$\rho_b = 1.0863 - 3.971m + 4.81m^2 \quad (4.16)$$

$$r = 0.921$$

Where:

ρ_k is kernel density.
ρ_b is bulk density.
m is moisture content.

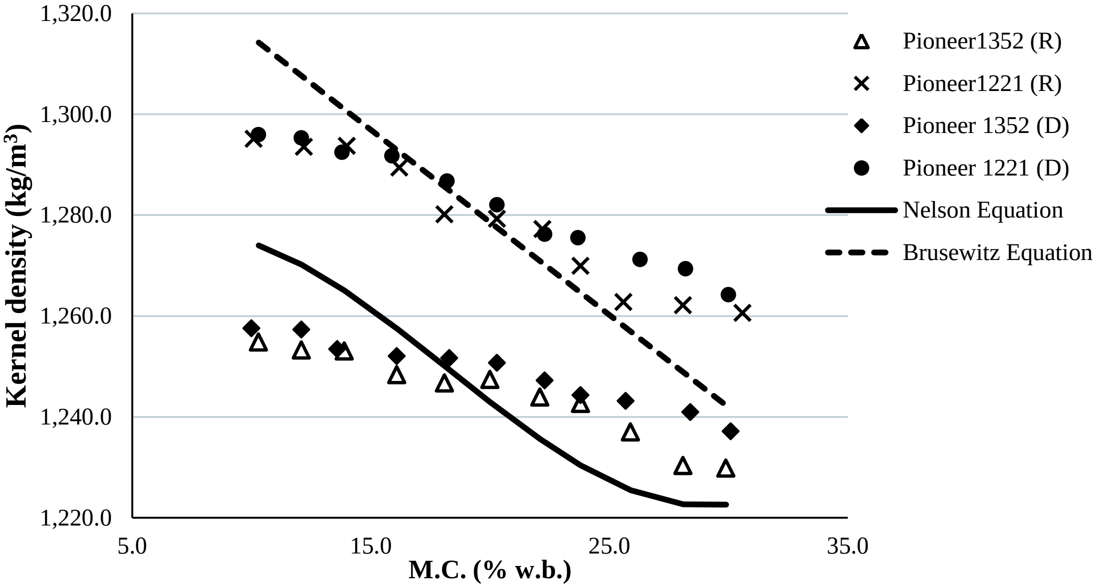


Figure 4.8. Kernel density and moisture content relationship comparisons with Nelson's (1980) and Brusewitz (1975) equations.

(R) - Rewetting from 10% to 30% moisture in 2% increments.
(D) - Drying from 30% to 10% moisture in 2% decrements after artificial rewetting to 30%.

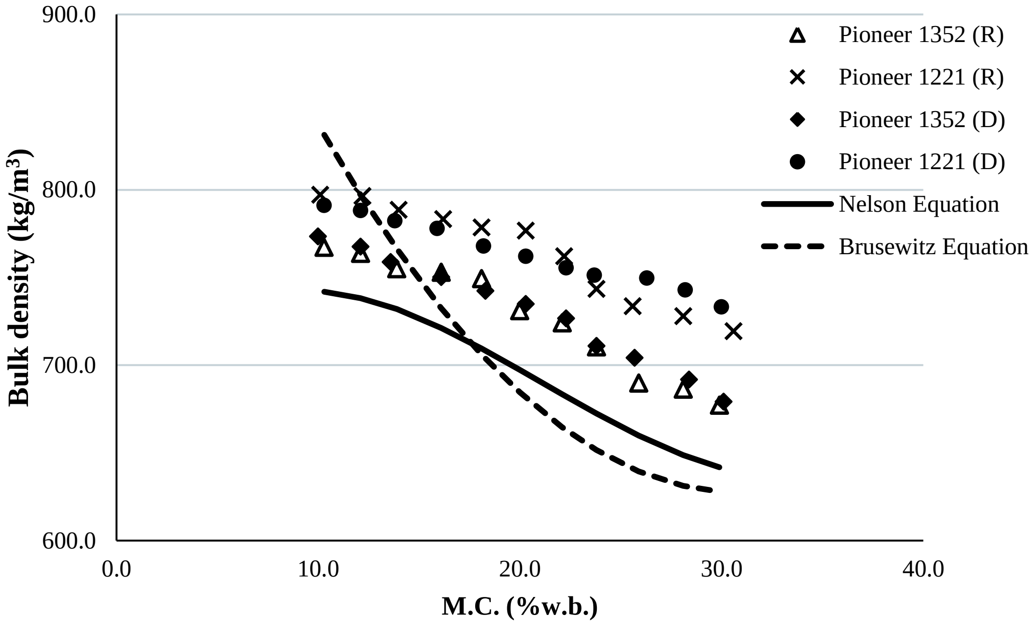


Figure 4.9. Bulk density and moisture content relationship comparisons with Nelson's (1980) and Brusewitz (1975) equations.

