

**NET ENERGY OF WHEAT-CORN DISTILLERS DRIED GRAINS WITH  
SOLUBLES FOR GROWING PIGS AS DETERMINED BY THE  
COMPARATIVE SLAUGHTER, INDIRECT CALORIMETRY, AND  
CHEMICAL COMPOSITION METHODS**

By

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A Thesis submitted to the Faculty of Graduate Studies of

The University of Manitoba

In partial fulfilment of the requirements

for the degree of

**MASTER OF SCIENCE**

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## ABSTRACT

Two experiments were conducted to determine the net energy (**NE**) of wheat-corn distillers dried grains with solubles (**wcDDGS**) using the comparative slaughter (**CS**), the indirect calorimetry (**IC**) and the chemical composition (**CH**) methods. Based on the CS method, NE values of 2,407, and 2,424 kcal/kg DM were obtained for wcDDGS included at 15% and 30%, respectively. For the IC method, the NE values of 2,407, and 2,403 kcal/kg DM were obtained for wcDDGS included at 15% and 30%, respectively; corresponding values for the CH method were 2,536 and 2,197 kcal/kg DM, respectively. It is concluded that NE value of wcDDGS ranges from 2,367 kcal/kg DM to 2,416 kcal/kg DM depending on the method used. As the values obtained from the various methods were not different, the average NE value for the wcDDGS evaluated was 2,396  $\pm$  25.71 kcal/kg DM.

## **DEDICATION**

This thesis is dedicated to my parents, Isaac Olatoyemu Ayoade and Esther Awuke Ayoade and to my siblings, Elizabeth, Dupe, Tunji, Racheal, Taiwo, Kehinde, Idowu, and Tunde.

## ACKNOWLEDGEMENTS

I owe a deep and sincere gratitude to my advisor, Dr. C. M. Nyachoti for his support and guidance throughout my MSc. programme. His constructive criticism and contribution in improving my presentation and writing skills are highly cherished and appreciated. I would also like to thank other members of my advisory committee, Drs. B. Slominski, G. Crow and S. Cenkowski for their contributions and valuable suggestions during committee meetings.

I wish to appreciate the Husky Energy, Agri-Food Research and Development Initiative, Western Economic Diversification, and Manitoba Pork Council for funding this project.

Thanks to Drs E. Kiarie, F. Opapeju, T. Woyengo and J. Heo for their guidance and for taking time to answer my numerous questions, Messias, Neijat, Rose, Atta and Nijitha for helping with slaughter of pigs and chamber measurements.

Appreciation goes to all administrative and technical staff of the Department of Animal Science, Robert Stusky, Dawin Ramos, Akin Akinwumi, Bernard, Margaret Ann Baker, Carol schlamb, Kathy Graham, Cathy Plouffe, Mei Ding, Atanas Karamano, Lisa Rigaux, Anna Rogiewics, Karen Carrette, Harry Muc, Prakash Sharma, and Janice Haines.

I appreciate Drs. Akinremi and Ige (Soil Science) for linking me with my Advisor.

I wish to express gratitude to my friends and church members in Winnipeg for their support and assistance throughout my MSc. programme. Special thanks to my brother-in-law, Emmanuel Ige and family for accommodating me on my arrival in Winnipeg and for helping with the admission processing.

Finally, my sincere appreciation goes to my fiancé, Temidayo Adewole for his constant support and encouragement.

I give glory to God Almighty who was, is, and is to come.

## **FOREWORD**

This thesis was written in a manuscript format and it is composed of two manuscripts. Manuscript I was partly presented at the 2011 ASAS/ADSA Joint Meeting, New Orleans, Louisiana. All manuscripts were written according to the guidelines for the American Society of Animal Science manuscript preparation. Authors to manuscript I are D. I. Ayoade, E. Kiarie, B. A. Slominski and C. M. Nyachoti, while authors to manuscript II are D. I. Ayoade, E. Kiarie, and C. M. Nyachoti.

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**LIST OF ABBREVIATIONS**

AA	Amino acids
ADF	Acid detergent fibre
ADFI	Average daily feed intake
ADG	Average daily gain
ATP	Adenosine triphosphate
ATTD	Apparent total tract digestibility
BW	Body weight
C	Carbon
CER	Calculated energy retention
CF	Crude fiber
CH <sub>4</sub>	Methane
CH	Chemical composition method
CO <sub>2</sub>	Carbon dioxide
CP	Crude protein
CS	Comparative slaughter
d	Day
DADF	Digestible acid detergent fiber
DC	Digestibility coefficient
DCP	Digestible crude protein
DDGS	Dried distillers grains with solubles

DE	Digestible energy
DEE	Digestible ether extract
DM	Dry matter
DMI	Dry matter intake
DRES	Digestible residual
EBW	Empty body weight
EDDM	Enzyme digestible ileal dry matter
EUDM	Enzyme undugested ileal dry matter
FC	Fermentable carbohydrate
FU	Feed unit
FUgp	Feed unit for growing pigs
FUgs	Feed unit for gestating sows
FUp	Feed unit for pigs
EDTA	Ethylenediaminetetraacetic acid
EE	Ether extract
Exp	Experiment
FHP	Fasting heat production
g	Gram
G:F	Gain to feed ratio
H	Hydrogen
h	Hour
HI	Heat increment
HP	Heat production

IC	Indirect calorimetry
IDC	Ileal digestible carbohydrate
kcal	Kilocalorie
kg	Kilogram
kJ	Kilojoules
ME	Metabolizable energy
ME <sub>m</sub>	Energy required for maintenance
MER	Measured energy retention
N	Nitrogen
NDF	Neutral detergent fiber
NE	Net energy
NE <sub>p</sub>	Net energy for production
NSP	Non starch polysaccharides
O <sub>2</sub>	Oxygen
ONEm	Total operational NE requirement for maintenance
P	Phosphorus
RDCF	Ileal digestible crude fat
RE	Retained energy
RQ	Respiratory quotient
SD	Standard deviation
SEM	Standard error of the mean
SID	Standardized ileal digestible
ST	Starch

VFA	Volatile fatty acids
wcDDGS	Wheat-corn distillers dried grain with solubles

## 1.0 GENERAL INTRODUCTION

The cost of feed is the most important cost of pig meat production representing more than 50% of the total cost and the energy component constitutes the greatest proportion of the feed (Noblet and Perez, 1993), thus energy becomes the most important nutrient in terms of cost. The primary goal of diet formulation is to accurately match energy supply to the pig's energy requirement for maintenance and productive functions. Energy supply below or above the pig's requirement may have an adverse effect on performance, quality of product and the environment (Chiba, 2000). Therefore, it is important to estimate precisely the energy value of feeds and to investigate the energy systems used to best meet the energy needs of pigs.

Swine diets can be formulated on a variety of energy systems which are (1) the digestible energy (**DE**) (2) the metabolizable energy (**ME**) and (3) the net energy (**NE**) systems. The DE is the dietary gross energy (**GE**) minus the GE of feces. Thus it accounts for loss of energy not digested and absorbed in the gut. The ME is the DE minus the GE of urinary and gaseous losses. Thus, it accounts not only for the fecal energy lost by the pig but also losses in urine and methane gas. The NE is the difference between ME and heat increment (**HI**). It thus accounts for losses in energy due to heat production (**HP**). Net energy is the energy retained by the animal for productive purposes.

Compared to the NE system, the DE and ME systems over-estimate the energy value to the animal of protein- and fiber-rich ingredients and under-estimate energy value

of starch and high fat ingredients (Noblet *et al.*, 1994a). This is because when diet ingredients contain high levels of fiber or protein, more energy is liberated as heat and thereby lost. The NE system accounts for the reduction in the efficiency of energy utilization from the hindgut whereas other systems do not. Similarly, because less heat is produced in metabolizing fat into energy, it is given a higher value than fiber when using the NE system (Noblet *et al.*, 1994a).

The NE system provides more accurate estimates of the energy available to the animal in an ingredient and the subsequent diet. Diets formulated using the NE system are typically lower in crude protein (CP) than those formulated using the DE or ME (Payne and Zijlstra, 2007). The decrease in nitrogen excretion as a result of lower CP results in a reduction in ammonia emissions and odour in barns which leads to improved animal performance and reduced environmental pollution (Canh *et al.* 1998). Using the NE system reduces diet cost because it leads to a reduction in the quantities of feedstuffs containing starch and an increase in feedstuffs containing fibre (Patience 2005; Payne 2006).

The evaluation of feedstuffs under the NE system would make it more economical to use co-products such as distillers dried grains with solubles (**DDGS**). It has been reported that DDGS has higher concentration of fibre than its parent grain due to the removal of starch during the fermentation process (Weigel *et al.*, 1997; Widyaratne and Zijlstra, 2007). Thus, DDGS should be evaluated on a net energy basis because of the increase in the amount of heat produced from fermentation in the hindgut of pigs. Combined with digestible amino acids and the ideal protein concept, a NE system will

allow nutritionists to formulate diets that provide the animal with the energy and amino acids that it needs for efficient and predictable growth and carcass performance.

Over the years in North America, nutritionists have been formulating pig diets using the DE or ME system. However, in European countries especially in The Netherlands and France, the NE system is widely used. Presently, in the North American pork industry, research data supporting the NE system is lacking (Patience and Beaulieu, 2005).

Although, the comparative slaughter (CS) technique is considered to be the gold standard methodology for determining the NE value of feeds and feedstuffs in animals, it is costly, time-consuming and requires large numbers of animals (van Milgen and Noblet, 2003). The indirect calorimetry (IC) method has been used to estimate heat production (HP) and all metabolic functions from activity and respiration (Noblet *et al.*, 1987; Bikker *et al.*, 1995), and it is a faster means of determining NE of feeds and feedstuffs. The IC system requires fewer animals and can be used for repeated measurement of energy balance. The NE value of feedstuffs can also be related to its chemical characteristics (Noblet and Perez, 1993). Net energy prediction equations for growing pigs based on the chemical composition measurements of the feeds have been developed (Noblet *et al.*, 1994a).

Therefore, it was hypothesised that the IC method of determining NE values of wheat-corn DDGS (**wcDDGS**) for growing pigs will give similar values to the CS technique and that these values will not be different from those obtained from Noblet *et al.* (1994a) prediction equation. To test this hypothesis, two experiments were conducted

with the overall objective of determining the NE of wcDDGS. The specific objectives of this study therefore were (1) to determine the NE value of wcDDGS for growing pigs using the CS, IC, and CH methods (2) to compare the CS, IC, and CH methods of determining NE of feeds and feedstuffs (3) to determine the effect of inclusion level on the NE value of wcDDGS.

## 2.0 LITERATURE REVIEW

### 2.1 INTRODUCTION

Energy is produced when the carbohydrates, fats and protein in feeds are metabolised by oxidative processes in the body. The carbon (C), hydrogen (H) and oxygen (O<sub>2</sub>) in carbohydrates, fats and protein react with inhaled O<sub>2</sub> to form carbon dioxide (CO<sub>2</sub>), water, and energy (Cromwell, 2001). In the body, much of the energy that is produced from this reaction is captured in the form of high-energy phosphate bonds. These bonds ultimately release this stored form of energy for functions which include synthesis of body protein and fat for growth, breathing, heartbeat, and other physical activities which keep the pig alive (Cromwell, 2001). Some of the energy produced when feed nutrients are metabolized is not retained by the body but is lost as heat. Some of this heat is used to maintain body temperature. The rest is dissipated from the body (Cromwell, 2001). The first and second law of thermodynamics hold that all forms of energy are quantitatively convertible to heat (Baldwin and Bywater, 1984) and hence all measurements of energy transactions are made and expressed in terms of heat energy or calories (Armsby, 1917). A calorie is the amount of heat required to raise the temperature of one gram of water from 14.5 to 15.5°C (Pond *et al.*, 1995). The joule is recommended as a unit at a higher level of abstraction expressing energy per se when different forms of energy (heat, work, electric energy) are added or subtracted. Therefore, heat and chemical energy should be expressed in calories (Kleiber, 1972).

Animal nutrition is focused on two forms of energy-chemical and heat (Oresanya, 2005). Carbohydrates, protein and fats have an average caloric value of 4.1, 5.7 and 9.4 kcal/g, respectively (Brafield and Llewellyn, 1982; Pond *et al.*, 1995). Often a particular feed ingredient may contain an excess of one or more nutrients and be deficient in others. Also, due to physiological factors in the gastrointestinal tract and digestible and metabolic inefficiencies, no single feed ingredient is used to supply the animal's requirement for nutrients (Oresanya, 2005). Therefore, there has been a concerted effort to quantitatively describe the energy value of the vast array of feed ingredients available for selection in practical swine diets (NRC, 1998).

## **2.2 BASIC DEFINITION OF ENERGY**

Energy is defined as the capacity of a physical system to perform work. Energy exists in several different forms which are heat, mechanical, light or electrical forms. According to the law of conservation of energy, the total energy in a system remains constant, although it may be transformed to another form. For example, mechanical energy such as kinetic energy of motion can be converted to heat energy, while potential energy can be converted to kinetic energy as the object moves. Chemical energy in plants can be converted to heat and chemical energy in animal products.

## **2.3 ENERGY NEEDS OF SWINE**

In order for animals to survive and grow, they need a supply of dietary energy. Dietary energy is used for maintenance and productive functions. Maintenance includes basal functions and involuntary activities such as muscle tone, feed digestion, blood

circulation, tissue replacement (Wenk *et al.*, 2000; Vestergren, 2001), cellular ion transportation for maintaining membrane potential and acid-base homeostasis (Baldwin and Bywater, 1984; Milligan and Summers, 1986). In addition, energy is required for homeothermal functions i.e. the maintenance of body temperature irrespective of the environment in which the pig is placed (Cole, 1995). Maintenance also involves the degradation of complex chemical substances into simpler substances that can be eliminated as waste products from the kidneys, digestive tract, lungs and skin (Oresanya, 2005). Under optimum conditions where thermoregulation, detoxification, immune, fever and stress responses are absent, energy available for maintenance is distributed into four parts for physical activity, cellular ion transport activity, protein turnover and other maintenance activity (e.g. waste elimination) (Verstegen, 2001).

