METABOLIC EFFECTS OF ZEOLITE AS NATURAL FEED SUPPLEMENT FOR GROWER PIGS

Yonghong Wan

Department of Animal Science
McGill University, Montreal

May, 2005

A Thesis Submitted to
McGill University
in Partial Fulfillment of the Requirements
of the Degree of Master of Science

Your file Votre référence
ISBN: 978-0-494-22777-0
Our file Notre référence
ISBN: 978-0-494-22777-0

Yonghong Wan **Animal Science**

M.Sc. Thesis

ABSTRACT

The rapid growth of Canadian pork industry has been challenged by its negative impact on the environment. To find an economical and promising solution to the environmental problems, 4% zeolite (90%+ clinoptilolite) were supplemented to a regular (100% crude protein (CP) and energy) or low CP and energy (90% CP and 90% energy or 90% CP and 85% energy) grower pig diets. Twenty male and twenty-four female grower pigs were used in two feeding experiments respectively, followed by a metabolic test with three batches of animals repeated to determine the metabolic effects of zeolite supplementation. Pig performance (body weight gain, daily feed intake and feed conversion ratio), and metabolic parameters (manure mass, feed intake, protein and energy conversion, as well as dry feed and organic matter retention) were evaluated. Zeolite supplementation at 4% to a regular diet for grower pigs had a positive but not significant ($P > 0.05$) effect on all pig performance and metabolic parameters, compared to the regular diet without zeolite. Among 4 rations, pigs on a regular diet with 4% zeolite performed consistently best throughout the entire trail, with decreased average daily consumption and reduced amount of feces, increased feed and organic matter retention in the gastrointestinal tract, improved feed as well as protein and energy conversion, and enhanced body weight gain. Moreover, zeolite supplementation at 4%, with 90% CP and 90% energy in grower pig diets, improved feed and protein and energy conversion rate, and increased body weight gain, when compared to those of pigs fed a regular diet without zeolite. However, a diet of 90% of CP and 85% of energy with 4% zeolite significantly ($P < 0.05$) increased feed consumption and the amount of feces produced, and decreased feed and organic matter retention in the gastrointestinal tract, thus reducing feed conversion rate. Therefore, 4% zeolite supplementation to the regular or low CP and energy (90% CP and 90% energy) grower pig diets could be a promising solution to the environmental problem challenging pork industry.

Key Words: Clinoptilolite, Pig, Pig Performance, Metabolic Parameter

Yonghong Wan **Zootechnie**
M.Sc. Thesis

RESUMÉ

Douze porcs males et vingt quatre porcs femelles furent utilisés pour effectuer des essais en cages métaboliques. Ces essais furent réalisés en deux phases et en trois groupes, pour déterminer l'effet de l'inclusion dans la moulée de 4% de zéolite (90%+ de clinoptilolite). Quatre moulées furent utilisées : une témoin, sans zéolite (ration 1); une contenant le même niveau d'énergie et de protéine, mais avec 4% de zéolite (ration 2); une contenant 90% de la protéine et 90% de l'énergie et avec 4% de zéolite (ration 3); la dernière avec 90% de la protéine et 85% de l'énergie et 4% de zéolite (ration 4). On observait les paramètres suivants de performance des porcs pendant les essais : gain de poids, consommation de moulée, et conversion alimentaires. On comparait aussi la production de fumier, le taux de conversion de protéine et d'énergie, et la rétention d'aliments et de matière organique.

Sauf pour la ration 4, l'ajout de 4% de zéolite à la diète des porcs avait un effet positif mais non significatif ($P < 0.05$) sur la performance alimentaire des porcs, comparativement à la diète sans zéolite. Pendant les trois essais, la ration avec zéolite améliorait de façon non significative, les résultats en diminuant le taux de consommation de moulée et de production de fèces, et en améliorant la rétention d'aliments et la conservions de moulée ainsi que de protéine et d'énergie. De plus, la ration 3 améliorait le taux de conversion de la moulée, de la protéine et de l'énergie, comparativement à la ration témoin. Par contre, la ration 4 augmentait significativement ($P < 0.05$) le taux de consommation de moulée et de production de fumier et diminuait significativement ($P < 0.05$) le taux de rétention de des aliments et de la matière organique et le taux de conversion alimentaire.

ACKNOWLEDGEMENTS

First of all, I would like to offer a very special thank you to my thesis supervisor Dr. Xin Zhao for his guidance, support and time spent during my master's work, especially when I met difficulties or lost my confidence in the program, he always immediately encourage me to continue. I also want to thank Dr. Suzelle Barrington, for her significant instruction for the experiment design, data analysis and thesis writing.

I wish to express my sincerest appreciation to my wife Li Yang, who supported me and took care of my family during my graduate studies, and gave birth to my second daughter. I also wish to thank my parents and brothers who understood and encouraged me to continue studying.

I would like to acknowledge the Federation des Producteurs de Porcs du Quebec for supporting me financially and KMI inc. for supplying the zeolite throughout the experiment.

Thanks to all professors and working staff especially Mr. Jan Pika and fellow graduate students especially Ms. Heather Watt in the Department of Animal Science for providing me help and sharing spare times during my studies. Thanks to Mr. Denis Hatcher and all the staff at the McGill Pig Research Complex for their time and help with handling the pigs.

Finally, I want to thank the kind and indispensable secretaries of the Departments of Animal Science and Bioresources Engineering for helping me out with the McGill administrative tasks.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF SYMBOLS AND ABBREVIATIONS

AA	Amino Acid
ADG	Average Daily Body Weight Gain (kg/d)
ADFI	Average Daily Feed Intake (kg/d)
Al	Aluminum
ANOVA	Analysis of Variance
As	Arsenic
AU	Animal Unit
C	Carbon
Ca	Calcium
Cd	Cadmium
Cdn	Canadian
CEC	Cation Exchange Capacity
CH_4	Methane
CLI	Clinoptilolite
Co	Cobalt
CO_2	Carbon Dioxide
COD	Chemical Oxygen Demand
CP	Crude Protein
Cr	Chromium
CRD	Completely Randomized Design
Cs	Cesium
Cu	Copper
DM	Dry Matter
DFR	Dry Feed Retention
ECR	Energy Conversion Rate
FCR (F/G)	Feed Conversion Rate (Feed intake/body weight gain)
Fe	Iron
GIT	Gastrointestinal Tract
H_2S	Hydrogen Sulfide
IBW	Initial Body Weight
ICP	Inductively-Coupled Plasma Emission Spectroscopy
K	Potassium
LSM	Least Square Mean
Meq	Milliequivalent
Mg	Magnesium
Mo	Molybdenum
MUC	Montreal Urban Community
N	Nitrogen
Na	Sodium
NH_3	Ammonia

NH_4^+	Ammonium
Ni	Nickel
NIST	National Institute of Standards and Technology
NSERC	Natural Science and Engineering Research Council of Canada
NSP	Nonstarch Polysaccharides
OM	Organic Matter
OMR	Organic Matter Retention
P	Phosphorus
Pb	Plumbum
PCR	ProteinConversion Rate
RCB	Randomized Complete Block
S	Sulfur
SAS	Statistical Analysis System
SD	Standard Deviation
Se	Selenium
SEM	Standard Error Mean
Si	Silicon
SiO_4	Silica
Sr	Strontium
SZA	Sodium Zeolite A
TFI	Total Feed Intake
TFE	Total Feces Excretion
TFR	Total Feed Retention
TKN	Total Nitrogen
TS	Total Solid
TUE	Total Urine Excretion
VFA	Volatile fatty acid
XRD	X-Ray Diffraction
Zn	Zinc

CHAPTER I

Introduction

1.1 General Introduction

The pork industry is one of the world's most important agri-food industries. Pork meat is the commodity most consumed in regions which have been experiencing rapid economic growth, including Asia and Latin American. Pork is also the preferred meat throughout most of Europe and North America. In Canada, the livestock industry annual grosses are more than $15 billion (Barrington, 2001). Amongst livestock industries, pig production is the third most important agricultural sector in Canada. It provides more than $3 billion of economic activity annually. This industry has grown rapidly over the last few years. The annual number of pigs in the market has dramatically increased since 1995, from 17 million hogs marketed in Canada in 1995 to more than 30 million heads in 2003. At present, there are more than 15,470 pork producers in Canada. Quebec, Ontario and Manitoba are major pork producers and 77.7% of all pigs marketed in 2003 came from these provinces (Statistics Canada, 2003). Prior to 2002, Quebec was the largest pig producing province.

Although several factors such as high feed grain prices, poor margins and reduced domestic consumption negatively affect the domestic pig market in Canada, the pork industry is still the most promising among all agricultural sectors. Several provinces have taken measures to expand their pig production in the past few years and have made great progress. From 1995 to 2003, pig production has increased by 47.8 % in Quebec, 81.5 % in Ontario, 162.3 % in Manitoba, 73 % in Saskatchewan and 71.5 % overall in Canada (Statistics Canada, 2003). This expansion is partially due to increasing international pork consumption demands.

Canadian pork is recognized internationally for its high quality, thereby allowing Canada to export 30% of its annual pork production to more than 55 different countries, with the greatest demand coming from the United States and Asia (Statistics Canada, 1996; Statistics Canada, 1997). The total amount of exported pork increased dramatically from 350 million kg in 1995 to 923 million kg in 2003, which represents an increase from $997 million (Canadian) in 1995 to $2.3 billion (Canadian) in 2003 (Statistics Canada, 2003). Furthermore, Canada exports live pigs to other countries, with the US being the main importer, due to high quality genetics. Canadian pork producers also directly supply weanling pigs to their US counterparts. The total export of live pigs has been steadily increasing since 1994, from 0.9 million live pig heads in 1994 to more than 5 million in 2002. This represents an increase from $100 million (Canadian) in 1994 to $487 million (Canadian) in 2002 (Statistics Canada, 2003). Moreover, some Canadian operations have joint ventures in the US for feeding pigs with further arrangements with US packers. In 2004, Canada was the second largest pork exporter in the world.