(R) - Rewetting from 10% to 30% moisture in 2% increments.
(D) - Drying from 30% to 10% moisture in 2% decrements after artificial rewetting to 30%.

Kernel density can also be estimated from known compositions of food material. The equation is shown below (Stroshine, 2012).

$$\rho_s = \frac{1}{\sum_{i=1}^{n} \frac{m_i}{\rho_i}} \quad (4.17)$$

Where:

ρ_s is the kernel density.
ρ_i is the density of the "i"th components.
m_i is the mass fraction of the "i"th components.
n is the number of different components.

For corn kernels, all solid components can be lumped. Assuming m_i is 1 kg, and the density of water is 1000 kg/m^3. From Table 4.1., Pioneer 1352 corn hybrid was harvested at 31.5% moisture and had a kernel density of 1230.6 kg/m^3. Therefore, the proportion of solid components should be 68.5% (100 – 31.5). At this initial harvested moisture content, neither rewetting nor drying has been done artificially. Putting the known values in Equation 4.17, we can estimate ρ_i as shown below:

$$1230.6 = \frac{1}{\frac{0.315}{1} + \frac{0.685}{\rho_i}} \tag{4.18}$$

Where ρ_i is the density of the solid lump.

For Pioneer 1352, the density of the solid lump can be calculated by solving for ρ_i in Equation 4.18, which results in 1376.6 kg/m^3. Having solved for ρ_i, the kernel density at various moisture contents during rewetting and drying after artificial rewetting can be calculated using Equation 4.17. Likewise, for Pioneer 1221, the density of the solid lump was calculated with the kernel density (1275.1 kg/m^3) at moisture content 17.5%, to be 1354.1 kg/m^3. For both corn hybrids, the kernel density at different moisture contents in both rewetting and drying treatments were calculated with Equation 4.17 and presented in Table 4.10. Figure 4.10 and 4.11 shows plots of the kernel density calculated using Equation 4.18 compared with data in this research. For Pioneer 1352 hybrid, the estimated kernel density tended to come close to agreement with the research data above 25% moisture, while it was the reverse for Pioneer 1221, where kernel density estimates were close to the research data from 10% to 18%, after which deviations increased.

Table 4.10. Kernel density (K.D.) using by Equation 4.15.

Hybrid	Rewetting		Drying after artificial rewetting	
	M.C. (%)	K.D.(kg/m^3)	M.C. (%)	K.D.(kg/m^3)
Pioneer 1352	10.3	1325.2	30.1	1236.4
	12.1	1316.6	28.4	1243.6
	13.9	1308.1	25.7	1255.1
	16.1	1297.9	23.8	1263.4
	18.1	1288.8	22.3	1269.9
	20.0	1280.2	20.3	1278.8
	22.1	1270.8	18.3	1287.8
	23.8	1263.4	16.1	1297.9
	25.9	1254.3	13.6	1309.5
	28.1	1244.9	12.1	1316.6
	29.9	1237.3	10.0	1326.6
Pioneer 1221	10.1	1307.3	30.0	1224.1
	12.2	1298.0	28.2	1231.2
	14.0	1290.1	26.3	1238.7
	16.2	1280.6	23.7	1249.3
	18.1	1272.5	22.3	1255.0
	20.3	1263.3	20.3	1263.3
	22.2	1255.4	18.2	1272.1
	23.8	1248.9	15.9	1281.9
	25.6	1241.6	13.8	1291.0
	28.1	1231.6	12.1	1298.5
	30.6	1221.7	10.3	1306.5