### **2.3.1 Energy for maintenance**

Maintenance is defined as the requirement of nutrients for the continuity of vital processes within the body so that the net gain or loss of nutrients by the animal is zero (ARC, 1981). In reality, this definition may be suitable only for mature, non-pregnant, non-lactating animals. Growing pigs fed to maintain constant weight deposited protein at the expense of body fat (Black, 1974; Campbell, 1988; Wiesemuller *et al.*, 1988; Kolstad and Vangen, 1996). Thus, growing pigs will not be in constant energy balance considering the higher heat of combustion of fat that is lost in exchange for protein gain. The energy required for maintenance (**ME<sub>m</sub>**) includes the needs of all body functions and moderate activity (NRC, 1998). These requirements are usually expressed on a metabolic basis, which is defined as body weight raised to the power of 0.75 ( $BW^{0.75}$ ). Other

exponents have been suggested as more appropriate: 0.67 (Heusner, 1982); 0.60 (Noblet *et al.*, 1989); and 0.42 (Noblet *et al.*, 1994a). The exponent 0.75 has been suggested as not appropriate for estimating a constant  $ME_m$  value per unit of metabolic body size over the growing period because it underestimates  $ME_m$  for the growing pig (Tess *et al.*, 1984; Thorbek *et al.*, 1984). Noblet *et al.* (1991) proposed 0.60 as the best exponent for predicting  $ME_m$ . The most commonly used exponent is 0.60 (Noblet *et al.*, 1994a).

### **2.3.2 Energy for growth**

It is generally assumed that greater priority is given to maintenance than growth. Energy supplied above the need for maintenance is partitioned into protein and lipid synthesis (Kolstad *et al.*, 2002). Lean tissue and growth rate respond in a linear manner with energy intake up to a point at which protein deposition is at a maximum (Close, 1996). Additional energy supplied beyond this point will produce a large increase in lipid deposition with little or no increase in lean tissue deposition. Lipid deposition, on the other hand, increases at a greater rate above the capacity point for lean deposition than below it (Close, 1996).

## **2.4 CLASSIFICATION OF ENERGY SYSTEMS FOR SWINE**

### **2.4.1 Gross Energy**

Gross energy is the energy liberated when a substance is burned in a bomb calorimeter. It describes the amount of energy available in the feed and it is the simplest measure of energy. It is also the maximum amount of energy that is available for use by the animal (Ewan, 2001). If the chemical composition of a feed is known, GE can be

predicted fairly accurately (NRC, 1998). The following relationship was reported by Ewan (1989) for predicting GE (kcal/kg) from ether extract (**EE**), crude protein (**CP**) and ash.

$$GE = 4,143 + (56 \times \%EE) + (15 \times \%CP) - (44 \times \%Ash) \dots\dots\dots [2.1]$$

Not all GE is absorbed by the pig; some is lost in the fecal matter. Growing pigs rarely retain more than 50% of the GE (van Milgen and Noblet, 2003). Although for most diets, 80 to 90% of GE is digested, not all this energy is available for metabolism as energy will be lost in the urine and as methane. Therefore, GE is a poor estimate of energy content in feed for the pig.

#### **2.4.2 Digestible energy**

Digestible energy refers to the energy available after fecal energy has been subtracted from GE (Fig. 2.1). Apparent indigestible energy is a major variable in the evaluation of feed ingredients (NRC, 1998). The ratio (x 100) between DE and GE, called the digestibility coefficient (DC) of energy varies between 70 and 90% for most pig diets and between 0 and 100% for ingredients (Noblet and Henry, 1993). These variations are associated with differences in fecal digestibility of the nutrients constituting organic matter (Noblet and Henry, 1993). With regard to crude protein and crude fat, their DC varies between 60 and 95% according to their chemical characteristics and their origin, while soluble carbohydrates (starch and sugars) are 95 to 100% digestible (Noblet and Henry, 1993). Most of the variation of energy DC is associated with the presence of fiber (defined as the sum of non-starch polysaccharides (**NSP**) and

lignin) which is less digestible and also reduces the apparent fecal digestibility of crude protein and fat (Noblet and Shi, 1993). The digestive utilization of fibre is variable. For example, Chabeauti *et al.* (1991) found DC of total NSP in wheat and straw, wheat bran, sugar beet pulp and soybean hulls equivalent to 16, 46, 69, and 79%, respectively. Based on these observations, the reduction of DC of energy with dietary fiber addition will vary with the tested fibrous material. Agricultural Research Council (1981) and Morgan and Whittemore (1982) suggested that DE is preferable in describing the energy content of swine feeds because it is easily and precisely determined. Digestible energy is considered more exact for the description of pig feeds because it is largely free of animal effects (Morgan and Whittemore, 1982). In addition, DE values are available for most of the commonly used feeds. In diet formulation, it is assumed that DE is additive (i.e. the energy contribution per unit of feed is constant and independent of the other components of the diet). This assumption has proven correct in some studies (Whittemore and Moffat, 1976; Young *et al.*, 1977). However, in the conventional scheme of energy utilization, DE is apparent, not true, because fecal metabolic energy is not considered (NRC, 1998). Digestible energy will be different for a very young pig which has not developed the appropriate digestive enzymes to deal with the feed, or for older pigs with active fermentative bacteria in their ceca (Morgan and Whittemore, 1982).

### **2.4.3 Metabolizable energy**

Metabolizable energy is the energy available for growth or productive purposes and other life processes. It is obtained by subtracting urinary and gaseous (methane) energy from DE (Fig. 2.1). In growing pigs, energy lost as methane is usually relatively

low; an average of 0.4% of DE intake was measured by Noblet *et al.* (1994a) on 41 diets. However, this percentage ranged from 0.1 to 1.2%, the latter value being obtained with diets which contained soybean hulls or sugar beet pulp. In that situation, the methane energy loss represented about 5% of the DE of the raw material. In most situations, the ME: DE ratio is relatively constant and equivalent to about 0.96 (Noblet and van Milgen, 2004). However, the ratio is not acceptable when dietary CP content and/or protein retention are either low or high. In a study by Noblet *et al.* (1990), ME:DE ratios ranged from 100% for animal fat to 98% for cereals, 93 to 96% for protein sources and 90 to 92% for fibrous protein sources. For most practical swine diets in North America, ME is 94 to 97% of DE, with an average of 96% (Farrell, 1979; ARC, 1981). Metabolizable energy allows comparison of energy utilization with other species, but it is influenced by the growth characteristics of the animal and the level and source of protein in the diet (Morgan and Whittimore, 1982). If protein fed to pigs is of poor quality or in excess, ME decreases because the amino acids not used for protein synthesis are catabolised and used as a source of energy, and the nitrogen is excreted as urea. Therefore, as the nitrogen content of the urine increases, the energy losses in the urine increase and the ME of the diet decreases.

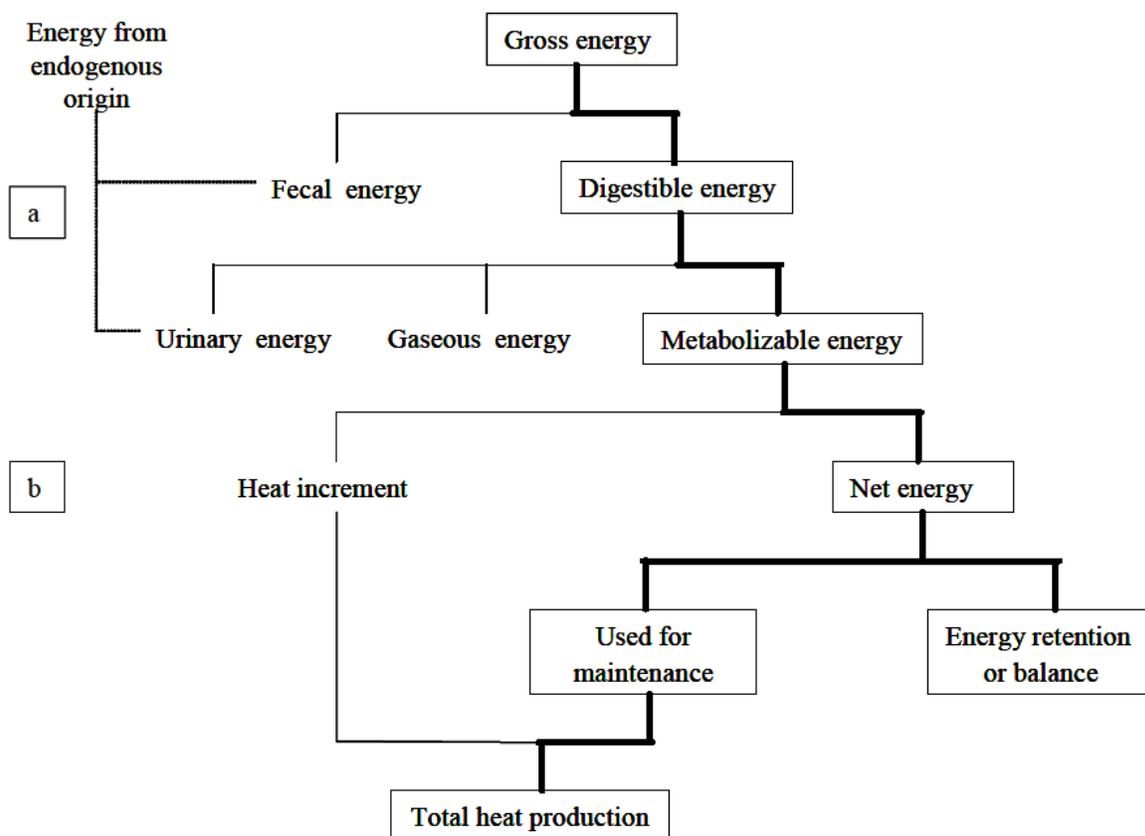
#### **2.4.4 Net energy**

Net energy is the difference between ME and heat increment (**HI**) (Birkett and de Lange, 2001). The HI is the amount of heat released because of the energy costs of the digestive and metabolic processes. The energy of the HI is not used for productive processes but can be used to maintain body temperature in cold environments. Net

energy, therefore is the energy that the animal uses for maintenance ( $NE_m$ ) and production ( $NE_p$ ). The energy used for maintenance is also dissipated as heat, so that total heat production is the sum of HI and  $NE_m$  (NRC, 1998). If energy is required to maintain body temperature or excess activity,  $NE_p$  is reduced. Net energy is the best indication of the energy available to an animal for maintenance and production (Noblet *et al.*, 1994a). In most NE based feed evaluation systems, energy of digestible nutrients (i.e, crude protein, crude fat, starch, sugars, and dietary fiber) is used to calculate the NE content of a diet or feed ingredient (Noblet *et al.*, 1994a). To determine NE, energy retention or heat production (HP) needs to be measured. Energy retention can be measured with the CS technique or by measuring the carbon-nitrogen balance. For pigs fed conventional diets and kept at thermoneutral temperatures, the ratio of NE to ME ranged from 0.66 to 0.75 (Thorbeck, 1975). The efficiency of ME utilization for energy gain and maintenance in growing pigs varies from 27% for wheat middling to 69% for corn to 75% for soybean oil (Ewan 1976; Philips and Ewan, 1977; Pals and Ewan, 1978). Noblet *et al.* (1994a) reported efficiencies of energy utilization of 90, 82, 80, 72 and 60% for rapeseed oil, cornstarch, sucrose and mixtures of protein and fibre sources, respectively, for pigs ranging in weight from 45 to 150 kg. Net energy values for growing pigs are calculated as the sum of fasting heat production and retained energy (Noblet *et al.*, 1994a) whereas retained energy is measured as the difference between ME intake and HP obtained in respiration chambers (Noblet *et al.*, 1994a). Net energy is the only system in which energy requirements and diet energy values are expressed on the same basis which is independent of the feed (Noblet and Henry, 1993).

## **2.5 HEAT PRODUCTION AND ENERGY RETENTION IN SWINE**

As reviewed by van Milgen and Noblet (2003), all ME not retained by the animal is lost as heat. The retained energy can be measured directly by the CS technique and the metabolic processes responsible for the energy supply to the body can be determined by measuring the HP of the animal. The quantity of energy in the oxidative processes of carbohydrates, fat, protein and short chain fatty acids can be measured as heat. Heat production can be measured by means of various calorimetry methods, either directly in a calorimeter or indirectly by measuring the gas exchange from the animal.



**Figure 2.1 Classical description of energy utilization.** A) Energy from endogenous sources contribute to fecal energy, Metabolizable energy = digestible energy minus urinary energy (with a portion from endogenous source) and gaseous energy; b) Total heat production = Heat increment plus maintenance energy. Adapted from Oresanya (2005)

## **2.5.1 Methods for quantitative determination of heat production and energy retention in swine**

Heat production may be measured by direct or indirect calorimetry. Energy retention, the actual part of feed energy retained by the animal, may be measured by either the comparative slaughter technique or by carbon-nitrogen balance (Adeola, 2001). Although, the CS method requires simple equipment, it requires labour and gives an estimate of the average energy retention over a longer period of time (van Milgen and Noblet, 2003).

### ***2.5.1.1 Comparative slaughter technique***

The most direct estimate of the energy retained in the body of an animal would be obtained as the difference between a determination of the body composition at the beginning and again at the end of a period of time which is clearly impossible in an animal (Blaxter, 1989). An alternative is to determine the body composition of a precisely similar animal at the beginning of the period and of the experimental animal at the end (Blaxter, 1989). This method of determining energy retention is termed the comparative slaughter method. Energy retention is the difference between the body energy contents of the initial and final slaughter groups (Kil *et al.*, 2011).