1.2 Impact of Pork Industry

The rapid growth of the pork industry has brought a special challenge to pork producers. Because of the impact of the pork industry on the environment, rural and urban communities have opposed its development. The complaints pertain to serious air, water and soil environmental pollution caused by the pig production despite the public's- liking for a variety of delicious pork products.

1.2.1 Present Situation of Pig Farms

In North America, the structure of the livestock industry has changed over the past thirty years. The number of cattle and sheep has dropped by 17% while the number of pigs and chickens has increased by 7.5% and 34%, respectively (Barrington, 2001). Meanwhile, livestock producers have been gradually focusing on improving production levels and expanding the size of their enterprise. The consequence is that net production has greatly increased, especially for milk and eggs (20 and 30%, respectively). This

indicates that the yield per animal has increased significantly. Unfortunately, the quantity of manure produced has increased exponentially due to increased use of higher feed nutrient content and the low digestive capability of most livestock (Barrington, 2001). Furthermore, intensive livestock operations have created regions of concentrated animal wastes, imposing more stress on the local environment.

The structure of the Canadian pork industry has been changing toward fewer numbers of production farms with a concomitant increase in herd size per farm. The number of pig farms has decreased greatly since 1976. The reported number of pig farms in Canada was 63 602 in 1976, 36 472 in 1986, 29 592 in 1991, 21 105 in 1996 and 15 472 in 2001 (Statistics Canada, 2001). Accordingly, the total number of Canadian pig farms decreased by 26.7 % between 1996 and 2001. However, the number of pigs in Canada increased by 26.4 % within the same period, from 11.04 million in 1996 to 13.96 million in 2001. As a consequence, the average number of pigs per farm unit has increased tremendously (993.4 % from 1976 to 2001), from an average of 91 pigs per farm in 1976 to 995 pigs per farm in 2001 (Statistics Canada, 2001). Furthermore, this trend is expected to continue as the proportion of large pig operations in Canada increase while smaller operations disappear.

Ontario, Quebec and Manitoba are the three major pig producing provinces, accounting for more than 75 % of pig production in Canada. Similar changes in pig production have happened in these three provinces over the past years. The number of pig farms has greatly decreased between 1976 and 2001 with a decline of 79.9 % in Ontario, 73.2 % in Quebec and 79.4 % in Manitoba. Meanwhile, the number of pigs sent to the market has increased, between 1991 and 2003, by 96.7% in Ontario, 64.3 % in Quebec, 239.6 % in Manitoba (Statistics Canada, 2003). As a result, the average number of pigs per farm unit has significantly increased by 703% in Ontario, 780% in Quebec and 1449% in Manitoba from 1976 to 2003. The average number of pigs per farm unit reached its highest level in 2001, with 827 pigs per farm in Ontario, 1567 in

Quebec and 1555 in Manitoba (Statistics Canada, 2001).

Despite the increase in animal production, the average livestock (Animal Unit = AU) density is not particularly high in Canada due to the large land area. Counted among the countries with the highest livestock densities are China, Denmark, The Netherlands, and Japan (Barrington, 2001). In these countries, average livestock densities exceed 1 AU per ha, whereas the total average density in Canada is 0.19 AU/ha. Nevertheless, the average livestock density exceed or come close to exceeding 1 AU/ha in some Canadian regions, such as in the Southern Ontario, and in the Lanaudière (North East of Montréal), l'Estrie (Eastern Townships) and the Chaudière-Appalaches (South of Québec City) regions of Québec (Barrington, 2001). Actually, expansion of the pork industry in Ontario and Québec has been limited by the availability of land for the spreading of manure. It is, therefore, reasonable to expect that, in the future, most of the growth in the Canadian pork industry will occur in Western Canada.

Most modern pig operation generates huge quantities of manure due to intensification of the industry over the past years, in the world as well as in Canada. Land spreading is still the most practical method of disposing of manures. Pig manure is recognized as a valuable source of crop nutrients. Recently, it has also been recognized as a significant source of alternative fuel such as methane. The proper use of manure nutrients is an integral part of a sustainable pork operation. The exact amount of land required for a pig production unit to fully utilize the nutrients can be calculated based, in a large part, on the crop to be grown and its nutrient needs as well as what the pigs are fed and the amount and composition of the manure produced. Basically, a sustainable pig farm requires about 0.25ha/sow for a farrow-to-finish unit. However, this changes with the type of farm. For instance, a farm will require about 1.1ha/sow if it raises only pregnant sows, lactating sows, nursing piglets, and nursery pigs. For a grower pig operation, finishing pigs require, on average, 0.025ha/pig, since finishing pigs produce

more manure than other stages of production (John and Wilson, 2003).

Most importantly, manure should be applied to crop field based on the nutrient requirements of crops. Manure management has not followed changes occurring in the development of pig farms. The problem arising is that the average number of pigs per farm unit has rapidly increased in Canada, especially in Ontario, Quebec and Manitoba, and has resulted in heavier applications of pig manure on limited land acreage. Accordingly, environmental problems ensued. The potential for pig-related pollution can be classified into two categories: contamination of soil and water or air pollution.

1.2.2 Soil and Water Pollution

Throughout the world, the general public is concerned with the environmental pollution caused by the pork industry. Soil and water pollution result from feedlot run-off, poor manure storage and heavy land applicaitons.

Basically, pig manure contains nutrients which are mainly composed of nitrogen (N), phosphorus (P) and potassium (K). In Canada, livestock manure represents some 1 080, 675 and 1 120 million tons of N, P and K, respectively. Although some N is lost through handling, all the P and K remain (Garcia Moreno, 1993). The N:P:K ratio in pig manure is about 10:9:8. Different crops have a varied ability to absorb these nutrients. For example, corn needs a ratio of 10:4:10 (John and Wilson, 2003). When a producer applies the appropriate amount of pig manure to the cropland, usually, P will accumulate on the cropland because the plants utilize N and K at higher rates than they do P. Since P is considered the limiting factor in applying livestock manure to land for years, P over-fertilization has occurred followed by soil P saturation and surface water loading through soil erosion and some leaching. Simard et al. (1995) found that, several times annually, the P levels in many large rivers of Québec exceed the surface water limit of 0.03 mg/l for potential eutrophication. When a producer applies much more pig manure than that required by plants on a limited tilled land area, accumulation of

excessive amounts of nutrients including N, P and K in this specific area of land will happen.

Over fertilization of N has both a short and long-term effect on land. On a short-term basis, excessive N is rapidly washed into rivers due to its high solubility. On a long-term basis, soluble N can reach and contaminate underground waters or be lost via denitrification (N_2O). The latter situation has unfortunately happened in some regions in Canada. High livestock densities in some regions such as Ontario, Quebec, Manitoba, Alberta and British Columbia, have led to excess amounts of manure nutrients over a limited arable land. These manure surpluses have been confirmed in the B.C. Fraser Valley, the Lethbridge region in Alberta, Southern Ontario and the Québec regions of St Hyacinthe, L'Assomption and Beauce (Barrington, 2001).

In fact, Quebec has the highest pig density per tilled surface in Canada: the average number of pigs per farm unit was 1 567 heads in 2001, while the density was 200 pigs, versus 33 pigs elsewhere in Canada, per 100ha of cultivated land fertilized with manure in 1997 (Statistics Canada, 1997). In comparison, Saskatchewan produced about 1.1 million finishing pigs on an arable land base of 26.7 million ha, with the density of 4.2 pigs per 100ha of cultivated land. Nonetheless, Quebec's situation is far less severe than that of the Netherlands where there were 375 pigs per 100ha of rural land or than that of Denmark where almost 20 million pigs were produced on an arable land base of 2.5 million ha (Meyer, 1997; Carlton Trail Regional Hog Information, 2005). As a result of high pig density, pork producers in some regions of Quebec do not have enough land to apply the pig manure while staying within the range of the plant's nutrient requirements. In 1996, the Quebec Ministry of the Environment and Wildlife conducted a study to check the capacity of agricultural land for receiving animal wastes (organic fertilizer) from Quebec farms. Quebec's nine largest river basins including Chaudiere, Yamaska, L'Assomption, Etchemin, Richelieu, Saint-Francois, Nicolet, Bayonne, and Boyer were chosen due to their dense animal population. Furthermore, the agricultural lands in

these basins were known to be over-fertilized in terms of P and N. This study reported that manure applications over-fertilized P in the agricultural land of such basins by a factor of 183%, except for that of Richelieu. In terms of N, 196% over fertilization occurred in 4 (Chaudiere, Etchemin, Bayonne and Boyer) of the 9 basins. The producers were found to apply manure to only 29% of the cultivated land. In addition, farmers in these basins continue to apply a lot of mineral fertilizer, besides the manure, thus resulting in over fertilization of P and N by 167% and 133%, respectively (Ministere de l'Environnement et de la Faune du Quebec, 1996).

Heavy land applications of manure can lead to serious river and underground water contamination. In Canada, it was estimated that 25 to 30% of the population depends on underground water for drinking water supply. Water pollution can kill fish and other wildlife and can pollute drinking water to the point where it is not safe for drinking or other uses (John and Wilson, 2003). Betcher et al. (1996) performed a study on 1300 domestic wells in rural regions of Ontario. One or more water contaminants were found in approximately 40% of the wells, and the concentration of the contaminant exceeded the acceptable limit for drinking water. More interestingly, a correlation was found between the occurrence of bacteria in wells, specifically fecal coliforms, and the proximity to a farm where manure was routinely applied.

1.2.3 Air Pollution and Manure Odor

The trend toward high-density confinements for pigs has increased tremendously in recent years. Such practice has created concentrated sources of odor emissions and air pollution. An increased frequency of odor-related complaints has been noted in areas where pig production facilities are more intense. Pig manure releases more than 100 odorous compounds presenting a risk for air contamination. These substances are divided into two groups: malodorous substances and toxic substances (Canadian Pig Council, 1996). Toxic substances such as methane and hydrogen sulphide, which are emitted during manure fermentation, are dangerous for humans and animals especially

inside pig buildings or beside manure pits. They have been known to have irritating and sometimes asphyxiating effects causing many deaths (Schulte, 1997). Although these substances are found in relatively high concentrations at certain sites on a pig operation, such as around storage facilities, they are diluted in the atmosphere and do not cause a direct health problem for nearby populations.