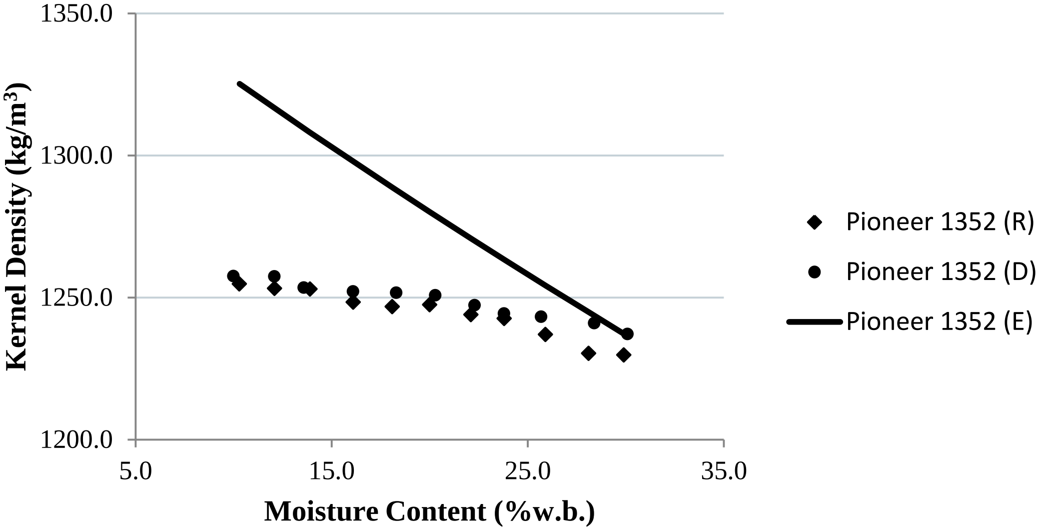


Figure 4.10. Plots of research data and Equation 4.15 estimates of kernel density changes with moisture content for corn hybrid, Pioneer 1352.

(R) - Kernel density in rewetting treatment.
(D) - Kernel density in drying after artificial rewetting treatment.
(E) - Kernel density calculated using Equation 4.15.

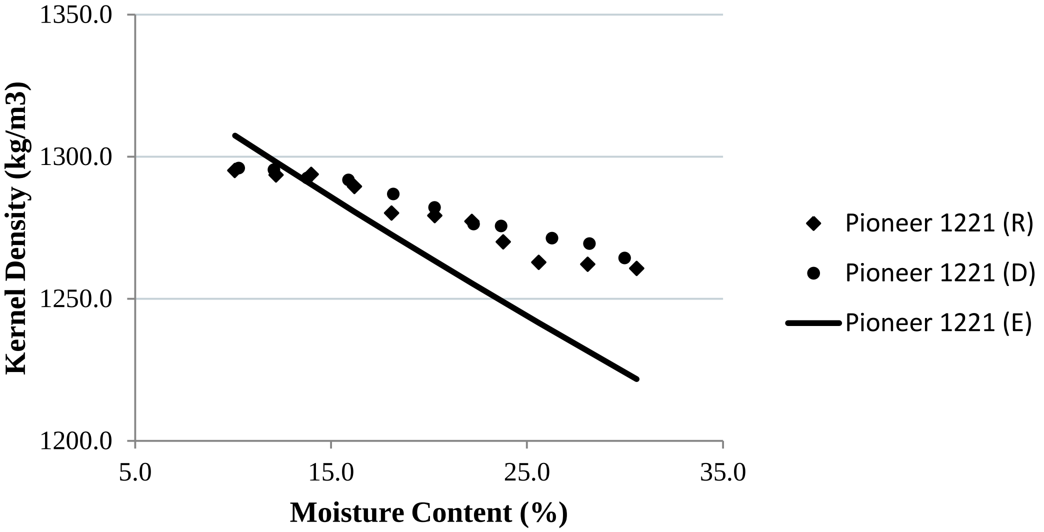


Figure 4.11. Plots of research data and Equation 4.15 estimates of kernel density changes with moisture content for corn hybrid, Pioneer 1221.

(R) - Kernel density in rewetting treatment.
(D) - Kernel density in drying after artificial rewetting treatment.
(E) - Kernel density calculated with Equation 4.15.

4.2 Effects of Mechanical Damage Levels on Bulk and Kernel Density

Different mechanical damage levels were created by mixing mechanically damaged kernels with sound kernels. Kernel and bulk density were measured at different mechanical damage levels. The results are shown in Table 4.11 with average values and their standard deviation presented.

Table 4.11.Means of kernel and bulk density at various mechanical damage levels.

Damage Level (%)		0	15	30	50	100
Pioneer 1352	Kernel Density[a] (kg/m^3)	1252.2 (28.1)	1281.5 (11.1)	1288.7 (8.87)	1298.3 (30.7)	1338.5 (11.5)
	Bulk Density[b] (kg/m^3)	749.3 (3.88)	738.7 (4.51)	737.0 (1.59)	719.8 (2.74)	697.5 (2.21)
Pioneer 1221	Kernel Density[a] (kg/m^3)	1291.0 (9.22)	1310.7 (23.9)	1318.3 (10.6)	1346.4 (7.08)	1377.9 (24.3)
	Bulk Density[b] (kg/m^3)	787.3 (1.70)	774.4 (3.11)	762.6 (2.98)	756.1 (1.47)	709.4 (3.61)

[a] Average kernel density for 9 replicates (standard deviation in parenthesis)
[b] Average bulk density for 3 replicates (standard deviation in parenthesis)

For both hybrids, the kernel density increased as the mechanical damage level increased. When measuring the kernel density with a multipycnometer, cracks in the kernel enabled the gas to penetrate inside the kernel. Chang (1987) found that there was 13.3% interior kernel porosity in corn kernels and the true density was larger than the apparent density. The cracks would make the interior pores accessible and lead to a smaller volume. As a result, the kernel density increased as mechanical damage level increased. However, the bulk density decreased as the mechanical damage level increased. This was most likely caused by differences in packing and void volume when smaller broken kernels fill the voids between whole kernels as the mechanical damage increased.

