CHARACTERIZATION OF CHEMICAL COMPOSITION, MILLING PROPERTIES AND CARBON DIOXIDE DIFFUSIVITY RESULTING FROM EARLY HARVEST CORN AND CORN STOVER

BY

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DISSERTATION

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Abstract

The increasing demand of corn as food and fuel sources has increased the competition for feedstock between livestock and ethanol industries. Developing an effective corn harvesting, storage and utilization system can help reduce the competition for the limited corn supply in the current period. The overall goal of this study was to examine the feasibility of early harvest of corn and corn stover and determine the implications of this proposed practice on corn processing characteristics, chemical composition and nutritive values. Effective diffusion coefficient of CO$_2$ through bulk corn at different moisture contents and temperatures were measured to a better understand CO$_2$ movement during corn storage, which could help develop a CO$_2$ monitoring system for corn storage.

Corn plants were harvested at different maturity stages in 2009 and 2010 and were quantitatively evaluated in terms of moisture contents, dry matter yields, compositions and processing characteristics of corn and corn stover. The dry matter yield of corn increased rapidly until reaching corn maturity and remained stable after maturity, with an average yield of 11.1 t/ha over the two year study. For corn stover, the two year average dry matter yield was 14.8 t/ha at the beginning of the study (filling stage) and decreased to 13.2 t/ha at corn physiological maturity and further decreased throughout corn dry down. Effects of corn harvest moisture content on dry grind ethanol processes were observed on fermentation characteristics. The final ethanol concentration from corn with harvest moisture content of 54% (110 days after corn planting, kernel dent stage) was 0.5 to 1.2 percentage points higher than that from mature corn with lower harvest moisture contents. Corn harvest moisture content affected compositions and nutritive values of corn, corn stover and dried distiller grain solubles (DDGS). As corn harvest moisture content decreased, neutral detergent fiber, acid detergent fiber and lignin concentrations in corn stover increased while crude protein concentrations decreased. These results showed that the whole corn plant could be most efficiently used if the corn and corn stover were harvested as soon as corn reached physiological maturity.

Early harvested corn poses a problem of storability due to its high moisture content. Effective CO$_2$ diffusion coefficients in bulk corn at various temperatures (10, 20 and 30°C) and corn moisture contents (14.0, 18.8 and 22.2% w.b.) were determined to help
develop a CO₂ monitoring system for corn storage. The diffusion coefficient measurements were conducted using a diffusion cell surrounded by a water jacket, which was used to control the temperature of the bulk corn in the diffusion cell. A source term (CO₂ respiration rate) was introduced in the diffusion equation to account for the CO₂ production by corn during the diffusion process. The corn respiration rate increased when temperature and corn moisture content increased. As respiration rate increased, it had a larger effect on the diffusion pattern when measuring the effective diffusion coefficient. The effective diffusion coefficients of CO₂ through bulk corn ranged between $3.10 \times 10^{-6}$ and $3.93 \times 10^{-6}$ m²/s, depending on temperature and moisture contents. As temperature increased from 10 to 30°C, the diffusion coefficient of CO₂ through bulk corn increased from $3.21 \times 10^{-6}$ to $3.76 \times 10^{-6}$ m²/s, respectively. As corn moisture content increased from 14.0 to 18.8%, the effective diffusion coefficient through bulk corn decreased from $3.59 \times 10^{-6}$ to $3.39 \times 10^{-6}$ m²/s, respectively. There was no difference observed in the effective CO₂ diffusion coefficient when corn moisture content increased from 18.8 to 22.2%.

**Keywords:** Corn, harvest moisture, corn processing, carbon dioxide, effective diffusion coefficient
To my parents and wife
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Chapter 1. Introduction

The increasing demand of corn as food and fuel sources has intensified the competition between livestock and ethanol industry. Developing an effective corn harvesting, storage and utilization system can help reduce the competition for the limited corn supply in the current period. In 2011-2012, the ethanol industry produced more than 53.2 billion liters of ethanol with 5011 million bushels of corn, which accounted for 40.3% of the total corn production (USDA, 2012). The rising price of gasoline will stimulate the ethanol industry to increase productivity. Livestock (ruminants and non-ruminants) are expected to consume about 50% of corn production each year (USDA, 2012). Unless there is a sharp increase in production or decreases in exports and other uses of corn, the severe competition between these two industries would remain. Because ruminants consume 35.6% of the 4,548 million bushels of corn fed to livestock in the United States each year (Sewell et al., 2009), efficient utilization of the non-grain portion of the corn plants as ruminant feed and grain for ethanol will allow the optimization of both food and fuel production. On the other hand, postharvest losses due to spoilage during corn storage remain a major problem in the world, developing of an effective corn storage monitoring system would increase the end-use of corn.

Traditionally corn and corn stover were harvested after corn reached physiological maturity and was dry (18-25% w.b.) (Unless noted otherwise, all the moisture content values are reported on a wet basis.). The cost of drying makes harvesting corn early not economically feasible. However there are several reasons why higher moisture, or early, harvest of corn and corn stover would be advantageous. If the corn and corn stover were harvested as soon as the corn reached physiological maturity, the stover would be less lignified and more responsive to chemical treatment to enhance digestibility (Weaver et al., 1978; Russell, 1986; Cone and Engels, 1993). With early harvest, corn stover yield would be higher (Pordesimo et al., 2004a, 2004b; Shinners and Binversie, 2007); the weather losses (early frost, storm etc.) of corn would be lower; and corn quality losses due to mold and insect invasion would be reduced. Early harvest of corn and corn stover also has advantages including better weather early in the harvest season, longer daylight length (so work can be done more efficiently and safely) and longer harvest window.
Furthermore, early removal of corn from the field may allow the production of a second crop that is nitrogen fixing. For example, hairy vetch has been shown to fix nitrogen over winter, producing about 50-200 kg of nitrogen per hectare (Smith et al., 1987; Dabney et al., 2011). This cover crop would also reduce soil erosion and increase soil organic matter, which would allow a higher fraction of corn stover to be removed from the field.

Harvesting corn at or before physiological maturity may be advantageous to the processing of the corn if it can be stored wet. First, the amylose/amylopectin ratio in corn starch increases as corn matures (Shannon and Garwood, 1984; Li et al., 2007). Resistant starch levels in corn starch are highly correlated with amylose levels (Berry, 1986). For the corn ethanol industry, higher resistant starch in corn leads to decreased ethanol yields. Second, with corn maturation, the starch granules in the endosperm are increasingly surrounded by protein bodies and embedded in a dense protein matrix (Philippeau and Michalet-Doreau, 1997), which would limit the action of starch hydrolytic enzymes during liquefaction. Therefore, starch in corn harvested at or before maturity is easier to hydrolyze into sugars by enzymes, thereby causing higher ethanol conversion efficiency in the dry grind process.

Early harvested corn poses a problem of storability due to its high moisture content, which needs strong postharvest management to reduce the loss during handling, transportation, and storage. Postharvest losses due to spoilage during storage remain a major problem around the world. Early detection of corn spoilage will reduce corn losses, help prevent production of noxious mycotoxins in the food chain, and avoid financial loss by applying timely management (e.g. stirring, aeration, etc.) (Ileleji et al., 2006). Utilizing thermal cables in storage structures for temperature monitoring has been the traditional method for detecting heat in bulk grain; however, because of the low thermal diffusivity of bulk grain, a single temperature measurement is usually not sensitive enough (Singh et al., 1983). Measured temperature cannot be easily interpreted due to the influence of the ambient air fluctuation. For example, temperature of 25°C of bulk grain may mean there is a possible spoilage spot in winter, or it may mean the bulk grain is heated by the ambient air in the summer. Previous work (Maier et al., 2006) has shown that monitoring of the head space of a bin with a carbon dioxide (CO₂) sensor can lead to
earlier detection of microbial degradation of grain than temperature measurement alone. As diffusion is expected to be one of the major factors in gas movement through bulk grain, it is important to determine the effective diffusion coefficient of CO$_2$ through bulk grain.

The overall goal of this study was to examine the feasibility of early harvest of corn and corn stover and determine the implications of this proposed practice on corn processing characteristics; chemical composition and nutritive values of the corn and its stover; and storability of high moisture corn. The specific objectives of the project were to

1. determine the harvest date influence on dry matter yields and moisture contents of corn and corn stover in the U.S. Midwest Corn Belt;

2. investigate the effect of corn harvest moisture on corn processing characteristics, including wet milling and dry grind fermentation processes;

3. determine the chemical compositions and nutritive values of corn, corn stover and dried distillers grain solubles (DDGS) at various corn harvest moisture contents and determine the harvest time to maximize corn plant utilization; and

4. determine the respiration rate and the effective diffusivity of CO$_2$ through bulk corn as a function of temperature and moisture content for developing corn storage monitoring system based on CO$_2$ measurements.
Chapter 2. Literature review

2.1 Dry matter yield and moisture contents of corn and corn stover at harvest

Corn is a tall, annual plant from the grass family. It has a fibrous root system and an erect stalk with a single leaf at each node and leaf in two opposite ranks. Each leaf consists of a sheath surrounding the stalk and an expanded leaf blade connected to the sheath by a blade joint. Normally, the tassel is located at the top of the main stalk and the ears are located at the end of short branches. The ears, surrounded by husks, grow to contain 500-1,200 developed kernels arranged in rows along a cob (Farnham et al., 2003). The corn plant biomass mainly contains five parts: roots, stalks, leaves, husks, cob and corn. Corn stover is defined as the above-ground portions of the corn plant, including stalks, leaves, husks and cob, but excluding corn (Wyman, 2003).

2.1.1 Dry matter yield of corn and corn stover at harvest time

After fertilization is complete, the main function of the plant is to develop corn ears. Nutrients are translocated from corn stover to corn kernels before kernels reach their physiological maturity. Corn kernels stop taking nourishment from the plant and generally complete the yield-making process about the same time a “black layer” develops at the tip of the kernel, where the kernel attaches to the cob (Erickson and Valentin, 2008). As harvest time delays, corn stover loses biomass primarily due to the nutrient translocation from stover to kernel before kernel physiological maturity (Center et al., 1970) and physical loss of leaf and husk as they become dry and brittle after kernel physiological maturity (Liu et al., 2009).

The effect of harvest time on dry matter yield of the corn stover has been widely studied. Cummins (1970) in Georgia stated the total dry matter yield of corn stover declined by 19% from 4 September to 26 September. Russell (1986) in Iowa collected corn stover samples at varying intervals from three weeks before to five weeks after corn physiological maturity and found that dry matter yield of stover decreased at 0.16 g per day for each corn plant. Studies in Tennessee reported that the dry matter yield of the corn stover peaked at the time of kernel physiological maturity, and decreased by almost 40% when harvest was delayed 90 days after kernel maturity (Pordesimo et al., 2004a,
Shinners and Binversie (2007) found the dry matter yield of corn stover decreased by around 20% between August and October over a three-year period study in Wisconsin. In China, Liu et al. (2009) reported corn stover yield reduced by 48% when harvest time was delayed from kernel filling stage to the following spring. There is strong evidence from previous studies that dry matter yield of corn stover decreased at various levels as harvest time delayed.

### 2.1.2 Moisture contents of corn and corn stover at harvest time

Moisture is a critical factor for efficient and safe collection, processing, drying and storage of both corn and corn stover. High moisture corn and corn stover are expensive to dry and spoil readily. High moisture biomaterials are also prone to spontaneous combustion and fires (Jenkins and Sumner, 1986). For an individual field, the optimum time to harvest is largely a balance of drying expenses from an early harvest and of usually greater field losses if waiting for the corn to dry. Understanding the effects of harvest time on the moistures of corn and corn stover would help corn producers to manage the optimum time to harvest and minimize field loss.

Corn kernel moisture content decreases as the kernel develops through the blister stage (~85% M.C.), milk stage (~80% M.C.), dough stage (~70% M.C.), dent stage (~55% M.C.), and physiological maturity (~30-40% M.C.). Prior to physiological maturity, decreases in kernel moisture occur from a combination of the accumulation of dry matter (such as starch) plus the actual water loss via evaporation. After physiological maturity (kernel moisture at 30-40%), decreases in kernel moisture are primarily due to water loss via evaporation (Nielsen, 2009).

The average kernel drying rate largely depends on weather and varies considerably from year to year. Corn kernel in the field would dry very little when weather conditions are cool and wet, but can lose more than one percentage point of moisture in a warm and dry day. Studies in recent years showed that the average corn kernel drying rate ranges between 0.4-1.0 percentage points per day in the U.S. Corn Belt (Table 1). Corn kernel drying rate in the U.S. Southern Corn Belt (Tennessee) is higher than that in the U.S. Northern Corn Belt (Wisconsin), which was probably due to the higher temperature and lower humidity weather in the U.S. Southern Corn Belt.
Table 1. Corn kernel drying rate at various locations (studies from 2004 to present).

<table>
<thead>
<tr>
<th>Corn drying rate (percentage points per day)</th>
<th>Location</th>
<th>Moisture range</th>
<th>Corn varieties</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.4-1.0</td>
<td>Illinois, U.S.</td>
<td>57% -15%</td>
<td>Pioneer 32D78, Pioneer P0916XR</td>
<td>Huang et al., (2012a)</td>
</tr>
<tr>
<td>0.4-0.6</td>
<td>Indiana, U.S.</td>
<td>40% - 15%</td>
<td>-</td>
<td>Nielson, (2011)</td>
</tr>
<tr>
<td>0.5</td>
<td>Wisconsin, U.S.</td>
<td>40% - 20%</td>
<td>Pioneer 35R58, Kaltenberg 6789, Agri-Gold 6382, DeKalb 570RR</td>
<td>Shinners et al., (2007)</td>
</tr>
<tr>
<td>0.8</td>
<td>Tennessee, U.S.</td>
<td>40% - 15%</td>
<td>Pioneer 32K64, Pioneer 32K61</td>
<td>Pordesimo et al., (2004b)</td>
</tr>
</tbody>
</table>

Corn stover moisture content is typically regarded as twice that of the corn kernel (Nielsen, 1995). Pordesimo et al. (2004b) showed that the total stover to corn moisture ratio was 2.0-2.5 with corn moisture between 18% and 31% in the U.S. Southern Corn Belt, Tennessee. Shinners and Binversie (2007) reported that the total stover to corn ratio averaged 2.15 during the typical harvest period in the U.S. Northern Corn Belt, Wisconsin. Relative high moisture in stover makes artificial drying uneconomical (Kaminsky, 1989). Nielsen (1995) suggested that the stover moisture should not exceed 30% M.C. for optimum harvesting and needs to be 20% or less for dry storage. Field drying of corn stover is common in the U.S., but it still has some drawbacks such as the low collection efficiency, high soil contamination and insect invasion. Harvesting corn stover at moisture contents greater than 50% and ensiling it immediately after harvest is an alternative. Shinners et al. (2007, 2011) found that ensiling resulted in lower dry mater losses and more uniform product moisture compared to dry stover bales stored outdoors.

2.2 Composition of corn and corn stover at harvest

2.2.1 Structure and composition of corn

Corn structure

The corn kernel is composed of three main parts: germ, endosperm, and pericarp (Figure 1). The tip cap is the conical, fibrous structure that remains attached to the kernel
(Wolf et al., 1952). During corn development, photosynthesis products are moved into the developing kernel through the tip cap.

Corn endosperm is the tissue produced inside the corn kernel around the time of fertilization. It surrounds the embryo and provides nutrition in the form of starch. The endosperm constitutes 82-84% of the kernel dry weight and is composed of 86-89% starch by weight (Earle et al., 1946). The corn endosperm could be divided into two parts: horny endosperm and floury endosperm. Horny endosperm is hard and translucent, whereas floury endosperm is soft and opaque. The starch granules in horny endosperm are polygonal shaped and tightly packed with few or no air spaces; and the starch granules in floury endosperm are spherical and loosely packed. The germ, which is the reproductive part that germinates to grow to a plant, is composed of the embryo and scutellum, making up 10-12% of the kernel dry weight. The germ contains 81-85% of the total corn oil, most of which is located in scutellum (Earle et al., 1946), making it a good source of vegetable oil for food ingredients and pharmaceutical uses. The pericarp is the CO2 layer of a corn kernel and it protects the endosperm and the germ from microorganism invasion. It makes up to 5-6% of the kernel dry weight and its main composition is fiber.

**Figure 1.** Structure of corn kernel.
Corn composition

The composition of the whole kernel is of utmost importance in both the corn processing plants and animal feed industry. The main composition of the mature kernel includes starch, fat, protein, fiber, sugar and ash (Earle et al., 1946).

Starch, which accounts for approximately 68-74% of a mature kernel by weight, is the primary source of stored energy in corn (Earle et al., 1946). It is synthesized in special organelles called amyloplasts (Badenhuizen, 1965). The starch consists of two types of molecules: the linear and helical amylose and the branched amyllopectin. Yellow dent corn starch generally contains 23-25% amylose and 75-77% amyllopectin. The amylose/amylopectin ratio in corn starch varies during kernel development. The amylose content of corn starch increased from 9.2% on 14 days after pollination (DAP) to 24.4% on 45 DAP (mature and dried) (Shannon and Garwood, 1984; Li et al., 2007).

Ingle et al. (1965) determined changes in dry matter and composition of corn kernel during development. The development of the embryo was found to be slower than the development of the endosperm, beginning at 15 DAP and proceeding at a linear fashion. The sugar content in the kernel peaked at 18 DAP and decreased thereafter. The decrease in soluble sugar content was a result of conversions from sugar to starch during kernel development. The protein content deposition began at the beginning of kernel maturing, accelerated at 15 DAP, and leveled off at 30 DAP. The second increase in protein content of the kernel, occurring at 40 DAP, was indicative of the production of specialized storage protein (McKee, 1958). The decrease in amino acid content after 28 DAP indicated these components are utilized for the protein production.

2.2.2 Composition of corn stover at harvest

In the animal feed industry, neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL) and crude protein (CP) are measures of the nutritive value of forage. NDF is the percentage of fiber in a forage sample which is not soluble in a neutral detergent solution and it is a good indicator to predict intake by animals consuming the feedstuff. ADF is the percentage of fiber in a forage sample which is insoluble in a weak acid and it is a very important value for forages because it closely
relates to the ability of animals to digest the forage. ADL is a polymer of phenyl propane units, which is the indigestible part of the forage. Crude protein, measured by multiplying total nitrogen by 6.25, is another measure of the forage nutritive value. Crude protein includes both the true protein and non-protein nitrogen. Because of the low crude protein content (3.5-8.7%) (Darby and Lauer, 2002; Lewis et al., 2004; Lee et al., 2007), corn stover is usually supplemented with protein or mixed with other high protein products (e.g., DDGS, soybean meal) to increase its nutritive value (Sewell et al., 2009).

The corn stover composition changes during corn kernel development and dry down. Weaver et al. (1978) reported that NDF concentrations in corn stover increased with kernel maturity. Irlbeck et al. (1993) found that corn stover harvested 28 days post-physiological maturity had higher concentrations of NDF and ADF but lower concentrations of crude protein than stover harvested at its physiological maturity. The decrease in crude protein concentration was also reported by other studies (Lewis, et al., 2004; Darby and Lauer, 2002). The decrease in crude protein concentration appears to be the result of continued carbon assimilation during corn kernel maturing, even though nitrogen uptake was probably completed, thereby diluting plant nitrogen concentration (Wiersma et al., 1993). Several studies reported lignin concentration in corn stover increased with kernel development and dry down (Cone and Engels, 1993; Pordesimo et al., 2005; Liu et al., 2009).

2.3 CO₂ coefficient measurement through bulk corn

2.3.1 Apparatus to measure effective diffusivity of CO₂ through bulk grain

High respiration of bulk corn in the spoilage spot increases CO₂ concentration locally and CO₂ tends to diffuse through the interstitial air to the rest of the storage bin under the influence of the concentration gradient. Due to its simplicity, Fick’s Law is the most popular approach to describe the diffusion process:

\[ J_A = -D \frac{\partial \phi}{\partial x} \] (1)
where \( J_A \) is the diffusion flux, \( \nabla \varphi \) is the species concentration gradient and \( D \) is the diffusion coefficient or diffusivity. The effective diffusion coefficient \( (D_e) \) describes diffusion through the pore spaces of porous media. In porous media, part of the total volume is occupied by a solid phase. The diffusion gas molecules have to follow a tortuous path in the porous matrix, which provides extra resistance to the gas movement and slows the gas diffusion rate. The effective diffusion coefficient through porous media can be estimated as follows:

\[
D_e = \frac{D \varepsilon \delta}{\tau}
\]  

where \( \varepsilon \) is the porosity of the porous media, \( \tau \) is the tortuosity of the porous media, and \( \delta \) is the constrictivity. Since it is difficult to precisely measure the tortuosity of the porous media, the effective diffusion coefficient is often measured directly through the experiments for practical applications.