Odors produced result from the digestive process of microorganisms in pig manure (Zhu, 2000). Generally, fresh manure releases a limited amount of odors. However, the microbial activity that developes within the first 12h of storage of pig manure creates significant amounts of odorous gases. The gases and odours in the building are transferred by the ventilation system into the atmosphere and cause pollution. Several of the 300 or so gases emitted by manure are detectable at concentrations of one part per billion (Barrington, 2001).

It is known that odors can impact the well-being of workers and animals (Schiffman, 1998). High levels of odorous compounds have been reported to reduce growth performance and increase susceptibility to disease in confinement housing (Roderick et al., 1998). Furthermore, odor can elicit a wide range of physiological responses that range from irritation of the eyes, nose, and throat, to nausea, headache, vomiting, and even disturbance, annoyance, and depression (Roderick et al., 1998). Importantly, pig manure-related odours can induce a psychological response and sensory effects in human beings (Schiffman, 1998). Actually, the psychological response and sensory effects of human beings regarding manure odor depend on several factors such as human emotion and memory, sociological aspects and the economic dependence of the human on pig production. Perceptions of odors by neighbors and government regulation of odor emissions can have serious economic consequences for farmers. Odor, like noise, is a nuisance or disturbance and complaints are normally handled by provincial or local authorities.

Some of the most important classes of odorous compounds commonly associated with pig production are volatile fatty acids (VFAs), mercaptans, esters, carbonyls, aldehydes, alcohols, ammonia, and amines (USDA, 2003). Among all odorous compounds, ammonia can create the strongest odor near manure storage or building sites, not necessarily because it is very easily detected but because it is produced in large quantities. In fact, ammonia produced by the pork industry represents a large proportion of total ammonia emissions. Kay and Lee (1997) reported that the U.K.'s agricultural sector produces around 198 x 10^6 kg of NH_3 per year, in which 23 x 10^6 kg come from the pork industry. In Canada, NH_3 emissions from agriculture amount to 468 Gg (thousand tonnes) of N per year, about 90% of total NH_3 emissions. Of this, 76 Gg (thousand tonnes) comes from pork industry (Janzen et al., 2003). The ammonia produced stays in the air for a short period of time and then falls to the ground or is transformed into other pollutants such as ammonium nitrate and ammonium sulfate. Excessive ammonia emissions cause acid rain thereby disturbing different ecosystems, damaging forests, acidifying fragile ecosystems and increasing the risk of river and lake eutrophications (Roderick et al., 1998; Williams and Nigro, 1997). Importantly, ammonia emissions can also be transformed into ammonium aerosol, which can be harmful to human health when present in high concentrations.

In the past decades, the dietary supplementation of zeolite has been studied as one way to attenuate the negative environmental impact of the pork industry while increasing productivity and performance. Zeolite, as crystalline with large structural cavities and a mineral collector, has special chemical and physical, as well as biological properties. Zeolite can lose and gain water reversibly without changes in crystal structure, selectively adsorb and exchange extraframework cations, immobilize microorganisms and improve the ammonia /ammonium ion equilibrium (Mumpton, 1999). Several studies showed a positive effect of zeolite supplementation by improving the production performance of animals and the digestibility of some feed nutrients, reducing odor emissions in the manure of treated pigs. It is likely that zeolite

supplementation to the pig diets could be an economically viable and potentially promising solution to the environmental problems challenged by the pork industry.

1.3 Objectives

The main objectives of this research program were to study the effects on pig performance and feed digestion, of adding 4% zeolite (90+% clinoptilolite) to the ration of growing pigs. Based on results from previous laboratory studies, 4% zeolite was supplemented to different rations. The rations used in this study were 100% CP & 100% energy without or with 4% zeolite; 90% CP& 90% energy with 4% zeolite, and 90% CP & 85% energy with 4% zeolite.

The research was divided into two phases to measure pig performance and metabolic parameters.

The first phase consisted of observing the following parameters while the experimental pigs were raised in growing/finishing pens:

1.The average daily feed intake (ADFI) of the pigs;

2.The average daily body weight gain (ADG) of the pigs;

3.The feed conversion rate (feed/gain, FCR) of the pigs;

The second phase consisted of monitoring the following parameters while the experimental pigs were held in individual metabolism cages:

1.The total feed intake (TFI) of individual experimental pig;

2.The total manure excretion volume including feces and urine (TFE, TUE);

3.The total manure retention (TFR) in the gastrointestinal tract (GIT);

4.The total dry feed retention (DFR) and organic matter retention (OMR) of feeds;

5.The protein conversion rate (PCR) and energy conversion rate (ECR) of feeds.

CHAPTER II

Literature Review

To effectively reduce the environmental impact of pig manure, it is essential to understand the characteristics and handling methods of pig manure. This chapter will also review different solutions used by pork producers to decrease the impacts of this manure on the environment.

2.1 Characteristics of Pig Manure

Upon ingestion of feed, pigs convert these foods into valuable products such as meat, blood, and body tissues as well as into unavoidable and less desirable waste products such as manure (Roderick et al., 1998). Manure is a combination of feces, urine and added products such as water, wasted feed, hair, and bedding (John and Wilson, 2003). Due to the unbalanced digestibility of certain nutrients in the feed, it is unavoidable that some nutrients remain in the manure, especially in the feces and urine. According to John and Wilson (2003), manure is rich in carbon (C), nitrogen (N), phosphorus (P) and potassium (K) as well as sulfur (S). The application of manure to cropland can help to maintain soil organic matter levels, improve soil tilth and water-holding characteristics as well as control soil erosion. When the soil is tilled and fields are sown, a portion of the organic matter is oxidized and lost from the field.

Manure carbon is primarily found in the form of partially digested plant materials as a nutrient. C is utilized by direct spreading of solids or semi-solids or released into the air in various froms such as carbon dioxide (CO_2) or methane (CH_4). Meanwhile, N, at about 0.5 mg/kg of manure, is found in the forms of elemental nitrogen, ammonia, ammonium and organic nitrogen such as urea and various metabolites of urea (John and Wilson, 2003). Not all N is available to the crop during the first year of application.

Ammonia is immediately available to the crop while organic N is slowly released to the crop and can have a significant impact in the N-supplying power of the soil if allowed to build up over several years of repeated manure applications. About 25 to 30% of the organic N is mineralized and available to the crop in the first year and the remainder becomes available over the next three years, at a decreasing rate.

Phosphorus is an essential element for plant growth and is required to maintain profitable crop production. With the approximate concentration of 0.4 mg/kg of manure, P exists in the form of phytate and in the plant-available form of P_2O_5. Approximately 50% of P in manure is immediately available for crop uptake (Liu et al., 1998).

Potasium, in the manure, exists in the available form of K_2O. Usually, the addition of K to soils through manure applications is of little benefit to crop production because most soils, except for sandy ones, have naturally high levels of K. In contrast, S levels are generally low in manure, usually between 0.1 to 0.4 kg/m^3. The availability of the S in manure is variable, so soil tests and fertility strip trials should be performed before applying additional sulfur fertilizer with manure.

Of the five main elements present in pig manure, the two principal nutrients of concern are N and P (Tamminga, 1996). Nitrogen is of concern because of its impact both on the inside and outside barn environment. Pig production has been recognized as a major source of ammonia emission, which is a noxious gas for humans and animals and contributes to bad odor and acidification of the environment. The main component of ammonia emission originates from urea in urine. Fecal N is less volatile than urinary N, because fecal N is chemically bound within proteins or other compounds (NRC 1998; Ruurd et al., 2001). Over-supplementation of diets with nutrients to ensure maximum pig performance results in excessive amounts of nutrients excreted in feces and urine. Phosphorus is also excreted in urine and feces, and could have a major impact on the environment and the economy if not managed properly (Cromwell et al., 1993).

It is a common knowledge that pig feces, urine, and respiration and fermentation gases are excreted in solid, liquid, and gaseous forms, respectively. After excretion, manure can be handled as liquid slurry, a semi-solid, or a dry form. When the manure contains 15% solids or less, it has characteristics of a liquid, whereas, it may have characteristics of a solid when it contains more than 20% solids. The concentration of nutrients in the manure varies, depending on the form of manure and the method of storage and handling. For example, in liquid pits, N and P are present at approximately 4.23 g/L and 3.22 g/L, respectively, but in lagoons their concentration is only about 0.48 g/L and 0.29 g/L, respectively (John and Wilson, 2003). On the other hand, it is found that dry manure poses a lower risk of environmental pollution than liquid manure, since dry manure, when truly dry, has less of an odor than liquid manure and is less likely to flow toward a waterway than liquid manure. Therefore, in the United States, pig farms are not allowed to discharge waste into waterways such as stream, lakes, or underground aquifers. Unfortunately, due to economic reasons, most pig manure is collected as slurry in channels beneath perforated floors. Along with increasing livestock density and its concomitant increase in manure production, manure is usually diluted to a water content exceeding 92% in order to facilitate its management. Typical pig manure water levels, outside of the barn are 93 %, whereas when handled as a solid, water content of this manure would be around 80% (Barrington, 2001). In addition, when stored over the winter in an open outdoor pit, manure water content increases to 95 %.

2.2 Soil and Water Impact

2.2.1 Soil Pollution by Pig Manure

Pollution of soil and land occurs when large volumes of pig slurry are applied to the land in excess of the fertilizer needs of the crop. Overapplication of manure to soil can result in the buildup of plant nutrients, phosphate and potash, as well as of toxic metals such as copper and zinc (Jongbloed and Lenis, 1998; Nielsen, 1987; Roderick et al.,

1998). The build up of phosphate and potash over time in the soils of some pig farms has been confirmed by soil analyses. In addition, prolonged overapplication of manure increases the risk of nitrate leaching, leading to an imbalance in soil chemistry and less biodiversity, thus resulting in reduced crop yields. Furthermore, when manures and slurries were heavily applied to land base, heavy metals accumulated in the top layer, raising potential risk for human and animal health (e.g., copper intoxication of sheep) and soil life (earthworms, microbiology) (Jongbloed and Lenis, 1998).