From the scatter plot, the relationships appear to be linear (Figure 4.12 and Figure 4.13). In order to obtain an insight of how mechanical damage level affected kernel and bulk density, linear regression ($\alpha=0.05$) was done using SAS (Table 4.12 and Table 4.13). The linear equations are shown below:

Pioneer 1352:

$$\rho_{\mathrm{kernel}} = 1.21\mathrm{MD} - 1528.17 \tag{4.19}$$

$$R^2 = 0.955$$

$$\rho_{\mathrm{bulk}} = 1.13\mathrm{MD} - 1464.99 \tag{4.20}$$

$$R^2 = 0.982$$

Pioneer 1221

$$\rho_{\mathrm{kernel}} = 1.13\mathrm{MD} - 1464.99 \tag{4.21}$$

$$R^2 = 0.976$$

$$\rho_{\text{bulk}} = -1.30\text{MD} + 1023.22 \tag{4.22}$$

$$R^2 = 0.984$$

Where:

MD = mechanical damage level (%).
ρ_{kernel} = kernel density.
ρ_{bulk} = bulk density.

From Figure 4.8, there is a positive correlation between kernel density and mechanical damage level for both hybrids. For bulk density and mechanical damage level the relationship is inverse, a negative correlation (Figure 4.9).

Table 4.12. Linear regression analysis of the kernel density at various mechanical damage levels.

Dependent Variable: Kernel Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.955	Intercept MD[a]	-1528.17 1.21	0.004 0.004
Pioneer 1221	0.976	Intercept MD[a]	-1464.99 1.13	0.002 0.002

[a] MD is the mechanical damage level.

Table 4.13. Linear regression analysis of the bulk density at various mechanical damage levels.

Dependent Variable: Bulk Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.982	Intercept MD[a]	1419.77 -1.90	0.001 0.001
Pioneer 1221	0.984	Intercept MD[a]	1023.22 -1.30	0.001 0.001

[a] MD is the mechanical damage level.

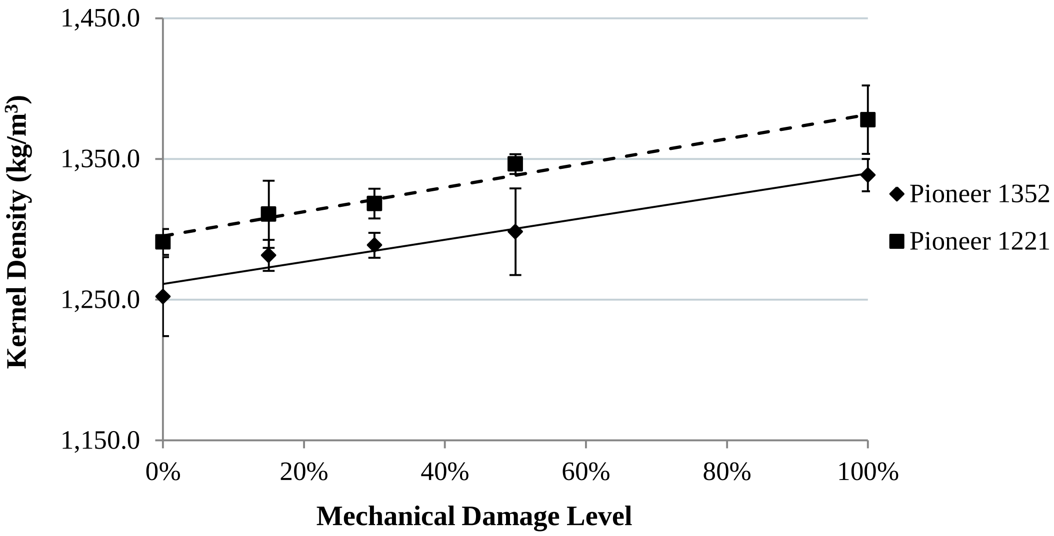


Figure 4.12. Kernel density changes with mechanical damage level.