### ***2.5.1.2 Carbon-Nitrogen Balance technique***

This method is based on the assumption that the only energy-yielding compounds stored by the body are fat and protein and that these have fixed chemical composition and enthalpies of combustion (Blaxter, 1989). The C-balance includes the measurement of

carbon in feed and that voided in feces, urine, CO<sub>2</sub> and CH<sub>4</sub>, while the N-balance is based on the measurement of N in feed, feces and urine. The C-balance gives the total amount of C retained in the body and the amount of C retained in fat can be calculated by subtracting the amount of C retained in protein as determined by the N-balance.

### 2.5.1.3 Indirect Calorimetry

Heat production is closely correlated to O<sub>2</sub> consumed, CO<sub>2</sub> produced, CH<sub>4</sub> produced, and urinary N produced (Adeola, 2001). Noblet *et al.* (1987) presented strikingly similar values for HP of pigs that were determined by IC or by CS techniques as 201 and 217 kcal/d/BW<sup>0.75</sup>, respectively. Most studies on HP in animals and man have been made by indirect methods in which heat is not determined directly but calculated according to various methods (Christensen *et al.*, 1988). This method involves measurements of O<sub>2</sub> intake, CO<sub>2</sub> and CH<sub>4</sub> production and N excretion in urine. Heat production is then calculated from the following empirical relationship (Brouwer, 1965), assuming 1 kcal = 4.1855 kJ:

$$HP = 16.18 \times O_2 + 5.023 \times CO_2 - 2.17 \times CH_4 - 5.989 \times UN \dots \dots \dots [2.2]$$

Where HP is in kJ; O<sub>2</sub>, CO<sub>2</sub>, CH<sub>4</sub> in litres; UN is urinary N in g. This method is termed the RQ method because it is based on the determination of the respiratory quotient (RQ = litres CO<sub>2</sub>/litres O<sub>2</sub>), has been used, often in reduced form (without correction for CH<sub>4</sub> and UN) (Christensen *et al.*, 1988). Another indirect method is to calculate HP as the difference between metabolizable energy (ME) and total energy retained in the body:

$$HP = ME - RE \dots \dots \dots [2.3]$$

Where HP = heat production, ME = metabolizable energy and RE = retained energy and all parameters are in kJ.

Retained energy is based on measurements of the C and N balances, assuming that all energy is retained either as fat or protein. It is generally believed that identical values are obtained by direct and indirect calorimetry (Blaxter, 1962) and also between the indirect principles according to the RQ or CN method (Thorbek, 1975). Calorimetry has the advantage over the CS technique in that it can be used to measure energy balance over successive short periods of time, even within days.

## 2.6 FASTING HEAT PRODUCTION IN SWINE

The energy expended in the fasting animal is represented by the fasting heat production (**FHP**). Determination of FHP on non-producing adult animals is the basis for the calculation of minimum quantity of NE, which must be supplied to the animal to keep it in energy equilibrium (Chandramoni *et al.*, 1999). Measurement of FHP provides a useful basis of reference for other phases of energy metabolism (Chandramoni *et al.*, 1999). Net energy systems use FHP as an estimate of the maintenance energy requirement (Noblet *et al.*, 1994a). During fasting, energy from body reserves is mobilized in order to generate adenosine triphosphate (ATP) for essential functions. However, normally-fed growing animals will seldom mobilise body reserves (other than glycogen) in order to supply energy for essential functions. Measured values of activity-free FHP range from 700 to 800 kJ/kgBW<sup>0.60</sup>/d in growing pigs (Le Bellego *et al.*, 2001; van Milgen *et al.*, 2001; Le Goff *et al.*, 2002).

## 2.7 COMPARISON OF ENERGY SYSTEMS FOR SWINE

It is extremely important to use the same energy system for expressing the diet energy values and the pig's energy requirements (Noblet, 2007). The energy value of protein-rich or fibrous feeds is overestimated when expressed on a DE or ME basis. On the other hand, animal fat is underestimated when expressed on a DE or ME basis. For instance, Noblet *et al.* (1993) reported a similar DE value for wheat and soybean meal (16.17 and 16.35 MJ/kg DM, respectively). However, soybean meal contained 34% less NE compared with wheat (8.02 vs. 12.14 MJ/kg DM) (Table 2.1). Similarly, the NE value of sunflower meal was only 53% of its DE value (8.02 and 16.35 MJ/kg DM for

NE and DE, respectively) compared with 75% obtained with wheat (12.14 and 16.17 MJ/kg DM for NE and DE, respectively) (Table 2.1). On the other hand, although wheat and tapioca contained a fairly comparable DE concentration (16.17 and 15.84 MJ/kg DM, respectively), the NE value of tapioca was 6% higher than that of wheat (12.91 and 12.14 MJ/kg DM, for tapioca and wheat, respectively) (Table 2.1). These values clearly indicate that DE underestimates the energy value of starch ingredients and overestimate that of high protein ingredients. Several growth trials conducted with variable dietary fat or crude protein levels has shown that the energy cost of growth or daily energy requirement are independent of diet composition when expressed on a NE basis (Noblet, 2007). On the other hand, on a DE or ME basis, the energy cost is decreased when CP content is decreased or fat content is increased (Sauvant *et al.*, 2004). This shows that DE and ME overestimate the energy value of protein and underestimates the energy value of fat. Therefore, unlike the NE system, the DE and ME systems are unable to predict the performance of pigs.

**Table 2.1** Digestible (DE), metabolizable (ME), and net energy (NE) in MJ/kg DM of selected feed ingredients

Ingredients	DE	ME	NE
Wheat	16.17	15.81	12.14
Barley	15.09	14.76	11.51
Maize	15.81	15.28	12.42
Tapioca	15.84	15.58	12.91
Sweet potatoes	12.25	14.63	12.27
Soybean meal	16.35	15.28	8.02
Peas	16.22	15.67	11.05
Animal fat	29.83	29.59	29.32
Maize distillers	11.54	10.79	7.39

Adapted from Noblet *et al.* (1993).

## **2.8 NET ENERGY SYSTEMS**

Net energy systems have been developed in France, The Netherlands, and Denmark. The French and the Dutch systems are based on NE values from animal experiments and prediction equations, whereas the Danish system is based on the potential physiological energy (**PPE**) released from ATP bonds at the cellular level of pigs (Stewart, 2005).

### **2.8.1 The French NE System**

The French NE system was developed by Noblet *et al.* (1994a). The NE values of 61 diets were measured in 45-kg growing boars using an IC system. The amounts of DE before the end of the ileum and in the hindgut were measured for each diet. Regression equations for predicting dietary NE content were calculated and a total of 11 prediction equations were developed (Table 2.2). In the present study, NE is calculated from DE; therefore, only equations 3, 4, and 5 are used.

**Table 2.2.** Equations for prediction of NE (kcal/kg DM) of diets for growing pigs (Noblet *et al.*, 1994a)

Equation
1 NE = 2.73 x DCP + 8.37 x DEE + 3.44 x ST + 0 x DADF + 2.93 x DRES
2 NE = 2.69 x DCP + 8.36 x DEE + 3.44 x ST + 0 x DCF + 2.89 x DRES
3 NE = 0.843 x DE - 463
4 NE = 0.703 x DE + 1.58 x EE + 0.47 x ST - 0.97 x CP - 0.98 x CF
5 NE = 0.700 x DE + 1.61 x EE + 0.48 x ST - 0.91 x CP - 0.87 x ADF
6 NE = 0.870 x ME - 442
7 NE = 0.730 x ME + 1.31 x EE + 0.37 x ST - 0.67 x CP - 0.97 x CF
8 NE = 0.726 x ME + 1.33 x EE + 0.39 x ST - 0.62 x CP - 0.83 x ADF
9 NE = 2,796 + 4.15 x EE + 0.81 x ST - 7.07 x Ash - 5.38 x CF
10 NE = 2,790 + 4.12 x EE + 0.81 x ST - 6.65 x Ash - 4.72 x ADF
11 NE = 2,875 + 4.38 x EE + 0.67 x ST - 5.50 x Ash - 2.01 x (NDF - ADF) - 4.02 x ADF

CP, crude protein; EE, ether extract; CF, crude fibre, ST, starch; SG, sugar; ADF, acid detergent fiber; DCP, digestible CP; DEE, digestible ether extract; DADF, digestible acid detergent fibre; DRES, digestible residual = digestible organic matter - (DCP + DEE + ST + DADF) all in g/kg DM.

### 2.8.2 The Dutch NE System

The Central Bureau Livestock Feeding (**CVB**) developed the NE system in The Netherlands. The CVB used a variation of one of Noblet's calculated NE prediction equations (Stewart, 2005). In this system, the concentration of starch and sugars was determined by the enzymatic method using amyloglucosidase (**ST-Am**) whereas the French system used the polarimetric procedure of Ewers ( **ST-Ew**). The ST-Am gives lower values compared with the ST-Ew (Blok, 2006). The CVB calculated the digestible crude fat fraction in the ingredient using digestible ether extract with an acid hydrolysis method as opposed to the ether extract method used by Noblet *et al.* (1994a). The equations developed by the CVB are presented below:

$$NE = 2.58 \times DCP + 8.63 \times DEE + 3.23 \times ST + 3.04 \times SG + 2.27 \times DRES \dots \dots \dots [2.4]$$

$$NE = 2.58 \times DCP + 8.63 \times DEE + 3.23 \times \text{ileal ST} + 2.92 \times \text{ileal SG} + 2.27 \times DRES \dots \dots [2.5]$$

Where NE = net energy (kcal/kg), DCP = digestible CP, DEE = digestible ether extract using acid hydrolysis, ST = starch, SG = sugar, and DRES = digestible residual ((digestible organic matter – (DCP + DEE + ST + digestible ADF))

### 2.8.3 The Danish NE System

Boisen and Verstegen (1998) proposed a new concept called “the physiological energy” for estimating the NE value of pig feeds. This concept is based on the combination of in vitro digestion methods for evaluating the ATP potential production

from the components and biochemical coefficients for evaluating the ATP potential production from components (Noblet, 2000). The energy evaluation system of the Scandinavian feed units (FU; an energy value equal to approximately one kg of barley) is based on the physiological energy value of an ingredient or on its ATP equivalents (Oresanya, 2005). In 2002, a new system that requires analysis of enzyme digestible ileal dry matter (EDDM) replaced the old feed unit for pig (FU<sub>p</sub>). The system is based on the following equation:

$$\text{FU}_{\text{gp}} \text{ per kg DM} = [9.9 \times \text{RDCP} + 31.7 \times \text{RDCF} + \text{factor} \times \text{IDC} + 7.0 \times \text{FC} - 28 \times \text{EUDMi}] / 7375 \dots [2.6]$$

$$\text{FU}_{\text{gs}} \text{ per kg DM} = [9.9 \times \text{RDCP} + 26.1 \times \text{RDCF} + \text{factor} \times \text{IDC} + 9.0 \times \text{FC} - 28 \times \text{EUDMi}] / 7540 \dots [2.7]$$

where FU<sub>gp</sub> is feed unit for growing pig; FU<sub>gs</sub> is feed unit for gestating sow, RDCF is ideal digestible crude fat, IDC is ileal digestible carbohydrate, FC is fermentable carbohydrate and EUDMi is enzyme undigested ileal dry matter, all g/kg DM.

Compared to the old system, the new system (equations 2.6 and 2.7) has separate units for growing pigs and gestating sows (FU<sub>gp</sub> and FU<sub>gs</sub>, respectively). With the new system, the energy value of grains has increased, while that of protein supplements has decreased (Oresanya, 2005).

#### **2.8.4 Comparison of net energy systems**

Several equations for prediction of NE of feeds are available (Schiemann *et al.*, 1972; Just, 1982; Noblet *et al.*, 1994a; CVB, 1994). All published NE systems for pigs combine the utilization of ME for maintenance and for growth (Just, 1982; Noblet *et al.*, 1994a; 1994b). There is a good correlation in the estimated NE values and ranking of major feed ingredients between the French and the Dutch NE system (de Lange and Birkett, 2004). Noblet and van Milgen (2004) who compared other NE systems to the French system indicated the NE Schiemann, NE Just, and NE Dutch are approximately 94, 83, and 96 % of the NE French, respectively, for several diets. These average differences are mainly due to differences in estimates of fasting heat production and diet composition (Noblet, 2000). In the Danish system, there is difficulty in implementing the *in vitro* digestion methods and also, this approach assumes that energy is used exclusively for ATP production – which is not the case for growing pigs (Noblet, 2000). The system proposed by Noblet *et al.* (1994a) is based on a large set of measurements and the results have been validated in some trials (Le Bellego *et al.*, 2001; van Milgen *et al.*, 2001). Equations 3, 4, and 5 of Noblet *et al.* (1994a) enables the calculation of NE from DE. In North America, DE values for majority of feedstuffs are available and most nutritionists utilize the DE in diet formulation.

#### **2.9 DISTILLERS' DRIED GRAINS WITH SOLUBLES IN SWINE DIETS**

Distillers dried grains with solubles (DDGS) is the main co-product after fermentation of starch in ethanol production, which is now being used as a feedstuff in

swine diets (Stein *et al.*, 2006). In the United States and Eastern Canada, corn is the main grain used for ethanol production, therefore corn DDGS is primarily produced. However, the cooler climate of Western Canada is not suitable for growing corn grain (Boila and Ingalls, 1995). Thus, the Western Canadian ethanol industry utilizes wheat or wheat blended with corn to meet the demands of legislation requiring the use of ethanol-blended gasoline.