2.2.2 Water Pollution by Pig Manure

Water pollution by pig waste was thought to be responsible for 9% of the total water pollution incidents attributable to agriculture (Nielsen, 1987). Usually, slurry storage is the most common source of pollutants entering waterwarp due to poor storage management, such as overfilling stores and failing to turn off pumps in time when filling slurry spreaders. Runoff from yards and washing water are the next most important causes of water pollution, probably because of carelessness and failure to separate clean rainwater from roofs and open areas from contaminated concrete areas. The major concern for ground or surface water pollution, originating from land application of excessive quantities of nutrients, has focused on N and P levels. Nitrate leaching is considered a major N pollution concern on livestock farms. Ammonia toxicity to fish and altered effectiveness of chlorination are additional concerns related to N pollution (Roderick et al., 1998). According to Canadian Water Quality Guideline (CWQG), the interim to protect freshwater life and marine life is 13 and 16 milligrams of nitrate per litre of water respectively (Environment Canada, 2005).

Phosphorus entering surface waters from runoff of pig manure left on the soil surface can cause and promote the process of water eutrophication (Smith et al., 2004; Tabbara, 2003). Advanced or accelerated eutrophication of surface water leads to problems for fisheries, recreation, industry, and even drinking, because P can stimulate the growth of undesirable algae and aquatic plants and even promote their blooms. Their subsequent senescence and decomposition result in an increased oxygen demand

that interferes with the welfare of fish and wildlife (Daniel et al., 1998; Roderick et al., 1998). These impairments can have serious effects on local or regional economics.

2.3 Characteristics of Pig Manure Odor

2.3.1 Odorous Compounds in Pig Manure

Understanding the odorous compounds and their sources in pig manure is necessary for putting into place effective control measures in pig production. A considerable amount of research has been conducted to determine odorous compounds in pig manure (O'Neill and Phillips, 1992a; Roderick et al., 1998). In general, odorous compounds are produced and accumulated during collection, handling, storage, and spreading of pig manure. The main source is anaerobic protein and carbohydrate degradation. Microbial activities are normally considered to be responsible for malodor generation from stored pig manure slurry (Zhu, 2000). In all, 168 chemical compounds have been identified in pig odors (O'Neill and Phillips, 1992b). Over 30 of these compounds have an odor detection threshold at a concentration under or equal to 0.001 mg/m3. These compounds can be classified into four different chemical classes: VFAs, indoles and phenols, volatile sulfur-containing compounds and ammonia and volatile amines.

Volatile fatty acids, produced from deamination of amino acids, mainly consist of acetic, propionic, butyric, iso-butyric, valeric, iso-valeric, caproic, and capric acids. The pH in the intestinal tract of pigs is normally maintained between pH 6–7, which is suitable for deamination of amino acids resulting in production of VFAs, CO_2, H_2, as well as ammonia (Zhu, 2000). Furthermore, anaerobic microbial fermentation contributes substantially to VFA production. The fermentation gases (CO_2 and CH_4), in particular, come from structural carbohydrates in the feed. Methane is odorless, but it significantly contributes to the greenhouse effect due to the large quantities produced. In the United States, it is estimated that CH_4 emissions from animal waste (50 % from pig) are responsible for 15% of total CH_4 production from human activities (Roderick et

al., 1998).

Phenolic compounds are mainly produced from microbial degradation of tyrosine and phenylalanine. When dietary protein concentration is increased, a higher level of amino acid fermentation occurs in the colon as indicated by urinary phenol excretion and fecal ammonia concentrations. But when increasing the amount of fermentable carbohydrate in the diet, this effect was largely reduced.

Sulfur-containing compounds are produced from reduction of sulfate and metabolism of sulfur-containing amino acids. Odorous compounds with the lowest detection threshold generally contain sulfur (O'Neill and Phillips, 1992b).

Ammonia volatilization is the main source of air pollution eminating from pig manure storages and is closely related to odor. Ammonia, a very pungent and irritating gas, is emitted in large quantities and most likely results from the decarboxylation of amino acids during the storage of fresh manure and from urea and nitrates as well as amino acid deamination (Zhu, 2000). Ammonia can also be incorporated into polyamines, such as putresine and cadaverine, which are major components of the odor. Generally, ammonia emissions start right after the manure is excreted and continue after it is spread on land.

2.3.2 Factors Affecting Manure Odor

The extent of manure odor depends on the handling systems employed during collection, storage and spreading (Miner, 1999; Zhu et al., 1997a). For example, it is usually believed that manure odor is strongest with liquid handling. Liquid manure systems can be anaerobic or aerobic. According to John and Wilson (2003), when a manure pond is deeper than 0.6 m, especially deeper than 1.22 m, the lower portion of the pond will be anaerobic. It is known that anaerobic and aerobic ponds have a different odor. Basically, an aerobic pond has a more constant yet less intense odor

year-round. In contrast, anaerobic ponds may have little odor at some times, and very strong odor at other times, i.e., as seasons change.

Extended storage time has very important effects on pig manure odor production. After excretion, solid and liquid animal waste is subjected to microbial conversions (mainly anaerobic), which start within 24 hours and convert organic substrates into microbial biomasses and soluble and gaseous end points (Roderick et al., 1998; Schiffman, 1998). O'Neill and Phillips (1991) reported that the concentration of malodorous compounds greatly increased upon anaerobic storage over a 24 hour period. Phenol levels increased by 140%, while indoles and total sulphides increased by 160% and 1350%, respectively. Interestingly, it was found that odor emission inside the building was reduced by 50% when slurry was removed, in comparison with that of the room with the slurry stored under slotted floor (Guingand et al., 1997).

Production of manure odor is obviously affected by the content of the feeds, such as the particle size of the diet, dietary supplemental enzymes, dietary protein and fermentable fibre, and dietary phosphorus content (Adeola et al., 1995; Jongbloed and Lenis, 1992; Ruurd et al., 2001; Sutton et al., 1999). Some of related research papers will be discussed in the following section of control measures.

Furthermore, production of manure odor from slurries is affected by environmental conditions, i.e., temperature, oxygen content, humidity, and air exchange rates and by pH, buffering capacity, and dry matter content of the slurry (Sutton et al., 1999).

2.4 Control of Environmental Impact of Pork Industry

2.4.1 Control of Soil and Water Pollution

Improved feeding strategies or nutritional management can reduce soil and water pollution from pig wastes. In order to control soil and water pollution, N, P and K concentrations in pig manure have to be controlled and maintained within the range of

crops' requirements. The most effective approach is to reduce the total amount of manure and/or to decrease its nutrient content. Dietary manipulation has been shown to be an effective method of achieving these goals. These manipulations include the formulation of diets on an ideal protein basis, reduction of crude protein (CP) content of diets, increasing the digestibility of the feedstuffs, and supplementation with feed additives, etc (Kornegay and Verstegen, 2001; Lee and Kay, 1997).

Amino acid (AA) supplementation of low-CP diets decreases N excretion from 3.2% to 62%, depending on the size of the pig, level of dietary CP reduction, and initial CP level in the control diet (Kerr et al., 1995). Carter et al. (1996) and Sutton et al. (1996) reported that a reduction of 3 % (from 13 to 10%) units in CP and a supplementation of the corn-soybean meal growing-finishing diets with AAs decreased total N excretion by approximately 28 to 36% in freshly excreted manure. When Sutton et al. (1997) fed growing-finishing pigs a corn-soybean diet with 10% CP supplemented with synthetic essential AAs and 5% cellulose, the excretion of N in urine, along with the total N and ammonia in fresh manure (urine and feces) were reduced by 49 % and 33 %, respectively. Meanwhile, dry matter content of the manure was increased by 50% compared to a 13% CP control diet. Ammonium and total N concentrations in stored manure were reduced by 73 and 35%, respectively. Hobbs et al. (1996) showed that reducing the CP from 21 to 14% in growing diets and from 19 to 13 % in finishing diets, alongside synthetic AAs supplementation, reduced N excretion by 40 % and reduced concentrations of the majority of odorants in the slurry. In addition, the volume of slurry and N content was reduced by 28 % and 40 %, respectively, when feeding a low CP (16.5 %) diet supplemented with synthetic essential AAs instead of feeding a regular commercial 22.5 % CP diet (Kay and Lee, 1997). All these studies demonstrate that synthetic AA supplements reduce manure N levels. However, supplementation of fiber sources in pig diets may decrease growth performance, and the feeding of a reduced-CP, AA-supplemented diet has been shown, in some cases, to be detrimental to growth performance and carcass traits (Kerr et al., 1995; Tuitoek et al., 1997). Moreover, the

cost of these synthetic AAs makes this option less attractive.

Increasing the digestibility of the feedstuff can also decrease the amount of nutrients excreted in manure. According to Jongbloed and Lenis (1992), the P in cereal grains has a low digestibility of 20 % to 40 %, indicating that 40 to 60% of the P is undigested and excreted in the feces. In contrast, organic P in meat and bone meal and inorganic P in monocalcium phosphate and dicalcium phosphate have a comparatively higher digestibility of 70 to 80%. Furthermore, supplementation with enzymes such as cellulase and phytase in the feed are confirmed to be effective methods for increasing the digestibility of the feedstuffs. Valencia (1996) fed piglets with a phytase-supplemented diet for 4 weeks and found that supplemental microbial phytase in a low P corn-soybean meal diet released some minerals from the phytic acid complex, improving animal performance, reducing need for inorganic P supplementation, and reducing P excretion in the feces. Jongbloed et al. (1991) reported that in a corn-soybean-wheat pig diet, phytase increased the P digestibility by 36%. Furthermore, it was reported that phytase can not only reduce P content in manure by 5%, but it can also decrease N content by 5 % in the manure. In this study, cellulase addition was showed to reduce N and P by 5 and 25-30 % respectively (Williams and Kelly, 1994).