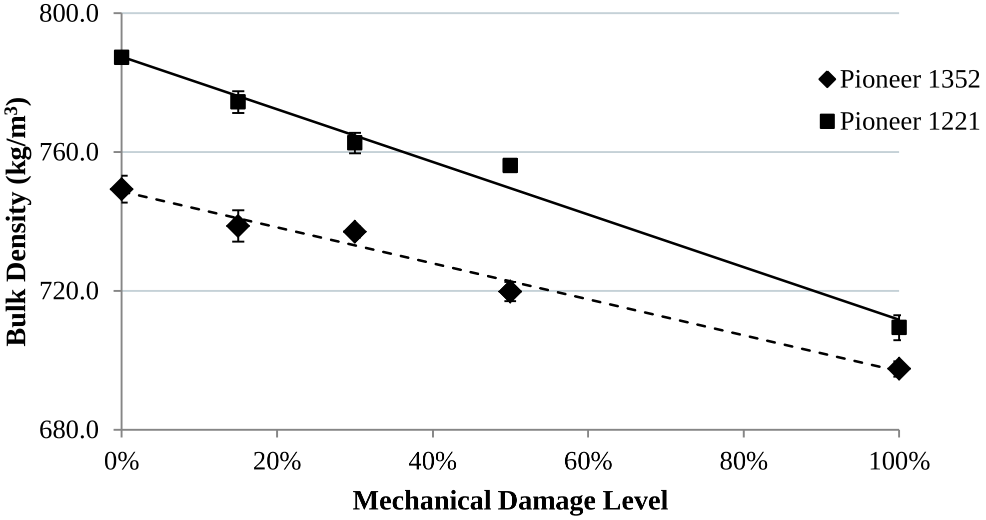


Figure 4.13. Bulk density changes with mechanical damage level.

4.3 Effects of Insect Damage on Bulk and Kernel Density

In this research objective, there were two treatments. In the first test, simulated insect damage was done artificially by drilling holes (one hole per kernel) in the corn kernels. The results of the effects of the artificial damage on bulk and kernel density are shown in Table 4.14.

Table 4.14. Effects of artificial damage level on bulk and kernel density.

Artificial Damage Level (%)		0	15	30	50	100
Pioneer 1352	Kernel Density[a] (kg/m^3)	1249.3 (26.7)	1252.1 (31.1)	1256.8 (1.93)	1281.5 (14.8)	1313.9 (34.2)
	Bulk Density[b] (kg/m^3)	749.2 (1.04)	744.3 (1.82)	731.7 (3.46)	726.5 (4.24)	698.6 (2.30)
Pioneer 1221	Kernel Density[a] (kg/m^3)	1289.3 (1.07)	1292.1 (12.4)	1296.8 (25.5)	1310.7 (42.0)	1330.2 (14.1)
	Bulk Density[b] (kg/m^3)	801.2 (4.06)	797.1 (3.68)	786.6 (2.15)	764.7 (2.49)	746.9 (4.27)

[a] Average kernel density for 9 replicates (standard deviation in parenthesis)
[b] Average bulk density for 3 replicates (standard deviation in parenthesis)

For both hybrids, the scatter plots were linear (for both kernel density and bulk density) (Figure 4.14 and Figure 4.15). One-way ANOVA was conducted by using PROC REG procedure using SAS. The details are shown in Table 4.15 and Table 4.16. The linear equations for both kernel and bulk density are given below.

Pioneer 1352:

$$\rho_{\mathrm{kernel}} = 1.40\mathrm{AD} - 1734.93 \tag{4.23}$$

$$\mathrm{R}^2 = 0.965$$

$$\rho_{\mathrm{bulk}} = 2.28\mathrm{AD} - 2927.75 \tag{4.24}$$

$$\mathrm{R}^2 = 0.988$$

Pioneer 1221

$$\rho_{\mathrm{kernel}} = -1.94\mathrm{AD} + 1458.67 \tag{4.25}$$

$$\mathrm{R}^2 = 0.981$$

$$\rho_{\mathrm{bulk}} = -1.65\mathrm{AD} + 1324.15 \tag{4.26}$$

$$\mathrm{R}^2 = 0.954$$

Where:
AD = artificial damage.
ρ_{kernel} = kernel density.
ρ_{bulk} = bulk density.

Above equations show that as the artificial damage level increased, the kernel density increased. This appears similar to the relationship of kernel density with mechanical damage, which increased due to the ability of the gas (nitrogen) used for kernel density determination using the multipycnometer to penetrate the inner pores of the kernels through the drilled holes. The bulk density decreased as artificial damage increased due to the weight losses of kernels from drilling, while still maintaining the same kernel and bulk volumes. For both kernel density and bulk density, there was a linear relationship with artificially induced insect damage level.

Table 4.15. Linear regression analysis of the kernel density at various artificially induced insect damage levels.

Dependent Variable: Kernel Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.965	Intercept AD[a]	-1734.93 1.40	0.003 0.003
Pioneer 1221	0.981	Intercept AD[a]	-2927.75 2.28	0.001 0.001

Table 4.16. Linear regression analysis of the bulk density at various artificially induced insect damage levels.