Singh et al. (1984) determined the effective diffusion coefficient of CO\(_2\) through wheat, rapeseed, oats and corn by a steady state method. In Singh et al.’s study, effective diffusivity of CO\(_2\) though bulk corn was measured only at 20% corn moisture content and 10 °C. The apparatus used by Singh et al. (1984) for measuring CO\(_2\) diffusivity consisted of a gas chamber and a cylindrical grain column (Figure 2a). One end of the grain column was connected to the gas chamber and the other end of the grain column was connected to the atmosphere. It was also assumed that the concentration at the outer end of the grain column is equal to the atmospheric CO\(_2\) concentration. The CO\(_2\) was injected into the gas chamber, and diffused through the cylindrical column filled with grain. By measuring the CO\(_2\) concentrations along the cylindrical column when the CO\(_2\) diffusion reached its equilibrium state, the diffusion coefficient of CO\(_2\) in bulk grain could be calculated. The main advantage of the steady state method is that it can provide the direct measurement of the effective diffusion coefficient. However, since measurements are possible only after equilibrium is reached, steady state methods are usually slow; several days often being required for one measurement (Flegg, 1953). During such long tests, pressure inside the chamber may change, which would affect the diffusion coefficient. Thereafter, Singh et al. (1985) developed a transient method to determine the
effective diffusion coefficient of CO$_2$ through bulk wheat. The results obtained compared well with the previous data published by Henderson and Oxley (1944).

Shunmugam et al. (2005) determined the effective diffusion coefficient of CO$_2$ through bulk storage of wheat, barley and canola at different temperatures, moisture contents, porosities and directions of gas flow, using an apparatus (Figure 2b) similar to Singh et al. (1984). Shunmugam et al. (2005) used a transient diffusion model to measure the effective diffusion coefficient. Gas samples were drawn at designed locations of the grain column each hour for six hours during CO$_2$ diffusion and were analyzed using a gas chromatograph. The measured data at each time interval were inserted into the time dependent governing equation

\[ \frac{\partial c}{\partial t} = D_e \frac{\partial^2 c}{\partial x^2} + S_o \] (3)

where $c$ is concentration of CO$_2$ at time $t$ and location $x$, $D_e$ is the effective diffusion coefficient of CO$_2$ through bulk grain, and $S_o$ is the sorption of CO$_2$ into the grain mass. The CO$_2$ diffusion coefficient was obtained by solving the governing equation using a finite different method. Compared to the steady state method, the transient method is faster but the mathematical interpretation of the results is more tedious. As a drawback in their research, drawing the gas samples from bulk grain would induce the local gas movement in the grain column, which makes the diffusivity measurements less accurate. Further, they neglected grain respiration, which would largely affect the diffusion pattern when grain moisture content is high.
Figure 2. Schematic of the apparatuses used to measure the rate of diffusion of carbon dioxide through grains and oilseeds (adapted from Singh et al., 1984 (Figure a) and Shunmugam et al., 2005 (Figure b)). Each of the apparatus consisted of a gas chamber and a grain column. For the apparatus a, one end of the grain column was connected to the gas chamber and the other end of the grain column was connected to the atmosphere. And it was assumed that the concentration at the inner end of the grain column is equal to the gas chamber CO$_2$ concentration and the concentration at the outer end of the grain column is equal to the atmospheric CO$_2$ concentration. For the apparatus b, the outer end of the grain column was insulated from the atmosphere, therefore, CO$_2$ flux between atmosphere and grain column was zero ($\frac{dc}{dx} = 0$).

2.3.2 Other diffusion apparatus to measure gas diffusivity through porous media

Several other diffusion apparatuses have been used to measure the effective gas diffusion coefficient through porous media (Ball et al., 1981; Dudukovic, 1982). Ball et al. (1981) considered a closed system in which a diffusion column was mounted between two closed chambers to measure the gas diffusion in soil (Figure 3a). One of the chambers was injected with gas A while the other chamber was filled with gas B. Since the start of the diffusion process, the concentration of gas A in each chamber was recorded as a function of time. The effective diffusion coefficient was determined either by an analytical method method. Dudukovic (1982) proposed using a cylindrical diffusion column with pellet materials between two open gas chambers (Figure 3b). Pure gas B (carrier gas) flowed initially across both the lower and upper chamber of the
apparatus. Gas A was introduced into the lower gas chamber, and the concentration of the gas A was measured as a function of time at the exit of the upper gas chamber (or sometimes at exits of both chambers). The effective diffusivity was then determined by matching a suitable mathematical model with observed concentrations. The first measurement method (Figure 3a) required the target gas to be well mixed in the chambers at all times. The second measurement method (Figure 3b) solved the mixing problem in the gas chamber and allowed greater flexibility in operation. However, extra care was needed for the pressure balance between two chambers. In order to accurately measure the effective diffusivity, pressures on both sides of the chambers should be maintained equal so that transfer through the columns is by diffusion alone. The moving gas in the chambers could produce pressure difference between the chambers, thereby causing convective movement of gas through porous media. The summary of the diffusion cells used to measure gas diffusivity are listed in Table 2.

Figure 3. Schematic of the diffusion apparatuses to measure gas diffusivity through porous media. a) apparatus used by Ball et al. (1981) and b) apparatus used by Dudukovic (1982). Both of apparatuses were designed based on the transient diffusion methods.
### Table 2. Description of selected apparatus for measuring gas diffusion coefficient in porous media

<table>
<thead>
<tr>
<th>Methods</th>
<th>Mathematical model</th>
<th>Media</th>
<th>Advantages</th>
<th>Disadvantages</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>One gas chamber— with other end open (Figure 2a)</td>
<td>( D_e = \frac{Q \cdot \Delta L}{S \cdot \Delta \mathcal{C} \cdot t} )</td>
<td>wheat, rapeseed, oats, corn</td>
<td>Direct measurement of diffusion coefficient</td>
<td>Time consuming; Easily affected by atmospheric pressure fluctuations</td>
<td>Singh et al. (1984);</td>
</tr>
</tbody>
</table>
| One chamber – with other end sealed (Figure 2b) | \( \frac{\partial \mathcal{C}}{\partial t} = D_e \frac{\partial^2 \mathcal{C}}{\partial x^2} + q \)  
1. \( \mathcal{C}(x, t) = \varphi(t) \) at \( x = 0 \)  
2. \( \frac{\partial \mathcal{C}}{\partial x} = 0 \) at \( x = L \)  
3. \( \mathcal{C}(x, 0) = f(x) \) | wheat, barley, canola | Faster than the steady state method; Closed system immune to atmospheric fluctuations | More complex mathematical model; Need to measure gas concentration along the diffusion cell | Shunmugam et al. (2005); Paes et al. (2011) |
| Two closed chambers (Figure 3a)              | \( \frac{\partial \mathcal{C}}{\partial t} = D_e \frac{\partial^2 \mathcal{C}}{\partial x^2} \)  
Boundary and initial conditions  
1. \( V \frac{\partial \mathcal{C}}{\partial t} = S D_e \frac{\partial \mathcal{C}}{\partial x} \) at \( x = 0 \)  
2. \( V \frac{\partial \mathcal{C}}{\partial t} = -S D_e \frac{\partial \mathcal{C}}{\partial x} \) at \( x = L \)  
3. \( \mathcal{C}(x, 0) = f(x) \) | soil, sand, steel wool, glass beads | Only few measurements required; Easy to operate; Closed system immune to atmosphere fluctuation | More complex mathematical model than the one chamber method; Require evenly distribution of the target gas in the chambers | Dye & Dallavalle (1958); Ball et al. (1981); Jones et al. (2003); Reible and Shair (1982); Allaire et al. (2008) |
| Two open chambers (Figure 3b)                | \( \frac{1}{\theta_1}(C_i - C_1) + \frac{P_1}{\theta_1 B_i} \frac{\partial C_1}{\partial x} = \frac{\partial C_1}{\partial t} \) \( \mathcal{C}(x, t) \) \( \mathcal{C}(x, t) \)  
\( \frac{1}{\theta_2}(C_r - C_2) + \frac{P_2}{\theta_2 B_i} \frac{\partial C_2}{\partial x} = \frac{\partial C_2}{\partial t} \) \( \mathcal{C}(x, t) \) \( \mathcal{C}(x, t) \) | soil | Do not require evenly distribution of the target gas in the chamber; Greater flexibility; | Difficult to balance the pressure between the two chambers | Dudukovic (1982) |

---

*a* \( \mathcal{C}(x, t) \): \( \text{CO}_2 \) concentration at distance \( x \) and time \( t \); \( L \): length of the diffusion column; \( S \): area of the cross section of the diffusion column; 
\( V_i \): volume of the gas injection chamber; \( C_i(t) \): \( \text{CO}_2 \) concentration in the injection chamber at time \( t \); \( V_r \): volume of the gas receiving chamber; \( C_r(t) \): \( \text{CO}_2 \) concentration in the gas receiving chamber.
Chapter 3. Effect of harvest date on dry matter yield and moisture content of corn and corn stover

3.1 Introduction

Traditionally, corn and corn stover were harvested after corn physiological maturity and at low moisture content levels due to high cost of drying. However, if corn and corn stover were harvested at high moisture contents before dry down, the stover would be less lignified and more responsive to chemical treatment to enhance digestibility (Cone and Engels, 1993). Also with early harvest of corn stover, corn stover yield would be higher due to the reduced physical loss of leaf and husk as they became dry and brittle after corn maturity (Shinners and Binversie, 2007). Harvesting corn at or before physiological maturity may be advantageous to corn processing if it can be stored wet, because starch in early harvested corn is more digestible by enzymes (Shannon and Garwood, 1984; Hane and Robyt, 1984; Philippeau and Michalet-Doreau, 1997). Furthermore, early removal of corn from the field may allow the production of a second crop that is nitrogen fixing. High moisture is the main problem for early harvesting corn and corn stover. High moisture corn and corn stover spoil readily, and it is a safety hazard when they are moldy.

In order to systematically evaluate the feasibility of early harvest corn and corn stover, the first step is to characterize dry matter yield and moisture contents of corn and corn stover at different maturity stages. Therefore, the specific objectives were 1) to determine dry matter yields and moisture contents of corn and corn stover before, during and after corn maturity; 2) to ascertain effect of mold on corn quality after corn kernel physiological maturity and effect of corn plant lodging.

3.2 Materials and methods

3.2.1 Test site and material

Corn was grown at the University of Illinois at Urbana-Champaign, Agricultural and Biological Engineering Farm, located in Urbana, Illinois, USA, with a geographical coordinate at 40.07044 N, 88.21034 W. In 2009, a regular dent hybrid 32D78 (Pioneer Hi-Bred International, Johnston, Iowa) was chosen due to its high potential yield and
high total fermentables for dry grind ethanol. In 2010, another dent hybrid P0916XR was chosen for its high potential yield. These hybrids had approximately similar RM (relative maturity) that was recommended for Central Illinois by the seed company (Pioneer Hi-Bred International, Johnson, Iowa). The relative maturity (RM) for 32D78 and P0916XR were 116 and 109, respectively. The growing degree days (GDDs) required for these two hybrids to reach physiological maturity were 1572 and 1461, respectively.

The corn crops were planted on 22 May 2009, which was deemed late for the central Illinois area and due to the frequent rainfalls and low temperature in April and May. Delayed planting postponed the corn maturity and cool weather in the fall affected the corn in-field drying rate. In 2010 the corn crops were planted on 21 April. In both years, the corn crops were planted on 76 cm (30 inch) row spacing and 15.6 cm (6 1/8 inch) rank spacing in a field with a size of 0.4 ha (1 acre). The target plant population for both years was 75,680 plants per hectare.

3.2.2 Weather conditions
Corn plant growth, yield and drying rate are largely dependent on accumulated heat input and precipitation during the growing and harvest season (Pordesimo et al, 2004a; Shinners and Binversie). Ambient air temperature and precipitation at the experimental location were obtained from National Oceanic and Atmospheric Administration (NOAA) (http://www.noaa.gov/). Growing-degree days were calculated using the following equation:

\[
\text{GDDs} = \left(\frac{T_{\text{max}} + T_{\text{min}}}{2}\right) - 10 \text{ °C}
\]

where \(T_{\text{max}}\) is the daily maximum temperature (with an upper limit of 30°C) and \(T_{\text{min}}\) is the daily minimum temperature (with a lower limit of 10°C) (McMaster and Wilhem, 1997; Darby and Lauer, 2002).

3.2.3 Sample collection
In 2009, delayed planting date and lower heat input (compared with an average year) postponed the corn maturity date to September. Corn plants were harvested bi-weekly between August and November, representing corn kernel filling, mature and dry down
In 2010, corn reached its maturity in August and was harvested weekly between 29 July and 10 September. The frequency of sampling was due to the higher in-field drying rate of corn in 2010.

For each harvest, corn plants were collected in a 3.05 m × 1.30 m rectangular plot. All plants from the plot were pooled together to form a representative sample. Three replications at different sites (designated southwest, southeast, and north of the experimental field) were conducted for each harvest (Figure 4). The plants were cut by hand at 10 cm above ground and transported to the laboratory for processing. Ears were separated from the plants, followed by the separation of the corn from the ears by hand, yielding three fractions: corn, cob and stalk & leaf fraction. The husk was added to the stalk and leaf fraction. It was observed that the moisture content of leaves was different from that of stalks and husks. To get a uniform moisture content sample, the stalk & leaf was chopped with a laboratory-scale chipper/shredder (Model 410, Troy-Bilt LLC, Cleveland, OH). The cobs were separately chopped to get a uniform sample for moisture content determination. The fractions of corn, chopped cob, chopped stalk & leaf were weighed separately.

**Figure 4.** Sampling locations and area in the ABE farm at University of Illinois at Urbana-Champaign. The size of each sampling location was 3.05 m × 1.30 m (L × W). (Figure from Google satellite map, accessed in April, 2012).
3.2.4 Moisture, dry matter yield, lodging percentage and moldy kernel percentage determination

The moisture content of corn kernels was measured at 103°C for 72 h using a convection oven (AACC International, 2000a). The chopped cob and chopped corn stover were first dried at 49 °C for 24 h followed by 135 °C for 2 h (AACC International, 2000b). The wet basis moisture contents and dry matter content of all sub-fractions were then calculated. The number of corn plant falling on ground in each harvest plot was recorded. The corn plant lodging percentage was determined as a fraction of the number of falling down corn plants in the total number of corn plants in each harvest plot. In our study, the corn plant lodging included both stalk lodging and root lodging. Corn kernels which had visible mold were separated by hand and weighed. The fraction of corn with mold (moldy kernel percentage) was calculated as a percent of total corn dry mass (Anderson et al., 1972).

3.3 Results and discussion

3.3.1 Weather conditions

Accumulative growing degree days (AGDDs) and precipitation information in 2009, 2010 and 10-year average during corn kernel filling, mature, and dry down stages are shown in Figure 5a and 5b. These weather parameters would affect not only stover and corn dry matter yield, but also in-field drying rates. The AGDDs in 2009 were lower than that in 10-year average; while the AGDDs in 2010 were higher than that in 10-year average (Figure 5a). The inadequate accumulative heat input in 2009 postponed the corn mature date and reduced the in-field drying rate of corn. The accumulated precipitation in 2009 was above the 10-year average; while in 2010, the accumulated precipitation was below the 10-year average before mid-June, and went above the 10-year average after mid-June, due to the frequent rainfalls in June (Figure 5b).
Figure 5. Accumulated growing-degree days (AGDDs) (a) and accumulated precipitation (b) during corn growing and harvest period for 2009, 2010 and 10-year average.

3.3.2 Corn and stover dry matter yield and distribution

In 2009, the corn yield increased rapidly until reaching physiological maturity when corn moisture was 40% (Figure 6a). After maturity, the corn dry matter yield became stable until the end of the study, ranging between 11.4 and 11.9 t/ha, with an average of 11.5 t/ha. In 2010, corn reached its physiological maturity when corn moisture was 35% (Figure 6b). After corn maturity, the corn dry matter yield ranged between 10.6 t/ha and 11.1 t/ha with an average of 10.8 t/ha. Difference in corn yield between 2009 and 2010 was observed, which could be due to the differences in corn varieties and weather conditions.
In 2009 and 2010, the dry matter yields of corn stover were greatest at the beginning of the study and decreased throughout the whole study. In 2009, stover yield decreased by 34% from the beginning to the end of the study (from 15.4 t/ha to 10.1 t/ha) (Figure 6a). In 2010, corn stover yield decreased by 17% from the beginning to the end of the study (from 14.1 t/ha to 11.6 t/ha) (Figure 6b). The loss of biomass was mainly due to nutrients translocation from corn stover to corn before corn physiological maturity (Center et al., 1970) and physical loss of leaf and husk as they became dry and brittle after corn maturity (Liu et al., 2009). This phenomenon was also reported by other studies during different lengths of study period. Cummins (1970) in Georgia stated the total dry matter yield of stover declined by 19% from 4 September to 26 September. Studies (Pordesimo et al., 2004a, 2004b) in Tennessee reported that the dry matter yield of the stover peaked at the time of corn physiological maturity, and decreased by almost 40% when harvest was delayed until winter. Shinners and Binversie (2007) found the dry matter yield of stover decreased by about 20% between August and October over a 3-year period study in Wisconsin. The higher dry matter loss in stover in 2009 than that in 2010 might be due to several reasons. Firstly, in 2009, the length of study period was 94 days (from 21 August to 23 November), while the length of study period in 2010 was only 43 days (from 29 July to 10 September). A longer study period would obviously lead to a higher dry matter loss in stover. Secondly, the greater rainfall in 2009 would also tend to increase the loss of leaf and husk after plant maturity. Thirdly, variety difference in corn plants could also affect the corn stover yield. Compared to stalk & leaf fraction, dry matter loss in the cob during the entire study period was lower (less than 10%), both in 2009 and 2010. The loss of cob more likely represented deterioration of the dry matter by cellular respiration, microbial action or insect activity.
Figure 6. Dry matter yield of corn plant fractions in seven harvests in 2009 (a) and 2010 (b). The husk was added to the faction of stalk & leaf fraction.
3.3.3 Corn and corn stover moistures and field drying rates

Moisture content of corn and corn stover is important because it affects the selection of harvest equipment and storage methods, which would finally affect the cost of corn and corn stover collection. In 2009, the moisture content of corn decreased rapidly before corn physiological maturity, from 72% to 40%, with an average drying rate at 0.8 percentage points per day (Figure 7a). After corn maturity, the drying rate of corn was lower, with an average of 0.3 percentage points per day. At the end of the study period (23 November), the corn moisture was around 21%. In 2010, the moisture content of corn decreased steadily throughout the study period, from 57% to 15% (Figure 7b). The corn moisture was 35% at its maturity stage (19 August). The average drying rate of corn was almost 1 percentage point per day. The drying rates found in this study were different than those reported by other studies. Pordesimo et al. (2004b) reported a drying rate of 0.8 percentage points per day in the U.S. Southern Corn Belt, Tennessee. Shinners and Binversie (2007) found that the drying rate was approximately 0.6 percentage points per day in the U.S. Northern Born Belt, Wisconsin. Nielson et al. (2009) in Indiana reported that corn would dry approximately 0.25-0.5 percentage points per day in late October to early November. The main reason for different drying rates could be due to the differences in weather conditions and daylight length at different locations.