As mineral supplementation, zeolite has been found to increase feed efficiency, reduce ammonia volatilization and control odors (Mumpton, 1999). The applications of zeolite in animal industry will be discussed in detail in the next section.

2.4.2 Control of Odor Pollution

Controlling ammonia emissions and odors from pig manure can be achieved in different ways: by manipulating the pig's diet, controlling the building environment, applying different manure treatments, improving the design of manure storage tanks and finally, using less odorous spreading methods (Airoldi et al., 1993; Barrington, 1996; Burnett and Dundero, 1970). Different measures should be taken depending on

the characteristics of manure at the different stages of manure production and handling.

2.4.2.1 Control of Odor from Housing

Manure odor is released into the atmosphere by the ventilation system. In order to control potential manure odor, three factors including the ventilation system, floor design, and dust control have to be considered.

Generally, the major carriers of odors in the pig building are gases from manure, dust, and water vapor. A well designed and managed ventilation system should be adequate to prevent the buildup of all three carriers, as well as decrease the total bacteria. For example, the application of sidewall ventilation is thought to be able to move large volumes of air, dilute the concentration of particles inside the building as well as in the air exhausted from the building (Pig Odor Task Force, 1995). Although, the application of ventilation systems has been very effective in controlling the impact of pig manure odors, these effects are mainly limited to the inside of the building for the pigs and workers.

In order to control the environmental impact of manure odor, it is essential that the air exhausted from the pig building is clean and odorless. In pig production, several methods, such as a biofilter, a bioscrubber, a thermal incinerator, a catalytique incineration, an absorption system and diffusion chimneys have been developed. For the biofilter (Young et al., 1997) and bioscrubber (Dong et al., 1997; Siemers and Van den Weghe, 1997), when the odorous air from pig buildings passes through a filter, the inside biological materials could breakdown volatile compounds into carbon dioxide, water, mineral salts and other harmless products. Thus, 90% or more of the volatile organic compounds can be removed. In addition, they create no secondary pollution, and are efficient in treating low concentrations of odorants (less than 20 ppm) (Pig Odor Task Force, 1995). However, the use of biofilters and bioscrubbers are not practical in pig production since they increase production costs.

Floor design can have a large impact on odors generated from a piggery. In general, solid concrete floors with scrapers or small flush gutters are easy to increase the production of odor compared to wet floors. In contrast, wet, manure covered surfaces emit more ammonia and other odorous compounds than slotted floors. Amongst all types of slotted floors, a pit ventilation system is best for maximally controling dust and air contaminants, since "pit ventilation" ensures that fresh air is available at the animal level, and keeps the slats dry (Choiniere et al., 1997; Lavoie et al., 1997).

Dust is generated from feed, manure, and the animals themselves. It can carry gases and adsorb odors from within the building (Carpenter, 1986). High dust concentrations can also be a health risk for workers in pig facilities as well as the pigs (Hoff et al., 1997). Reynolds et al. (1996) reported respiratory problems among pig workers after they were exposed to ammonia and total dust levels of 7.5 ppm and 2.5 mg/m^3. Lau et al. (1996) observed that the pig's body weight increased 0.04 kg/day while the incidence of lung score decreased 35 to 40 % and snout score reduced 25 to 40 %, when the level of dust was reduced by 20 to 52% with an electrostatic precipitator filter. Furthermore, because dust can carry pathogenic microorganisms (Carpenter, 1986), it is a very important medium for transmission of some diseases when it enters air currents. Therefore, by controlling dust in the buildings, the quality and amount of odor carried outside by the ventilation systems could be greatly reduced. Meanwhile, the concentrations of some malodorous gases, i.e. ammonia and hydrogen sulphide, inside pig facilities could also be decreased.

The first principle for controling dust inside the pig facility is to clean interior building surfaces regularly. The "all-in, all-out" style of management is a commonly used method for modern commercial pig production facilities to reduce dust levels. An alternative way to control dust is to use oil in the feed and on the floor, as well as to spray pigs with oil or water. In a study conducted by Takai et al. (1993), spraying

rapeseed oil at a concentration of 5 to 15 ml per pig daily on the floors of a piglet room reduces respirable dust by 76%. In another study performed by Zhang et al. (1994), the respirable and inhalable dust was reduced by 75% with a mineral oil on the floor of a grower finishing unit. Perkins and Feddes (1996) also reported a reduction of dust, by 73%, during a 24 hour period following the weekly application of 24 ml/m^3 of mineral oil on the floors of a pig farrowing unit. Addition of oil to dry pig rations can significantly reduces the amount of dust in a building. When 5% soybean oil was supplemented to a starter diet, the dust concentration was reduced by 45% (Gore et al., 1986). When tallow was included into the pig's ration at a level of 2.5 and 7.5%, aerial dust was decreased by 21 to 53% (Chiba et al., 1985, 1987).

2.4.2.2 Manure Treatment during Storage

In order to take appropriate measures to control the environmental impact of pig manure storage, the length of time manure spends in storage must be taken into account (Nicholson et al., 2002). These measures, which can be applied either individually or in combination, include using tank covers, composting, aeration, anaerobic digestion, aerobic digestion, separation and pit additive applications.

Obviously, manure tank covers can reduce the emission of odor, ammonia and hydrogen sulfide. The effect depends on the characteristics of covering materials applied. Effective odor control of above-ground concrete and enamelled steel tanks could be achieved using a variety of cheap covers. For example, more effective control was obtained by erecting a light wooden/metal framework, which was used to support a strong plastic sheet (Nielsen, 1987).

Aerobic digestion is a good method for reducing offensive odors (AI-Kanani et al., 1992; Berg and Hornig, 1997; Burton, 1992). The basic principle of aeration is to provide, by whatever means, enough dissolved oxygen to aerobic bacteria so that they can actively decompose the odorous compounds into chemically stable materials, such

as oxidizing carbohydrates to carbon dioxide and water, thereby reducing both pathogens and odor (Williams et al., 1984; Williams et al., 1989; Sneath et al., 1992). Some bacteria initially convert N compounds to ammonium and then to nitrites by nitrification. Importantly, aerobic treatment of wastes does not produce VFAs and various other compounds associated with very offensive odors. In a study conducted by Sneath et al. (1992), odors could be reduced by 70% using a farm scale aerobic digester. Once the solids are removed, slurries treated aerobically become more stable and produce fewer odors when they are subsequently stored and applied to land than treated before. Sneath et al. (1992) reported that a 1.5% DM slurry could remain stable and odorless for up to 30 days following a 4-day aerobic treatment. The main disadvantage of aerobic treatment is that it generally requires power to aerate the materials.

Anaerobic lagoons use anaerobic microorganisms to convert biodegradable organic materials to odorless gases, such as methane and carbon dioxide, and nonbiodegradable solids (Zhu, 2000). In a laboratory scale batch, chemical oxygen demand (COD) of the manure was reduced by 73% and the manure was relatively odorless when using a psychrophilic anaerobic digestion (Masse et al., 1997).

Many pit manure additives are available for treating or preventing odors in animal facilities, manure storage tanks, and lagoons (Zhu et al., 1997a). Most of these products can be classified into 5 different categories: masking agents, counteractants, digestive deodorants, adsorbents and chemical deodorants. Masking agents are mixtures of aromatic oils used to cover up an objectionable odor with a more desirable one (Pig Odor Task Force, 1995). According to the report by Ritter (1989), counteractants are aromatic oils that cancel or neutralize an odor so that the intensity of the mixture is less than that of its constituents. Both products are effective in short-term control of wastes since they are quickly broken down by bacteria in lagoons and tanks. Digestive deodorants contain bacteria and/or enzymes that eliminate odor and suppress gaseous pollutants by their biochemical digestive processes, and are effective to break down

solids, reduce the release of ammonia and conserve N when the added bacteria become predominance in the manure. Adsorbents such as sphagnum peat moss, limestone and zeolite have a large surface area to adsorb the odor-causing chemicals before they are released into the environment. Chemical deodorants have two categories: one being strong oxidizing agents such as hydrogen peroxide, potassium permanganate, and ozone which can chemically oxidize odor-causing compounds, and the other being germicides such as orthodichlorobenzene chlorine, formaldehyde, and paraformaldehyde which can alter or eliminate bacterial action responsible for odor production. Each of these groups has its strengths and limitations. But all these chemicals are corrosive and harmful to the environment.

2.4.2.3 Odor Control during Spreading

The largest number of complaints concerning odor from pig manure by the public come within the first 24 hrs after spreading and when slurry or solid manure remains on the surface for several days. In fact, odor is most intense during the first few hours after spreading and decreases exponentially with time, with small daily fluctuations. The complaints generally come from residents living within 900m downwind of the field on which manure has been spread; however, under unusual weather conditions, the complaints can come from as far away as 3000m downwind (Nielsen, 1987). In general, 70% of complaints about pig odor come from manure spreading, 10% from the pig building and the remaining 20% from the storage facility (Pig Odor Task Force, 1995). Obviously, spreading manure on top of the soil can cause high odors; however, odor can be reduced if manure slurries or lagoon sludges are injected or incorporated into the soil immediately after application. Therefore, to reduce odor emissions and ammonia volatilization, manure should be spread as close as possible to the ground or, even better, directly incorporated into the soil. In a study conducted by Morken and Sakshaug (1997), manure could be injected into the soil to a depth of 5 to 10 cm by using a new direct ground injector which pressurizes manure into a series of 13 mm nozzles placed directly on the ground. The results showed that ammonia volatilization was reduced by

up to 90% and all possible sources of run-off were removed. Unfortunately, injection is not the universal answer, since it will not work either in soil with high water table or in soil which is very dry, hard or stoney. The choice of system depends on the farm situation and has to be set against the additional costs and risk of complaints.