Dependent Variable: Bulk Density				
Hybrid	R-Square	Variable	Parameter Estimate	Pr > |t|
Pioneer 1352	0.988	Intercept AD[a]	1458.67 -1.94	0.005 0.005
Pioneer 1221	0.954	Intercept AD[a]	1324.15 -1.65	0.004 0.004

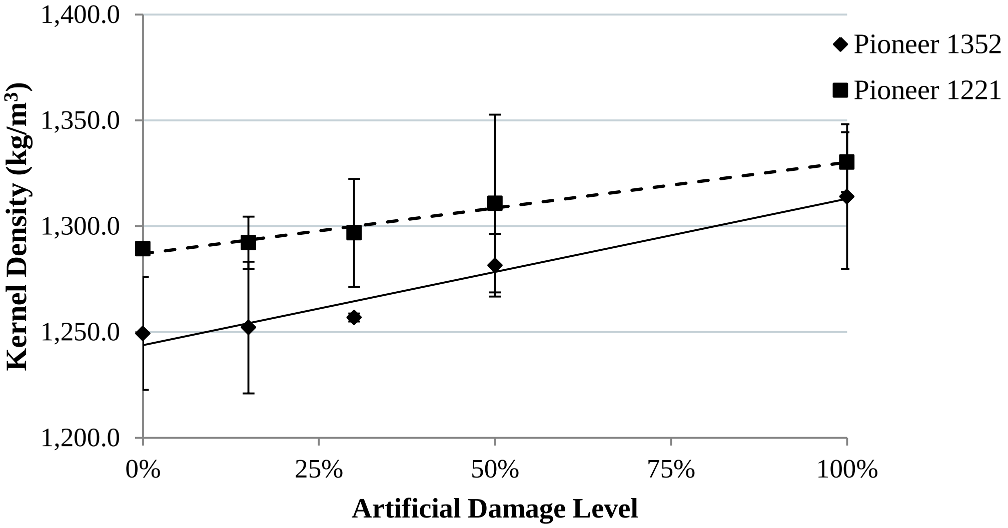


Figure 4.14. Kernel density changes with artificially induced insect damage levels.

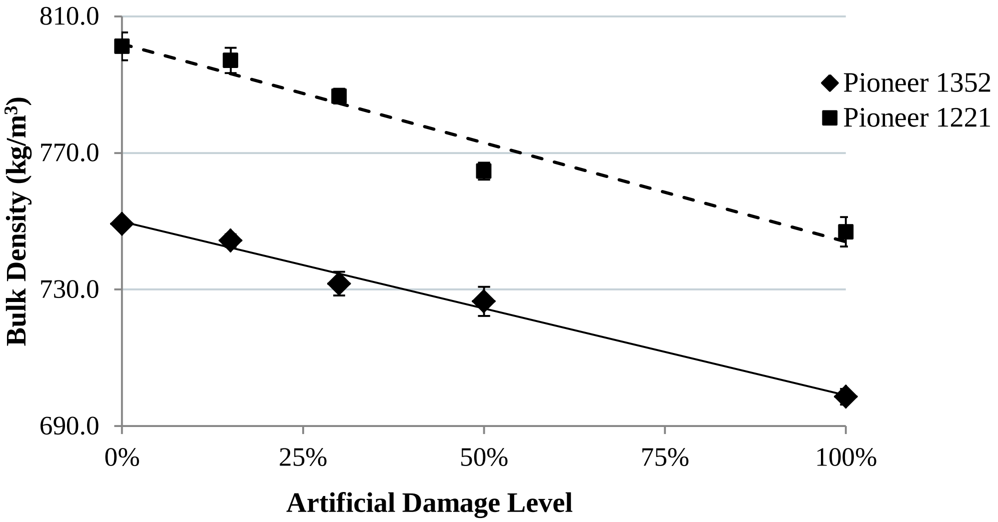


Figure 4.15. Bulk density changes with artificially induced insect damage levels.

The second treatment in this section was actual maize weevil infestation damage. Kernel density was measured at different life stages of the maize weevil that would be expected to be in a corn kernel after incubation for a given period of time based on the maize weevil life cycle. The results are shown in Table 4.17.

Table 4.17. Kernel density of corn kernels at different maize weevil life stages.