Wheat DDGS has a higher content of gross energy, a higher protein and fibre content and a drastically reduced starch content compared to wheat grain (Nyachoti *et al.*, 2005; Widyaratne and Zijlstra, 2007; Cozannet *et al.*, 2010). This nutritional profile provides an opportunity to use DDGS as a protein feedstuff in livestock feeding to mitigate feed cost. Distillers dried grains with solubles contains high fibre, which may have a negative effect on its nutritive value for swine (Emiola *et al.*, 2009). Previous studies with DDGS (Nyachoti *et al.*, 2005; Widyaratne and Zijlstra, 2007; Lan *et al.*, 2007) have shown that nutrient digestibilities were generally lower for DDGS than for its corresponding grains possibly due to the high fiber content. Thacker, (2006) reported that BW gain, feed intake and nutrient digestibility decreased with increasing level of wheat DDGS when fed to growing-finishing pigs. Inclusion of corn DDGS up to 30% in growing-finishing pig diets resulted in a decrease in growth rate (Cromwell *et al.*, 1993; Whitney *et al.*, 2006). However, Cook *et al.* (2005) and DeDecker *et al.* (2005) observed no effect when corn DDGS was included up to 30%. The inconsistent effects of DDGS on growth performance of growing-finishing pigs may be due to the variation in the chemical composition of the DDGS samples among studies.

Dietary fiber content has direct effects on digestive physiology (Larsen *et al.*, 1994; Eggum, 1995). Before reaching the fermentation site in the pigs, the cecum and colon, fiber through its physicochemical properties (water-holding, cation-exchange, adsorption and gel-forming properties) will exert diverse physiological actions along the gastrointestinal tract. The extent to which this occurs depends on the chemical nature of fiber (source and treatment), the way in which it is chemically associated with other compounds, the concentration and feeding level, the physiological state of the animal and the transit time in the gut (Dierick *et al.*, 1989).

### **2.9.1. Concentration of energy in DDGS for growing pigs**

Spiehs *et al.* (2002) reported DE values ranging from 3,879 to 4,084 kcal/kg DM for corn DDGS. Nyachoti *et al.* (2005) found that DE content of wheat DDGS averaged 3,225 kcal/kg DM. A similar DE value of 3,488 kcal/kg DM was reported by Cozannet *et al.* (2010) for wheat DDGS. In an experiment to evaluate the nutritional value of DDGS derived from corn, wheat, and a blend of wheat and corn, Widyaratne and Zijlstra, (2007) found that total tract DE was higher for corn DDGS (4,292 kcal/kg DM) than wheat-corn DDGS (4,038 kcal/kg DM), wheat DDGS (4,019 kcal/kg DM) and wheat samples (3,807 kcal/kg DM). For corn DDGS obtained from 10 sources, Stein *et al.* (2006) reported DE values ranging from 3,382 to 3,845 kcal/kg DM and Pedersen *et al.* (2007) reported DE values ranging from 3,947 to 4,593 kcal/kg DM. Wheat DDGS has been reported to contain 3,273 kcal of ME/kg DM (Cozannet *et al.*, 2010). Pedersen *et al.* (2007) reported a higher average value of 3,897 kcal of ME/kg DM for corn DDGS. Cozannet *et al.*

(2010) reported a NE value of 2,133 kcal/kg DM for wheat DDGS. Net energy of wheat-corn DDGS has not been determined.

## **2.10 CONCLUSION**

There is increased use of DDGS in swine diets as a result of the growth of the ethanol industry. The NE system is believed to be more accurate in expressing the energy value of a feedstuff than the DE and ME systems, because DE and ME systems tend to overestimate the energy value of protein and fiber-rich feedstuffs and underestimate the energy value of fat (Noblet *et al.*, 1994a). The NE of wheat-corn DDGS is yet to be determined and it is necessary to measure its NE content to more accurately express its energy value.

### 3.0 MANUSCRIPT I

#### GROWTH AND PHYSIOLOGICAL RESPONSES OF GROWING PIGS TO WHEAT-CORN DISTILLERS DRIED GRAINS WITH SOLUBLES

**3.1 ABSTRACT:** Gaining a detailed knowledge on the impact of a feedstuff on pig growth and physiological responses is critical for its effective utilization in swine nutrition. Thus, the purpose of this study was to investigate the effect of distillers dried grains with solubles derived from co-fermentation of wheat and corn (**wcDDGS**) on performance, carcass and visceral organ weights, whole body O<sub>2</sub> consumption, and heat production (**HP**) in growing barrows. The experimental diets were: (A) corn-soybean meal diet (**Control**), (B) Control + 15% wcDDGS, and (C) Control + 30% wcDDGS. All diets were formulated to meet the NRC (1998) nutrient specifications for 20 to 50 kg pigs. In Exp. 1, 48 pair-housed pigs of average initial BW  $18.6 \pm 1.5$  kg (mean  $\pm$  SD) were allotted based on BW to the 3 diets (n = 8). Pigs had free access to water and feed for a 28-d period during which ADG and ADFI were monitored weekly. Thereafter, 1 pig/pen was killed to measure carcass and visceral organ weights. Overall, wcDDGS linearly decreased ( $P < 0.05$ ) ADFI and ADG but had no effect on G:F ( $P > 0.10$ ). The ADFI was 1.55, 1.45 and 1.36 kg/d for diets A, B and C, respectively; corresponding values for ADG were 0.79, 0.75 and 0.67 kg/d. A linear decline ( $P = 0.01$ ) in eviscerated hot carcass weight was observed as dietary wcDDGS increased, specifically, pigs fed 30% wcDDGS had 11.2% lower carcass weight relative to those fed the control diet. However, diet had no effect ( $P > 0.10$ ) on the weights of any of the visceral organs. In

Exp. 2, 18 pigs of average initial BW of  $20.4 \pm 2.4$  kg (mean  $\pm$  SD), individually housed in metabolism crates were fed the 3 diets ( $n = 6$ ) at  $550$  kcal ME  $\text{kg BW}^{-0.60} \text{d}^{-1}$  for a 16-d period followed by measurement of  $\text{O}_2$  consumption and  $\text{CO}_2$  production over a 24-h period using an indirect calorimeter system. There was no effect of diet ( $P > 0.10$ ) on whole body  $\text{O}_2$  consumption and  $\text{CO}_2$  production and HP. In conclusion, increasing wcDDGS content in growing pig diets linearly reduced ADFI, ADG, and eviscerated hot carcass weight but had no effect on G:F, visceral organ weights and heat production.

**Key Words:** growth performance, heat production, oxygen consumption, pig, visceral organ weights, wheat-corn DDGS

### 3.2 INTRODUCTION

With the rapid growth of the ethanol industry, distillers dried grains with solubles (DDGS), the main co-product after fermentation of starch has become increasingly available as a feed ingredient for swine feeds. In the United States, corn is the main grain used for ethanol production, however, in Western Canada, wheat is used alone or in combination with corn as feedstock for ethanol production (Cozannet *et al.*, 2010). There are considerable published data on the performance of pigs fed corn DDGS (Whitney *et al.*, 2006; Xu *et al.*, 2010) and wheat DDGS (Thacker, 2006; Emiola *et al.*, 2009). However, there appears to be a lack of information on growth performance of pigs fed DDGS obtained from co-fermentation of wheat and corn (wcDDGS).

Distillers dried grains with solubles has higher fibre content than its parent grain due to the removal of starch during the fermentation process (Weigel *et al.*, 1997;

Widyaratne and Zijlstra, 2007). Dietary fiber has been reported to increase the length and weight of the gastrointestinal tracts in pigs (Anugwa *et al.*, 1989) and broiler chickens (Jorgensen *et al.*, 1995). Differences in visceral organ weights are highly correlated with differences in heat production (**HP**) in animals (Koong *et al.*, 1985). The relationship between visceral organ size and HP has important implications for food animal production, because nutrients are diverted from the growth of edible carcass parts in favour of visceral organ development. Information on visceral organ weights, whole-body oxygen (**O<sub>2</sub>**) consumption, and HP in pigs fed diets containing DDGS is not known.

We hypothesised that feeding up to 30% wcDDGS to growing pigs will reduce growth performance, increase visceral organ weights and whole-body O<sub>2</sub> consumption and heat production. Therefore, the objective of this study was to determine the effect of wcDDGS on growth performance, carcass and visceral organ weights and on whole body O<sub>2</sub> consumption and heat production of growing pigs.

### 3.3 MATERIALS AND METHODS

All experimental procedures were reviewed and approved by the University of Manitoba Animal Care Protocol Management and Review Committee and pigs were cared for according to the guidelines of the Canadian Council on Animal Care (CCAC, 2009).

Wheat-corn DDGS (1:1, wt/wt) was obtained from Husky Energy ethanol plant in Minnedosa, MB, Canada. The experimental diets were: (A) corn-soybean meal diet (**Control**), (B) 15% wcDDGS diet, and (C) 30% wcDDGS diet (Table 3.1). Diets were

formulated to meet or exceed NRC (1998) nutrient specifications for pigs in the BW range 20 to 50 kg. All diets were pelleted. The pigs used in both experiments were Genesis (Yorkshire-Landrace female  $\times$  Duroc male) obtained from the Glenlea Swine Research Unit, University of Manitoba.

**Table 3.1.** Ingredient, calculated, and analysed compositions of the experimental diets<sup>1</sup>

Item	Control	15% DDGS	30% DDGS
Ingredient, %			
Corn	67.9	61.4	54.8
Soybean meal (45%)	27.2	18.7	10.0
wcDDGS <sup>2</sup>	0.0	15.0	30.0
Soybean oil	1.3	1.3	1.7
Salt	0.5	0.5	0.5
Monocalcium phosphate	0.96	0.7	0.52
Limestone	1.05	1.23	1.4
HCL-Lys	0.06	0.22	0.41
DL-Met	0.01	0.0	0.0
L-Thr	0.01	0.0	0.0
Vitamin-mineral premix <sup>3</sup>	1.0	1.0	1.0
Calculated provisions			
ME, kcal/kg	3,294	3,265	3,265
SID Lys/ME, g/mcal <sup>4</sup>	2.6	2.6	2.6
CP, %	18.2	18.2	18.1
SID Lys, %	0.87	0.86	0.86
Analysed composition			
CP, %	17.6	17.4	17.8
DM, %	87.8	88.7	88.6
Gross energy, kcal/kg	3,899	4,002	4,135
Ash, %	4.3	4.3	4.5
NDF, %	9.3	10.5	18.8
ADF, %	3.9	5.4	6.3

<sup>1</sup>as fed basis

<sup>2</sup>wcDDGS = Wheat-corn distillers dried grains with solubles (1:1, wt:wt). Analysed composition of wcDDGS: CP = 29.1%, DM = 90.4%, Gross energy = 4,785 kcal/kg, Ash = 4.8%, NDF = 36.5%, ADF = 16.7%, Ether extract = 9.2%, Starch = 0.9%.

<sup>3</sup>Supplied the following per kg of finished feed: vitamin A, 5,000 IU; vitamin D, 300 IU; vitamin E, 40 IU; vitamin K, 4 mg; choline, 350 mg; pantothenic acid, 14 mg; riboflavin, 7 mg; folic acid, 2 mg; niacin, 21 mg; thiamin, 1 mg; vitamin B6, 4.5 mg; biotin, 0.10 mg; vitamin B12, 0.025 mg, Cu, 10 mg; Zn, 60 mg; Fe, 120 mg; Mn, 10 mg; I, 0.4 mg; Se, 0.3 mg.

<sup>4</sup>SID = standardized ileal digestible.

### ***3.3.1 Growth Performance and Organ Weight Study (Exp. 1)***

Forty-eight barrows with an average initial BW of  $18.6 \pm 1.5$  kg (mean  $\pm$  SD) were housed 2 pigs per pen on the basis of BW. Dietary treatments were assigned to pens ( $n = 8$ ) in a completely randomised design. Pigs had unlimited access to feed and water throughout the duration of the experiment. Body weight and feed consumption were monitored weekly. Room temperature was maintained between 22 and 24°C throughout the 28-d experimental period. Thereafter, pigs were slaughtered over a 4-d period. On each d of slaughter, pigs were weighed, anaesthetized by an intramuscular injection of ketamin:xylazine (20:2mg/kg; Bimeda-MTC Animal Health Inc., Cambridge, ON, Canada) and killed by an intravenous injection of sodium pentobarbital (50 mg/kg of BW; Bimeda-MTC Animal Health Inc., Cambridge, ON, Canada). The carcass was split down the midline from the groin to the chest cavity and the visceral organs were removed. The carcass, visceral organs (stomach, intestine, liver, lungs, heart, kidneys, and spleen), and blood were weighed, and processed separately. The digestive tracts were separated from other organs, emptied of digesta and weighed.

### 3.3.2 Indirect Calorimetry Study (Exp. 2)

Eighteen barrows with an average initial BW of  $20.4 \pm 2.4$  kg (mean  $\pm$  SD) were obtained from the Gleanlea Swine Research Unit, University of Manitoba. Pigs were individually housed in adjustable metabolism crates (1.8 x 0.6 m) with smooth transparent plastic sides and plastic-covered expanded metal sheet flooring in a temperature-controlled room (23 to 24°C). The study was conducted in two consecutive periods (9 pigs per period) using the same facility and similar experimental conditions and procedures because only 3 respiration chambers were available for the present study. Each period lasted for 25 d. In each period, pigs were allocated to the 3 diets in a completely randomized design to give three pigs per diet. During the first 16 d, pigs were housed in metabolism crates and were fed their respective diets at  $550 \text{ kcal ME} \cdot \text{kg BW}^{-0.60} \cdot \text{d}^{-1}$ , which was close to *ad libitum* intake (Noblet *et al.*, 1994a) and based on BW on d 1, 5, 10, and 15. Throughout the experiment, pigs were fed once daily at 0830 h and had free access to water.

On d 17, three pigs were transferred to the respiration chambers for a 24-h heat measurement. The second and third sets of three pigs were transferred to the respiration chambers on d 20 and 23, respectively. On each d, pigs were transferred into the respiration chambers within 1 h of finishing their daily feed allocation. Oxygen consumption and CO<sub>2</sub> production were monitored over a 24-h period. Water was freely available in the chambers and urine voided during the 24-h period was collected, weighed, sub-sampled and stored at -20°C until required for N analysis. Pigs were kept at

23 to 24°C and personnel movement around the chambers was limited to avoid disturbances.