Several countries have implemented regulations about the timing of manure-slurry applications, the locations, methods, and rates of application. The spreading of pig manure is typically limited to spring and early summer when crops are growing rapidly. In the Netherlands, manure has to be incorporated into the soil within 24 hours after spreading by regulations. Most importantly, pig producers must analyze the amounts of P and N contained in animal wastes and balance them with the estimated needs of the crop that willl be sown on that particular field (Pig Odor Task Force, 1995).

2.4.2.4 Feed Manipulations

As mentioned above, primary odor-causing compounds result from excess degradable proteins and lack of specific fermentable carbohydrates during microbial fermentation. The concentrations of odorous compounds in the manure vary, depending on the variety of pigs and the diet fed. As a general rule, N is the key ingredient in ammonia and many other odorous compounds. When the amount of protein in the diet is poorly balanced or protein is fed in excess of what can be efficiently utilized, the animal will excrete the excess in its feces and urine (Baidoo, 1996). The higher the N content of pig manure the greater its potential odor. If N in the manure is reduced by 100 units, the odor level will be decreased by 75 units (Swine Odor Task Force, 1995). Therefore, how to manipulate the diet to increase nutrient utilization and reduce excretion products, and how to enhance microbial metabolism in the GIT to reduce excretion of odorous compounds in the manure have become hot topics and urgent research areas for pig producers and scientists all over the world. There are several approaches such as adding essential AAs and complex carbohydrates, increasing the digestibility of proteins, changing feeding style and adding odor absorbers, enzyme and

microbes.

By substituting synthetic AAs for traditional protein sources, N excretion by pigs and odor production can be decreased significantly. Hobbs et al. (1996) reported that the concentrations of a majority of odorants in the slurry were reduced when diets were supplemented with synthetic AAs and CP was decreased from 21 to 14% in growing diets and from 19 to 13 % in finishing diets. In an in vitro study, ammonia emissions from manure was reduced by 79 % and 58 % in diets supplemented with synthetic AAs and when CP was decreased from 16 to12% in growing diets and from 14 to 10% in finshing diets, respectively (Turner et al., 1996). On average, synthetic AAs reduce ammonia emissions by 40% and thus decreasing the odor emissions by 30% (Denis, 1999).

The addition of fermentable carbohydrates in the pig ration could also reduce odor emission. The principle is to reduce N excretion in urine, as urea, and shift the N excretion in feces to a form of bacterial protein, resulting in a change in the ratio of N excretion in urine and feces and thus reducting ammonia volatilization (Sutton et al., 1999). Complex carbohydrates such as β-glucans, nonstarch polysaccharides (NSP), and specific oligosaccharides were reported to influence endogenous N excretion at the terminal ileum and microbial fermentation in the large colon, resulting in increased bacterial protein production and altered VFA production (Canh et al., 1997; Sutton et al., 1999).

Supplementation of some enzymes, i.e., cellulases and phytases, in the feed has been demonstrated to be effective to reduce the pig manure odor production (Cromwell et al., 1993). For example, use of proteolytic enzymes in feed processing or as dietary supplements in the diet has been reported to be able to improve protein digestibility and subsequently reduce odor emission (Swine Odor Task Force, 1995).

The change of feeding style affects the emission of some odorous compounds in pig manure. It was found that odorous compounds including NH_3, sulfides, VFA, phenols, and indoles in pig slurry were modified by liquid feeding (Hobbs et al., 1997). Slurry from weanling pigs fed a 4:1 or 3:1 water to feed ratio diet contained only 13% and 31% odorous compounds, respectively, compared to the control dry feed.

Odor absorbents such as calcium bentonite, sagebrush and charcoal could be added to the pig's diet to absorb ammonia produced in the GIT of pig (Swine Odor Task Force, 1995). In addition, zeolite has been reported to be a potential odor absorbent to reduce odor emissions from pig manure. Moreover, several reports have shown that zeolite could be an effective diet additive to improve pig performance.

2.5 Zeolite

Since the discovery of Zeolite in 1756, it had been considered by geologists to be fairly large crystals with vugs and cavities and as a mineral collector. In the late 1950s, the natural zeolite was identified as crystalline, hydrated aluminosilicates containing positively charged metallic alkali ions and alkaline earth elements (Pecover, 1987). The crystals are characterized by SiO_4 tetrahedron where all four corner oxygen ions are shared with a central tetrahedral ion of silicon atoms (Si) or aluminum atoms (Al), single or double rings and a larger symmetrical polyhedra which form an infinite, open, three-dimensional framework. Because of its structure, zeolite has interconnecting channels and large voids which are capable of trapping molecules of proper dimensions, without significant structural deformation (Mumpton and Fishman 1977). With this in mind, synthetic zeolites are also referred to as molecular sieves.

2.5.1 Characteristics of Zeolite

Because of its large structural cavities and entry channels, zeolite has special chemical and physical properties, i.e., it can lose and gain water reversibly without changes in the crystal structure. In addition, because some Si^{4+} in the structure are replaced by trivalent aluminum atoms (Al^{3+}), causing increasing deficiency in positive

charge, zeolite can selectively adsorb and exchange extraframework cations. Normally, the cation-exchange capacity (CEC) of a zeolite is between 2 to 4 milliequivalentsyg (meqyg), about twice the CEC of bentonite clay, depending on the amount of Al which substitutes for Si in the tetrahedral framework (Mumpton and Fishman, 1977). Usually, naturally occurring zeolite has a relatively small CEC (2.25 meqyg). Since its cation selectivity is $Cs^+ > Rb^+ > K^+ > NH_4^+ > Ba^{2+} > Sr^{2+} > Na^+ > Ca^{2+} > Fe^{2+/3+} > Al^{3+} > Mg^{2+} > Li^+$, one expects a release of Na^+ or Ca^{2+} and uptake of K^+ and NH_4^+ in the GIT (Mumpton , 1999).

In addition, zeolite minerals have been demonstrated to possess the biological properties, such as high capacity for immobilization of microorganisms and for improving the ammonia/ammonium ion equilibrium, that make zeolite capable to reduce the ammonia and ammonium ions in solution.

There are over 45 different zeolites. Among them, sodium zeolite A (SZA) and clinoptilolite (CLI) are often used (Cefali et al., 1995). The characteristics of these two zeolites are different, depending on the size of the openings between their lattice work. SZA is produced synthetically with a CEC of 700 meq/100g (Cook et al., 1982). It is rich in exchangeable Na (12.5%), but has the highest affinity for Ca. Thus, SZA has generally been used to improve the adsorption of Ca and to exchange ions in order to reduce the toxicity of excess salts, as can be found in poultry feed (Fethiere et al., 1994; Rolland et al., 1985) and in dairy cow diets (Enemark et al., 2003a,b; Thilsing-Hansen et al., 2002a, b; Jorgensen et al., 2001). For example, in chicks, SZA was able to counteract the adverse affects of excess dietary Ca (Elliott and Edwards, 1991; Watkins et al., 1989). In dairy cows, SZA can replace sodium bicarbonate supplementation to improve the digestibility of feed without any negative effect on rumen function (Johnson et al., 1988; Holthaus et al., 1996).

In contrast, the CEC of CLI is 120 meq/100g. It releases Na^+ or Ca^{2+} to take up K^+. Moreover, CLI has a special affinity for NH_4^+ and has been used to improve N absorption in feed (Mumpton and Fishman, 1977). In general, CLI has been used in pig feed.

2.5.2 Applications of Zeolite

2.5.2.1 General Applications

Since its discovery, zeolites have been used for a multitude of applications in industry, agriculture, veterinary medicine, sanitation and environmental protection because of their physicochemical properties. Based on their unique attractive adsorption, related molecular sieving, cation-exchange, dehydration–rehydration, and catalytic properties, natural zeolites, including those found in volcanic sedimentary rocks, have been used as building dimension stone, lightweight aggregate and pozzolans in cements and concretes, filler in paper, drying of acid-gases, separation of oxygen from air, uptake of Cs and Sr from nuclear waste, mitigation of radioactive fallout, energy exchangers in solar refrigerators, and deodorizing agents to remove malodors from shoes, athletic footwear, as well as pet litter to absorb water and odor-causing NH_3 from animal urine (Bundy et al., 1997; Mumpton, 1999).

According to Mumpton (1999), some zeolites are used for medical purpose. They can be applied, for example, as antidiarrheal remedies, as effective filters to remove NH_4^+ from the dialysate of kidney patients during hemodialysis, as buffers to reduce stomach acidity and to treat stomach ulcers in Cuba, as external powder to treat athlete's foot and to decrease the healing time of wounds and surgical incisions.

Since the late 1970s, natural zeolites have been used in large-scale cation-exchange processes such as clinoptilolite cationexchange columns to reduce the ammonia content in municipal wastewater and drinking water in many countries, such as Russia and Ukraine (Liberti et al., 1995).

Interestingly, natural zeolite is reported to have a high ability to improve the yield of growing crops and vegetables in agronomy and horticulture. It can be used as soil amendments to improve CEC and water absorption capacities, as soilless zeoponic substrates for greenhouses and space missions, and as dusting agents to kill aphids afflicting fruit trees (Mumpton, 1999). Furthermore, it has been found that natural zeolite has important uses in aquaculture such as removal of ammonium from hatchery, transport, and aquarium waters, generating oxygen for aeration systems in aquaria and during transport, and supplementing fish rations.

2.5.2.2 Applications of Zeolite in Animal Production

Since 1965, a variety of zeolites have been used as dietary supplements for various species in several countries. CLI, being a zeolite of the heulandite group and the most abundant zeolite in nature, is the most widely used natural zeolite with animals.