Hybrid	Kernel Density (kg/m^3) of	Maize Weevil Life Stage				
		Initial[a]	Egg	Larva[b]	Pupae[b]	Adult[b]
Pioneer 1352	Infested kernels	1250.6 (40.4)	1251.6 (49.7)	1249.5 (38.2)	1247.8 (47.8)	1250.1 (90.1)
	Un-infested kernels (control)	1250.6 (30.6)	1251.9 (33.3)	1252.7 (10.3)	1253.1 (33.3)	1253.5 (93.3)
	Moisture Content (% w.b.)	15.1%	14.7%	12.5%	11.8%	10.7%
Pioneer 1221	Infested kernels	1286.4 (86.7)	1289.8 (22.2)	1288.5 (74.2)	1286.9 (43.5)	1290.8 (64.1)
	Un-infested kernels (control)	1286.4 (86.7)	1289.7 (42.8)	1292.4 (52.1)	1292.8 (80.3)	1294.4 (57.1)
	Moisture Content (% w.b.)	15.0%	14.3%	12.6%	12.1%	10.9%

[a] Kernel density of un-infested kernels before adding maize weevil.

[b] Represents life stages that have significant difference between infested kernels and control kernels.

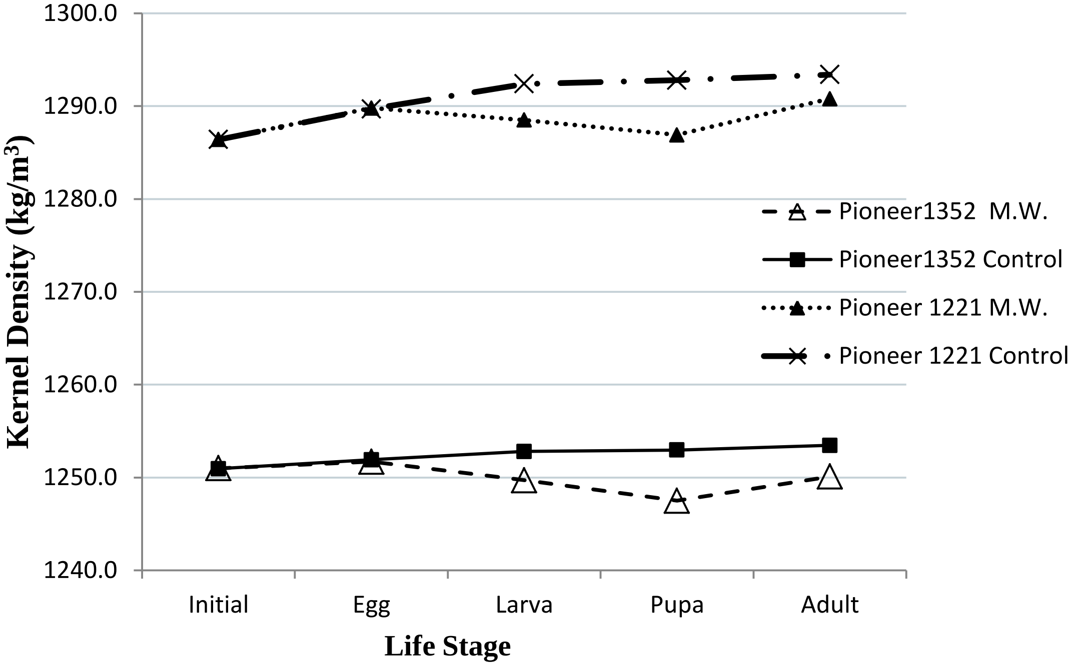


Figure 4.16. Kernel density of infested kernels and control (non-infested) kernels at different maize weevil life stages.

The moisture content of kernels decreased from about 15% to 11% moisture level due to equilibration with environment low relative humidity inside the chamber (30 °C and 30% RH). The moisture changes led to kernel density changes as was seen with the slight increase in kernel density as moisture decreased from the initial to the adult stage (about 50 days) for the control jars, which had no maize weevil infestation. Since the control jar with un-infested kernels were in the same environment (the chamber) as the infested kernels, they should have the same kernel density changes caused by the moisture content changes. Therefore, we can conclude with a great degree of certainty that the decrease in kernel density seen from the larva stage was most likely caused by infestation of this stage growing inside the corn kernel. As development of the maize weevil progressed into adult, the kernel density decreased at the pupa stage and then increased at the adult stage (Fig. 4.16.). Unfortunately, no images where collected of the infested kernels to conclusively verify the size, life stage and space occupied by the infesting weevil. However, these results prompt the need for further work in this area, especially if kernel density could be used as a distinguishable measure for internal infestation in grains. It is also important to note that the artificially induced infestation by drilling a hole per kernel did not truly reflect actual insect infestation.

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In order to see whether the kernel density changes caused by insect damages were significantly different from un-infested kernels (control sample), a paired t-test was conducted using Excel. The result shows that there is significant difference in kernel density between infested kernels and the control kernels in the following life stages: larva, pupa, and adult. This is consistent with the result shows in Figure 4.16.