Moisture content of corn stover is usually much higher than that of corn. As a rule of thumb, corn stover moisture is twice that of the corn (Nielson, 1995; Buchele, 1975). In 2009, the moisture content of corn stover was around 76% at the beginning of the study (21 August), and decreased slightly between 21 August and 13 October (Figure 7a). After that, the moisture content decreased rapidly from 72% to 47%, with an average drying rate of 0.6 percentage points per day. In 2010, when the weather was warmer with less precipitation, the moisture content of stover remained stable at the beginning of the study (Figure 7b). After that, the moisture content decreased steadily throughout the study, from 73% to 55%, with an average drying rate of 0.5 percentage points per day. The leaf and husk in stover was the main source of moisture loss because of their large surface area and thin cross-section which promotes drying (Shinners and Binversie, 2007). In 2009, moisture content of cob decreased from 70% to 37% in the 94-day study period.
In 2010, moisture content of cob decreased from 61% to 27% in the 43-day study period. One interesting observation both in 2009 and 2010 is that moisture content of cob started to decrease rapidly when moisture content of corn was below 30%. This phenomenon might be due to the high moisture gradient between cob and corn when the moisture content of corn is low. The 2-year average results showed that when corn was at its maturity stage, the moisture contents of corn, stover and cob were 37.7, 71.1 and 54.5% respectively. The high moisture content in corn stover indicated that ensiling could be an effective storage option when the stover is harvested in a single-pass.
Figure 7. Moisture content (w.b.) of corn plant fractions in seven harvests in 2009 (a) and 2010 (b). The husk was included with the stalk & leaf fraction.
3.3.4 Field losses

Corn losses prior to harvest are known to increase the longer the crop stands in the field after reaching maturity. These losses are mainly due to wind, animals, lodging, senescence, microbial activity and biological degradation. Mold can form on corn ears during cool, wet growing seasons. The severity of the problem depends on variations in weather, crop genetics, and field management practices. High mold levels in corn can reduce the corn grade and storage life and affect livestock health and performance (Nielsen et al., 2009). Table 3 shows the moldy kernel and stalk lodging percentages in seven harvests during the 43-day study period in 2010. Corn mold became significant (1.7%) when corn moisture was 42%, which in this study was at R6 maturity stage. After that, the moldy kernel percentage increased steadily as corn moisture decreased. At the end of the study, at corn moisture of 15%, the moldy kernel percentage reached 4.1%, which could potentially reduce the corn quality from No.1 grade to lower grades and reduce the corn marketability (USDA, 2012).

In most years growers can get the crop out of the field early enough that corn plant lodging is not a major problem; however, lodging could be a major harvest loss when weather is severe, such as high wind and rainfall. This study showed that the lodging started when corn moisture was 35%, when the corn just reached its physiological maturity (Table 3). At that time, 1.2% of the corn plants were lodged. As harvest was delayed, lodging became greater. At the end of the study, at corn moisture of 15%, the stalk lodging percentage reached 3.6%.

Table 3. Corn field loss information during a 43-day study period in 2010.

<table>
<thead>
<tr>
<th>Corn moisture</th>
<th>57%</th>
<th>49%</th>
<th>42%</th>
<th>35%</th>
<th>27%</th>
<th>21%</th>
<th>15%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moldy kernels (%)</td>
<td>0</td>
<td>0</td>
<td>1.7</td>
<td>2.1</td>
<td>3.3</td>
<td>3.3</td>
<td>4.1</td>
</tr>
<tr>
<td>Stalk Lodging (%)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1.2</td>
<td>2.4</td>
<td>3.6</td>
<td>3.6</td>
</tr>
</tbody>
</table>

3.4 Conclusions

The two year average corn moisture content was 37.4% when reaching physiological maturity. At the same time, the average stover moisture was 67.7%. The dry matter yield of corn increased rapidly until reaching corn maturity and remained stable after
corn maturity, with an average yield of 11.1 t/ha over the two year study. For corn stover, the two-year average dry matter yield was 14.8 t/ha at the beginning of the study (filling stage) and it decreased to 13.2 t/ha at corn physiological maturity and decreased throughout corn dry down. During corn dry down period, moldy kernel percentage increased from 2.1 to 4.1%, and plant lodging percentage increased from 1.2 to 3.6%.
Chapter 4. Effect of corn harvest moisture on dry grind fermentation characteristics and DDGS composition

4.1 Introduction

Corn harvested at different moisture contents may exhibit different processing characteristics for ethanol industry, due to their differences in physical and chemical properties. Wolf et al. (1948) compared corn starch at different maturity stages from three corn cultivars, and found lower starch content in immature corn. Li et al. (2007) reported that the amylose content of endosperm starch increased from 9.2% on 14 days after pollination (DAP) to 24.4% on 45 DAP (mature and dried). Shannon and Garwood (1984) also reported that the level of amylose present in a developing kernel increases as the kernel matured. The resistant starch levels in corn starch is highly correlated with amylose levels (Berry 1986), because amylose forms a thermally stable starch resistant to enzymatic reactions to break down starch to glucose (Jane and Robyt 1984). Resistant starch present corn starch has been reported to have many health benefits, including prevention of colon cancer, type II diabetes, obesity and cardiovascular disease (Englyst et al., 1986; Robertson et al., 2005; Jiang et al., 2010). However, for corn-ethanol industry, higher resistant starch could lead to a decreased ethanol yield. Sharma et al. (2010) reported that 30%-amylose starch had higher resistant starch content at initial, after liquefaction and simultaneous saccharification and fermentation (SSF) than 0% amylose starch in dry grind process. Sharma et al. (2010) also found that higher resistant starch resulted in lower conversion of starch into sugars and hence to lower final ethanol yield. All of the above information lead to the hypothesis that corn harvested at early stage before corn dry down may have higher fermentability for ethanol. The objective of this study was to investigate the effect of corn harvest moisture content on dry grind fermentation characteristics and DDGS composition.

4.2 Materials and methods

4.2.1 Experimental materials

The corn hybrid 32D78 harvested at seven moisture contents in 2009 was used as the experimental material for dry grind fermentation. Corn samples were collected on 21
August, 09 September, 30 September, 13 October, 25 October, 09 November, and 23 November, at moisture contents of 73, 54, 40, 37, 30, 24 and 21% respectively. The corn kernels reached their physiological maturity at the moisture content of 40%.

4.2.2 Enzymes and yeast

Maxaliq™ ONE (Genencor, Palo Alto, CA) is a formation of α–amylase (E.C. 3.2.1.1) derived from Bacillus licheniformis and phytase (E.C. 3.1.3.26) derived from Trichoderma reesei. The density of the Maxaliq™ ONE enzyme is 1.15 g/ml. The optimum pH for this enzyme formulation is 5.4-6.0. The recommended temperature for the enzyme is 82-88°C. GC147 enzyme (Genencor, Palo Alto, CA) contains gluco-amylase (E.C. 3.2.1.3) and bacterial pullulanase (3.2.1.41). The density of the GC147 enzyme is 1.07 g/ml, with declared activity of 580 GTU/g (1 GTU = enzyme to liberate 1 g of reducing sugars, calculated as glucose/h from soluble starch substrate). The optimum pH and temperature for GC147 were 4.0-4.5 and 58-65°C, respectively. Yeast culture was prepared by dispersing 5 g of active dry yeast (Fleischmann’s, Fenton, MO) and 25 g of deionized water, and agitated at 90 rpm at 30°C for 20 min in an incubator shaker (Model C24, New Brunswick, NJ).

4.2.3 Dry grind laboratory process

A conventional dry grind process was conducted following a modified version reported by Wang et al. (2005) (Figure 8). Triplicate cleaned corn samples were ground finely using a Quaker City plate mill (Model 4-E, The Straub Co., Hatboro, PA). Ground corn moisture content was measured at 135°C for 2 h (AACC International 2000b). Ground corn weight 100 g (dry matter) was mixed with deionized water to obtain mash with a 30% dry solid content. The mash was adjusted to pH 5.7 using 10 N sulfuric acid solution. All experiments were performed in a 500 ml flask for agitation. Mash was incubated with 26 µL α–amylase (Maxaliq™ ONE) at 82°C for 90 min with agitation at 30 rpm. After 90 min, slurry temperature was decreased to 30°C and the pH was adjusted to 4.0 using 10 N sulfuric acid solution. An aliquot of 2 ml yeast culture, 50 µl gluco-amylase (GC147) and 0.5 ml urea were added to the mash prior to a simultaneous saccharification and fermentation (SSF) to convert the starch to ethanol.
Fermentation was conducted at 30°C for 72 h with continuous agitation at 90 rpm. Fermentation was monitored by taking 1 ml slurry sample at 0, 2, 4, 6, 8, 10, 12, 24, 48 and 72 h, and measuring sugar and ethanol concentrations using an HPLC (Model 1515, Waters Corporation, Milford, MA; Aminex HPX-87 column, Bio-Rad Laboratories, Hercules, CA) by running the sample twice to get the average value. HPLC samples were prepared according to the procedure reported by Singh et al. (2005). Actual ethanol yields (ml/kg dry matter of corn) were calculated based on the final ethanol concentrations (72 h) and the amount of initial corn processed (dry matter). Ethanol conversion efficiencies were calculated as a ratio of the actual ethanol yield over the theoretical ethanol yield, which was based on the starch content of the corn. The total starch contents (including sugar contents) in the samples were used to calculate theoretical ethanol yields, assuming 1 g starch converted to 1.11 g of glucose and that 1 g glucose generated 0.511 g ethanol (Thomas et al., 1996; Wu et al., 2006). After 72 h fermentation, the mash was heated to 90°C for 3 h to vaporize the ethanol. Remaining materials were dried in a convective oven at 49°C for 72 h to produce DDGS. Moisture content and the dry matter content of DDGS were determined according to AACC Approved Method (44-19, AACC International, 2000b).

**Figure 8.** Flowchart of corn conventional dry-grind process.
4.2.4 Compositional analysis

Corn samples were sent to a commercial laboratory (Rock River Laboratory Inc., Watertown, WI) for compositional analysis, including crude protein (AOAC, 2003a), crude fat (AOAC, 2003b), neutral detergent fiber (National Forage Testing Association, 2002), and ash (AOAC, 2003c). Starch content was determined using a glucose analyzer (Model 2000, Yellow Spring Instrument, Yellow Springs, Ohio) and using an enzymatic external hydrolysis. Three replications were conducted for each composition analysis. The Duncan’s multiple range test was used for data analysis (SPSS 17.0, Somers, NY). The level selected to show statistical significance was 5% (p < 0.05).

4.3 Results and discussion

4.3.1 Corn composition

Corn harvested at moisture contents of 73% and 54% had lower starch concentrations than corn harvested at lower moisture contents (Table 4), indicating the starch accumulation was not completed. Glucose concentration was highest (6.1%) when corn was harvested at moisture content of 73% and decreased to 2.5% when corn was harvested at moisture content of 54%, because some glucose was converted to starch during corn maturation (Ingle et al., 1965). Total protein concentration decreased from 9.5 to 7.8% when corn moisture dropped from 73% to 54%, due to the faster synthesis of starch than protein (Bressani & Conde, 1961). During the same time period, fat concentration in corn increased from 2.1% to 3.7%. The increase in fat concentration during kernel maturation was also reported by Ingle et al. (1965). After corn reached physiological maturity at moisture content of 40%, no changes in composition were observed.
Table 4. Composition of corn kernel harvested at different moisture contents\(^a\).

<table>
<thead>
<tr>
<th>Corn harvest M.C. (%, wb)</th>
<th>72.5 ± 0.8</th>
<th>54.4 ± 1.0</th>
<th>40.2 ± 0.4</th>
<th>36.8 ± 1.0</th>
<th>30.4 ± 0.4</th>
<th>24.0 ± 1.1</th>
<th>21.2 ± 0.2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Starch (%, db)</td>
<td>57.1 ± 3.2c(^b)</td>
<td>67.9 ± 2.7b</td>
<td>70.0 ± 1.5ab</td>
<td>70.0 ± 1.6ab</td>
<td>72.2 ± 1.1a</td>
<td>70.4 ± 1.6ab</td>
<td>69.6 ± 2.3ab</td>
</tr>
<tr>
<td>Sugar (%, db)</td>
<td>6.1 ± 0.3a</td>
<td>2.5 ± 0.2bc</td>
<td>2.3 ± 0.3b-d</td>
<td>2.8 ± 0.3b</td>
<td>1.4 ± 0.4e</td>
<td>1.8 ± 0.6de</td>
<td>2.0 ± 0.0c-e</td>
</tr>
<tr>
<td>Protein (%, db)</td>
<td>9.5 ± 0.8a</td>
<td>7.8 ± 0.4b</td>
<td>7.9 ± 0.3b</td>
<td>7.8 ± 0.6b</td>
<td>7.8 ± 0.1b</td>
<td>7.6 ± 0.2b</td>
<td>7.7 ± 0.5b</td>
</tr>
<tr>
<td>Fat (%, db)</td>
<td>2.1 ± 0.1b</td>
<td>3.7 ± 0.3a</td>
<td>3.8 ± 0.1a</td>
<td>4.1 ± 0.1a</td>
<td>3.9 ± 0.1a</td>
<td>3.9 ± 0.2a</td>
<td>3.9 ± 0.4a</td>
</tr>
<tr>
<td>NDF (%, db)</td>
<td>9.6 ± 0.9a</td>
<td>8.3 ± 0.5ab</td>
<td>7.0 ± 0.6bc</td>
<td>7.4 ± 1.1bc</td>
<td>7.0 ± 0.1bc</td>
<td>6.6 ± 0.4c</td>
<td>7.4 ± 0.7bc</td>
</tr>
<tr>
<td>Ash (%, db)</td>
<td>2.6 ± 0.3a</td>
<td>1.8 ± 0.1b</td>
<td>1.4 ± 0.0c</td>
<td>1.6 ± 0.2bc</td>
<td>1.5 ± 0.0bc</td>
<td>1.3 ± 0.1c</td>
<td>1.3 ± 0.2c</td>
</tr>
</tbody>
</table>

\(^a\) All compositions (except for moisture) are expressed as % dry solids and are means of three observations.

\(^b\) Values followed by the same letter in the same row are not significantly different (p > 0.05)
4.3.2 Fermentation results
Ethanol profile and yields

For the first 8 h of fermentation, ethanol profiles were comparable (Figure 9 a, b). After 8 h, corn harvested with 73% M.C. had a visible lower ethanol concentration compared to corn harvested with any other moisture contents (Figure 9 a, b), which was likely due to the lower starch concentration (57.1%) and higher fiber concentration (9.6%) in corn with 73% harvest M.C. (Table 4). This is similar to the results previously reported by Jennings et al. (2002a) and Huang et al. (2012b), who found the starch yield in wet-milling process from pre-matured corn was lower than that from matured corn. Although corn harvested with 54% M.C. had a lower starch content (67.9%) compared to matured corn (69.6 to 72.2% starch content), the final ethanol concentrations for the corn with 54% harvest M.C. was 0.5 to 1.2 percentage points higher (Table 5). The result indicated that starch in corn with 54% harvest M.C. had higher fermentability than that in matured corn with lower harvest moistures. This was also verified by the residual starch content in DDGS, where the residual starch in DDGS for pre-maturity corn (harvest M.C. of 73 and 54%) was lower than that for matured corn (Figure 11). Lower residual starch content in DDGS indicated a higher starch to sugar conversion during liquefaction, and thereby leading to a higher ethanol yield. Lately, profit margins in dry grind ethanol production are shrinking due to the high corn price. Increasing ethanol yield for each kilogram of corn, even if only one percent, would have a great impact in a commercial ethanol plant.

The highest ethanol conversion efficiency (98.5%) was achieved when corn was harvested with 73% M.C., followed by corn harvested with 54% M.C., whose conversion efficiency was 93.2%. The ethanol conversion efficiencies for matured corn (harvest M.C. of 40 to 21%) were lower, between 83.2 and 88.3% (Table 5). Higher ethanol conversion efficiency for pre-maturity corn (harvest M.C. of 73 and 54%) could be due to several reasons: first, pre-matured corn had lower amylose/amylopectin ratios in corn starch (Shannon and Garwood, 1984; Li et al., 2007), which lead to lower resistant starch after liquefaction and fermentation (Sharma et al., 2010). Second, with corn maturation starch granules in endosperm are increasingly surrounded by protein bodies and embedded in a dense protein matrix (Khoo and Wolf, 1970; Philippeau and Michalet-Doreau, 1997), which would limit the action of starch hydrolytic enzymes during
liquefaction. Also, as corn kernel matures, starch granules become more compact, losing their round shape and becoming more polygonal (Evans, 1941). Compact starch granules are more difficult to hydrolyze by enzymatic action, compared with loose ones (Wang et al., 2010). Therefore, starch in pre-matured corn is easier to hydrolyze into sugars by enzymes, causing higher ethanol conversion efficiency in the dry grind process. Higher ethanol conversion efficiency has potential benefits such as a lower raw material (corn) cost and a higher ethanol concentration, therefore reducing the size of equipment in downstream processing (Wang et al., 1999).

**Figure 9.** Concentrations of ethanol during fermentation. a) Corn at kernel prematurity and maturity stages, with harvest M.C. of 73, 54 and 40%; b) Corn at kernel post-maturity stage, with harvest M.C. of 37, 30, 24 and 21%.
Glucose profile

For the corn harvested with 73% M.C., initial glucose concentration in the slurry was 2.5% (w/v), which increased to 9.1% at 6 h, then exponentially dropped to a negligible amount at the end of the fermentation (72 h) (Figure 10 a). For the corn harvested thereafter with lower moisture contents, initial glucose concentration was lower (approximately 1%), which increased to the peak concentration at 8 h, then exponentially dropped (Figure 10 a, b). The corn harvested with 73% M.C. had the lowest peak concentration (9.1% w/v) and the corn harvested with 54% had the highest peak concentration (11.6% w/v). Wang et al. (2005) showed that the peak glucose concentration occurs at 4 h, which was earlier compared with this study (6-8 h). This could be due to different corn particle size distributions and different enzymes used for liquefaction and saccharification.

After 72 h simultaneous saccharification and fermentation, final glucose concentrations from corn harvested with all moisture contents (except for 30%) were less than 0.5% (Figure 10, Table 5), suggesting the fermentations were complete. Final glucose concentration from corn harvested with 30% M.C. was 0.7% (Table 5), indicating an insufficient fermentation. The insufficient fermentation was also expressed in final ethanol concentration and ethanol yield. Final ethanol concentration and ethanol yield from corn harvested with 30% M.C. was lower than that from corn harvested after kernel maturity (harvest M.C. of 40, 37, 24 and 21%) (Table 5). The reason for the insufficient fermentation needed to be further investigated.
Figure 10. Concentrations of glucose during fermentation. a) Corn at pre-maturity and maturity stages, with harvest M.C. of 73, 54 and 40%; b) Corn at post-maturity stage with harvest M.C. of 37, 30, 24 and 21%.
Table 5. Final ethanol concentrations, ethanol yields and ethanol conversion efficiencies in dry grind process with corn harvested at different moisture contents\textsuperscript{a}.

<table>
<thead>
<tr>
<th>Corn harvest M.C.</th>
<th>73%</th>
<th>54%</th>
<th>40%</th>
<th>37%</th>
<th>30%</th>
<th>24%</th>
<th>21%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol conc.\textsuperscript{b} (%, v/v)</td>
<td>15.8 ± 0.1\textsuperscript{c}</td>
<td>17.9 ± 0.3\textsuperscript{a}</td>
<td>17.4 ± 0.3\textsuperscript{b}</td>
<td>17.1 ± 0.0\textsuperscript{bc}</td>
<td>16.7 ± 0.2\textsuperscript{c}</td>
<td>17.3 ± 0.2\textsuperscript{b}</td>
<td>17.2 ± 0.2\textsuperscript{b}</td>
</tr>
<tr>
<td>Ethanol yield (mL/kg)</td>
<td>374.6 ±4.0\textsuperscript{bc}</td>
<td>397.0 ± 6.5\textsuperscript{a}</td>
<td>386.4 ±6.9\textsuperscript{ab}</td>
<td>382.2 ±1.2\textsuperscript{bc}</td>
<td>371.1 ±12.3\textsuperscript{c}</td>
<td>381.5 ±3.9\textsuperscript{bc}</td>
<td>379.5 ±4.8bc</td>
</tr>
<tr>
<td>Ethanol conversion efficiency (%, db)</td>
<td>98.5 ± 1.1\textsuperscript{a}</td>
<td>93.2 ± 1.5\textsuperscript{b}</td>
<td>88.3 ± 1.6\textsuperscript{c}</td>
<td>86.8 ± 0.3\textsuperscript{c}</td>
<td>83.2 ± 2.8d</td>
<td>87.2 ± 0.9\textsuperscript{c}</td>
<td>87.5 ± 1.1c</td>
</tr>
<tr>
<td>Glucose\textsuperscript{b} (%, db)</td>
<td>0.1 ± 0.1\textsuperscript{c}</td>
<td>0.0 ± 0.0\textsuperscript{c}</td>
<td>0.0 ± 0.0\textsuperscript{c}</td>
<td>0.4 ± 0.2\textsuperscript{b}</td>
<td>0.7 ± 0.3\textsuperscript{a}</td>
<td>0.0 ± 0.0\textsuperscript{c}</td>
<td>0.2 ± 0.2\textsuperscript{c}</td>
</tr>
</tbody>
</table>

\textsuperscript{a} All data are means of three observations.

\textsuperscript{b} Ethanol and glucose concentration at the 72 h of fermentation.