### ***3.3.3 Sample Preparation and Chemical Analysis***

Urine samples were thawed and pooled for each pig and sieved through cotton gauze and filtered through glass wool. The urinary N content was determined by the combustion method (method 990.03; AOAC, 1990) using a Leco N analyzer (Model CNC-2000; Leco Corporation, St. Joseph, MI) and EDTA as a calibration standard. Diet samples were analysed for DM, ash, GE, CP, NDF, and ADF. Dry matter and ash were determined according to AOAC (1990; 942.05) and GE was determined using a Parr adiabatic O<sub>2</sub> bomb calorimeter. Crude protein (N x 6.25) was determined by the combustion method (AOAC, 1990; method 990.03) as described above. The ADF and NDF contents were determined according to the method of Goering and Van Soest (1970).

### ***3.3.4 Calculations and Statistical Analysis***

Heat production was calculated as described by Brouwer (1965), excluding the correction for CH<sub>4</sub> production:

$$HP = 3.87 \times O_2 + 1.20 \times CO_2 - 1.43 \times \text{urinary N} \dots\dots\dots [3.1]$$

Where HP = heat production (kcal), O<sub>2</sub> = oxygen consumption (L), CO<sub>2</sub> = carbon dioxide production (L), and urinary N (g)

The Respiratory quotient (**RQ**) was calculated as the ratio of CO<sub>2</sub> production to O<sub>2</sub> consumption.

Data were subjected to analysis of variance using the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). For Exp. 2, effects of diet and period were included in the statistical model. Orthogonal polynomials were used to determine linear effect of diet and a probability of  $P < 0.05$  was considered significant.

### **3.4 RESULTS AND DISCUSSION**

#### ***3.4.1 Chemical Composition of wcDDGS and Experimental Diets***

The CP content of the wcDDGS used in this study was 29.1% (as-fed basis; Table 3.1). This value is lower than that (39.0%) reported by Widyaratne and Zijlstra (2007) for wcDDGS. Difference in CP content in wcDDGS may be due to difference in blending ratio of wheat and corn (the ratio of wheat to corn in wcDDGS used in this study was 1:1 as opposed to 4:1 used by Widyaratne and Zijlstra (2007)), differences in source or cultivars of the original cereal grain or differences in the fermentation process (Nyachoti *et al.*, 2005). This fact is also reflected in fibre content. In the current study, NDF and ADF of wcDDGS were 36.5% and 16.7%, respectively. Widyaratne and Zijlstra (2007) reported a value of 28.2% and 17.9% for NDF and ADF, respectively.

#### ***3.4.2 Growth performance and organ weight study (Exp. 1)***

The effects of dietary treatment on ADG, ADFI, and G:F of pigs are presented in Table 3.2. Overall ADFI linearly decreased ( $P < 0.05$ ) with increasing wcDDGS inclusion. Similarly, Widyaratne and Zijlstra (2007) observed a lower ADFI in pigs fed

wcDDGS-containing diets compared with those fed a wheat-based control diet. Dietary fiber can result in reduced feed intake due to an increase in dietary bulkiness and water-holding capacity which may cause early satiety (Kyriazakis and Emmans, 1990, 1995). Dietary fiber in DDGS consists mainly of insoluble dietary fiber (Urriola *et al.*, 2010) that may increase the bulkiness and water-holding capacity of the diet (Potkins *et al.*, 1991; Cherbut *et al.*, 1994). Inclusion of wcDDGS may also have altered taste or smell of the diet progressively reducing diet palatability (Whitney and Shurson, 2004; Avelar *et al.*, 2010). Also, in growing pigs, a diet with low fibre content will be preferred as this results in maximum intake of available nutrients and energy.

Overall, ADG linearly decreased ( $P < 0.05$ ) with wcDDGS inclusion (Table 3.2). This observation is in agreement with the results of Cromwell *et al.* (1993), Whitney *et al.* (2006) and Linneen *et al.* (2008) who reported that inclusion of up to 30% corn DDGS in growing-finishing pig diets resulted in a decrease in growth rate. The daily intake of digestible Lys was lower for pigs fed diets with wcDDGS than those fed the control diet, mainly due to a lower feed intake, because the diets were formulated to an equal digestible lysine content. The reduced digestible Lys intake may have constrained protein accretion of pigs fed diets with wcDDGS (Friesen *et al.*, 1994). Throughout the entire study, there was no effect of dietary treatment on G:F. Also, in wk 1, 3, and 4, there was no effect of diet on ADG. This is an indication that pigs consuming diets containing wcDDGS converted feed to gain similarly to those consuming the control diet.

There was a linear reduction ( $P < 0.05$ ) in slaughter weight and eviscerated carcass weight with increasing dietary wcDDGS inclusion (Table 3.3). Similarly,

Cespedes (2009) found that live BW was lower in growing pigs fed corn DDGS diet than for pigs fed the corn-soybean control diet. Also, hot carcass weight and slaughter weight were greater in pigs fed the control diet than for pigs fed the corn DDGS diet. The reduction in slaughter weight and eviscerated carcass weight of pigs fed diets with wcDDGS inclusion is attributable to reduced nutrient digestibility. Digestibility studies by Widyaratne and Zijlstra (2007) using wcDDGS showed a reduction in apparent total tract digestibility of GE and CP with 40% dietary inclusion of wcDDGS. Reduction in slaughter weight and carcass weight may be the result of lower feed intake observed in the pigs fed wcDDGS diets compared with the control diet. Whitney *et al.* (2006) found that including 20 or 30% corn DDGS in the diet of growing pigs resulted in similar feed intakes but lower growth rate compared with 0 or 10% inclusion. Pond *et al.* (1989) found that daily gain and daily feed intake were depressed by high levels of fiber. Also, Nyachoti *et al.* (2000) found that final BW was lower in pigs fed a diet with alfalfa meal compared with diets with lower dietary fiber content.

Higher weight of digestive organs was expected for pigs fed diets containing wcDDGS compared with those fed the control diet because of the higher fiber content. It is well known that increase in dietary fiber results in an increase in the empty weight of the gut (Dierick *et al.*, 1989). In the large intestine, undigested feed components and endogenous secretions are fermented by micro-organisms and short chain fatty acids are produced and absorbed which results in an increase in gut weight. In the present study, there was no effect of wcDDGS on weight of visceral organs (g/kg of empty BW). Similarly, Cespedes (2009) found that organ weights were not different among treatments

when high protein corn DDGS, conventional corn DDGS, and a corn-soybean control diet were fed to growing pigs. This observation also agrees with Hochstetler *et al.* (1959), who reported that dressing percentage was not affected by feeding high fiber diets containing cellulose, oat bran, or alfalfa. Gargallo and Zimmerman (1981) reported that different levels of dietary sunflower hulls did not have an effect on the weight of empty intestines. Contrary to this observation, increased intestinal mass has been reported when high fibre diets are fed to pigs (Kass *et al.*, 1980). Also, Nyachoti *et al.* (2000) found that pigs fed high fiber diets (NDF: 19.3-21.4%) had heavier visceral organs relative to empty BW compared with those fed the control diet. In the latter 2 studies, the increase in cecum and colon weights of pigs fed high fiber diets might have resulted from the distension and lengthening of the hindgut where most of the fibrous feed components are digested via microbial fermentation (Pond *et al.*, 1988; Jorgensen *et al.*, 1996). Also, the increase in visceral organ weights in the above studies was mainly due to the response of digestive organs, including the intestines, liver and kidneys, to an increased feed intake (Bikker *et al.*, 1995). In the present study, pigs fed wDDGS diets had lower feed intake compared with those fed the corn-soybean meal control diet. Lowered feed intake will reduce substrate availability for microbial fermentation in the hindgut which may contribute to the lack of a diet effect on digestive organ weights.

As reviewed by Wenk (2001), pigs do not have the same ability to utilize dietary fibre from different sources and therefore responses of pigs to high-fibre diets also depend on the type of dietary fibre. Indeed, Michel and Rerat (1998) reported that a diet containing 10% sugar beet fibre but not wheat bran increased hindgut fermentation and

therefore the absorption of VFA. Therefore, it is not only the level of dietary fibre that is important but also the type and source may play a significant role in digestion and absorption. For DDGS, various factors including fermentation procedures, type of additives and the amount of solubles blended with distillers dried grains may affect the response of pigs relating to hindgut fermentation and consequently, the weights of digestive organs.

**Table 3.2.** Growth performance of growing pigs fed wheat-corn DDGS-containing diets<sup>1</sup> (Exp 1)

Item	Control	15% DDGS	30% DDGS	SEM	P-value <sup>2</sup>
Initial BW, kg	18.60	18.61	18.55	0.38	0.925
Final BW, kg	39.88	39.56	37.36	0.79	0.029
ADFI, kg/d					
d 0 to 7	1.15	0.96	0.95	0.04	0.003
d 7 to 14	1.54	1.36	1.30	0.05	0.002
d 14 to 21	1.70	1.71	1.53	0.05	0.038
d 21 to 28	1.83	1.78	1.66	0.06	0.040
Overall	1.55	1.45	1.36	0.03	0.001
ADG, kg/d					
d 0 to 7	0.68	0.55	0.58	0.06	0.228
d 7 to 14	0.79	0.74	0.62	0.04	0.003
d 14 to 21	0.83	0.85	0.76	0.04	0.274
d 21 to 28	0.85	0.85	0.73	0.07	0.202
Overall	0.79	0.75	0.67	0.02	0.001
G:F					
d 0 to 7	0.59	0.57	0.61	0.04	0.717
d 7 to 14	0.51	0.55	0.48	0.03	0.438
d 14 to 21	0.49	0.49	0.50	0.02	0.627
d 21 to 28	0.46	0.48	0.44	0.04	0.619
Overall	0.51	0.52	0.49	0.01	0.303

<sup>1</sup>n = 8.

<sup>2</sup>Linear P-value.

There was a linear reduction ( $P < 0.05$ ) in final BW and eviscerated carcass weight with increasing dietary wcDDGS inclusion (Table 3.4). Similarly, Stein (2009) found that live BW was lower in growing pigs fed conventional DDGS and DDGS from uncooked corn diets than for pigs fed the corn-soybean basal diets. Also, hot carcass weight and slaughter weight were greater in pigs fed the basal diet than for pigs fed DDGS from uncooked corn. The reduction in slaughter weight and eviscerated carcass weight of pigs fed diets with wcDDGS inclusion is attributable to reduced nutrient digestibility. Digestibility study by Widyaratne and Zijlstra (2007) showed reduction in ATTD of GE and CP as dietary wcDDGS inclusion increased. Reduction in slaughter weight and carcass weight have been the result of lower feed intake observed in the pigs fed wcDDGS diets compared with the basal diet. Whitney *et al.* (2006) found that including 20 or 30% corn DDGS in the diet of growing pigs resulted in similar feed intakes but lower growth rate compared with 0 or 10% inclusion. Pond *et al.* (1989) found that daily gain and daily feed intake were depressed by high levels of fiber. Also, Nyachoti *et al.* (2000) found that final BW was lower in pigs fed diet with alfalfa meal compared with diets with lower dietary fiber content. Animals that reach satiety physically and nutritionally are less stressed and, therefore, physical activity may be reduced (Rijnen *et al.*, 2001) which may result in higher nutrient retention in the carcass.

There was no effect of wcDDGS on weight of visceral organs (g/kg of empty BW). Similarly, Stein (2009) found that, organ weights were not different among treatments when high protein corn DDGS, conventional DDGS and DDGS from uncooked corn were included in corn-soybean basal diet. This observation also agrees

with Hochstetler *et al.* (1959), who reported that the dressing percentage was not affected by feeding high fiber diets containing cellulose, oat bran, or alfalfa diets. Gargallo and Zimmerman (1981) reported that different levels of dietary sunflower hulls did not have an effect on the weight of empty intestines. Contrary to this observation, increased intestinal mass has been reported when high fibre diets are fed to pigs (Kass *et al.*, 1980). Also, Nyachoti *et al.* (2000) found that pigs fed high fiber diets (NDF: 19.3-21.4%) had heavier visceral organs relative to empty body weight compared with those fed the control diet. In these studies, the increase in the weights of cecum and colon of pigs fed high fiber diets might have resulted from the distension and lengthening of the hindgut where most of the fibrous feed components are digested via microbial fermentation (Pond *et al.*, 1988; Jorgensen *et al.*, 1996). Also, the increase in weight of visceral organ in the above studies was predominantly the response of digestive organs, including the intestines, liver and kidneys, to an increased feed intake, and representing increased metabolic activity (Bikker *et al.*, 1995). Koong *et al.* (1983) and Rao and McCracken (1992) reported increased weights of metabolically active organs when high fibre diets were fed to pigs. In the present study, pigs fed wCDDGS diets had lower feed intake compared with those fed the corn-soybean diet. Less feed intake will make less substrate available for microbial fermentation in the hindgut and thereby contributing to the lack of effect of diet on weights of digestive organs. As reviewed by Wenk (2001), pigs do not have the same ability to utilize dietary fibre from different sources. Responses of pigs to high-fibre diets also depend on type of dietary fibre. For example, Michel and Rerat (1998) fed 10% wheat bran and 10% sugar beet fibre to pigs. Sugar beet fibre increased hindgut fermentation but wheat bran did not. Therefore, it is not only the level of dietary

fibre that is important but the type or the source also plays a significant role in digestion and absorption.

**Table 3.3.** Carcass and visceral organ weights of growing pigs fed wheat-corn DDGS-containing diets<sup>1</sup> (Exp. 1)

Item	Control	15% DDGS	30% DDGS	SEM	P-value <sup>2</sup>
Slaughter weight, kg	39.0	37.8	35.5	0.93	0.017
Eviscerated carcass, kg	31.0	29.8	27.8	0.82	0.012
Total viscera, g/kg EBW <sup>3,4</sup>	116.8	117.0	121.0	3.28	0.381
Small intestine, g/kg EBW	34.8	35.2	37.1	1.26	0.225
Large intestine, g/kg EBW <sup>5</sup>	19.8	19.6	20.7	1.22	0.615
Stomach, g/kg EBW	7.9	7.7	8.2	0.42	0.643

<sup>1</sup>n = 8.