2.5.2.2.1 Improving Animal Performance

Several studies have demonstrated that zeolite supplementation can improve the production performance of animals and the digestibility of some nutrients in the feed (Pond, 1984, 1989; Pond et al., 1989; Poulsen and Oksbjerg, 1995). Nestrov (1984) extensively studied the effect of zeolite in the diets of beef cattle, sheep, pigs and poultry, and found an improved weight gain with the inclusion of zeolite in the diets of all animals studied. Furthermore, evaluation of the meat from these animals did not show any detrimental effect of zeolite supplementation on quality. Sweeny et al. (1984) demonstrated improved digestibility of N, organic matter (OM) and acid detergent fibre with a 5 % CLI supplementation to the diet of growing steers and heifers. Mumpton and Fishman (1977) showed improved body weight gain and efficiency of feed utilization when natural zeolites were added to the feed of pig (Papaioannou et al., 2002) and poultry (Olver, 1997). This is further supported by Vrzgula and Bartko (1984) who studied the effects of feeding 5% supplemental CLI diet in pigs. The results showed an increase in weight gain of 0.49 kg/week of pigs as compared to the control animals. In

addition, the pigs fed CLI produced less odoriferous feces and those with diarrhea produced firmer feces within 24 hours of testing. Meanwhile, they did not detect any unfavorable effects of CLI supplementation on the liver function of the test animals. Barrington and EI Moueddeb (1995) also reported an improved feed conversion by 0.15 kg of feed per kg of body weight gain with 5 % zeolite (77 % CLI) in pig feed.

However, not all experiments show improved weight gain with the addition of zeolite. Pond and Yen (1982) fed growing pigs with 5 % or 10 % CLI supplementation and did not show positive effects on body weight gain and feed conversion rates. In addition, some blood traits including Ca, Mg, P, alkaline, and protein concentration remained unchanged. In contrast, the weight gain of broiler chickens was slightly decreased with a diet of 5 % CLI than those on the normal diet (Mumpton, 1984). However, the apparent feed efficiency was increased and the mortality rate was decreased due to the zeolite diet. It seems that the effect of zeolites depends on the species and the geographical source of the involved zeolite, its purity and physicochemical properties (i.e., CEC), as well as the supplemental level used in the diets (Mumpton and Fishman, 1977; Pond et al., 1988). CLI with a CEC of 100 to 140 meq/100g and fed at levels ranging from 2 to 7.5% generally improve cattle and pig performances. When less than 1 % is fed, CLI and natural SZA had no effect (Ward et al., 1991). Different species of zeolites, along with their complexed physiological processes determine their effects on an animal's digestion and absorption of ingested diet constituents.

CLI can also improve the reproduction of pregnant sows. Ma et al. (1979) reported that providing a diet containing 5 % CLI to pregnant Landrace sows increased litter size at birth. However, in another test, addition of 5% CLI did not show any significant effect on embryo survival and total ovarian weight, 24 days after inseminating sows (Ma et al., 1984).

Kyriakis et al. (2002) added 2% CLI in sow diets throughout pregnancy and lactation periods. From this experiment, it was found that the treated sows/gilts produced larger litter sizes and higher mean piglet body weights at birth (increased by 13%) and at weaning (increased by 63%). In another study by the same author, addition of 5% CLI to the rations of pregnant sows during 20–90 days postmating also increased litter size at birth by an average of 1.78 piglets. In contrast, when feeding crossbred sows with 2.5% to 5% CLI diets from 2 to 3 weeks before mating, decreased ovulation rates along with normal embryo-survival rates were observed. Moreover, the dietary use of 4-7% zeolite (65% CLI) could increase litter size at both birth and weaning by 6% and 13.7%, respectively. In the experiment by Kyriakis et al (2002), there were no confirmed adverse side effects from CLI supplementation in diets of pregnant and lactation pigs and there were no alterations in the serum concentrations of certain vitamins (Vitamin E and A) and mineral elements (inorganic P, K, Cu, Zn).

2.5.2.2.2 Taking Up Ammonia and Other Toxic Agents

The addition of zeolite to feed can not only improve the performance, but it can also reduce odor emissions in the manure of treated animals and the accumulation of toxic substances in tissues. Since some types of zeolite, especially CLI, have a high affinity for N and sulphur compounds, they can adsorb the harmful ammonia produced by the intestinal bacteria and can probably slow down the passage of feed to the intestinal tract. Barrington and EI Moueddeb (1995) fed growing finishing pigs 5 % zeolite (77% CLI) and observed a 75% reduction of NH_3 volatilization on average. Consequently, odor levels were decreased by 1 point on a scale of 0 to 5. In the study conducted by Bartko et al. (1993), improved growth rate of animals along with reduced manure NH_3 and odor emissions were also observed with the inclusion of 5% CLI in the feed.

Ward et al. (1993) reported that zeolite prevented or minimized the number and severity of intestinal diseases due to the uptake of NH_4^+, produced by the deamination

of dietary proteins during the digestive processes, via the intestinal wall (Pond et al., 1988). Ammonia is a cell toxicant in animals, therefore maintaining its levels below toxic levels could reduce epithelial turnover in the intestinal tract, spare energy and promote better nutrient utilization. Furthermore, CLI can efficiently remove other toxic agents from the GIT of pigs (Shurson et al., 1984), e.g., p-cresol, which is produced by anaerobic degradation of tyrosine and is responsible for depressing effect. Milan et al (2001) also demonstrated that zeolite reduced the concentration of NH_3 and NH_4^+ in solution, and subsequently eliminated the inhibitory effect of these compounds. In an experiment by Bernall and Lopez-Real (1993), both synthetic and natural zeolites were introduced into the rumen of test animals to reduce the toxic effects of high NH_4^+ in ruminal fluids, when animal diets were mixed with nonprotein-nitrogens, such as urea and biuret. It was found that NH_4^+, produced by enzyme decomposition of the nonprotein-N, could be transfered immediately to the zeolite and then held for several hours until released by salivary Na^+ entering the rumen. This function allows rumen microorganisms to efficiently synthesize cellular protein for absorption by the animals' digestive systems.

In addition, an NH_4^+-containing zeolite may promote the growth of N-loving bacteria which contribute to the health of the animals. When used in adequate levels, CLI has a significant effect on adsorption of minerals and water in the GIT of animals, therby maintaining the quality of the meat or product. Also, zeolite in the gut system may adsorb deleterious heavy metals, and result in fewer or less severe stomach ailments (Mumpton, 1999).

2.5.2.2.3 Preventing Diseases

CLI has the ability to absorb aflatoxins in contaminated animal feeds and further protect livestocks and poultry against the toxic effects of mycotoxins and their carryover into animal products (Ortatatli and Oguz, 2001). Moreover, zeolite is efficient in preventing hypocalcaemia in dairy cows around calving (Thilsing-Hansen et al.,

2002a,b).

2.5.2.2.4 Animal-Waste Treatment

Natural zeolites could be used to reduce the malodor, increase N retention of animal waste and purify the methane gas produced by the anaerobic digestion of manure (Milan et al., 2001). Several million tons of zeolites have been used in deodorizing animal litter and barns in some countries (Mumpton, 1999).

2.6 Conclusion

As mentioned above, some research projects show positive effects of zeolite supplementation on the performance of grower pigs, while other research projects show no effects. Few research projects have identified the mechanisms by which zeolite improves feed digestion and reduces the environmental impact of pig production. The objective of this project is to provide knowledge on some of these missing elements.

CONNECTING STATEMENT

To find an economically, viable and promising solution to the environmental problems challenging the pork industry, zeolite (90%+ clinoptilolite) has been tested as a feed additive. Zeolite is known to improve the productivity of the animals while reducing the nutrient content of their manure. Zeolite has the ability to exchangeably adsorb ammonia while releasing minerals inside and slowing down the passage of feed through the gastrointestinal tract of pigs. The result is an increasing of feed digestion efficiency and a reduction in the environmental impact of manure. Chapter 3 presents a test conducted to measure the effects of zeolite as a feed additive on pig growth performance and metabolic processes.

Chapter 3 deals with a trial where zeolite (90%+ clinoptilolite) was used in the diet of growing pigs at level of 4% and using three different combinations of protein and energy levels. The effects on growth performance and metabolic parameters of pigs in growing/finishing pens and metabolic cages were monitored and compared.

This paper will be submitted for publication in Journal of Animal Science. The manuscript was authored by Yonghong Wan, Barrington, S.F. and Xin Zhao. The first author carried out entire experimental work, collected and analyzed the data and drafted the article. The second and third authors cosupervised the research and co-edited the article.

CHAPTER III

Metabolic Effect of Zeolite as Natural Feed Supplement for Grower Pigs

Y.H. Wan[1], S. Barrington[2], X. Zhao[1]

[1]Department of Animal Science and [2]Department of Bioresource Engineering
Macdonald Campus, McGill University
21, 111 Lakeshore Road, Ste. Anne de Bellevue
Québec, Canada H9X 3V9.

Presented to the Journal of Animal Science

Corresponding author: Xin Zhao
Department of Animal Science,
Macdonald Campus of McGill University,
21 111 Lakeshore, Ste Anne de Bellevue
Québec, H9X 3V9
514-398-7975 (tel)
514-398-7964 (fax)
xin.zhao@mcgill.ca

3.1 Abstract

Clinoptilolite (CLI), a type of zeolite, has been used as a feed additive to improve feed digestion efficiency of pigs and reduce the nutrient content of their manure. This study was designed to determine the metabolic response of pigs fed zeolite (90%+ CLI) supplemented rations. Twelve male and twenty four female growing pigs, with an average initial body weight (IBW) of 50.18 ± 2.95 kg and 48.95 ± 5.25 kg, respectively, were randomly allocated into four different dietary treatments in growing/finishing pens. Ration 1 was the control diet with no zeolite and 100% crude protein (CP) and energy requirements; ration 2 contained 4% zeolite with 100% CP and energy requirements; ration 3 contained 4% zeolite with 90% CP and energy requirements; and ration 4 contained 4% zeolite with 90% CP and 85% energy requirements. The metabolic test was carried out in 12 metabolic cages and repeated with three batchs of animals. After being placed in growing/finishing pens for two (batchs 1 and 2) or three (batch 3) weeks, three pigs under each treatment were randomly assigned into individual metabolic cages and observed for an additional 8 days. Daily feed consumption was measured. Feces and urines from each pig were daily collected and measured after 3 days for adjustment. Feces and urine samples were also analyzed for dry matter and organic matter content. All pigs were weighed before and after the metabolic cage period to calculate feed conversion and body weight gain. Our results demonstrated that ration 2 had a consistently positive, but not significant ($P > 0.05$), effect on pig performance and metabolic parameters, compared to ration 1. In addition, ration 3 also improved feed as well as protein and energy conversion rate, and increased body weight gain, when compared to ration 1. However, ration 4, significantly ($P < 0.05$) increased feed consumption and the amount of feces produced, and decreased feed and organic matter retention in gastrointestinal tract as well as feed conversion rate. Accordingly, zeolite supplementation at 4% to grower pig diets could be a potential promising method for pork producers to control the impact of pig manure on the environment.