\textsuperscript{c} Values followed by the same letter in the same row are not significantly different (p > 0.05).
4.3.3 DDGS composition

The effects of corn harvest moisture on DDGS composition were observed (Figure 11). Starch concentration of DDGS material increased as corn harvest moisture decreased. For corn harvested with 73% M.C., starch concentration in DDGS was 7.7% compared to corn harvested with 21%, which produced DDGS with 15.4% starch concentration. Higher residual starch concentration in DDGS indicated a lower sugar conversion efficiency during liquefaction, and thereby a lower ethanol conversion efficiency. The hardness of corn kernel increased with corn maturation and drying (data not shown), which increased the corn particle size during grinding (Wu, 1992). Increased corn particle size could lead to fewer fermentable sugars from starch hydrolysis and higher residual starch concentrations in DDGS samples (Naidu et al., 2007). With corn harvest M.C. decreased from 73 to 21%, CP concentrations in DDGS decreased from 29.4 to 24.9% and NDF concentration decreased from 26.6 to 20.6%. The likely reason for the decrease in CP and NDF concentration was the increased portion of residual starch in DDGS, making CP and NDF concentration proportionally smaller.

![Figure 11. Composition (% dry basis) of DDGS after dry grind process with corn harvested at different moistures.](image-url)
4.3.4 Ethanol yield per land use

Although corn with harvest moisture contents of 73 and 54% had higher ethanol conversion efficiencies, the dry matter yields of high moisture corn were lower, compared to corn harvested after physiological maturity. An appropriate way to evaluate the best corn harvest moisture is to compare the ethanol yield per land use at different corn harvest moisture contents. Ethanol yield per land use could be calculated by multiplying ethanol yield per kg of corn by dry matter yield of corn per hectare, which was shown in Figure 6. Corn harvest at moisture contents of 73 and 54% had low ethanol yield per land use due to the low dry matter yield of corn before physiological maturity. After corn reached physiological maturity at moisture content of 40%, the ethanol yield per land use reached highest level and then remained stable, ranging between 5000 and 6000 L/ha.

![Ethanol yield per land use at different corn harvest moisture contents.](image)

**Figure 12.** Ethanol yield per land use at different corn harvest moisture contents.

4.4 Conclusions

Effects of corn harvest moisture content on dry grind ethanol processes were observed on fermentation characteristics and DDGS composition. The final ethanol concentration from corn with harvest moisture content of 54% (110 days after corn planting, kernel dent stage) was 0.5 to 1.2 percentage points higher than that from matured corn with lower harvest moisture contents. Ethanol conversion efficiencies for
the corn with harvest moisture contents of 73 and 54% were higher than that for the corn with lower harvest moisture contents, however, ethanol yield per land use were lower due to the low dry mater yield of corn before physiological maturity. With the corn harvest moisture drying down from 73 to 21%, the residual starch concentration in DDGS increased while the CP and NDF concentrations in DDGS decreased.
Chapter 5. Effect of corn harvest moisture on wet milling yields and starch pasting properties

5.1 Introduction

Early harvest corn and corn stover has benefits in terms of high quality of corn, high dry matter yield of corn stover and long harvest time window. However, early harvested corn poses a problem of storability due to its high moisture content. High moisture corn spoils readily, and it is a safety hazard when moldy. Most corn drying systems today are designed to dry corn from the mid-twenty percent moisture range to 15%. Increasing average initial moisture content from 20-25% to 35-40 means more than doubles the amount of moisture needed to be removed, which leads to longer drying time and higher energy cost. To solve this problem, Eckhoff (2010) suggested freezing instead of drying corn for storage. The latent heat of fusion for water is 334 kJ/kg, and the latent heat of vaporization for water is 2260 kJ/kg. Therefore, there is an opportunity to save energy by freezing, if corn could be flash frozen and insulated well in a large building. Simulation results (data not shown) indicated that 10 to 15 centimeters of polyurethane foam wall would be good for frozen corn insulation. The frozen corn can be delivered to the wet milling plant or ethanol plant for further processing. The benefits of using freezing are skewed to greater production for the farmer but also have some advantages for the ethanol processor. Where water is scare, freezing 40% moisture content corn provides input water equal to 2.9 gallons of water per bushel, compared to dried corn (14.5% M.C.). This would cut process water usage in half or better. The frozen corn can also be used as a heat sink for the fermenter, reducing the cooling water load. In wet milling, 8 to 10 gallons of water are typically used in processing a bushel of corn. The addition of 40% moisture corn would reduce the amount of water needed in steeping to 5.1 to 7.1 gallons per bushel.

Jennings et al. (2002a, 2002b) harvested corn at different maturity stages with different moisture contents between 25% and 56%, and evaluated the effects of maturity on corn quality and wet milling properties of two selected corn hybrids. They found that maturity affected the starch yield and some of the starch pasting properties. The effect of frozen storage of corn on starch yields and quality from wet milling process is unknown.
The objective of this study was to evaluate the effect of harvest moisture content and frozen storage time on corn wet-milling yields and the pasting properties of the resulting starch.

5.2 Materials and methods

5.2.1 Experimental materials and chemical composition analyses

Triplicate samples of corn from the southwest, southeast and north regions of testing field in the ABE farm were collected on 05 August, 19 August, and 03 September 2010, which corresponded to 1233, 1432, and 1626 AGDDs after planting, representing corn pre-mature, mature and drying down stages. The moisture contents collected at these three stages were 49, 35 and 21%, respectively. Determining the chemical composition, moisture content, and starch content of the samples was carried out in the same way for samples used for the dry grind ethanol experiments.

5.2.2 Wet milling laboratory process

The corn samples were evaluated in triplicate using the 100 g wet milling procedure described by Eckhoff et al. (1996) (Figure 13). Samples for milling were steeped at 52°C for 24 h in 180 ml steep water, containing 2,000 ppm sulfur dioxide and 0.5% (w/w) lactic acid. At the completion of steeping, steepwater was drained into a 250 ml graduated cylinder, and unabsorbed steepwater volume was measured and dried to determine total steepwater solids (SWS) using a two-stage drying procedure (AACC International, 2000b). The steeped corn was milled in an equal volume of water (v/v) using a Waring type blender (Dynamic Corp. of America, New Hartford City, CT). The ground slurry was transferred along with 500 ml water to a tarred standard testing sieve (U.S. NO.7, 2.80 mm openings) placed at the bottom of a 10 L bucket. The bucket and sieve arrangement was shaken (Model RX-86, W.S. Tyler Co., Cleveland, OH) for 5 min. The material retained on the sieve contained whole and large broken germ pieces and large pieces of pericarp. The slurry that passed through the sieve was then finely ground in a Quaker City plate mill (Model No. 4-E, The Straub Co., Warminster, PA). After second grind, the slurry was allowed to settle for 45 min and approximately 750 ml water was decanted. The decanted slurry was sieved with a standard testing sieve (U.S. NO. 200, 75 µm openings) on the Ro-Tap testing sieve shaker for 5 min. The material
retained on the sieve after washing was collected as cellular fiber. The specific gravity of the slurry remaining after cellular fiber removal was adjusted to 1.04-1.045 by allowing the slurry to settle for 1 h. The decanted slurry was pumped onto the starch stable at a flow rate of 50 ml/min. The overflow, containing primarily small or light granule starch and protein, was collected as gluten. After tabling was completed, the starch was allowed to air dry overnight on the table. The dry matter yield for each fraction was determined using the two-stage drying procedure (AACC International, 2000b). Total fiber content was calculated as the sum of the coarse fiber and cellular fiber. Corn samples were wet milled immediately after collection and referred to as the control.

Since Ferrero et al. (1993) showed that rheological properties of corn starch were different between a short term and a long term frozen storage, the rest of corn was placed in a -10°C freezer for three days, to represent short term frozen storage, and five months, to represent long term frozen storage. The stability of the freezer temperature was ± 0.5°C. Relative humidity (RH %) of the storage freezer was measured using a relative humidity sensor (MicroDAQ.com, Ltd., Contoocook, NH) and was found to be 75 ± 5%. Samples were thawed at 4°C for 24 h prior to the wet milling processes. A full factorial experiment was performed in triplicate with each treatment to study the effect of corn harvest moisture and frozen storage time on wet milling yields and pasting properties of the resulting starch.
5.2.3 Starch pasting properties

Pasting properties of the resulting starch were determined in triplicate with a rapid visco analyzer (Model RVA 4, Newport Scientific, Warriewood, New South Wales, Australia) following the Standard 2 temperature profile. Samples were prepared by mixing 3.0 g starch (12% M.C.) in 25.0 ml deionized water. In Standard 2 RVA procedure, samples were stirred at 960 rpm for 60 s, followed by stirring at 160 rpm for 18 min. Once the sample reached equilibrium at 50°C, it was heated at a rate of 6°C/min to 95°C, maintained at 95°C for 5 min, cooled at 6 °C/min to 50°C, and held at 50°C for 5 min. Peak, trough, final, breakdown and setback viscosities were recorded, as well as pasting temperature and peak time. A two way analysis of variance (ANOVA) and Duncan’s multiple range test was used for data analysis (SPSS 17.0, Somers, NY). The level selected to show statistical significance was 5% (p < 0.05).
5.3 Results and discussion

5.3.1 Corn composition

Starch contents in corn at harvest M.C. of 35 and 21% were higher (> 2 percentage points) than that in corn at 49% harvest M.C., indicating the starch accumulation was not complete when corn moisture was 49% (Table 6). NDF content in corn decreased with corn moisture decreasing. No differences were observed for CP, fat and ash contents in corn at any harvest moisture.

Table 6. Corn compositions at three harvest moisture contentsa.

<table>
<thead>
<tr>
<th>Harvest M.C. (%)</th>
<th>Starchb</th>
<th>CP</th>
<th>NDF</th>
<th>Fat</th>
<th>Ash</th>
</tr>
</thead>
<tbody>
<tr>
<td>49.2 ± 0.4</td>
<td>72.6 ± 2.7bc</td>
<td>9.0 ± 0.3a</td>
<td>14.2 ± 1.1a</td>
<td>3.4 ± 0.2a</td>
<td>1.6 ± 0.1a</td>
</tr>
<tr>
<td>34.6 ± 0.9</td>
<td>75.7 ± 1.0a</td>
<td>8.9 ± 0.2a</td>
<td>12.2 ± 1.0b</td>
<td>3.5 ± 0.4a</td>
<td>1.5 ± 0.3a</td>
</tr>
<tr>
<td>21.0 ± 1.0</td>
<td>74.6 ± 1.8a</td>
<td>9.4 ± 1.0a</td>
<td>10.6 ± 0.9c</td>
<td>3.4 ± 0.0a</td>
<td>1.6 ± 0.2a</td>
</tr>
</tbody>
</table>

a. All compositions are expressed as % dry solids and are means of three observations.
b. The starch content was sugar included.
c. Values followed by the same letter in the same column are not significantly different (p > 0.05).

5.3.2 Yield of wet milled products

The effects of interaction of harvest moisture × frozen storage of all wet milled products were not significant (Table 7), indicating that differences for frozen storage of corn varied in a similar manner across harvest moistures. The harvest moisture affected all the yields of wet-milled products, except for the cellular fiber. Frozen storage affected the yields of germ, cellular fiber and coarse fiber.

Table 7. Significance of harvest moistures and frozen storage times on yield of wet-milled productsa.

<table>
<thead>
<tr>
<th>Factors</th>
<th>SWS</th>
<th>Germ</th>
<th>CF1</th>
<th>CF2</th>
<th>TF</th>
<th>Starch</th>
<th>Gluten</th>
</tr>
</thead>
<tbody>
<tr>
<td>Harvest M.C. × Frozen Storage</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
</tr>
<tr>
<td>Harvest M.C.</td>
<td>*b</td>
<td>*</td>
<td>ns</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>*</td>
</tr>
<tr>
<td>Frozen Storage</td>
<td>ns</td>
<td>*</td>
<td>ns</td>
<td>*</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
</tr>
</tbody>
</table>

a. Steep water solids (SWS), cellular fiber (CF1), coarse fiber (CF2), total fiber (TF).
b. *, significant at p < 0.05; ns, not significant.
The starch yield from corn at 21% harvest M.C. was 1.2 percentage point higher than that from corn at 49% harvest M.C. (65.3 ± 0.4 vs. 64.1 ± 0.5) (Table 8), which was likely due to the lower starch content in corn at 49% harvest M.C.. This is similar to the result reported by Jennings et al. (2002a), who found the starch yield increased by 2.9 percentage points during corn maturity. Based on the corn composition analysis (Table 6), 85-88% of the total starch was extracted by wet-milling processes, depending on the corn harvest moisture. The unrecoverable starch was lost to germ, fiber and gluten fractions (Eckhoff et al., 1999). Statistical analysis based on ANOVA test showed no significant effects of frozen storage on starch yield in wet-milling process (Table 8).

During steeping, soluble protein and sugars leach out, primarily from the germ. The yield of steep water solids decreased with corn harvest moisture decreasing (Table 8). The yield of steep water solids from corn at 49% harvest M.C. was 2.1 percentage points higher than that from corn at 21% harvest M.C. (5.8 ± 0.1 vs. 3.7 ± 0.2). The decreased yield of steep water solids could be attributed to the transformations of soluble protein and sugars into storage protein and starch during corn maturity (Ingle et al., 1965). No significant effect of frozen storage on yield of steep water solids was observed in this study (Table 7).

The germ yield increased by 1.9 percentage points when corn harvest moisture decreased from 49% to 21% (5.3 ± 0.1 vs 7.2 ± 0.2) (Table 8). This result would be expected because the corn embryo is much slower to develop than the endosperm (Kiesseelbach and Walker, 1952; Schel et al., 1984; Watson, 2003), and the embryo was not fully developed at the pre-maturity stage, when the corn moisture was 49%. Lower yield of germ from pre-mature corn was also reported by Jennings et al. (2002a), who found the yield of germ from pre-mature corn was 0.8 percentage points lower, compared with matured corn.

For corn harvested at 49% M.C., the coarse fiber yields from 3-day and 5-month frozen storage were 0.8 and 0.7 percentage points lower, respectively, than the coarse fiber yield from control (unfrozen corn) (Table 8). We hypothesized that the frozen storage made the pericarp fragile, and the fragile pericarp was easily broken during the first grind with a blender. All the broken pericarp flowed into the cellular fiber fraction, causing a higher yield of cellular fiber from the frozen storage corn. The total fiber
content decreased by 0.7 percentage points as corn harvest moisture decreased. This is similar to the results previously reported by Jennings et al. (2002a), who proposed the main reason was that the surface area of fiber becomes proportionately smaller as the other components of the maturing corn are deposited.
Table 8. Yield (% d.b.) of wet-milled products from corn at different harvest moisture content and frozen storage times.

<table>
<thead>
<tr>
<th></th>
<th>Unfrozen (Control)</th>
<th>3-day frozen storage</th>
<th>5-month frozen storage</th>
<th>Mean (across storage)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Starch (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>64.5 ± 0.3a</td>
<td>64.3 ± 0.6</td>
<td>63.6 ± 0.5</td>
<td>64.1 ± 0.5c</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>64.7 ± 0.4</td>
<td>64.3 ± 0.9</td>
<td>65.0 ± 0.3</td>
<td>64.6 ± 0.6b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>65.6 ± 0.4</td>
<td>65.4 ± 0.3</td>
<td>65.0 ± 0.5</td>
<td>65.3 ± 0.4a</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>64.9 ± 0.6a</td>
<td>64.6 ± 0.8a</td>
<td>64.5 ± 0.7a</td>
<td></td>
</tr>
<tr>
<td><strong>S.W. Solids (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>5.8 ± 0.1</td>
<td>6.0 ± 0.1</td>
<td>5.9 ± 0.1</td>
<td>5.9 ± 0.1a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>4.5 ± 0.1</td>
<td>4.7 ± 0.1</td>
<td>4.7 ± 0.0</td>
<td>4.6 ± 0.1b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>3.6 ± 0.2</td>
<td>3.7 ± 0.1</td>
<td>3.7 ± 0.1</td>
<td>3.7 ± 0.2c</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>4.6 ± 0.9a</td>
<td>4.7 ± 1.0a</td>
<td>4.8 ± 1.0a</td>
<td></td>
</tr>
<tr>
<td><strong>Germ (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>5.5 ± 0.1</td>
<td>5.3 ± 0.1</td>
<td>5.3 ± 0.1</td>
<td>5.3 ± 0.1b</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>7.3 ± 0.1</td>
<td>7.1 ± 0.1</td>
<td>7.0 ± 0.1</td>
<td>7.2 ± 0.2a</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>7.3 ± 0.2</td>
<td>7.1 ± 0.3</td>
<td>7.3 ± 0.2</td>
<td>7.2 ± 0.2a</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>6.7 ± 0.9a</td>
<td>6.5 ± 0.9b</td>
<td>6.6 ± 0.9b</td>
<td></td>
</tr>
<tr>
<td><strong>Cellular fiber (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>7.1 ± 0.2</td>
<td>7.8 ± 0.4</td>
<td>7.5 ± 0.2</td>
<td>7.5 ± 0.4a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>7.3 ± 0.2</td>
<td>7.8 ± 0.3</td>
<td>7.5 ± 0.3</td>
<td>7.5 ± 0.3a</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>7.8 ± 0.3</td>
<td>7.9 ± 0.7</td>
<td>7.7 ± 0.4</td>
<td>7.8 ± 0.4a</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>7.4 ± 0.3b</td>
<td>7.8 ± 0.4a</td>
<td>7.6 ± 0.2ab</td>
<td></td>
</tr>
<tr>
<td><strong>Coarse fiber (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>6.4 ± 0.2</td>
<td>5.6 ± 0.3</td>
<td>5.7 ± 0.2</td>
<td>5.8 ± 0.5a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>5.3 ± 0.2</td>
<td>5.0 ± 0.5</td>
<td>4.8 ± 0.6</td>
<td>5.0 ± 0.3b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>4.8 ± 0.3</td>
<td>4.8 ± 0.4</td>
<td>4.9 ± 0.4</td>
<td>4.8 ± 0.2b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>5.4 ± 0.8a</td>
<td>5.0 ± 0.4b</td>
<td>5.1 ± 0.5b</td>
<td></td>
</tr>
<tr>
<td><strong>Total fiber (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>13.5 ± 0.2</td>
<td>13.4 ± 0.2</td>
<td>13.2 ± 0.4</td>
<td>13.3 ± 0.5a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>12.5 ± 0.4</td>
<td>12.8 ± 0.4</td>
<td>12.3 ± 0.4</td>
<td>12.6 ± 0.4b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>12.5 ± 0.4</td>
<td>12.7 ± 0.6</td>
<td>12.6 ± 0.3</td>
<td>12.6 ± 0.4b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>12.9 ± 0.6a</td>
<td>12.9 ± 0.6a</td>
<td>12.7 ± 0.5a</td>
<td></td>
</tr>
<tr>
<td><strong>Gluten (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>11.2 ± 0.5</td>
<td>11.6 ± 0.3</td>
<td>10.9 ± 0.7</td>
<td>11.3 ± 0.5a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>10.4 ± 0.5</td>
<td>10.5 ± 0.3</td>
<td>10.2 ± 0.4</td>
<td>10.4 ± 0.3b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>10.9 ± 0.5</td>
<td>10.8 ± 0.5</td>
<td>10.4 ± 0.4</td>
<td>10.7 ± 0.5b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>10.8 ± 0.5a</td>
<td>11.0 ± 0.6ab</td>
<td>10.5 ± 0.6b</td>
<td></td>
</tr>
</tbody>
</table>

a. Values in the table are mean ± SE from three replicates.

b. Values followed by the same letter in the same column are not significantly different (p > 0.05).

c. Values followed by the same letter in the same row are not significantly different (p > 0.05).
5.3.3 Starch pasting properties

The harvest moisture, frozen storage and the interaction of harvest moisture × frozen storage significantly affected peak viscosities (Table 9). The peak viscosities of starch from corn at 49% harvest M.C. were higher than that from corn at 21% harvest M.C. (3824 ± 36 cp vs. 3520 ± 38 cp) (Table 10). This could be explained by the different amylose/amylopectin ratios in corn starch. Pre-mature corn with high moisture have higher amylopectin and lower amylose in the starch granule compared to matured corn with low moisture (Shannon and Garwood 1984). Since starch granular swelling is a property of amylopectin, and amylose actively inhibits swelling during gelatinization (Burt and Russell 1983; Tester and Morrison 1990, 1992), the pre-mature corn starch with higher amylopectin resulted in higher viscosities in the starch pastes.