<sup>2</sup>Linear P-value.

<sup>3</sup>Total visceral weight = sum of liver, lungs, kidneys, spleen, heart, emptied stomach, emptied small intestine, emptied colon and emptied cecum.

<sup>4</sup>Empty BW = weight of carcass + total visceral weight + weight of blood.

<sup>5</sup>Sum of colon and cecum.

### 3.4.3 Indirect calorimetry study (Exp. 2)

Oxygen consumption and CO<sub>2</sub> production were not affected by wDDGS inclusion. However, there was a linear decrease ( $P < 0.05$ ) in RQ with wDDGS inclusion (Table 3.4). Reduction in RQ observed in pigs fed wDDGS diets is an indication of higher O<sub>2</sub> consumption relative to CO<sub>2</sub> production. It shows that the corn-soybean diet (higher in starch) compared to the wDDGS diets (higher in fibre) required less O<sub>2</sub> for oxidation. This is consistent with the results of Noblet *et al.* (1994b) who observed that RQ increased when starch or sucrose was added and reduced when more fibre was included in the diet.

**Table 3.4.** Oxygen consumption, carbon dioxide production, respiratory quotient and heat production in growing pigs fed wheat-corn DDGS-containing diets<sup>1</sup> (Exp. 2)

Item	Control	15% wcDDGS	30% wcDDGS	SEM	P-value <sup>2</sup>
Initial BW, kg	20.4	20.6	20.2	1.03	0.910
Final BW, kg	31.8	31.5	30.5	1.70	0.608
Feed intake kg/d	1.20	1.07	1.02	0.07	0.095
O <sub>2</sub> , l/d	444	455	415	19.11	0.290
CO <sub>2</sub> , l/d	431	430	379	21.27	0.116
RQ <sup>3</sup>	0.96	0.94	0.91	0.01	0.005
HP, kcal BW <sup>-0.6</sup> d <sup>-1</sup>	298	308	287	14.42	0.581

<sup>1</sup>n = 6

<sup>2</sup>Linear p-value

<sup>3</sup>RQ = respiratory quotient represents the ratio of carbon dioxide output to oxygen intake (l:l, v/v)

Heat production was not affected by dietary wDDGS inclusion. However, the observed values (298, 308 and 287 kcal/kgBW<sup>-0.6</sup> for diets containing 0, 15, and 30% wDDGS, respectively) were similar to the value of 320 kcal/kgBW<sup>-0.6</sup> reported by Noblet *et al.* (1994b) for growing pigs. Previous studies showed greater heat production for fibre-rich diets (Dierick *et al.*, 1989; Noblet *et al.*, 1994a), so we expected higher HP for pigs fed diets with wDDGS. However, total HP includes HP associated with feed consumption, maintenance, thermal regulation and physical activity (NRC, 1998). Feed intake (preceding heat measurement) in pigs fed diets with wDDGS inclusion tended to be lower ( $P = 0.10$ ) compared to those fed the control diet (Table 3.4). Fuller and Boyne (1972) reported that the level of feed intake could have a large impact on total HP in pigs. Therefore, one may argue that the effect of wDDGS inclusion on HP was confounded by the reduction in feed intake for pigs fed diets with wDDGS inclusion. Physical activity can represent up to 10% of the total HP in growing pigs (Van Milgen *et al.*, 1998). The energy cost of physical activity can be divided into energy cost of sitting, standing, sitting up, standing up, and walking (Kelley *et al.*, 1978). Van Milgen *et al.* (1998) reported that positional movements as well as metabolic efficiency of movement contribute more to the energetic cost of physical activity than the act of rising itself. It has been demonstrated that high fibre diets for growing pigs decreased physical activity and consequently heat production (Shrama *et al.*, 1995). Although physical activity was limited in the respiratory chambers by limiting human distraction, it is possible that the lack of effect of diet on heat production observed in this study was due to a lower physical activity when pigs are fed high fibre diets.

The results of this study demonstrate that including up to 30% wDDGS in a corn soybean meal diet reduced growth performance and hot carcass weight but had no effect on visceral organ weights. Also, wDDGS had no effect on whole body O<sub>2</sub> consumption and heat production in growing pigs. The reduction in hot carcass weight with increasing wDDGS inclusion is attributable to a reduction in nutrient digestibility while the lack of effect on visceral organ weights, HP and whole-body O<sub>2</sub> consumption with wDDGS inclusion is attributable to the reduced feed intake observed in pigs fed diets containing 15 and 30% wDDGS. Consequently, up to 15% wDDGS can be fed to growing pigs with limited reduction in growth performance and carcass weight. Further study on the determination of rates of production and absorption of VFA in the cecum and colon of growing pigs fed diets containing wDDGS as it relates to the overall energy metabolism and contribution to maintenance energy requirement may be necessary.

## 4.0 MANUSCRIPT II

### NET ENERGY OF WHEAT-CORN DISTILLERS DRIED GRAINS WITH SOLUBLES AS DETERMINED BY INDIRECT CALORIMETRY,

### COMPARATIVE SLAUGHTER, AND CHEMICAL COMPOSITION METHODS

**4.1 ABSTRACT:** The NE content of wheat-corn distillers dried grains with solubles (wcDDGS) fed to growing pigs was determined using the comparative slaughter (CS), indirect calorimetry (IC), and chemical composition (CH) methods. The experimental diets were a corn-soybean meal diet (Control), 15% wcDDGS diet, and 30% wcDDGS diet. In Exp. 1, 56 barrows (18.5 kg BW) were used to determine the NE value of wcDDGS using the CS method. Pigs were initially placed in 8 groups (n = 7) based on BW and then 1 pig/group (n = 8) killed to obtain baseline body composition. The remaining 48 pigs were housed in pairs and allotted to the 3 diets (n = 8). Pigs had free access to feed and water for a 28-d period after which 1 pig/pen was slaughtered to determine final body composition. Based on the CS method, NE values of 2,407, and 2,424 kcal/kg DM were obtained for wcDDGS included at 15% and 30%, respectively. In Exp. 2, 18 barrows (20.4 kg BW) were used to determine the NE value of wcDDGS using the IC and CH methods. Pigs were individually housed in metabolism crates and were fed the 3 diets (n = 6) at 550 kcal ME/kg BW/d for a 16-d period. Feces and urine were collected from d 11 to 16 followed by measurement of O<sub>2</sub> consumption, CO<sub>2</sub> production and urinary N over a 36-h period using an IC system. For the IC method, the NE values of 2,407, and 2,403 kcal/kg DM were obtained for wcDDGS included at 15%

and 30%, respectively; corresponding values for the CH method were 2,536 and 2,197 kcal/kg DM, respectively. Similar ( $P > 0.10$ ) NE values for diets and wcDDGS were obtained for CS, IC, and CH methods. In conclusion, the NE value of wcDDGS ranged from 2,367 to 2,416 kcal/kg DM, depending on the method used. As the values obtained using the various methods were not different, the average NE value for the wcDDGS evaluated was  $2,396 \pm 25.71$  kcal/kg DM.

**Key Words:** comparative slaughter, indirect calorimetry, NE, pigs, wheat-corn distillers dried grains with solubles.

## 4.2 INTRODUCTION

In North America, dietary energy is usually expressed as DE or ME. However, DE and ME systems tend to overestimate the energy value of fibrous and high protein feedstuffs and underestimate the energy value of ingredients that have high concentrations of starch or fat (Noblet *et al.*, 1994a). These deficiencies in measurement of dietary energy are very important to the economics of pig production and there is therefore an increasing interest in using a system based on NE. Research data in support of NE system is lacking in North America. The NE system allows for a more effective use of high fibre co-products such as distillers dried grains with solubles (**DDGS**) as feedstuffs for swine.

Determination of NE of feedstuffs has been done using the comparative slaughter (**CS**) (Noblet *et al.*, 1987; Kil *et al.*, 2011) and indirect calorimetry (**IC**) (Noblet *et al.*,

1994a; Hansen *et al.*, 2006) methods. However, the CS method is labour intensive, and requires a large number of animals. The IC method requires fewer animals, takes relatively short period of time and can be used for repeated measurement of energy balance. (van Milgen and Noblet, 2003). Net energy prediction equations based on chemical composition measurements of feeds and feedstuffs have also been suggested (Noblet *et al.*, 1994a). The equations have not been widely adopted in North America because nutritionists have not developed confidence in the coefficients incorporated in those systems. Inclusion level of DDGS may affect the NE because increasing dietary inclusion level of DDGS has been shown to reduce ATTD of energy in pigs (Urriola and Stein, 2010). Thus, the objectives of this study were to (1) determine the NE value of wheat-corn DDGS (**wcDDGS**) for growing pigs using the CS, IC, and CH methods (2) compare the CS, IC, and CH methods of NE determination (3) determine the effect of dietary inclusion level of wcDDGS on the NE value.

### 4.3 MATERIALS AND METHODS

All experimental procedures were reviewed and approved by the University of Manitoba Animal Care Protocol Management and Review Committee and pigs were cared for according to the guidelines of the Canadian Council on Animal Care (CCAC, 2009).

Wheat-corn DDGS was obtained from Husky Energy, Minnedosa, MB, Canada. The experimental diets were a corn-soybean meal-based diet (**Control**), 15% wcDDGS diet, and 30% wcDDGS diet (Table 3.1, Manuscript I). Diets were formulated to meet or exceed NRC (1998) nutrient specifications for pigs in the BW range 20 to 50 kg. All diets

were pelleted. The pigs used in both experiments were Genesus (Yorkshire-Landrace female × Duroc male) obtained from the Glenlea Swine Research Unit, University of Manitoba.

#### ***4.3.1 Exp. 1: Comparative Slaughter***

Fifty-six barrows with an average initial BW of  $18.5 \pm 1.5$  kg (mean  $\pm$  SD) were placed into 8 groups ( $n = 7$ ) based on BW. At the start of the experiment, 1 pig/group ( $n = 8$ ) was slaughtered to determine initial body composition. The remaining 48 pigs were housed in pairs and randomly assigned to the 3 dietary treatments for a 28-d period. Pigs had unlimited access to feed and water throughout the duration of the experiment and room temperature was maintained between 22 and 24°C. On d 29, 30, 31 and 32, one pig per pen was slaughtered (2 pigs per treatment on each d). On each d of slaughter, pigs were weighed, anaesthetized by an intramuscular injection of ketamin:xylazine (20:2 mg/kg; Bimeda-MTC Animal Health Inc., Cambridge, Ontario, Canada) and killed by an intravenous injection of sodium pentobarbital (50 mg/kg of BW; Bimeda-MTC Animal Health Inc.). The carcass was split down the midline from the groin to the chest cavity and the viscera were removed. Care was taken to ensure all blood was collected from each pig, and weighed. The carcass was split into two equal halves and the right half was weighed and stored at -20°C. The digestive tracts were separated from other organs, emptied of digesta and weighed. All samples of blood and visceral organs were frozen at -20°C until grinding and further analyses.

### 4.3.2 Exp. 2: Indirect calorimetry

Eighteen barrows with an average initial BW of  $20.4 \pm 2.4$  kg (mean  $\pm$  SD) were individually housed in adjustable metabolism crates (1.8 x 0.6 m) with smooth transparent plastic sides and plastic-covered expanded metal sheet flooring in a temperature-controlled room (23 to 24°C).

The study was conducted in 2 consecutive periods (9 pigs per period) using the same facility and similar experimental conditions and procedures because only 3 respiration chambers were available for this study. Each period lasted for 25 d and in each period, based on initial BW, pigs were allocated to the 3 diets in a completely randomized design to give 3 pigs per diet. During the first 16 d, pigs were housed in metabolism crates and fed their respective diets at  $550 \text{ kcal ME} \cdot \text{kg BW}^{-0.60} \cdot \text{d}^{-1}$  (Noblet *et al.*, 1994a) and based on BW on d 1, d 5, d 10, and d 15. Throughout the experiment, pigs were fed once daily at 0830 h and had free access to water. The first 10 d were for adaptation and the next 6 d were for collection of feces and urine to determine DE and ME. Collection of urine commenced on the morning of d 11 and ended on the morning of d 15. From d 11 to d 16, feces was collected once daily in the morning and stored at -20°C. Urine was also collected once daily in the morning (in jugs containing 10 mL of HCl to minimise N losses) and weighed. A sample (10% of the total weight) was obtained, strained through cotton gauze and glass wool, and stored at -20°C.

On d 17, three pigs were transferred to the respiration chambers for 36 h of heat production measurement. The second and third sets of three pigs were transferred to the respiration chambers on d 20 and 23, respectively. On each d, pigs were transferred into

the respiration chambers within 1 h of finishing their daily feed allocation to measure oxygen ( $O_2$ ) consumption and carbon dioxide ( $CO_2$ ) production over a 24-h period (fed state) followed by another 12-h measurement period (fasted state). Water was freely available in the chambers and urine voided during the 24-h and 12-h periods were collected separately, weighed, sub-sampled and stored at  $-20^\circ C$  until required for N analysis. Pigs were kept at 23 to  $24^\circ C$  and personnel movement around the chambers was limited to avoid disturbances.

#### ***4.3.3. Sample Preparation and Chemical Analyses***

Fecal samples were oven dried at  $50^\circ C$  over a 5-d period, and finely ground before chemical analysis. Urine samples from metabolism crates and IC chambers were thawed and pooled separately for each pig, sieved with cotton gauze and filtered with glass wool. Frozen carcasses were cut into pieces using an electric band saw in order to fit into the grinder (Model H600, Hobart Manufacturing Co. LTD, Toronto, ON, Canada) and were ground through a 24 mm die. After grinding, carcasses were mixed in a 20 kg mixer (Model A200, Hobart Manufacturing Co. LTD, Toronto, ON, Canada) to ensure even distribution and to facilitate sampling. After 10 min of mixing, approximately 1 kg of carcass was collected and re-ground through a 12 mm die. This was freeze-dried and finely ground for chemical analysis. The body organs and the empty gastrointestinal tract were ground in a meat grinder (OMAS LR 37050, OMCAN-Stoney Creek, Hamilton, ON, Canada) through a 22 mm die. Stomach, small intestine, colon, cecum and liver were ground together and was marked as 'intestine' while the remaining organs (lungs, spleen, kidneys and others) were ground together and was marked as 'miscellaneous'.