CHAPTER 5. SUMMARY AND CONCLUSION

5.1 Restatement of Thesis Objectives

The overall goal of this research was to investigate kernel density and bulk density changes due to moisture content, mechanical damage and internal insect damage of corn kernels. Specifically, the objectives as stated in Chapter 1 were as follows:

1. To evaluate the effect of the following treatments on corn kernel density and bulk density:
    a. Drying corn kernels right after harvest from the field.
    b. Artificially rewetting naturally dried kernels from 10% to 30% moisture content in 2% point increments.
    c. Artificially rewetting naturally dried kernels to 30% moisture content, then drying from 30% to 10% in 2% point decrements.
2. To determine the effect of mechanical damage levels (broken and chipped kernels) induced using the grain breakage tester on corn kernel density and bulk density.

3. To determine the effect of internal insect infestation on corn kernel density and bulk density investigated by:
    a. Drilling holes in kernels to simulate internal insect infestation damage by emerged *Sitophilus zeamais* (maize weevil) adults.
    b. Actual life stages of *Sitophilus zeamais* infestation reared on corn kernels.

5.2 Results Summary

The first objective of this research was to evaluate the effect of moisture content changes on corn kernel density and bulk density of two corn hybrids, Pioneer 1352 and Pioneer 1221. The following three treatments were applied: 1) drying corn kernels right after harvested from the field, 2) artificially rewetting naturally dried kernels from 10% to 30% moisture content in 2% point increments, 3) artificially rewetting naturally dried kernels to 30% moisture content, then drying from 30% to 10% in 2% point decrements. In all three treatments, the relationships between 1) kernel density and moisture content and 2) bulk density and moisture content were analyzed. Additionally, the drying after rewetting process was compared with the rewetting process. All three treatments showed that both kernel and bulk density had a linear negative relationship with moisture content from 10% to 30%. Both corn hybrids followed similar trends, with Pioneer 1221 having a higher kernel and bulk density that Pioneer 1352.

In objective 2, the effect of mechanical damage levels (broken and chipped kernels) induced using a grain breakage tester on corn kernel density and bulk density was determined. The result shows that there was a positive linear relationship between kernel density and mechanical damage level. This might be caused by the cracks in the kernels, which allowed gas to penetrate into the external and internal pores when measuring the kernel density using a multipycnometer. There was also a negative linear relationship between bulk density and mechanical damage level, most likely due to packing from smaller broken kernels filling the voids between whole kernels.

In objective 3, the effect of internal insect infestation on corn kernel density and bulk density was investigated using two different treatments. The treatments involved 1) an artificially induced insect damage by drilling a hole per kernel to simulate emerged adults of maize weevil, and 2) actual infestation of corn kernels with adult *Sitophilus zeamais* (maize weevil) while determining kernel density at various life stages of development. The result shows that as the artificial damage level increased, the kernel density increased. The increase in kernel density can be explained by the reduction in kernel volume due to access by the gas into the internal pores of the kernel. The bulk density decreased as artificial damage increased due to weight loss of kernels from the drilled out endosperm while still maintaining the same kernel volume, and thus constant bulk volume (1 pint). The artificially drilled kernels did not accurately simulate changes in kernel and bulk density caused by internal insect infestation such as the maize weevil.

The result of the second treatment using maize weevil infested kernels showed that the kernel density did not change in the egg stage. However, for larva and pupa stages, the kernel density decreased and for adult stages, the kernel density increased compared with larva and pupa stages. Except for the egg stage, all other stages for the infested kernels had a smaller kernel density than the control (un-infested kernels). Further research needs to be conducted to conclusively explain these differences, and most importantly determine whether kernel density would be a good distinguishable measure for determining internal infestation in corn kernels.

5.3 Future Work

Several important issues were brought up by the results of this study. A better understanding of the hybrids genetics would have been useful in explaining the results of this investigation. Further studies should investigate more corn hybrids of known genetics background, especially with respect to other quality parameters such as hardness and chemical composition, which can be possibly correlated to kernel and bulk density. The relationship of kernel and bulk density to shrinkage under drying and rewetting cycles would be insightful and useful to grain science and industry. Additionally, other tests that should be pursued include kernel shape and size, and kernel hardness.

For the maize weevil infestation tests, the infestation level and life stages could not be definitely confirmed with additional verification methods. According to the literature, X-ray is the best way to know the internal infestation of maize weevils. Other methods using new powerful microscopes could also provide a low-cost verifiable means. If these tools are available, it would be better if the internal infestation level and life stage can be measured over the life stage sampling period for maize weevil. The results obtained so far definitely prompts further investigation on whether kernel density could be used as a reliable method to determine internal insect infestation of corn kernels by *Sitophilus zeamais* (maize weevil).

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