Table 9. Significance of harvest moistures and frozen storage times on starch pasting properties

<table>
<thead>
<tr>
<th>Factors</th>
<th>PV</th>
<th>TV</th>
<th>BV</th>
<th>FV</th>
<th>SV</th>
<th>P_time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Harvest M.C.</td>
<td>*b</td>
<td>ns</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>*</td>
</tr>
<tr>
<td>Frozen Storage</td>
<td>*</td>
<td>ns</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>ns</td>
</tr>
<tr>
<td>Harvest M.C.× Frozen Storage</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>ns</td>
<td>*</td>
<td>ns</td>
</tr>
</tbody>
</table>

a Peak viscosity (PV), trough viscosity (TV), breakdown viscosity (BV), final viscosity (FV), setback viscosity (SV), and peak time (P_time).

b *, significant at p < 0.05; ns, not significant.

The breakdown viscosity decreased by 13% as corn harvest moisture decreased from 49% to 21% (2336 ± 47 cp vs. 2029 ± 60 cp) (Table 10). This result would be expected because pre-mature corn starch with higher amylopectin has an initially higher peak viscosity, and followed by more rapidly viscosity breakdown during continued heating (Thomas and Atwell 1997).

No significant difference in setback viscosities was observed between the starch from 3-day frozen corn and control (unfrozen corn). However, setback viscosities of starch from 5-month frozen corn were lower than that of control (1574 ± 65 cp vs. 1828 ± 79 cp) (Table 10). It indicates that long-term frozen storage reduces the ability of starch to reassociate to form gels. Leaching of amylose is a requirement and main factor for reassociations of the dispersed starch molecules (Biliaderis 2009). Therefore, it is quite
possible that long-term frozen storage of corn inhibits the amylose leaching from starch granule during gelatinization and breaking down process. Jeong and Lim (2003) found that the freeze-thawed starch powders had higher setback viscosities. Jeong and Lim (2003) froze corn starch after wet-milling, while in this study, the corn kernel was frozen before wet-milling and therefore, their results cannot directly be compared with this study. Setback viscosities were also affected by interaction of harvest moisture × frozen storage (Table 9), indicating that frozen storage affects setback viscosities differently, depending on corn harvest moisture. For corn at 21% harvest M.C., the setback viscosity for 5-month frozen storage was 8% lower than that for the control, while the setback viscosities for 5-month frozen storage were 19% and 22% lower for corn at 35% and 49% harvest M.C., respectively. Long term (5-month) frozen storage also decreased the final viscosity of the resulting starch by 8%, compared to control (3063 ± 27 cp vs. 3317 ± 101 cp) (Table 10).

There was no effect of frozen storage on peak time during corn starch heating, while harvest moisture affected the peak time (Table 9). The peak time increased as corn harvest moisture decreased (Table 10). As the corn kernel matures and drying, the starch granules become more compact (Jennings et al., 2002b), thereby requiring more heat and time to gelatinize, and delaying the peak viscosity occurring time. The effects of harvest moisture and frozen storage were not significant for trough viscosity (Table 9), suggesting that it was the pasting property that was least affected.
Table 10. Pasting properties of starch from corn at different harvest moistures and frozen storage times

<table>
<thead>
<tr>
<th></th>
<th>Unfrozen (Control)</th>
<th>3-day frozen storage</th>
<th>5-month frozen storage</th>
<th>Mean (across storage)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Peak viscosity (cp)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>3802 ± 58a</td>
<td>3843 ± 18</td>
<td>3829 ± 15</td>
<td>3824 ± 36b</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>3452 ± 39</td>
<td>3524 ± 34</td>
<td>3590 ± 14</td>
<td>3517 ± 71b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>3511 ± 46</td>
<td>3495 ± 19</td>
<td>3556 ± 14</td>
<td>3520 ± 38b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>3588 ± 172b</td>
<td>3620 ± 169b</td>
<td>3658 ± 129a</td>
<td></td>
</tr>
<tr>
<td><strong>Trough (cp)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>1504 ± 63</td>
<td>1509 ± 52</td>
<td>1453 ± 11</td>
<td>1488 ± 49a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>1438 ± 8</td>
<td>1456 ± 49</td>
<td>1561 ± 35</td>
<td>1484 ± 65a</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>1523 ± 30</td>
<td>1496 ± 30</td>
<td>1455 ± 14</td>
<td>1491 ± 37a</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>1488 ± 52a</td>
<td>1487 ± 46a</td>
<td>1489 ± 57a</td>
<td></td>
</tr>
<tr>
<td><strong>Breakdown (cp)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>2298 ± 48</td>
<td>2334 ± 38</td>
<td>2376 ± 25</td>
<td>2336 ± 47a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>2001 ± 31</td>
<td>2068 ± 32</td>
<td>2029 ± 31</td>
<td>2032 ± 40b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>1988 ± 20</td>
<td>1999 ± 47</td>
<td>2102 ± 13</td>
<td>2029 ± 60b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>2095 ± 155c</td>
<td>2133 ± 157b</td>
<td>2168 ± 160a</td>
<td></td>
</tr>
<tr>
<td><strong>Final Viscosity (cp)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>3404 ± 96</td>
<td>3400 ± 62</td>
<td>3047 ± 28</td>
<td>3284 ± 187a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>3265 ± 39</td>
<td>3264 ± 43</td>
<td>3058 ± 11</td>
<td>3196 ± 107b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>3281 ± 111</td>
<td>3289 ± 66</td>
<td>3085 ± 28</td>
<td>3218 ± 120b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>3317 ± 101a</td>
<td>3318 ± 80a</td>
<td>3063 ± 27b</td>
<td></td>
</tr>
<tr>
<td><strong>Setback (cp)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>1901 ± 43</td>
<td>1890 ± 25</td>
<td>1595 ± 22</td>
<td>1795 ± 152a</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>1827 ± 31</td>
<td>1808 ± 75</td>
<td>1497 ± 44</td>
<td>1710 ± 167b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>1758 ± 83</td>
<td>1793 ± 91</td>
<td>1630 ± 16</td>
<td>1727 ± 97b</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>1828 ± 79a</td>
<td>1830 ± 76a</td>
<td>1574 ± 65b</td>
<td></td>
</tr>
<tr>
<td><strong>Peak Time (min)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>49% harvest M.C.</td>
<td>6.32 ± 0.00</td>
<td>6.27 ± 0.07</td>
<td>6.44 ± 0.03</td>
<td>6.32 ± 0.06c</td>
</tr>
<tr>
<td>35% harvest M.C.</td>
<td>6.47 ± 0.09</td>
<td>6.37 ± 0.02</td>
<td>6.46 ± 0.12</td>
<td>6.43 ± 0.09b</td>
</tr>
<tr>
<td>21% harvest M.C.</td>
<td>6.60 ± 0.07</td>
<td>6.62 ± 0.06</td>
<td>6.66 ± 0.10</td>
<td>6.62 ± 0.07a</td>
</tr>
<tr>
<td>Mean (across harvest M.C.)</td>
<td>6.46 ± 0.13ab</td>
<td>6.42 ± 0.16b</td>
<td>6.50 ± 0.15a</td>
<td></td>
</tr>
</tbody>
</table>

a. Values in the table are mean ± SE from three replicates.

b. Values followed by the same letter in the same column are not significantly different (p > 0.05).

c. Values followed by the same letter in the same row are not significantly different (p > 0.05).
5.4 Conclusions

As corn harvest moisture decreased from 49 to 21%, the yields of starch and germ increased by 1.2 and 1.9 percentage points respectively, while the yields of steep water solids, total fiber and gluten decreased at different levels. The frozen corn had lower coarse fiber yield but higher cellular fiber yields compared with control (unfrozen corn). For pasting properties of starch, the peak viscosity and breakdown viscosity decreased, while peak time increased as corn harvest moisture decreased. The setback and final viscosities of starch from long term frozen corn were lower than that from control (unfrozen corn).
Chapter 6. Effect of harvest moisture on composition and nutritive value of corn stover and DDGS

6.1 Introduction

The increasing use of corn for ethanol would intensify the competition for feedstock between ethanol plants and livestock feed industry. Because ruminants consume 35.6% of the 135 million tonnes of corn fed to livestock in the United States each year (Sewell et al., 2009), efficient utilization of the non-grain portion of the corn plant as ruminant food will allow the optimization of both food and fuel production. Sewell et al. (2009) found that when the thermochemically treated corn stover was with DDGS, the feed value enhanced stover (FVES) could be as nutritious as corn in ruminant diets. Traditionally corn and corn stover were harvested after the corn is dry in field. However, if the corn and corn stover were harvested earlier before dry down, the corn stover would have less acid detergent fiber (ADF) and lignin and higher in vitro dry matter digestibility (IVDMD) (Russell, 1986).

Previous studies have been reported the relationships between plant maturity and its nutritive values as animal food (Leask and Daynard, 1973; Russell, 1986; Irlbeck et al., 1993; Johnson et al., 1999; Lewis et al., 2004). This study linked the corn residues from the field with DDGS from the corn-ethanol production, and systematically evaluated the yield and nutritive values of the products. The objectives of this study were to determine the yields, compositions and nutritive values of corn stover, DDGS and FVES at different corn harvest moisture contents and to determine the harvest time to maximize the corn plant utilization.

6.2 Materials and methods

6.2.1 Experimental materials

The corn hybrid 32D78 (Pioneer Hi-Bred International, Johnson, IA) was planted on 22 May 2009. The corn and corn stover were harvested on 21 August, 09 September, 30 September, 13 October, 25 October, 09 November, and 23 November, which corresponded to 1049, 1217, 1402, 1432, 1467, 1514, and 1534 AGDDs after planting,
respectively. DDGS samples were collected from conventional dry grind process, which is described in section 4.2.

6.2.2 Compositional and nutritive analyses

Corn and corn stover samples were sent to a commercial laboratory (Rock River Laboratory Inc., Watertown, WI) for compositional analysis including acid detergent fiber (Method 973.18, AOAC, 2003), lignin (Method 973.18C, AOAC, 2003), crude protein (Method 990.03, AOAC, 2003), minerals (Method 985.01, AOAC, 2003) and neutral detergent fiber (National Forage Testing Association, 2002). Starch content was determined according to Yellow Spring Instrument glucose analyzer procedure (YSI 2000) and using an enzymatic external hydrolysis. The feed value enhanced stover (FVES) is a mixture of corn stover and DDGS, which could be as nutritious as corn in ruminant diets (Sewell et al., 2009). The dry matter yield of FVES was the sum of the dry matter yield of corn stover and DDGS. Chemical compositions of FVES were calculated based on the proportional composition in the stover and leaf, cob and DDGS samples. The energy and nutritive values were determined using the following equations (Undersander et al., 1993)

\[
TDN(\%DM) = 31.4 + 53.1 \times NEL \\
NEL \text{ (Mcal/Lb)} = 1.044 - 0.0124 \times ADF \\
ME = 0.01642 \times TDN
\]

\[
NEM \text{ (Mcal/Lb)} = -0.508 + (1.37 \times ME) - (0.3042 \times ME^2) + (0.051 \times ME^3) \\
NEG(\text{Mcal/Lb}) = -0.7484 + (1.42 \times ME) - (0.3836 \times ME^2) + (0.0593 \times ME^3)
\]

where TDN is the total digestible nutrients, NEL is the net energy of lactation, ME is the metabolizable energy, NEM is the net energy of maintenance, NEG is the net energy of gain. Dry matter yield of TDN per hectare (t/ha) was determined by the product of total digestible nutrients (% DM) and dry matter yield of biomass in each hectare using the following equation

\[
\text{TDN per hectare (t/ha)} = \text{TDN(\%DM)} \times \text{dry matter yield per hectare}
\]
Ducan’s multiple range test (SPSS 17.0, Somers, NY) were used to determine significant differences, if any, in chemical and nutritive values. The level selected to show statistical significance was 5% (p < 0.05).

6.3 Results and discussion

6.3.1 Chemical composition of corn stover

NDF concentrations in both corn stover and cob increased with corn harvest moisture decreasing (Table 11). The increase in NDF concentrations in corn stover was also reported by Weaver et al. (1978), who found that NDF concentrations in all plant parts increased with maturity except for corn which remained highly stable after maturity. During the same time period, ADF concentrations in corn stover and cob increased by 12.7 percentage points and 9.6 percentage points, respectively. The lignin concentrations in corn stover and cob increased by 1 percentage point and 2.2 percentage points when corn moisture decreased from 73 to 21%. The increase in lignin during and after corn maturity was also reported by Cone and Engels (1993) and Pordesimo et al. (2005). The increases in NDF, ADF and lignin concentrations indicated a decreasing digestibility of corn stover as animal food.

Crude protein (CP) concentrations in corn stover decreased from 7.4 to 3.3% as corn moisture decreased from 73 to 21%, and CP concentration in cob decreased from 3.7 to 1.6% (Table 11). The decrease in CP concentration was in consistent with previous studies (Lewis, et al., 2004, Darby and Lauer, 2002) and it appeared to result from continued carbon assimilation during corn maturing, even though nitrogen uptake probably was completed, thereby diluting plant nitrogen concentration (Wiersma et al., 1993). Average stover CP concentration in our study was lower than those reported by Lewis et al. (2004) and Darby and Lauer (2002), which could be attributed to different corn varieties and different amounts of nitrogen in the field. The composition of DDGS was shown in Figure 11, section 4.3.3.
Table 11. Chemical composition of corn stover from seven corn harvest moisture contents\(^a\).

<table>
<thead>
<tr>
<th></th>
<th>73%</th>
<th>54%</th>
<th>40%</th>
<th>37%</th>
<th>30%</th>
<th>24%</th>
<th>21%</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Stalk and leaf</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NDF, % DM</td>
<td>63.5d(^2)</td>
<td>67.9cd</td>
<td>69.2c</td>
<td>75.0b</td>
<td>78.1ab</td>
<td>78.3ab</td>
<td>80.3a</td>
</tr>
<tr>
<td>ADF, % DM</td>
<td>40.3bc</td>
<td>42.0c</td>
<td>43.5bc</td>
<td>48.7bc</td>
<td>52.2ab</td>
<td>54.0a</td>
<td>53.0a</td>
</tr>
<tr>
<td>Lignin, % DM</td>
<td>4.8c</td>
<td>4.3ab</td>
<td>4.7ab</td>
<td>5.0ab</td>
<td>6.1ab</td>
<td>6.4a</td>
<td>5.8ab</td>
</tr>
<tr>
<td>CP, % DM</td>
<td>7.4a</td>
<td>7.1a</td>
<td>4.7b</td>
<td>4.1bc</td>
<td>4.6b</td>
<td>4.0bc</td>
<td>3.3c</td>
</tr>
<tr>
<td><strong>Cob</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NDF, % DM</td>
<td>73.2c</td>
<td>72.7c</td>
<td>79.7b</td>
<td>83.9a</td>
<td>86.4a</td>
<td>87.5a</td>
<td>87.5a</td>
</tr>
<tr>
<td>ADF, % DM</td>
<td>36.8d</td>
<td>36.2d</td>
<td>41.8c</td>
<td>42.0c</td>
<td>43.4bc</td>
<td>44.2b</td>
<td>46.4a</td>
</tr>
<tr>
<td>Lignin, % DM</td>
<td>1.8d</td>
<td>2.3b-d</td>
<td>2.0cd</td>
<td>3.0a-d</td>
<td>3.6a-c</td>
<td>4.2a</td>
<td>4.0ab</td>
</tr>
<tr>
<td>CP, % DM</td>
<td>3.7a</td>
<td>2.4b</td>
<td>1.9bc</td>
<td>1.9bc</td>
<td>1.8c</td>
<td>1.9bc</td>
<td>1.6c</td>
</tr>
</tbody>
</table>

a. All data are means of three observations.

b. Values followed by the same letter in the same row are not significantly different (p > 0.05).

Mineral elements are essential for animals and for which signs of deficiency have been described by Underwood (1999). With corn maturity, all mineral element concentrations in corn stover decreased consistently, for example, potassium (K) concentration decreased by 0.55 percentage points from the beginning to the end of the study, followed by phosphorus (P) and magnesium (Mg), decreased by 0.12 and 0.1 percentage points, then by calcium (Ca) and sulfur (S), decreased by 0.08 and 0.04 percentage points, respectively (Table 12). The decrease in mineral elements in biomass was mainly due to nutrient leaching by rain and snow (Landström et al., 1996; Adler et al., 2006). Decrease in mineral elements with corn maturity showed a reduced nutritive value as animal food.
Table 12. Mineral compositions in the stalk and leaf fraction, cob and DDGS at seven corn harvest moisture contents\(^1\).

<table>
<thead>
<tr>
<th></th>
<th>73%</th>
<th>54%</th>
<th>40%</th>
<th>37%</th>
<th>30%</th>
<th>24%</th>
<th>21%</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Stalk and leaf</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca, % DM</td>
<td>0.40ab(^2)</td>
<td>0.41ab</td>
<td>0.43a</td>
<td>0.39a-c</td>
<td>0.38a-c</td>
<td>0.34bc</td>
<td>0.32bc</td>
</tr>
<tr>
<td>P, % DM</td>
<td>0.18a</td>
<td>0.14b</td>
<td>0.10c</td>
<td>0.10c</td>
<td>0.08bc</td>
<td>0.10b</td>
<td>0.06c</td>
</tr>
<tr>
<td>K, % DM</td>
<td>1.90a</td>
<td>1.82ab</td>
<td>1.83ab</td>
<td>1.89a</td>
<td>1.51cd</td>
<td>1.62bc</td>
<td>1.35d</td>
</tr>
<tr>
<td>Mg, % DM</td>
<td>0.27a</td>
<td>0.26a</td>
<td>0.22ab</td>
<td>0.20ab</td>
<td>0.17b</td>
<td>0.16b</td>
<td>0.17b</td>
</tr>
<tr>
<td>S, % DM</td>
<td>0.09a</td>
<td>0.08b</td>
<td>0.07bc</td>
<td>0.07c</td>
<td>0.06c</td>
<td>0.06c</td>
<td>0.05d</td>
</tr>
<tr>
<td><strong>Cob</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca, % DM</td>
<td>0.08ab</td>
<td>0.07ab</td>
<td>0.09a</td>
<td>0.06b</td>
<td>0.08ab</td>
<td>0.07ab</td>
<td>0.08ab</td>
</tr>
<tr>
<td>P, % DM</td>
<td>0.12a</td>
<td>0.06b</td>
<td>0.04bc</td>
<td>0.03c</td>
<td>0.03c</td>
<td>0.03c</td>
<td>0.02c</td>
</tr>
<tr>
<td>K, % DM</td>
<td>0.53bc</td>
<td>0.53bc</td>
<td>0.50c</td>
<td>0.49c</td>
<td>0.59a-c</td>
<td>0.70a</td>
<td>0.65ab</td>
</tr>
<tr>
<td>Mg, % DM</td>
<td>0.07a</td>
<td>0.04b</td>
<td>0.04bc</td>
<td>0.03c</td>
<td>0.03c</td>
<td>0.03c</td>
<td>0.03bc</td>
</tr>
<tr>
<td>S, % DM</td>
<td>0.05a</td>
<td>0.03b</td>
<td>0.03b</td>
<td>0.03b</td>
<td>0.03b</td>
<td>0.03b</td>
<td>0.03b</td>
</tr>
<tr>
<td><strong>DDGS</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca, % DM</td>
<td>0.11a</td>
<td>0.09a-c</td>
<td>0.08b-d</td>
<td>0.09a-c</td>
<td>0.09ab</td>
<td>0.07cd</td>
<td>0.07d</td>
</tr>
<tr>
<td>P, % DM</td>
<td>0.90a</td>
<td>0.87ab</td>
<td>0.80bc</td>
<td>0.81bc</td>
<td>0.75c</td>
<td>0.79c</td>
<td>0.81bc</td>
</tr>
<tr>
<td>K, % DM</td>
<td>2.60a</td>
<td>1.43b</td>
<td>1.03c</td>
<td>1.04c</td>
<td>1.00c</td>
<td>1.00c</td>
<td>1.04c</td>
</tr>
<tr>
<td>Mg, % DM</td>
<td>0.35ab</td>
<td>0.35ab</td>
<td>0.35ab</td>
<td>0.36ab</td>
<td>0.34b</td>
<td>0.33b</td>
<td>0.37a</td>
</tr>
<tr>
<td>S, % DM</td>
<td>1.13a</td>
<td>0.76b</td>
<td>0.60c</td>
<td>0.60c</td>
<td>0.54c</td>
<td>0.54c</td>
<td>0.58c</td>
</tr>
</tbody>
</table>

1. All data are means of three observations.
2. Values followed by the same letter in the same row are not significantly different (p > 0.05).