Approximately, 600 g sample was taken from the 'intestine' and 'miscellaneous' of each pig after thorough mixing and freeze-dried and samples were finely ground with a coffee grinder (Applica Consumer Products Inc., Miami Lakes, FL) for chemical analysis.

Diet and fecal DM was determined according to the AOAC (1990; method 925.09) by oven drying 5 g of sample at 105°C overnight. The DM content of body components (i.e. carcass, viscera and blood) was determined by freeze drying to a constant weight. The GE content of wcDDGS, diets, feces, carcass, viscera, blood and urine was measured using an adiabatic bomb calorimeter (model 6300, Parr Instrument, Moline, IL) which had been calibrated using benzoic acid as a standard. Nitrogen content in diets, feces, urine, and body components was determined using the combustion method (method 990.03; AOAC, 1990) using the LECO N analyser (model CNS-2000; LECO Corp., St. Joseph, MI) and CP was calculated as  $N \times 6.25$ . Crude fat in diet and ingredient samples was determined after hexane extraction (method 920.39; AOAC, 1990) in an extraction apparatus and in fecal samples after acidification with 9 N HCl followed by hexane extraction. Diets and feces were analysed for AIA according to McCarthy *et al.* (1974). Briefly, 10 g sample of feed or digesta was boiled for 30 min in 100 mL of 4M HCl, filtered through ashless filter paper (Whatman #41) with boiling water until free of acid and ashed for 6 h at 650°C. The ADF and NDF contents in diets were determined according to the method of Goering and Van Soest (1970) and ash content was determined according to AOAC (1990; method 942.05).

The urine samples collected from the metabolism crates were analysed for DM and GE. For the DM of urine, 1mL of each sample was mixed with 0.5 g of cellulose and

the weight of the resulting mixture recorded. The urine-cellulose mixtures together with samples of pure cellulose were dried in an oven at 50°C for 24 h. The GE was then determined on the dried urine-cellulose mixtures as described above and samples of pure cellulose, and the contents of the same in urine were calculated by the difference method (Fleisher *et al.*, 1981).

### **4.3.3 Calculations and Statistical Analyses**

#### **4.3.3.1 Exp. 1**

The total quantity of energy, protein, and lipids in each pig at slaughter was calculated from the sum of the energy, protein, and lipids in blood, viscera and carcass. Retention of energy, protein, and lipids was calculated as the difference between final quantity of energy, protein and lipids and the initial quantity of energy, protein, and lipids. The daily NE requirement for maintenance for each pig was calculated by multiplying the mean metabolic BW ( $\text{kg}^{0.6}$ ) by 179 kcal (Noblet *et al.*, 1994a). The mean metabolic BW of each pig was calculated as the average of the metabolic BW obtained weekly during the experimental period. The NE of each diet was calculated as the sum of the energy retained in the body and the total operational NE requirement for maintenance during the experimental period. The NE of wcDDGS at 15 and 30% inclusion levels were subsequently calculated using the difference method (Woyengo *et al.*, 2010).

#### **4.3.3.2 Exp. 2**

Heat production, fasting HP, retained energy, DM intake and NE were calculated using the following equations:

$$HP = 3.87 \times O_2 + 1.20 \times CO_2 - 1.43 \times \text{urinary N} \dots \dots \dots [4.1]$$

Where HP = heat production (kcal),  $O_2$  = oxygen consumption (L),  $CO_2$  = carbon dioxide production (L).

$$RE = ME - HP \dots \dots \dots [4.2]$$

Where RE = retained energy (kcal/d), ME = metabolizable energy intake, (kcal/d), HP = heat production (kcal/d).

$$DMI = \text{feed intake} \times \text{feed DM} \dots \dots \dots [4.3]$$

Where DMI = dry matter intake (kg), feed intake is in kg, and feed DM is in %.

$$NE = (RE + FHP) / DMI \dots \dots \dots [4.4]$$

Where NE = net energy (kcal/kg DM), RE = retained energy (kcal/d), FHP = fasting heat production (kcal/d), DMI = dry matter intake (kg).

Net energy was also calculated according to the following equations (Noblet *et al.* 1994a):

$$NE = 0.843 \times DE - 463 \dots \dots \dots [4.5]$$

$$NE = 0.703 \times DE + 1.58 \times EE + 0.47 \times ST - 0.97 \times CP - 0.98 \times CF \dots \dots \dots [4.6]$$

$$NE = 0.700 \times DE + 1.61 \times EE + 0.48 \times ST - 0.91 \times CP - 0.87 \times ADF \dots \dots \dots [4.7]$$

Where NE = net energy (kcal/kg DM), DE = digestible energy (kcal/kg DM), EE = ether extract (% DM), ST = starch (% DM), CF = crude fiber (% DM).

Digestible energy of wcDDGS at 15 and 30% inclusion levels were calculated using difference method (Woyengo *et al.*, 2010)

All data (Exp. 1 and 2) were subjected to the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). Effects of diet and period were included in the model for statistical analysis. The effect of period was not statistically significant ( $P = 0.21 - 0.71$ ); therefore, it was not included in the final model. Orthogonal polynomials were used to determine linear and quadratic effects of diet and a probability of  $P < 0.05$  was considered significant.

## 4.4 RESULTS

### 4.4.1 Exp. 1

The analysed composition of experimental diets and wcDDGS are presented in manuscript I. The analysed dietary CP values were similar to the calculated values. There was no effect of wcDDGS inclusion on RE and NE of diets. The RE values of diets containing 0, 15%, and 30% wcDDGS were 1,393, 1,351 and 1,305 kcal/kg, respectively; corresponding values for NE were 2,431, 2,427, and 2,429 kcal/kg, respectively. The NE content of wcDDGS included at 15% (2,407 kcal/kg DM) was not different ( $P > 0.10$ ) from the NE content of wcDDGS included at 30% (2,424 kcal/kg DM) (Table 4.1).

**Table 4.1.** Energy values of diets and wcDDGS by comparative slaughter method in growing pigs (Exp. 1)<sup>1</sup>

Item	Dietary treatment			SEM	P-value	
	Control	15% wcDDGS	30% wcDDGS		Linear	Quadratic
Empty BW, kg DM	13.14	12.56	11.90	0.410	0.043	0.930
Total protein, kg	6.34	6.17	5.71	0.182	0.023	0.526
Total lipid, kg	5.22	4.89	4.56	0.260	0.091	0.996
Total Energy, mcal	85.18	80.94	75.97	3.001	0.041	0.923
Total energy retained, mcal	53.26	53.00	44.05	4.450	0.158	0.434
Total ONE <sub>m</sub> , mcal <sup>2</sup>	39.6	38.7	37.8	0.70	0.076	0.964
Retained energy, kcal/kg DM <sup>3</sup>	1,393	1,351	1,305	67.61	0.370	0.980
NE of diets, kcal/kg DM <sup>4</sup>	2,431	2,427	2,429	68.58	0.985	0.976
NE of wcDDGS, kcal/kg DM <sup>5</sup>	-	2,407	2,424	323.13	0.159	-

<sup>1</sup>n = 8

<sup>2</sup>Total operational NE requirement for maintenance was calculated by multiplying the mean metabolic BW (kg<sup>0.6</sup>) of each pig by 179 kcal (Noblet *et al.*, 1994a) and the number of days on experiment.

<sup>3</sup>Retained energy over 28-d period = [(Total energy retained/28)]/DMI

<sup>4</sup>[Retained energy + ONE<sub>m</sub>]/DMI

<sup>5</sup>NE of wcDDGS was calculated using the difference method by subtracting the NE contribution of the basal diet from the NE of the diets containing 15% wcDDGS or 30% wcDDGS (Woyengo *et al.*, 2010).

#### 4.4.2 Exp. 2

Digestible energy and ME contents linearly decreased ( $P < 0.05$ ) with dietary inclusion of wcDDGS (Table 4.2). Heat production and fasting heat production were not affected by wcDDGS inclusion. The RE of diets containing 0, 15%, and 30% wcDDGS were 950, 787, and 888 kcal/kg, respectively and there was no difference ( $P > 0.10$ ) between them. The NE of diets containing 0, 15, and 30% wcDDGS were 2,586, 2,513 and 2,520 kcal/kg DM, respectively. The NE of wcDDGS included at 15% (2,407 kcal/kg DM) was not different ( $P > 0.10$ ) from the NE of wcDDGS included at 30% (2,403 kcal/kg DM). No effect of wcDDGS inclusion was observed on NE and efficiencies of NE (NE:ME and NE:DE) of diets. For the CH method, the NE of wcDDGS at 15% inclusion level (2,536 kcal/kg DM) was not different ( $P > 0.10$ ) from the NE of wcDDGS at 30% inclusion level (2,197 kcal/kg DM).

The average NE value of wcDDGS obtained from the CS method (2,416 kcal/kg DM) was similar ( $P > 0.10$ ) to that obtained from the IC method (2,405 kcal/kg DM) and to that obtained from the CH method (2,367 kcal/kg DM).

**Table 4.2.** Energy balance of pigs and energy values of diets and wcDDGS determined by the indirect calorimetry method (Exp. 2)<sup>1</sup>

Item	Dietary treatment			SEM	P-value	
	Control	15% wcDDGS	30% wcDDGS		Linear	Quadratic
DE, kcal/kg	3,497	3,475	3,362	36.17	0.022	0.330
ME, kcal/kg	3,065	3,017	2,906	45.25	0.029	0.581
HP, kcal/kg <sup>2</sup>	2,056	2,261	2,131	76.57	0.531	0.120
FHP, kcal/kg <sup>3</sup>	1,577	1,757	1,744	92.59	0.227	0.412
RE, kcal/kg <sup>4</sup>	950	787	888	98.85	0.663	0.297
NE, kcal/kg <sup>5</sup>	2,586	2,513	2,520	43.58	0.304	0.470
Efficiencies of NE						
NE/ME	0.85	0.83	0.87	0.01	0.180	0.135
NE/DE	0.74	0.72	0.75	0.02	0.454	0.081
NE of wcDDGS, kcal/kg <sup>6</sup>	-	2,407	2,403	62.87	0.651	-

<sup>1</sup>n = 6

<sup>2</sup>Heat production = [3.87 x O<sub>2</sub> + 1.20 x CO<sub>2</sub> – 1.43 x urinary N]/DMI

<sup>3</sup>Fasting heat production = [3.87 x O<sub>2</sub> + 1.20 x CO<sub>2</sub> – 1.43 x urinary N]/DMI

<sup>4</sup>Retained energy = (ME intake – HP)/DMI

<sup>5</sup>Net energy = (RE + FHP)/DMI

<sup>6</sup>NE of wcDDGS was calculated using the difference method by subtracting the NE contribution of the basal diet from the NE of the diets containing 15% wcDDGS or 30% wcDDGS (Woyengo *et al.*, 2010).

**Table 4.3.** Energy values of diets and wcDDGS (kcal/kg DM) determined by the chemical composition method

Item	Dietary treatment				P-value	
	Control	15% wcDDGS	30% wcDDGS	SEM	Linear	Quadratic
DE of diets	3,470	3,474	3,362	31.43	0.028	0.151
NE of diets <sup>1</sup>	2,447	2,451	2,364	22.00	0.015	0.117
NE of wcDDGS <sup>2</sup>	-	2,536	2,197	167.62	0.183	-

<sup>1</sup>Average calculated NE from equations 3.5, 3.6, and 3.7

<sup>2</sup>NE of wcDDGS was calculated using the difference method by subtracting the NE contribution of the basal diet from the NE of the diets containing 15% wcDDGS or 30% wcDDGS (Woyengo *et al.*, 2010).

**Table 4.4.** Comparison of NE of diets and wcDDGS (kcal/kg DM) between comparative slaughter, indirect calorimetry, and chemical composition methods

Item	Method of NE determination			SEM	P-value
	Comparative slaughter	Indirect calorimetry	Chemical composition <sup>1</sup>		
Diet					
Basal	2,430	2,586	2,447	23.28	0.885
15% DDGS	2,427	2,513	2,451	28.52	0.781
30% DDGS	2,429	2,520	2,368	24.63	0.644
Ingredient <sup>2</sup>					
15% DDGS	2,407	2,407	2,536	190.11	0.822
30% DDGS	2,424	2,403	2,197	82.11	0.866
Average NE of wcDDGS	2,416	2,405	2,367	107.85	0.678

<sup>1</sup>Average calculated NE from equations 3, 4, and 5 (Noblet *et al.*, 1994a)

<sup>2</sup>NE of wcDDGS was calculated using the difference method by subtracting the NE contribution of the basal diet from the NE of the diets containing 15% wcDDGS or 30% wcDDGS (Woyengo *et al.*, 2010).

## 4.5 DISCUSSION

The contents of CP, GE, ash, NDF, ADF, and EE of the DDGS sample in the present study compared favourably with that of Yanez *et al.* (2011) who used DDGS with the same proportions of wheat and corn feedstock as that used in the present study (i.e. 1:1; wt:wt). This nutritional profile confirmed that Canada Prairie Spring (CPS) wheat and corn were co-fermented in a 1- to -1 ratio (Yanez *et al.*, 2011). These values were intermediate to the average nutrient profile of corn DDGS (Stein and Shurson, 2009) and wheat DDGS from CPS wheat (Widyaratne *et al.*, 2009). However, the wcDDGS sample used in this study differed in CP and fiber contents from that reported by Widyaratne and Zijlstra (2007) and Nyachoti *et al.* (2005). Differences in CP and fiber contents in wcDDGS may be due to differences in blending ratio of wheat and corn (the ratio of wheat to corn in the wcDDGS used in this study was 1:1 as opposed to 4:1 used by Widyaratne and Zijlstra (2007)), differences in source or cultivars of the original cereal grain, or differences in the fermentation process (Spiehs *et al.*, 2002).