6.3.2 Feed value enhanced stover

FVES is a mixture of corn plant residues (corn stover) and DDGS from the fermentation of corn from the corn plants sampled. Both NDF and ADF concentrations in FVES were lowest when corn just reached maturity, and then consistently increased as corn moisture decreased (Table 13). Lower concentrations of NDF and ADF indicated higher digestibility of forages. Except for the first harvest date, CP concentration in FVES was stable throughout of the study period, between 9.2% and 9.8%, which was higher than that in corn kernel (8.8% in CP). The nutritive and energy values were calculated using the prediction equations based on ADF concentrations. Dry matter yield of TDN per hectare was highest when corn just reached its physiological maturity at a
corn moisture content of 40%. It was attributed to the fact that both the highest TDN concentration (% DM) and highest dry matter yield of FVES happened at corn physiological maturity stage. TDN yield in FVES decreased by 2.6 t/ha when corn moisture decreased from 40% (maturity stage) to 21%, which was due to the increase in ADF concentration and decrease in dry matter yield with delay in harvest. Furthermore, the highest NEL, NEM, NEG happened when corn and stover harvested when corn just reached its maturity (Table 13). The results in Table 13 indicated that if the corn and stover were harvested as soon as the corn reached maturity, the TDN value in FVES (11.2 t/ha) was 72% of that in the whole plant harvested at the corn moisture of 21.2% (w.b.) (15.6 t/ha). The corn portion of the corn plants could be used for the ethanol production.
Table 13. Chemical compositions and nutritive values in FVES in seven harvests in 2009.

<table>
<thead>
<tr>
<th></th>
<th>Corn M.C. (%)</th>
<th>73%</th>
<th>54%</th>
<th>40%</th>
<th>37%</th>
<th>30%</th>
<th>24%</th>
<th>21%</th>
<th>Corn³</th>
<th>Stover³</th>
<th>Whole plant³</th>
</tr>
</thead>
<tbody>
<tr>
<td>NDF¹, % DM</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>62.4bc</td>
<td>62.9a-c</td>
<td>60.2c</td>
<td>62.0a-c</td>
<td>62.3a-c</td>
<td>62.9ab</td>
<td>63.4a</td>
<td></td>
<td>7.4</td>
<td>81.3</td>
<td>41.3</td>
</tr>
<tr>
<td>ADF, % DM</td>
<td>38.4a-c</td>
<td>37.4bc</td>
<td>35.9c</td>
<td>37.9a-c</td>
<td>38.5a-c</td>
<td>40.4a</td>
<td>39.6ab</td>
<td></td>
<td>1.8</td>
<td>52.1</td>
<td>24.9</td>
</tr>
<tr>
<td>CP, % DM</td>
<td>8.2b</td>
<td>9.2ab</td>
<td>9.3ab</td>
<td>9.2ab</td>
<td>9.8a</td>
<td>9.8a</td>
<td>9.5ab</td>
<td></td>
<td>8.8</td>
<td>3.4</td>
<td>6.3</td>
</tr>
<tr>
<td>NEL (MJ/kg)</td>
<td>5.26ab</td>
<td>5.35ab</td>
<td>5.53a</td>
<td>5.26ab</td>
<td>5.26ab</td>
<td>4.98b</td>
<td>5.07b</td>
<td></td>
<td>8.30</td>
<td>3.69</td>
<td>6.18</td>
</tr>
<tr>
<td>NEM (MJ/kg)</td>
<td>5.72a-c</td>
<td>5.81ab</td>
<td>5.90a</td>
<td>5.72a-c</td>
<td>5.72a-c</td>
<td>5.53c</td>
<td>5.63bc</td>
<td></td>
<td>7.29</td>
<td>4.43</td>
<td>6.00</td>
</tr>
<tr>
<td>NEG (MJ/kg)</td>
<td>3.32a-c</td>
<td>3.32ab</td>
<td>3.51a</td>
<td>3.32a-c</td>
<td>3.32a-c</td>
<td>3.14c</td>
<td>3.23bc</td>
<td></td>
<td>10.33</td>
<td>2.12</td>
<td>6.55</td>
</tr>
<tr>
<td>TDN, % DM</td>
<td>61.6a-c</td>
<td>62.2ab</td>
<td>63.2a</td>
<td>61.9a-c</td>
<td>61.5a-c</td>
<td>60.3c</td>
<td>60.8bc</td>
<td></td>
<td>89.5</td>
<td>52.5</td>
<td>72.5</td>
</tr>
<tr>
<td>TDN per hectare (t/ha)</td>
<td>10.1ab</td>
<td>10.2ab</td>
<td>11.1a</td>
<td>9.3bc</td>
<td>8.7cd</td>
<td>8.1d</td>
<td>8.6cd</td>
<td></td>
<td>10.3</td>
<td>5.3</td>
<td>15.6</td>
</tr>
</tbody>
</table>

1. All data are means of three observations.
2. Values followed by the same letter in the same row are not significantly different (p > 0.05).
3. Values are based on the corn and stover harvested on 23 November, with harvest kernel moisture of 21.2%.
4. NDF, neutral detergent fiber; ADF, acid detergent fiber; CP, crude protein; NEL, net energy of lactation; NEM, net energy of maintenance; NEG, net energy of gain; TDN, total digestible nutrients.
6.4 Conclusions

Corn harvest moisture content affected chemical compositions and nutritive values of corn stover, DDGS and feed value enhanced stover. Neutral detergent fiber, acid detergent fiber and lignin concentrations in corn stover consistently increased with corn harvest moisture content decreasing. Crude protein concentrations decreased in both corn stover and DDGS with corn harvest moisture content decreasing. For feed value enhanced stover, both the highest dry matter yield and the highest total digestible nutrient concentration happened when the corn just reached physiological maturity. The study showed that the whole corn plant could be most efficiently used if the corn and corn stover were harvested as soon as corn reached physiological maturity.
Chapter 7. Determination of the respiration and effective diffusion coefficient of carbon dioxide through bulk corn

7.1 Introduction

Postharvest losses due to spoilage during grain storage remain a major problem around the world. Early detection of grain spoilage will reduce grain quantity and quality losses, decrease mycotoxin production in food chain, and avoid financial loss by applying timely management (Ileleji et al., 2006). Utilizing thermal cables in storage bin for temperature monitoring has been a traditional method for detecting grain spoilage, since microorganisms produce a large amount of heat in the spoilage location. However, temperature monitoring is usually not sensitive enough due to its low thermal diffusivities in bulk grain (Singh, 1983; Gonzales et al., 2009). Furthermore, measured temperature cannot be easily interpreted due to the influence of the ambient air fluctuation. For example, temperature of 25°C of bulk grain may mean there is a possible spoilage spot during storage in the winter months, or it may mean the bulk grain is heated by the ambient air in the summer. In addition to the temperature monitoring, studies reported that increases in carbon dioxide (CO₂) concentration in bulk grain is another indicator of grain deterioration (Steele et al., 1969; Seitz et al., 1982; Fernandez et al., 1985; Pronyk et al., 2004; Moog et al., 2010). The CO₂ concentration measured in a stored bulk can be compared to the CO₂ concentration of the ambient air (around 400 ppm) as a standard to interpret the readings (Singh et al., 1984). Muir et al. (1985) measured concentrations of CO₂ in interstitial air in 39 farm-stored bulks of wheat, rapeseed, barley and corn. Spoilage was confirmed by analyses of grain samples in 97% of the 34 bins having CO₂ concentrations greater than ambient air. More recently, studies reported that monitoring CO₂ concentration in the headspace of the storage bin with a CO₂ sensor can lead to earlier detections of grain spoilage compared to the temperature monitoring (Maier et al., 2006; Ileleji et al., 2006). CO₂ monitoring in bulk grain in silo bags is even more important since it is an indicator of whether hermetic conditions are being maintained.
In order to further develop effective and commercially feasible techniques for utilizing CO\(_2\) sensors for grain quality monitoring in storage bins and silo bags, knowledge of movement of CO\(_2\) in bulk grain is necessary. Since diffusion is one of the most important factors in gas movement in bulk grain, the effective diffusion coefficient \((D_e)\) of CO\(_2\) through bulk grain must be determined. Singh et al. (1984) determined the effective diffusion coefficient of CO\(_2\) through wheat, rapeseed, oats and corn by a steady state method and reported the dependence of the diffusion rate on temperature, moisture content and porosity. The main advantage of the steady state method is that it can provide the direct measurement of the effective diffusion coefficient. However, since measurements are possible only after equilibrium is reached, steady state methods are usually slow; several days are often being required for one measurement (Flegg, 1953). Thereafter, Singh et al. (1985) developed a transient method to determine the effective diffusion coefficient of CO\(_2\) through bulk wheat. Recently, Shunmugam et al. (2005) determined the effective diffusion coefficient of CO\(_2\) through bulk wheat, barley and canola by using a transient diffusion model.

All of the previous studies on measuring the effective diffusivity of CO\(_2\) through bulk grain did not consider the CO\(_2\) produced by grain respiration. The CO\(_2\) flux through bulk grain can be influenced by the CO\(_2\) production during the diffusion process, especially when the grain respiration rate was high at elevated temperatures and high grain moisture content. In these conditions, the CO\(_2\) production must be taken into consideration when measuring the effective diffusivity through bulk grain. Furthermore, to date, the effective diffusion coefficient of CO\(_2\) through bulk corn has been only reported by Singh et al. (1984) at only one temperature \((10^\circ\text{C})\) and one moisture content \((14.0\%)\). Moisture content and temperature of the bulk corn varies during corn storage period, especially when corn spoils. Hence, it is necessary to measure the CO\(_2\) diffusivity through bulk corn at different temperatures and grain moisture content. Therefore, the objective of this paper was to measure effective diffusion coefficient of CO\(_2\) through bulk corn at different temperatures and grain moisture content, with considerations of CO\(_2\) production by corn respiration during diffusion process.
7.2 Materials and methods

7.2.1 Sample preparation

Corn (P1395R, Pioneer Hi-Bred International, Johnston, IA) harvested at 22.2% moisture content in October 2012 was used for the experiment. The wet corn was dried at 49°C in a convection oven to 18.8 and 14.0% moisture content. The moisture content of corn was determined according to the ASAE Standard S352.2 (1988) based on the mass difference before and after oven drying. Prepared samples were transferred to a plastic bag, sealed and placed in a -10°C freezer. Prior to each test, the samples were warmed to the designated temperature in an incubator. Because porosity has a close relationship with the gas diffusivity (Shunmugam et al., 2005), the porosity of the bulk corn was determined using the following equation:

\[
\text{Porosity (\%)} = \left(1 - \frac{\text{bulk density}}{\text{unit density}}\right) \times 100\%
\]  

where bulk density of the corn was determined by the volume of the grain column and the mass of corn placed into the grain column; unit density was determined according to the methodology of volume complementation. The differences of the unit densities and porosities among corn with three moisture content were within ± 2% (Table 14).

Table 14. Unit densities and porosities of corn with different moisture content.

<table>
<thead>
<tr>
<th>Corn moisture content</th>
<th>14.0%</th>
<th>18.8%</th>
<th>22.2%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unit density (kg/m³)</td>
<td>1235.5 ± 5.8</td>
<td>1224.0 ± 5.4</td>
<td>1229.9 ± 7.1</td>
</tr>
<tr>
<td>Porosity (%)</td>
<td>35.1 ± 0.3</td>
<td>34.5 ± 0.3</td>
<td>34.8 ± 0.4</td>
</tr>
</tbody>
</table>

7.2.2 Diffusion measurement

Diffusion apparatus

A diffusion apparatus (Figure 14) was designed and fabricated to measure the effective CO₂ diffusion coefficient through bulk corn based on the concept by Reible and Shair (1982). The apparatus consisted of two cylindrical gas chambers connected by a
cylindrical grain column. Cumberland and Crawford (1987) reported that the diameter of the chamber should be at least 10 times larger than the size of the granular media to provide a representative volume. Since a corn kernel is approximately 0.5 cm in volumetric equivalent diameter, a 10.2 cm diameter and 20 cm length grain column was designed in order to minimize the kernel size and column edge effects. The two identical cylindrical gas chambers measuring 19.1 cm diameter and 15 cm length were made of acrylic tubes with 6.4 mm wall thickness. Bulk corn was placed within the grain column. To hold the bulk corn in place, wire mesh was fixed on both ends of the grain tube. A retractable seal (11 cm diameter) was made from soft rubber to block gas diffusion between gas chamber A and grain chamber before the diffusion test. Two 0.5 cm diameter holes were drilled in gas chamber A, one for gas injection and one for gas vent. A 2 cm diameter hole was drilled in chamber A & B and in the grain column for installing CO₂ gas probes. A digital manometer (Extech HD755, Extech Instruments, Nashua, NH) connected between the gas chamber A and gas chamber B was used to monitor any pressure difference across both chambers during testing. A low speed motor fan (ebm-papst Inc., Farmington, CT) was installed in gas chamber A & B to mix gas. The temperature of the apparatus was controlled by putting a cooling/heating water jacket (plastic tubing) around the gas chambers and the grain column using a circulating water bath with chiller. The seals of the apparatus were tested by injecting a high CO₂ concentration (10,000 ppm) gas and continuously measuring CO₂ concentration for 12 h. Seal tests showed that the leakage of CO₂ from the apparatus was less than 0.2% CO₂ per hour. Since each diffusion test was 4 h, the gas leakage was less than 0.8% during the whole test and was deemed acceptable.

The CO₂ sensors (GMT220 CO₂ transmitters and GMT 221 CO₂ probes, Vaisala Inc., Helsinki, Finland) used consisted of three parts, a detection probe, a transmitter and a cable (Figure 15). The sensor had an accuracy of ± 1.5 % of range + 2% of reading and it has been used to measure the CO₂ concentrations by other studies (Tang et al., 2003; Sydney et al., 2010). The detection probe was a silicon based, non-dispersive infrared (NDIR) sensor, with 1.85 cm diameter and 10.9 cm length. The measurement range of
the sensor was 0 to 11,000 ppm, with a response time of 20-30 seconds. The sensor measured CO$_2$ concentrations based on the diffusion through a hydrophobic membrane and provides a linear analog output as a function of CO$_2$ concentration. The operation temperature and humidity ranges of the sensor are -20 to 60°C and 0 to 100% RH, respectively. The CO$_2$ probes were installed in the gas chambers A and B and in the grain column to measure concentrations as a function of time during each diffusion test. After receiving the signal from the probe, the transmitter sent an output signal to a data acquisition module (Personal Daq/56, Measurement Computing Corporation, Norton, MA), and data were recorded every 1 min.

**Figure 14.** An apparatus for CO$_2$ diffusivity measurement. a) schematic and b) fabrication. CO$_2$ gas was injected into gas chamber A. After allowing gas to be well mixed in chamber A, the retractable seal was opened to start the diffusion process. CO$_2$ gas diffused from chamber A to chamber B through bulk corn in grain column. CO$_2$ sensors were used to monitor the CO$_2$ concentrations over time during the diffusion process. The CO$_2$ diffusivity was tested by a transient method. The diffusion apparatus was designed based on the concept by Reible and Shair (1982). Compared to the one designed by Reible and Shair (1982), the diffusion apparatus in this study added a cooling/heating system, electrical sensors to monitoring CO$_2$ change and had different sizes of the chambers and the column.
Figure 15. (cont.) fabricated apparatus CO$_2$ diffusivity measurement.

Figure 16. A CO$_2$ sensor module (GMT220 CO$_2$ transmitters and GMT 221 CO$_2$ probes, Vaisala Inc., Helsinki, Finland) used to measure CO$_2$ concentration in the gas chambers during diffusion. The sensor had an accuracy of ±1.5 % of range + 2% of reading. The detection probe was a silicon based, non-dispersive infrared (NDIR) sensor, with 1.85 cm diameter and 10.9 cm length. The measurement range of the sensor was 0 to 11,000 ppm, with a response time of 20-30 seconds. (Figure from http://www.vaisala.com)
Diffusion test procedures

To achieve a desired porosity, a 1.3 kg of corn was loaded into the column in six increments and after each increment, the grain column was shaken for 30 sec by hand. The retractable seal separating the grain chamber and the gas chamber was closed before each diffusion test. Gas chamber A was flushed with CO$_2$ (10,000 ppm, balanced with air) from a compressed gas cylinder after conditioning the gas to the desired relative humidity by bubbling gas through a saturated salt solution (Figure 16). After the CO$_2$ gas was well mixed by the low speed motor fan, the retractable seal was opened to start the diffusion process. The sensors installed on the apparatus measured CO$_2$ concentrations at 1 min intervals for 4 h from the start of the diffusion (seal opening).

The desired relative humidity was chosen based on the equilibrium moisture contents for corn at different relative humidity (ASAE, 2007). Equilibrium relative humidity for corn at moisture contents of 14.0, 18.8 and 22.2% were 81, 68 and 48%, respectively. The saturated salt solutions chosen to control the desired relative humidity were potassium chloride, potassium iodide and magnesium nitrate, respectively.

**Figure 17.** Sketch of the experimental setup for the diffusion tests. CO$_2$ gas from a compressed gas cylinder was bubbled through saturated salt solution to get the desired relative humidity. Water bath temperature controller was used to control the temperature of the corn in grain column by putting a water jacket (plastic tubing) around the gas chambers and the grain column.
7.2.3 Corn respiration measurement

Corn respiration measurement method in this study was similar to the one described by Dillahunty et al. (2000). The respiration measurement apparatus consisted of three main components: a gas pump, a conical flask containing salt solution and a plastic quart jar containing corn kernels (Figure 17). These three main components were connected with plastic tubing. A hole with a 2 cm diameter was drilled into the lid of a plastic quart jar, which was used to insert the CO₂ sensor probe. During measurement, the plastic quart jar was placed in a temperature controlled water bath. To prevent CO₂ leakage, the sensor ports and the lid were sealed with silicone sealant.

A 100 g sample of corn at the specified moisture content was placed in the plastic quart jar which was flushed with fresh air using a gas pump. The air from the gas pump was bubbled through saturated salt solution to reach the designed relative humidity before being allowed to flush the corn in the quart jar. After 6 min of flushing, the quart jar was completely sealed with stoppers. CO₂ produced by corn respiration was allowed to accumulate in the sealed jar. The sensor started to measure CO₂ concentrations at 1 min interval. The amount of CO₂ that accumulated inside the jar was used to calculate the corn respiration rate. Respiration rates were expressed as mg CO₂ produced by each kg of corn for each hour and they were calculated by the following equation (Dillahunty et al. 2000):

\[
\text{Respiration rate (mg CO}_2/\text{kg Corn/h)} = \frac{\Delta \% \text{ CO}_2 \text{ (mg/mL)} \times \text{volume of headspace in jar (mL)}}{\text{Corn sample mass (kg)} \times \text{time (hr)}}\]

Each test at various temperature and grain moisture conditions was replicated three times.
Figure 18. Schematic diagram of the respiration measurements. Corn sample was placed in the plastic quart jar, which was flushed with fresh air using a gas pump. The salt solution was used to control the relative humidity of air flushed into the jar. CO₂ produced by corn respiration accumulated in the sealed jar and the sensor was used to measure the CO₂ concentration.

7.2.4 Mathematical model and effective diffusion coefficient determination

The transient diffusion of CO₂ through bulk corn could be described by the 1-D modified Fick’s second law with a source term \( q \):

\[
\frac{\partial C}{\partial t} = D_e \frac{\partial^2 C}{\partial x^2} + q
\]  

(13)

where \( C \) is CO₂ concentration (mole/m³), \( t \) is time (s), \( D_e \) is the effective diffusion coefficient of CO₂ through bulk corn (m²/s), \( x \) is the distance (m) from the left of the grain column (interface between the gas chamber A and the grain column), and \( q \) is the corn volumetric respiration rate (mole/m³). For the conditions of the experimental apparatus (Figure 18), the initial conditions are

\[
C(x, 0) = C_0 \text{ at } -H \leq x \leq 0
\]  

(14a)

\[
C(x, 0) = C_1 \text{ at } 0 < x < L
\]  

(14b)

\[
C(x, 0) = C_2 \text{ at } L \leq x \leq L + H
\]  

(14c)
where \( C_0, C_1, C_2 \) are the \( \text{CO}_2 \) concentrations at the starting time \((t = 0)\) of the diffusion process in chamber A, grain chamber and chamber B, respectively. \( L \) is the length of the grain column (m) and \( H \) is the length of the gas chamber (m). And boundary conditions of the experimental setup are

\[
V_1 \frac{\partial C}{\partial t} = S D_e \left( \frac{\partial C}{\partial x} \right)_{x=0} \quad \text{at} \quad x = 0 \tag{15a}
\]

\[
V_2 \frac{\partial C}{\partial t} = -S D_e \left( \frac{\partial C}{\partial x} \right)_{x=L} \quad \text{at} \quad x = L \tag{15b}
\]

where \( V_1 \) and \( V_2 \) are the volumes of the gas chamber A and B, respectively, and \( S \) is the cross-section area of the diffusion chamber.

**Figure 19.** Schematic of boundary and initial conditions for the two-chamber diffusion cell. \( C_A \) and \( C_B \) are \( \text{CO}_2 \) concentrations in chamber A and chamber B at different time during the diffusion process.

Equation 13 along with the associated initial and boundary conditions (Equations 14 and 15) was numerically solved by using the Crank-Nicolson implicit finite difference method (Shunmugam et al., 2005). The grain column was divided into 200 spatial elements \((\Delta x)\), with 1 mm each. The time step \((\Delta t)\) was chosen as 0.5 s.
The discretization of the partial differential equation was shown as:

$$\frac{c_{j+1}^{n+1} - c_j^n}{\Delta t} = \frac{1}{2} D \left[ \frac{c_{j+1}^{n+1} - 2c_j^{n+1} + c_{j-1}^{n+1}}{\Delta x^2} + \frac{c_{j+1}^n - 2c_j^n + c_{j-1}^n}{\Delta x^2} \right] + q \quad (16)$$

where \( j \) and \( n \) are the spatial and time index, respectively.

Let \( R = \frac{D \Delta t}{2 \Delta x^2} \), and rearrange the equation:

$$-RC_{j-1}^{n+1} + (1 + 2R)C_j^{n+1} - RC_{j+1}^{n+1} = RC_{j-1}^n + (1 - 2R)C_j^n + RC_{j+1}^n + q \Delta t \quad (17)$$

The discretization of the boundary equation at \( x = 0 \) was shown as:

$$V_1 \frac{c_{0}^{n+1} - c_0^n}{\Delta t} = \frac{1}{2} SD \left[ \frac{c_{1}^{n+1} - c_0^{n+1}}{\Delta x} + \frac{c_1^n - c_0^n}{\Delta x} \right] \quad (18)$$

Let \( r_1 = \frac{SD \Delta t}{2V_1 \Delta x} \), and rearrange the equation:

$$C_0^{n+1} = \frac{r}{1+r} C_1^{n+1} + \frac{r}{1+r} C_1^n + \frac{1-r}{1+r} C_0^n \quad (19)$$

The discretization of the boundary equation at \( x = L \) was shown as:

$$V_2 \frac{c_{m+1}^{n+1} - c_{m+1}^n}{\Delta t} = \frac{1}{2} SD \left[ \frac{c_{m+1}^{n+1} - c_m^{n+1}}{\Delta x} + \frac{c_{m+1}^n - c_m^n}{\Delta x} \right] \quad (20)$$

Let \( r_2 = \frac{SD \Delta t}{2V_2 \Delta x} \), and rearrange the equation:

$$C_{m+1}^{n+1} = \frac{r}{1+r} C_m^{n+1} + \frac{r}{1+r} C_m^n + \frac{1-r}{1+r} C_{m+1}^n \quad (21)$$

The effective diffusion coefficient of CO\(_2\) through bulk corn was determined by minimizing the average percent error between the predicted CO\(_2\) concentrations by solving the PDE equation and the measured CO\(_2\) concentrations by installed CO\(_2\) sensors in both gas chamber A and gas chamber B at time from 1 min to 240 min. The measured CO\(_2\) concentration in grain chamber was not used in determining the diffusion coefficient, since the CO\(_2\) concentration in grain chamber exceeded the sensor detection limit during
the diffusion process when corn respiration rate was high (Figure 21). The average percent error was calculated by the following equation:

\[
\text{Average percent error} = \frac{1}{n} \sum_{t=1}^{n} \left| \frac{C_{\text{simulated}} - C_{\text{measured}}}{C_{\text{measured}}} \right| \times 100\% \tag{22}
\]

where \( n \) is the measurement time in minutes. The algorithm developed by MATLAB software to determine the effective diffusion coefficient was shown in Figure 19. A program code in MATLAB language is attached in Appendix A.

![Flow chart](image)

**Figure 20.** Flow chart for computer algorithm to determine the effective diffusion coefficient \( D_e \) by minimizing the average percent error between simulated and measured \( \text{CO}_2 \) concentrations.

### 7.2.5 Assumptions and limitations of the measurement model

Several assumptions were made in the simplified model:

1) Diffusion happened only in the x-direction.
2) No viscous flow in bulk corn, due to the pressure balance during diffusion test.
3) Ideal mixing both in the gas chamber A and B was achieved, so that the concentration in the chambers is uniform.

4) The CO₂ adsorption by corn was negligible.

The first assumption was used and validated by other studies (Ball et al., 1981; Singh et al., 1984; Jones et al., 2003; Shumugam et al., 2005). The second assumption was validated experimentally by monitoring the pressure difference across the two gas chambers using a digital manometer. The higher the pressure difference between the two ends of the diffusion column, the greater chances for viscous flow in the column. The pressure difference during the experiment was maintained at a negligible value (pressure < 1Pa) during the diffusion process. The third assumption is critical and has been discussed by Ball et al. (1981), Reible and Shair (1982), Singh et al. (1984) and Jones et al. (2003). Since diffusion of CO₂ in air is much larger than that in the porous media, this assumption is usually valid. Reible and Shair (1982) applied a small hand-driven fan to help keep the gas well mixed. In our experiment, a low speed fan was used to mix the gas in both gas chambers. The fourth assumption is based on the work done by Singh et al. (1984), who reported that the CO₂ adsorbed by wheat and rapeseed during 45 h was less than 5% of the amount of CO₂ diffused during the same period. In our experiment, it was assumed that the adsorption was negligible during a 4 h diffusion test.

7.2.6 Experimental design and statistical analysis

The variables studied were temperature (10, 20 and 30°C) and corn moisture content (14.0, 18.8 and 22.2%). A full factorial completely randomized design (3 temperature × 3 moisture content × 3 replicates) was used in this experiment. Tukey multiple range test was used for data analysis (SAS 9.2, SAS Institute Inc., Cary, NC). The level selected to show statistical significance was 5% (p < 0.05).

7.3 Results and discussion

7.3.1 Corn respiration rate

Differences were found in the respiration rates of corn at different moisture content and temperatures (Figure 20). Respiration of corn increased as moisture content
increased, which is in general agreement with published results with various grains (Bailey, 1940, Ragai and Loomis, 1953; Fernandez et al., 1982 and Dillahunty et al., 2000). The respiration rates of corn with moisture content of 14.0% were low, between 0.16 and 0.54 mg CO$_2$/kg corn/h and rose exponentially as moisture content increased. For example, at 20°C, the respiration rate of corn increased approximately 100 times from 0.31 mg CO$_2$/kg corn/h at 14.0% moisture content to more than 30 mg CO$_2$/kg corn/h at 22.2% moisture content. Corn at 14.0% moisture content is considered safe for bulk storage while corn containing 22.2% moisture is too wet for safe storage and favorable for deterioration. According to Shelled Corn Storage Time Table made by Bern et al. (2002), corn containing 22.2% moisture could be safely stored for only 10-13 days at 20°C. As would be expected, respiration of corn increased with temperature at each moisture level (Figure 20). The rise in respiration nearly doubled with each 10°C temperature increase. The respiration rates in this experiment were lower than those from Bailey (1940), who reported that corn respiration rates were between 0.3 and 9.1 mg CO$_2$/kg corn/h when corn moisture content was between 11 and 17%. This could possibly be due to the corn variety differences during a 70-year time span. The difference could be also due to the sample treatments. Bailey used dried corn samples and re-wetted them to the desired moisture, while we used freshly harvested corn in our experiment. Previous studies reported that freshly harvested corn had lower respiration rates than re-wetted corn (Ragai and Loomis, 1953; Fernandez et al., 1982).
Figure 21. Respiration rates of corn at three temperatures and three moisture content levels. The respiration rates were based on the dry matter of corn. Corn respiration rate increased when temperature and grain moisture content increased.

7.3.2 Effective diffusion coefficient of CO$_2$ through bulk corn

Once the respiration rates (CO$_2$ producing rates) of corn were obtained, they can be introduced into the modified diffusion model to calculate the effective diffusion coefficient of CO$_2$ through bulk corn. As respiration rate increased, it had a larger effect on the diffusion pattern from chamber A to chamber B. Three cases with different measured respiration rates from Figure 20 were illustrated: low respiration rate ($q = 0.3$ mg CO$_2$/kg corn/h) for 14.0% moisture corn at 20°C, moderate respiration rate ($q = 4.7$ mg CO$_2$/kg corn/h) for 18.8% moisture corn at 20°C, and high respiration rate ($q = 30$ mg CO$_2$/kg corn/h) for 22.2% moisture corn at 20°C. Figure 21 shows the measured CO$_2$ concentrations in chamber A, chamber B and the grain column at above three cases. The influence of respiration rate on the concentration values is clearly seen from these results. For the low respiration rate (case 1), CO$_2$ concentration in chamber A decreased while CO$_2$ concentration in chamber B increased. For the moderate respiration rate (case 2), CO$_2$ concentration in chamber A decreased slower than that in case 1, while CO$_2$ concentration in chamber B increased faster. This is due to the high CO$_2$ concentration built up in the grain column between these two gas chambers, which decreased concentration gradient from the chamber A to the grain column but increased
concentration gradient from the grain column to the chamber B. For the high respiration rate (case 3), the influence of respiration becomes more significant. CO$_2$ concentration in chamber A decreased in the first 15 min and then turned to increase due to the high CO$_2$ concentration built up in the grain column. CO$_2$ concentration in chamber B increased much faster than that in case 1 and case 2. Therefore, with only considering the respiration effect in the model, one can get an accurate prediction of the CO$_2$ diffusion in bulk grain.

Dry matter loss of corn during 4 h diffusion test can be calculated based on the corn respiration rate. In case 1, when corn moisture content and temperature were 14.0% and 20°C respectively, corn respiration rate was 0.3 mg CO$_2$/kg corn/h. According to the simple aerobic respiration equation

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + 2816 \text{kJ}$$

the dry matter loss was 0.8 mg/kg corn during 4 h diffusion test, which was less than $1 \times 10^{-4}$% of the total corn placed in the grain column. When corn moisture content was high at 22.2% and at 20°C temperature, corn respiration rate was 30 mg CO$_2$/kg corn/h. The dry matter loss was 82 mg/kg corn during 4 h diffusion test, which was less than 0.01% of the total corn placed in the grain column. Therefore, the dry matter loss during the 4 h diffusion test was in a negligible level. In case 3, the CO$_2$ concentration in grain column was estimated between 1-2% (since it reached sensor’s detection limit), and the O$_2$ concentration decreased from 19-20% according to the aerobic respiration equation. Whether corn aerobic respiration would be depressed at this high CO$_2$ concentration and low O$_2$ concentration remains unknown and needs further experiment to answer it.
Figure 22. Measured CO$_2$ concentrations in chamber A, chamber B and in the middle of the grain column at three levels of respiration rates: low respiration rate (q = 0.3 mg/kg/h) for 14.0% moisture corn at 20°C, moderate respiration rate (q = 4.7 mg/kg/h) for 18.8% moisture corn at 20°C, and high respiration rate (q = 30 mg/kg/h) for 22.2% moisture corn at 20°C. As respiration rate increased, it has a larger effect on the diffusion pattern from chamber A to chamber B during the diffusion test. At high respiration rate (case 3), CO$_2$ concentration in the grain chamber reached sensor’s detection limit (11,000 ppm) at approximately 160 min, and the measured data after 160 min were invalid.
The effective diffusion coefficients of CO$_2$ through bulk corn were in the range of $3.10 \times 10^{-6}$ to $3.93 \times 10^{-6}$ m$^2$/s, depending on moisture and temperature conditions (Table 15). Singh et al. (1984) reported the effective diffusion coefficient of CO$_2$ through bulk corn was $3.02 \times 10^{-6}$ m$^2$/s at 14% corn moisture and 10°C temperature. In our study, the diffusion coefficient of CO$_2$ through bulk corn was $3.30 \times 10^{-6}$ m$^2$/s at 14.0% corn moisture and 10°C temperature. These two diffusion coefficients were in good agreement, considering the difference in porosity and probable differences in kernel size and shape. Xu et al. (2002) suggested that effective diffusivity of CO$_2$ in stored grain can be approximated by multiplying the normal mass diffusivity of CO$_2$ in air by the porosity of grain if the matrix of medium is assumed not to affect gas diffusivity significantly. The results in this study showed that this approximation can result in an overestimate of the effective diffusivity in the stored corn. The diffusion coefficient of CO$_2$ in air could be calculated by the gas kinetic theory developed by Hirschfelder et al. (1949). At 10°C, the diffusion coefficient of CO$_2$ in air is calculated to be $13.8 \times 10^{-6}$ m$^2$/s. By Xu et al.’s approximation, the estimated effective diffusivity of CO$_2$ in bulk corn would be $4.7 \times 10^{-6}$ m$^2$/s, which is about 50% higher than the measured value in this study.

As temperature increased from 10 to 30°C, the diffusion coefficient increased from $3.21 \times 10^{-6}$ to $3.76 \times 10^{-6}$ m$^2$/s, by 17%. According to the kinetic theory of gas, an increase in temperature increases the velocity of gas molecules of the interstitial air and hence increases the CO$_2$ diffusion rate through bulk corn. This finding agrees with the study by Shumugam et al. (2005, who reported that the CO$_2$ diffusion through bulk grain (wheat, barley and canola) increased at different levels as temperature increased from 5 to 40°C. As corn moisture content increased from 14.0 to 18.8%, the effective diffusion coefficient through bulk corn decreased from $3.59 \times 10^{-6}$ to $3.39 \times 10^{-6}$ m$^2$/s. But as corn moisture content further increased from 18.8 to 22.2%, there was no difference observed on the effective diffusion coefficient. Overall, the effect of corn moisture content was not as large as the effect of temperature on the CO$_2$ diffusion coefficient.
Table 15. Diffusion coefficients (10^{-6} m^2/s) of CO\textsubscript{2} through bulk corn at various temperature and moisture content.

<table>
<thead>
<tr>
<th>Moisture (w.b.)</th>
<th>10°C</th>
<th>20°C</th>
<th>30°C</th>
<th>Mean across Temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td>14.0%</td>
<td>3.30 ± 0.00\textsuperscript{a}</td>
<td>3.53 ± 0.06</td>
<td>3.93 ± 0.07</td>
<td>3.59 ± 0.32\textsuperscript{a}\textsuperscript{b}</td>
</tr>
<tr>
<td>18.8%</td>
<td>3.10 ± 0.10</td>
<td>3.43 ± 0.10</td>
<td>3.63 ± 0.11</td>
<td>3.39 ± 0.27\textsuperscript{b}</td>
</tr>
<tr>
<td>22.2%</td>
<td>3.23 ± 0.15</td>
<td>3.50 ± 0.20</td>
<td>3.70 ± 0.12</td>
<td>3.48 ± 0.23\textsuperscript{ab}</td>
</tr>
<tr>
<td>Mean across moisture</td>
<td>3.21 ± 0.10\textsuperscript{c}</td>
<td>3.49 ± 0.05\textsuperscript{b}</td>
<td>3.76 ± 0.16\textsuperscript{a}</td>
<td></td>
</tr>
</tbody>
</table>

a. Values in the table are mean ± SE from three replicates.
b. Values followed by the same letter in the same column are not significantly different (p > 0.05).
c. Values followed by the same letter in the same row are not significantly different (p > 0.05).

7.4 Conclusions

The corn respiration rate increased when temperature and grain moisture content increased. As respiration rate increased, it had a larger effect on the diffusion pattern when measuring the effective diffusion coefficient. Effective diffusion coefficients of CO\textsubscript{2} through bulk corn ranged between 3.10 × 10^{-6} and 3.93 × 10^{-6} m^2/s, depending on temperature and moisture conditions. As temperature increased from 10 to 30°C, the diffusion coefficient of CO\textsubscript{2} through bulk corn increased by 17%. As corn moisture content increased from 14.0 to 18.8%, the effective diffusion coefficient through bulk corn decreased, but there was no difference observed in the effective diffusion coefficient when corn moisture content increased from 18.8 to 22.2%.
Chapter 8. Conclusions and recommendations

8.1 Conclusions

The feasibility of early harvest of corn and corn stover was examined in terms of dry matter yields, processing characteristics, chemical compositions and nutritive values. For corn harvested with high moistures, its storability was investigated by developing a CO$_2$ monitoring system during storage. In line with the four specific objectives stated in the Introduction, the following are the major conclusions.

1. The dry matter yield of corn increased rapidly until reaching corn maturity and remained stable after corn maturity, with an average yield of 11.1 t/ha over the two year study. For corn stover, the 2 year average dry matter yield was 14.8 t/ha at corn kernel filling stage and decreased to 13.2 t/ha at kernel physiological maturity and further decreased throughout kernel dry down. During corn dry down period, moldy kernel percentage increased from 2.1 to 4.1%, and plant lodging percentage increased from 1.2 to 3.6%.

2. Effects of corn harvest moisture content on dry grind ethanol processes were observed on fermentation characteristics and DDGS composition. The final ethanol concentration from corn with harvest moisture content of 54% (110 days after corn planting, kernel dent stage) was 0.5 to 1.2 percentage points higher than that from matured corn with lower harvest moisture contents. Ethanol conversion efficiencies for the corn with harvest moisture contents of 73 and 54% were higher than that for the corn with lower harvest moisture contents. For the corn wet milling process, as corn harvest moisture content decreased from 49 to 21%, the yields of starch and germ increased by 1.2 to 1.9 percentage points respectively, while the yield of steep water solids, total fiber and gluten decreased.

3. Corn harvest moisture content affected compositions and nutritive values of corn stover, DDGS and feed value enhanced stover (FVES). As corn harvest moisture content decreased, neutral detergent fiber, acid detergent fiber and lignin
concentrations in corn stover increased while crude protein concentrations decreased. The study showed that the whole corn plant could be the most efficiently used if the corn and stover were harvested as soon as corn reached physiological maturity.

4. Corn respiration rate increased when temperature and grain moisture content increased. As respiration rate increased, it had a larger effect on the diffusion pattern when measuring the effective diffusion coefficient. Effective diffusion coefficients of CO$_2$ through bulk corn ranged between $3.10 \times 10^{-6}$ and $3.93 \times 10^{-6}$ m$^2$/s, depending on temperature and moisture conditions. As temperature increased from 10 to 30°C, the diffusion coefficient of CO$_2$ in bulk corn increased. As corn moisture content increased from 14.0 to 18.8%, the effective diffusion coefficient through bulk corn decreased, but there is no difference observed in the effective diffusion coefficient when corn moisture content increased from 18.8 to 22.2%.

8.2 Recommendations for future work

Three potential issues were identified that would benefit from further investigation:

1. The results in this study showed that corn harvested at high moisture contents have higher ethanol conversion efficiencies compared to corn with lower harvest moisture contents. Tests in this study were only performed in one variety of the corn. It is possible that the harvest moisture effects on conversion efficiencies would vary among different varieties of corn. Therefore, it would be beneficial to repeat tests with different varieties of corn.

2. The results showed that corn harvested earlier with high moisture content had advantages. However, there are still some questions, such as how should we store high moisture corn? Dr. Eckhoff and I proposed the freezing concept to store the high moisture corn to save the cost and energy. However, data are needed to show this concept on a large scale. Also, will corn ethanol facilities accept high
moisture corn? An economic analysis of the whole process using high moisture corn needs to be performed to show if using high moisture corn is a profitable venture.