Digestible energy value of wcDDGS obtained in the present study was 3,305 kcal/kg DM (Data not shown). Cozannet *et al.* (2010) reported a similar average DE value of 3,349 kcal/kg DM for 17 samples of wheat DDGS fed to growing pigs. Also, Nyachoti *et al.* (2005) reported a similar DE value of 3,371 kcal/kg DM for wheat DDGS. For corn DDGS, similar DE values of 3,490 and 3,100 kcal/kg DM were reported by Spiehs *et al.* (2002) and Jacela *et al.* (2011), respectively. Stein *et al.* (2006) reported DE values ranging from 3,382 to 3,845 kcal/kg DM for corn DDGS.

Net energy values of wcDDGS obtained in the present study from the various methods ranged from 2,367 to 2,416 kcal/kg DM. These values are consistent with that reported (2,220 kcal/kg DM) for corn DDGS (NRC, 1998). Also, Cozannet *et al.* (2010) reported a similar NE value of 2,133 kcal/kg DM for wheat DDGS. Fu *et al.* (2004) and Hastald *et al.* (2004) reported NE value of 2,610 kcal/kg DM for corn DDGS. Jacela *et al.* (2010, 2011) reported calculated NE values of 2,131 and 2,045 kcal/kg DM for corn DDGS and high-protein corn DDGS, respectively.

Similar ( $P > 0.05$ ) NE values were obtained from the CS, IC, and CH methods. Kiesling *et al.* (1973) reported 22% higher NE value of sorghum grain for yearling steers with the IC method compared with the CS method. The reason for this discrepancy between cattle and pigs could be that physical activity in the respiration chambers was lower for cattle than for pigs because of their higher BW. Lower physical activity reduces HP which leads to an increase in NE value. Also the IC system used by Kiesling *et al.* (1973) was the old technology IC system which could be of lower sensitivity compared with new technology system. In the current study, NE was calculated from DE because DE values of feedstuffs are easily and routinely determined. Also, in North America, DE values of most feedstuffs are available (NRC, 1998).

In the various methods, dietary inclusion level of wcDDGS did not affect the NE value of wcDDGS. In theory, with greater inclusion of dietary fiber, a greater proportion of the diet was expected to be degraded in the hindgut where short chain fatty acids are produced and absorbed with a lower metabolic efficiency compared with energy digested in the small intestine (Noblet *et al.*, 1989). Accordingly, it was expected that increasing

inclusion level of fiber will decrease NE of wcDDGS. However, we did not detect any difference in the NE of wcDDGS included at 15 or 30%. Therefore, the results of this experiment indicate that the proportions of dietary fiber that are absorbed in the small intestine and in the hindgut are similar for diets containing 15 and 30% wcDDGS.

If determined NE values for the diets are expressed as percentages of DE, values between 72 and 75% are obtained for all diets. These values are in close agreement with the average value of 71% previously reported for diets containing between 1 and 11% fat by Noblet *et al.*, (1994a). If the same calculation is completed for wcDDGS, values of 73 and 72% are obtained for the IC and CS methods, respectively. Lower values of 64 and 66% were generated from the data of Cozannet *et al.* (2010) and Jacela *et al.* (2010) for wheat DDGS and corn DDGS, respectively. The reason for this difference may be because the DDGS used in the current study was lower in CP (32.2% - DM basis) compared with that used by Cozannet *et al.* (2010) (36.1 % - DM basis) and that used by Jacela *et al.* (2010) (40.8% - DM basis). The efficiency of conversion of CP for NE is lower than other components such as ether extract and starch (Noblet *et al.*, 1994a).

Retained energy values of diets were higher in the CS method (1,393, 1,351 and 1,305 kcal/kg for diets containing 0, 15, and 30% wcDDGS respectively) compared with the IC method (950, 787, and 888 kcal/kg for diets containing 0, 15, and 30% wcDDGS, respectively). The reason for this difference could be an over-estimation of HP by the Brouwer's equation. The Brouwer's coefficients have been obtained in respiration trials with ruminants (Brouwer, 1958) and these mean values may not be applicable to pigs (Quiniou *et al.*, 1996). Heat production obtained in the present study ranged from 2,056

to 2,261 kcal/kg. Noblet *et al.* (1994b) reported a similar HP value of 2,062 kcal/kg of feed for growing pigs. Values of FHP obtained in this present study were 1,577, 1,757 and 1,744 kcal/kg DM for the control, 15% wDDGS, and 30% wDDGS diets, respectively. These values are similar to that obtained by Noblet *et al.* (1994a): 179 kcal/kg BW<sup>0.6</sup> for 35 kg pigs (equivalent to 1,517 kcal/kg DM of feed).

In conclusion, the NE value of wDDGS for growing pigs obtained from the present study was 2,416 kcal/kg DM for the CS method, 2,405 kcal/kg DM for the IC method, and 2,367 kcal/kg DM for the CH method. As the values obtained using the various methods were not different, the average NE value for the wDDGS evaluated was 2,396 ± 25.71 kcal/kg DM. Therefore, for routine NE determination of feeds and feedstuffs, one may use the CH method. Dietary wDDGS inclusion did not affect the NE value.

## 5.0 GENERAL DISCUSSION

Distillers dried grains with solubles is produced by the fuel ethanol industry and is available for inclusion in diets fed to swine. The increasing supply of DDGS suggests that evaluation of its nutritional value for swine is worthwhile to support the development of cost-effective feeding programs (Widyaratne and Zijlstra, 2007). During recent years, several research projects have been completed to investigate the feeding value of DDGS (Nyachoti *et al.*, 2005; Slominski *et al.*, 2008; Cozannet *et al.*, 2010). Crude nutrient concentrations, energy and nutrient digestibility values, and effects of including DDGS in diets fed to swine of different categories have been investigated (Cook *et al.*, 2005; Pedersen *et al.*, 2007; Lan *et al.*, 2007). Only a few studies have been conducted to determine growth performance and nutrient digestibility of wDDGS (Widyaratne and Zijlstra, 2007 and Yang *et al.*, 2010) and no work has been done to determine the NE value of wDDGS for pigs. Widyaratne and Zijlstra (2007) reported reduced growth performance without effect on feed efficiency and an ileal energy digestibility of 66.5% for wDDGS included at 30% in grower-finisher pig diet. Yang *et al.* (2010) reported ileal dry matter and crude protein digestibility of 69.7 and 69.2%, respectively, for wDDGS. To gain a detailed knowledge of wDDGS as a feedstuff for swine, it is important to look into its NE value and also to determine its effect on growth performance, carcass and visceral organ weights, whole body O<sub>2</sub> consumption, and heat production when fed to growing pigs.

The wDDGS used in this study had 29.1% crude protein, 90.4 % dry matter, 4,785 kcal/kg gross energy, 4.8% ash, 9.2% ether extract, 36.5% NDF and 16.7% ADF.

Dietary inclusion of wcDDGS reduced ADFI and ADG but had no effect on feed efficiency for growing pigs. The reduction in ADFI in pigs fed diets containing wcDDGS is attributable to a reduction in palatability and increased bulk density and water holding capacity (Kyriazakis and Emmans, 1990, 1995; Avelar *et al.*, 2010). The lack of effect on feed efficiency is an indication that pigs consuming diets containing wcDDGS converted feed to gain similarly to those consuming the basal diet. Dierick *et al.* (1989) reported that dietary fiber depressed the growth of pigs and decreased the feed to gain ratio depending on the source and level of the fiber.

Dietary inclusion of wcDDGS caused a linear reduction in eviscerated carcass weight. This is attributable to the reduction in feed intake and reduction in DE observed in manuscript II. The amount of energy retained in the body depends on the amount of digestible energy (Noblet and van Milgen, 2004). Cespedes (2009) similarly found that live BW was lower in growing pigs fed corn DDGS than for pigs fed the corn-soybean basal diet. The ability of the pig to maintain DE intake would appear to be the major factor influencing weight gain of pigs consuming high-fiber diets (Dierick *et al.*, 1989). There was no effect of wcDDGS on the relative weight of visceral organs. For diet containing as much as 18.8 % NDF content as in the case with the diet containing 30% wcDDGS used in the present study, higher weights of organs especially cecum and colon where undigested feed components and endogenous secretions are fermented by microorganisms and short chain fatty acids and are absorbed was expected. It is well known that increase in dietary fiber results not only in an increase in the weight and volume of the gut, but also in an increase in the empty weight of the gut (Kass *et al.*, 1980; Dierick

*et al.*, 1989; Nyachoti *et al.*, 2000). As gut tissue and gut contents are elements of body weight, they will affect the maintenance requirement and the efficiency of utilization of ME (Burrin *et al.*, 1990). Diets with high fibre content caused a significant increase in the secretion of endogenous fluids and increased secretion of these fluids is associated with a higher activity of secretory organs resulting in the enlargement of such organs (Wenk, 2001). Factors such as, how much solubles were added to the distiller's grains, the completeness of the fermentation process and the quality control of the fermentation process could affect the nature of dietary fibre in DDGS thereby causing a lack of effect on weight of visceral organs. Also, reduced feed intake in pigs fed wcDDGS diets could be the reason why there was no effect on organ weight. When feed intake is reduced, less substrate will be available for microbial fermentation and less VFA will be produced and absorbed in the hindgut.

No effect of wcDDGS on whole body  $O_2$  consumption and HP was observed in this study. However, the values of 298, 308, and 287 ( $\pm 14.42$ ) kcal/kgBW<sup>-0.6</sup> for diets containing 0, 15, and 30% wcDDGS respectively, obtained in the current study were similar to that (320 kcal/kg BW<sup>-0.6</sup>) reported by Noblet *et al.* (1994a) for growing pigs. Previous studies (Dierick *et al.*, 1989; Noblet *et al.*, 1994a) observed greater HP for high fibre diets. In the current study, the desired feeding level (550 kcal ME  $\cdot$ kg BW<sup>-0.60</sup> $\cdot$ d<sup>-1</sup>) was not achieved for some pigs in the wcDDGS diet group and this could have confounded the effect of wcDDGS on HP because the level of feed intake could have a large impact on HP in pigs (Fuller and Boyne, 1972). Noblet *et al.* (1994a) also reported

that the expected feed intake was not achieved for some diets because of their high fiber content.

The NE value of wcDDGS obtained for growing pigs from the present study was 2,416 kcal/kg DM of feed for the CS method, 2,405 kcal/kg DM of feed for the IC method, and 2,367 kcal/kg DM of feed for the CH method (Manuscript II). These values are in close agreement (SE = 107.85) which indicate that NE of feeds and feedstuffs can be determined accurately by the IC procedure and that the CH method can be used for routine NE determination. In both the IC and CS procedures, diet had no effect on NE value. This is in agreement with the observation in Manuscript I in which there was no effect of diet on HP.

Measurement of energy retention has been carried out extensively to evaluate the nutritional characteristics of diets and feedstuffs or to study the energy requirements of pigs using the balance technique or by the CS technique (Just *et al.*, 1983; Neergaard, 1981). Measurement of energy retention can be obtained either by IC studies, where retained energy is calculated as the difference between ME intake and HP estimated from gas exchanges or by CS where energy retained corresponds to the difference in body energy between experimental pigs at the end of the experiment and control pigs slaughtered at the beginning of the experiment (Quiniou *et al.*, 1996). Because of undetected and additive losses of feed and feces, energy retention would be overestimated by the balance or IC technique while losses by the CS technique would underestimate energy retention (Neergaard, 1981; Just *et al.*, 1982). The difference in energy retention, under continuous measurement of HP, has been investigated in a study

on piglets (Verstegen *et al.*, 1993) which indicates a 7.5% discrepancy between techniques. Usually, energy retention assessed by the CS technique is considered to be closer to the true retention than the balance technique (Kotarbinska and Kielanowski, 1969; Davidson and Williams, 1968). In the present study, RE was lower in the IC method than the CS method. An explanation of the observed difference could be an over-estimation of HP by the Brouwer equation. The coefficients of Brouwer's equation were obtained in respiration trials with ruminants (Brouwer, 1958) and therefore these mean values may not be applicable to pigs (Quiniou *et al.*, 1996).

## 6.0 SUMMARY AND CONCLUSION

Inclusion of up to 30% wcDDGS in a corn-soybean meal based diet reduced growth performance and carcass weight but had no effect on visceral organ weights, whole body O<sub>2</sub> consumption and HP for growing pigs. Likewise, wcDDGS inclusion had no effect on RE and NE of diet for both the IC and the CS methods. Similar NE value of wcDDGS was observed for the IC, CS, and CH methods. As the values obtained using the various methods were not different, the average NE value of the wcDDGS evaluated was  $2,396 \pm 25.71$  kcal/kg DM. According to the results of the present research for the NE of wcDDGS (1:1; wt/wt), it can be concluded that the results from the IC method using the Columbus Instrument are as reliable as ones obtained by the CS method. Also, for routine NE determination, one can use the chemical composition method. Further research is suggested to:

1. Validate the NE value of wcDDGS obtained from this study with growth performance trials.
2. Determine the effect of wcDDGS on the production and absorption of volatile fatty acids in the cecum and colon of pigs and intestinal microbial population. This will shed light on why wcDDGS inclusion had no impact on weights of cecum and colon.
3. Determine the effect supplementing diets containing wcDDGS with exogenous enzymes on NE value for growing pigs.
4. Develop a nutritional profile for wcDDGS from various ethanol producing plants.
5. Investigate the effect of varying the ratio of wheat and corn feedstock blend on the NE value of wcDDGS.

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