3. Having a good estimate of the effective CO₂ diffusivity through bulk corn is the first step to better understanding CO₂ movement in corn storage bins and hermetic storage bags. Mathematical models and field studies needs to be conducted in the future to develop a CO₂ monitoring system for corn storage.
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Available at:


Appendix A. Matlab code for calculating effective diffusivity

% solving PDE dc/dt = De*du^2/dx^2 + q
% were q is CO2 producing rate
% boundary conditions: De*du/dt = V/S*du^2/dx^2  at x = 0
% -De*du/dt = V/S*du^2/dx^2  at x = 0.2
% initial conditions:  u(x,0) = 0.44 mole/m3(10,000 ppm) at x = 0
% solving method: Crank_Nilcoson_Methods
% Measure the first four hours (4*3600s = 14400s)

clc
clear
L = 0.2; % grain chamber length
dx = 0.001; % length step
dt = 0.5; % time step
TT = 4 % simulated time in hours
TTS = TT*3600; % simulated time in seconds
D1 = 0.1908 % diameter of the gas chamber
D2 = 0.1016 % diameter of the grain chamber
H1 = 0.15 % Length of the gas chamber

k=1; % calculation index

V2 = 0.004294624 % Volume of the gas chamberB (sink chamber)
V1 = 0.00416336 % of the gas chamber A (source chamber)

S = pi()/4*D2^2 % cross-section area of the grain chamber

% initial value
x = [dx:dx:L-dx];
n = length(x);

% read the experimental measured data from excel
% EM is the experimental measured database
% EM(:,1) time in minutes
% EM(:,2) time in seconds
% EM(:,3) CO2 concentration in chamber A
% EM(:,3) CO2 concentration in chamber B

% ==============input==================
EM = xlsread('diffusion5.xlsx');% read files from excel

q1 = 0.53 % corn respiration rate (unit:mg/kg/hr, dry basis)
u1st = EM(1,3) % Initial CO2 concentration in Chamber A (ppm)
u2st = EM(1,4) % Initial CO2 concentration in Chamber B (ppm)
u3st = EM(1,5) % Initial CO2 concentration in bulk corn (ppm)\
\% == input ==
q = q1*(4.3e-6) \% nothing but unit convert from mg/kg/hr to mole/m^3/s

Average_Perc Pret Perv_e = 1e7 \% define the first step Error_sum

for De = 0 : 1e-7 : 9e-6

uend = u2st/22703; \% init CO2 concentration in chamber B (mole/m^3)
uporous = u3st/22703; \% init CO2 concentration in chamber B (mole/m^3)

delta_u = (uporous - uend)/(L/2.0);

for i = 1:n
u(i,1)= uend + delta_u*(200-i)*dx;
end

Q = q*dt*ones(n,1); \% CO2 respiration rate vector
u0 = u1st/22703; \% init CO2 concentration in chamber A (mole/m^3)

R = De*dt/(dx^2)/2;

r1 = De*S*dt/V1/dx/2;
r2 = De*S*dt/V2/dx/2;

\% algorithm_Crank_Nicolson_Methods
\% matrix A without b.c.
\% A*U(n+1) = B*U(n) + C \% C is the previous initial u0 and uend

A = sparse(diag(-R*ones(n-1,1),1)+
... diag((1+2*R)*ones(n,1))+diag(-R*ones(n-1,1),-1));

\% add boundary conditions
\% De*du/dt = V/S*du^2/dx^2 \at x = 0
\% -De*du/dt = V/S*du^2/dx^2 \at x = 0.2

A(1,1) = (1 + 2*R - R*r1/(1+r1));
A(n,n) = (1 + 2*R - R*r2/(1+r2));

B = sparse(diag(R*ones(n-1,1),1)+
... diag((1-2*R)*ones(n,1))+diag(R*ones(n-1,1),-1));

\% add boundary conditions
\% De*du/dt = V/S*du^2/dx^2 \at x = 0
\% -De*du/dt = V/S*du^2/dx^2 \at x = 0.2

B(1,1) = (1 - 2*R + R*r1/(1+r1));
B(n,n) = (1 - 2*R + R*r2/(1+r2));
% time steps number
s = TTS/dt;
% Create matrix for each time concentration at x = 0;
C0 = zeros(s,1);
% Create matrix for each time concentration at x = L;
Cend = zeros(s,1);
% index i
i = 1;
C0(i,1) = u0;
Cend(i,1) = uend;
% A*U(n+1) = B*U(n) + C %%% C is the previous initial u0
for t = dt:dt:s*dt
    ST = u(1,1); % store the previous u(1,1)
    SP = u(n,1); % store the previous u(n,1)
    BU = B*u;
    BU(1,1) = BU(1,1)+2*R/(1+r1)*u0; % BU = B*u + C
    BU(n,1) = BU(n,1)+2*R/(1+r2)*uend;
    RU = BU + Q; % add the CO2 source term (respiration)
    u = A\RU;
    u0 = r1/(1+r1)*u(1,1) + r1/(1+r1)*ST +(1-r1)/(1+r1)*u0;
        %calculate next
    uend = r2/(1+r2)*u(n,1) + r2/(1+r2)*SP+(1-r2)/(1+r2)*uend;
        %calculate next
    i = i+1;
    C0(i,1) = u0; % store u0 at each time step
    Cend(i,1) = uend; % store uend at each time step
end

% PART 2_Calculate the residues between simulated and measured data
% Measured data is from matrix EM
% Simulated data is from matrix C0 and Cend
% Start with C0
% totally there are 241 measured data points
% 0 min, 1min, 2min ....240 min

error_1 = zeros(241,0);
error_2 = zeros(241,0);
CendPPM = Cend*22703; %Convert the unit mol/m3 to ppm
C0PPM = C0*22703; %Convert the unit mol/m3 to ppm
for j = 2:241
    Cmeas = EM(j,4); %Cmeas is the measured data by CO2 sensors
    jt = EM(j,2); %jt is the time in seconds
nt = jt/dt;  % nt is the index in vector C0
Csimu = CendPPM(nt,1);  % Csimu is the simulated data
error_1(j,1) = (Cmeas - Csimu)/Cmeas/241;  % store the difference at each step
end

for j = 2:241
    Cmeas = EM(j,3);  % Cmeas is the measured data by CO2 sensors
    jt = EM(j,2);  % jt is the time in seconds
    nt = jt/dt;  % nt is the index in vector C0
    Csimu = C0PPM(nt,1);  % Csimu is the simulated data
    error_2(j,1) = (Cmeas - Csimu)/Cmeas/241;  % store the difference at each step
end

Average_Percent_Error = sum(abs(error_1))+ sum(abs(error_2))/2  % total error from all time steps

if Average_Percent_Error < Average_Percent_Error_Previous
    uend_Previous = uend;
    u0_Previous = u0;
    u_Previous = u;
    CendPPM_Previous = CendPPM;
    C0PPM_Previous = C0PPM;
else
    De_fit = De - 1e-7
    break;
end
end

%======exact data at every 1 min, and stored in another matrix======%
C_united = zeros(241,5);
for i = 1 : 241
    C_united(i,1) = EM(i,1);
    C_united(i,2) = EM(i,3);
    C_united(i,3) = EM(i,4);
end

nstep = (EM(2,2)-EM(1,2))/dt;
for i = 1 : 241
C_united(i,4) = C0PPM_Previous((i-1)*nstep+1,1);
C_united(i,5) = CendPPM_Previous((i-1)*nstep+1,1);
end

% plotting concentrations at each time step at x = 0 and x = L
%(time scale second)
figure (1)
tf = [0:dt:s*dt];
plot(tf,C0PPM_Previous,'LineWidth',2,'color','red')
hold all
plot(tf,CendPPM_Previous,'LineWidth',2,'color','red')
dtm = EM(2,2);
tfm = [0:dtm:240*dtm];
plot(tfm,EM(:,3),'LineWidth',1,'color','blue');
plot(tfm,EM(:,4),'LineWidth',1,'color','blue');
hold off
hleg1 = legend('Simulated','Simulated','Measured','Measured');
title('measured and simulated CO2 conc.')
xlabel('diffusion time (sec)')
ylabel('CO2 concentration (ppm)')

% plotting concentrations at each time step at x = 0 and x = L
%(time scale hour)
figure (2)
tf = [0:dt/3600:s*dt/3600];
plot(tf,C0PPM_Previous,'LineWidth',2,'color','red')
hold all
plot(tf,CendPPM_Previous,'LineWidth',2,'color','red')
dtm = EM(2,2)/3600;
tfm = [0:dtm:240*dtm];
plot(tfm,EM(:,3),'LineWidth',1,'color','blue');
plot(tfm,EM(:,4),'LineWidth',1,'color','blue');
hold off
hleg1 = legend('Simulated','Simulated','Measured','Measured');
title('measured and simulated CO2 conc.')
xlabel('diffusion time (hr)')
ylabel('CO2 concentration (ppm)')

% plotting concentrations at final step from x = 0 to x to L
%(time scale hour)
figure (3)
n = 1:length(HEM);
uppm = 22703*u_Previous;
plot(x,uppm,'LineWidth',2,'color','red')
title('simulated CO2 conc.from x = 0 to x = L')
xlabel('diffusion time (sec)')
ylabel('CO2 concentration(ppm)')
Appendix B.
Finite element modeling of carbon dioxide and temperature distributions in stored corn

B1. Introduction

The CO$_2$ movement in bulk corn can be primarily considered as the diffusive and convective transport of CO$_2$ through the interstitial spaces of the bulk corn where corn itself acts as a source for the CO$_2$ through respiration. In the diffusion transport, the CO$_2$ moves from high concentration field to low concentration to diminish concentration gradients and at the same time the respiration of corn at the spoilage spot tends to maintain such gradients, and thus, the problem of CO$_2$ movement becomes dynamic in nature. In North America, corn is harvested in the fall and placed in the storage bins at high temperatures after drying. As winter proceeds, the temperature of corn near the outer wall would decrease, but the temperature of corn in the center of the storage bin would remain high, due to the low thermal diffusivity of the bulk corn. Such temperature gradients would induce the natural convections in the grain storage bins. Moreover, the time varying ambient temperature makes the whole transport an unsteady process, which makes the system more complex.

In this section, a finite element model (FEM) was built to predict the CO$_2$ movement and distribution in stored corn, based on the measured corn respiration rate and CO$_2$ diffusivity through bulk corn in Chapter 7. In this model, only the diffusion of the CO$_2$ movement is considered. Compared to the comprehensive diffusion-convection model, the model built based on the diffusion only is simpler and needs less computational time since it does not involve the bulk fluid flow computation. However, this model would be a good starting point in developing a physical understanding of CO$_2$ movement process in bulk corn.

B2 Model development

Transient CO$_2$ transport and temperature changes in porous media can be described by partial differential equations based on the mass and energy conservation theorem. All
conservation equations developed in this study assumed a continuum. In other words, the bulk corn in the storage bin is evenly distributed in the storage bin. Mass conservation for CO$_2$ is given as:

$$\frac{\partial C_j}{\partial t} = \nabla \cdot (D_e(T) \nabla C_j) + r(T) \quad \text{(B1)}$$

where $C_j$ is CO$_2$ concentration in mol/m$^3$, $D_e(T)$ is effective diffusion coefficient through bulk corn as a function of temperature ($T$) in m$^2$/s, $t$ is diffusion time in s. $\frac{\partial C_j}{\partial t}$ represents the change in CO$_2$ concentration in time in bulk corn and $\nabla \cdot (D_e(T) \nabla C_j)$ on the right side of the equation represents the net CO$_2$ diffusion flux through bulk corn. $r(T)$ represents the source term, the CO$_2$ generation rate (mole/m$^3$·s) by corn respiration, which is dependent on temperature ($T$). In this equation, the sorption term is neglected. The energy equation is expressed as:

$$\left(\rho_{\text{bulk}} c_{\text{bulk}} \right) \frac{\partial T}{\partial t} = \nabla \cdot (k_{\text{bulk}} \nabla T) + \rho_{\text{bulk}} q_H r(T) \quad \text{(B2)}$$

where $\rho_{\text{bulk}}$ is corn bulk density in the storage bin in kg/m$^3$, $c_{\text{bulk}}$ is specific heat of bulk corn (J/kg·°C), $k_{\text{bulk}}$ is thermal conductivity through bulk corn in W/m·K and $q_H$ represents heat generated by each mole of CO$_2$ produced by corn respiration in kJ/mole [CO$_2$]. $\left(\rho_{\text{bulk}} c_{\text{bulk}} \right) \frac{\partial T}{\partial t}$ represents temperature change in bulk corn, $\nabla \cdot (k_{\text{bulk}} \nabla T)$ represents the total energy transfer due to thermal diffusion, and $\rho_{\text{bulk}} q_H r(T)$ is the source term, representing the heat production due to respiration. Thus, the energy conservation equation is linked with the mass conservation equation through the source term.

The $q_H$ value is based on the simple aerobic respiration equation (Cofie-Agblor et al., 1997):

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + 2816kJ$$
Thus, if the CO$_2$ respiration rate $r(T)$ in mole/m$^3$·s is known, according to this stoichiometric respiration equation, the heat released can be calculated as:

$$q = \frac{28.16 \text{ kJ}}{6 \text{ mole}} r(T) = q_H r(T)$$

(B3)

Therefore, $q_H$ was determined as 469.3 kJ/mole [CO$_2$].

The thermal properties of the bulk corn were determined from the relationships reported by ASABE Standards (ASAE, 2003). The thermal conductivity of the bulk corn (W/m·K) is expressed as:

$$k_{bulk} = 0.1409 + 0.00112 \text{ M.C.}$$

(B4)

And the specific heat of the bulk corn (kJ/kg·K) is expressed as:

$$c_{bulk} = 1.4651 + 0.0356 \text{ M.C.}$$

(B5)

where M.C. is moisture content of the corn in wet basis.

**B3 A case study for CO$_2$ and heat transfer in storage bin**

An example of a storage bin with corn was studied in this section. CO$_2$ diffusion through bulk corn in a circular steel bin with a concrete floor, 6 m in diameter and 4 m in height was modeled in this study. The capacity of the storage bin was approximately 140 m$^3$ (90 tonne for corn). It was assumed that the bin was filled with 14% moisture content corn, except for a spoilage spot. The spoiling corn was assumed to be a cylinder with 1 m in diameter and 0.3 m in height, located in the center of the steel bin (Figure B1). The spoiling corn was assumed to have a moisture content of 22%, with a high respiration rate of $1.25 \times 10^{-4}$ mole/m$^3$·s according to the measured data in Figure 16. For initial conditions, the temperature in the entire storage bin was assumed to be a uniform $15 \text{ °C}$, and the CO$_2$ concentration was assumed to be the same as that in the ambient air of 400 ppm (0.0176 mol/m$^3$).
Figure B1. Schematic figures of the storage bin and boundary conditions assumed for the model.

Corn respiration rate, \( r(T) \), is a dependent variable of temperature and moisture content. The moisture content of corn in this study was assumed to be 14\%, and it was assumed that the moisture did not change during the storage period. Therefore, the corn respiration rate was dependent on the temperature only. The data measured in Chapter 7 was used to estimate the respiration rates of corn at different temperatures by an exponential growth model (Figure B2).
Figure B2. Measured and predicted respiration rate of corn at different temperatures. The respiration rates of corn at different temperatures were estimated by an exponential growth model.

The effective CO$_2$ diffusion coefficient in bulk corn at various temperatures and moisture contents were measured in chapter 7, Table 15. Moisture content did not have an effect on the effective diffusion coefficient, but the temperature had an effect on the effective diffusion coefficient ($p < 0.05$). Therefore, it was assumed that the effective CO$_2$ diffusion coefficient was dependent on temperature only. A linear regression model was used to predict the effective diffusion coefficient at different temperatures (Figure B3).
The concrete floor of the bin and steel bin wall were assumed to be impermeable to CO\textsubscript{2} diffusion. Thus, the boundary conditions for the bin floor and wall were expressed as:

\[
\frac{\partial c_j}{\partial n} = 0
\]  

In reality, the top surface of the grain is open to atmosphere under a ventilated roof. Due to the high diffusivity of CO\textsubscript{2} in air, it is reasonable to assume that CO\textsubscript{2} leaving the top surface of the grain diffuses away instantaneously. Therefore, the CO\textsubscript{2} boundary condition for the top surface of the grain was expressed as:

\[
c_j = c_a
\]  

where \(c_a\) is the CO\textsubscript{2} concentration in the atmosphere, which equals to 400 ppm (0.0176 mol/m\textsuperscript{3}). The temperature \(T\) of the bin wall and the top grain surface was specified to be the same as the varying atmosphere temperature \(T_a(t)\). The temperature boundary condition for the bin wall was expressed as:

\[
T = T_a(t)
\]
In this study, the daily average dry bulk temperatures at Illinois/Champaign from 1 October to 30 October were obtained from NOAA (http://www.noaa.gov/) and are based on the period of 2002 to 2012. Daily variations of the average temperature are presented in Figure B4.

![Temperature vs Dates in October](image)

**Figure B4.** Averaged daily atmosphere temperature from 1 October 1 to 30 October in a period between 2002 and 2012.

The concrete floor was assumed to be adiabatic based on other studies (Khankari et al., 1995). Based on this assumption, the boundary condition on bin floor was expressed as:

$$\frac{\partial T}{\partial n} = 0$$  \hspace{1cm} (B9)

**B4 Numerical simulation methods**

The heat and CO₂ transport equations developed above are coupled, unsteady in nature and involve variable transport properties, making finding analytical solutions difficult. A finite element software COMSOL Multiphysics™ was used to numerically solve the partial differential equation system. All boundaries at storage bin wall were considered to be impermeable to CO₂ transport and the wall temperature was considered
to be equal to the ambient temperature. These boundary conditions made the calculation domain (storage bin) axisymmetric, and hence calculations could be performed in a two dimensional axisymmetric cylindrical coordinate \((r, z)\) system (Figure B5). By using the two-dimensional axisymmetric system, the computational requirements were dramatically reduced.

![Diagram](image)

**Figure B5.** Reduction of a three dimensional coordinate system to a two dimensional coordinate system.

The storage bin with bulk corn was initially meshed by 2,449 non-uniform triangle and quadrilateral elements by COMSOL Multiphysics™. The initial mesh was refined at the edge of the storage bin and at the pocket of corn spoilage location to catch up the large temperature and CO\(_2\) concentration gradients during heat and CO\(_2\) diffusion. The mesh refinement improved the accuracy of the simulation. It increased the number of grid cells to 4,479 (Figure 16). The COMSOL Multiphysic MUMPS Linear solver was used to solve this two dimensional unsteady state model to predict the heat and CO\(_2\) transport in the storage bin. The convergence criteria assigned by COMSOL Multiphysics were retained for all simulations. The total time span of the simulation is 30 days (from 1 October to 30 October) and the time step of the simulation was 1 h.
Figure B6. Two dimensional mesh for the axisymmetric storage bin.

**B5 Simulation results**

CO₂ concentrations and temperatures were high in the spoilage spot, because of the high respiration rate inside. CO₂ and heat in the spoilage spot diffused into the rest of the storage bin at different rates according to the diffusion coefficients (Figure B7). At the end of 1 day storage period, CO₂ concentrations inside the spoilage spot increased dramatically from 400 ppm (ambient concentration) to approximately 35,000 ppm. At a location about 1 m away from the spoilage spot, the CO₂ concentration was approximately 3,500 ppm. Although the temperature inside the spoilage pocket increased from 15 to 17.5°C at the end of 1 day, the temperature at the location 1 m away from the spoilage spot had little change due to the low thermal diffusivity through bulk corn. At the end of 10 day storage period, the CO₂ concentration inside the spoilage pocket increased to more than 55,000 ppm. Because of its high diffusivity through bulk corn, the CO₂ concentration in the entire storage bin increased to more than 5,000 ppm. The temperature inside the spoilage spot increased to approximately 26 °C. At a location of 0.5 m away from the spoilage spot, the temperature changed slightly from 15.0 to 15.6°C,
which is difficult to be detected with temperature sensors, such as a thermal couples typically used in grain bins. Thermocouples have a typical accuracy of ± 2 °C. At the end of 3- day storage period, the temperature near the bin boundaries decreased due to the decreasing ambient temperature in late October. However, the temperature inside the spoilage spot kept increasing due to the large amount of heat produced by respiration. The temperature in the spoilage spot reached to 31 °C although the ambient temperature was only 8 °C. The large temperature gradient between the inside bin and outside bin boundaries would increase the heat conduction. The CO₂ concentration in the entire storage bin increased to more than 6,000 ppm. Overall, CO₂ diffused much faster in the storage bin than heat. The simulation results in this study agree with the report published by Singh and Wallace (1965), who stated that a single temperature measurement must be within about 0.5 m of an active spoilage spot to detect the grain spoilage. Further, Ileleji et al. (2006) conducted field study by comparing CO₂ concentrations and temperatures monitoring and concluded that temperature cables alone was not a reliable indicator of stored grain conditions and CO₂ sensors could be used as an additional complimentary tool for stored grain management.
(a) 1 day storage period

(b) 10 day storage period

(c) 30 day storage period

Figure B7. Distributions of CO$_2$ concentration and temperature at different storage period.