3.2 Introduction

The Canadian pork industry has rapidly developed over the past decade. Since 1995, the annual number of pigs produced has dramatically increased by 71.5 %. Furthermore, due to Canadian pork products being internationally recognized for its high quality, the total mass of exported meat and live animals has steadily increased by 164% and 456%, respectively, between 1995 and 2003 (Statistics Canada, 2003). Along with increased production, the number of pig farms in Canada has decreased by 27% between 1996 and 2001 (Statistics Canada, 2001). As a result, the average number of pigs per farm unit has tremendously increased (994%) between 1976 and 2001 (Statistics Canada, 2001). The changes in pig production structure have resulted in a smaller number of larger pig enterprises having a larger impact on environment. The production and spreading of excessive dosage of manures on tillable land has resulted in serious environmental problems involving soil, water and air resources. In reality, environmental pollution has become a development bottleneck for the Canadian pork industry.

Supplementation of zeolite in pig diets has been reported to reduce the environmental impact of the manures. Zeolite is a crystalline, hydrated aluminosilicate containing positively charged metallic ions and alkaline earth elements (Pecover, 1987). The crystals are characterized by a SiO_4 tetrahedron where all four corner oxygen ions are shared with a central tetrahedral ion of silicon atoms (Si) or aluminum atoms (Al), and a larger symmetrical polyhedra which forms an infinite, open, three-dimensional framework (Mumpton and Fishman 1977). Because of the large structural cavities and the entry channels in the structure, zeolite has special chemical and physical properties. Zeolite can lose and gain water reversibly without changes in crystal structure. Since some Si in the structure are replaced by trivalent aluminum atoms (Al^{3+}) resulting in positive charge deficiencies, zeolite can selectively adsorb and exchange extraframework cations. Normally, the cation-exchange capacity (CEC) of a zeolite is between 2 to 4 milliequivalents/gram (meq/g), about twice the CEC of bentonite clay,

depending on the amount of Al substituting for Si in the tetrahedral framework (Mumpton and Fishman, 1977). In addition, zeolite, as a mineral collector, has interesting biological properties, such as high capacity for immobilizing microorganisms and for improving the ammonia/ammonium ion equilibrium. Thus, zeolite is capable of reducing the ammonia and ammonium ions in solution (Mumpton, 1999).

There are over 45 different types of zeolites. Clinoptilolite (CLI), being the most abundant zeolite in nature, is the most widely used natural zeolite in animal studies because of its structural stability under acidic conditions. Zeolite supplementation can improve the production performance of animals and the digestibility of some nutrients in the feed. Nestrov (1984) extensively studied the effect of adding zeolite in the diets of beef cattle, sheep, pigs and poultry, and reported improvement of weight gain. Furthermore, there were no negative effects on the meat of these animals. Sweeny et al. (1984) demonstrated that 5% CLI in the diet of growing steers and heifers improves the digestibility of proteins, organic matter and acid detergent fibre. Mumpton and Fishman (1977) showed improved body weight gain and efficiency of feed utilization when natural zeolites were added to the feed of pig and poultry. This is further supported by Vrzgula and Bartko (1984) who studied the effects of supplementing pig diets with 5% CLI and reported an increase in weight gain of 0.49 kg/week of pigs, as compared to the control animals. In addition, the pigs fed CLI produced less odoriferous feces and those with diarrhea produced firmer feces within 24 hours of testing. Meanwhile, no unfavorable effects of CLI supplementation were observed on the liver function of the experimental animals. Barrington and El Moueddeb (1995) also reported an improved feed conversion of 0.15kg of feed per kg of body weight gain with 5 % zeolite (77 % CLI) in pig feed.

Furthermore, the supplementation of zeolite to feed can reduce odor emissions in the manure of treated animals and the accumulation of toxic substances in tissues. Since

some types of zeolite, especially CLI, have a high affinity for nitrogen and sulphur compounds, they can adsorb the harmful ammonia produced by the intestinal bacteria and can probably slow down the passage of feed through the intestinal tract. Barrington and EI Moueddeb (1995) fed growing finishing pigs 5% zeolite (77% CLI) and observed a 75% reduction of NH_3 volatilization. Consequently, odor levels were decreased by 1 point on a scale of 0 to 5. Bartko et al. (1993) observed an improvement in growth rate of animals along with reduced manure NH_3 and odor emissions with the diet inclusion of 5% CLI. Since NH_3 is a cell toxicant to animals, maintaining its levels below toxic levels could reduce epithelial turnover in the intestinal tract, spare energy and promote better nutrient utilization. Because of a reduction of odor emission produced by the gastrointestinal track (GIT), zeolite can further prevent or minimize the number and severity of intestinal diseases (Pond et al., 1988; Shurson et al., 1984; Ward et al., 1993).

Moreover, CLI was shown to efficiently remove other toxic agents from GIT of pigs (Shurson et al., 1984). In addition, an NH_4^+-adsorbing zeolite may promote the growth of N-loving bacteria which contribute to the health of the animals. Also, CLI in GIT may adsorb deleterious heavy metals, and result in fewer or less severe stomach ailments (Mumpton, 1999). Furthermore, natural zeolites can reduce malodors, increase N retention of animal waste and purify methane gas produced from the anaerobic digestion of manure (Milan et al., 2001). In fact, several million tons of zeolites have been used in deodorizing animal litter and barns in some countries.

It is plausible that addition of zeolite, especially CLI, to the pig diet could retard the passage of feed and improve the digestibility of nutrients in the GIT. Consequently, the total mass of excretions along with the nutrient content should be reduced. However, very few studies have reported the metabolic effects of zeolite as a feed additive on pigs. The objective of this study was to determine how pig performance and metabolism were affected when pigs consumed a diet supplemented with 4% zeolite (90% CLI).

3.3 Materials and Methods

3.3.1 The Experimental Piggery

The studies were performed at the experimental pig unit of the Macdonald Campus of McGill University, in Ste-Anne-de-Bellevue (Montreal, Quebec) during the summer of 2004. This piggery housed 50 sows, which provided the growing pigs used in this study.

The experiment was run in a growing/finishing room and a metabolic room. The growing/finishing room measured 14.75m by 3.80m and had a ceiling height of 3.05m. There were four pens located along one wall in the growing/finishing room, each pen measuring 3.00m in length by 1.84m in width. The floor of all pens was fully slatted and pigs were allowed to have ad libitum access to standard upright feeders located inside of the pens and to the waterer with a nipple. Each pen held six animals at a density of $0.92m^2$/pig. Two fans ventilated the growing/finishing room, each measuring 300mm and 400mm in diameter, and were controlled by a common thermostat. The ventilation system produces a ventilation rate of 20.0L/s/pig with two fans in operation.

The metabolic room measured 16.25m by 7.6m, with a ceiling height of 3.05m. Three fans, each measuring 300mm, 400mm and 600mm in diameter, controlled by a common thermostat, were used to ventilate the room. During the experimental period, four thermostats were placed in the four corners of the metabolic room to ensure a balanced temperature throughout of 24°C. The ventilation system produces a ventilation rate of 20.0 and 48 L/s/pig with the two smaller fans and all three fans running, respectively. For the metabolic studies, subjects were housed in stainless steel metabolic cages. Totally, there were twelve cages randomly placed in the room.

3.3.2 Experimental Material

3.3.2.1 Experimental Zeolite

The experimental zeolite contains over 90% CLI and was supplied by the KMI mine of Nevada, USA. It has a CEC of 1.6 to 2.1 meq/gm. The CLI percentage of the experimental zeolite was determined using X-Ray Diffraction (XRD) by core Laboratories Canada Ltd. of Calgary, Canada (Table 3.1). The characteristics of this zeolite including its chemical and physical properties and heavy metal content are presented in Table 3.2.

Table 3.1: Bulk composition of experimental zeolite

Element	Formulate	Weight (%)
Quartz	SiO_2	Trace
Plagioclase	$NaAlSi_3O_8$ - $CaAl_2Si_2O_8$	Trace
Calcite	$CaCO_3$	1
Dolomite	$[CaMg]CO_3$	1
Clinoptilolite	$KNa_2Ca_2(Si_{29}Al_7)O_{72}{\cdot}24H_2O$	98
Opal	$SiO_2 .nH_20$	0
Muscovite/Illite	$KAl_2[AlSi_3O_{10}][OH]_2$	0

Determined using XR Diffraction by core Laboratories Canada Ltd. of Calgary, Canada using a 95% pure reference sample.

3.3.2.2 Experimental Rations

The experimental feed was manufactured by Agribrands Purina Canada Inc, of St-Hubert, Québec. The basal diet was based on a standard pig ration of corn as the main energy source and soybean meal as the main protein source. Energy was also supplemented using vegetable and animal fat. Four types of diets were used in the experiment (Table 3.3). The first two rations (R1 and R2) were formulated to meet the nutrient requirements of the National Research Council (NRC,1998) for grower pigs, while the third (R3) offered 90% of the crude protein (CP) and the energy requirement, and the fourth (R4) offered 90% of the CP and 85% of the energy requirements, respectively. Zeolite (90%+ CLI) was incorporated into rations R2, R3 and R4 at inclusion rate of 4 %.

For each diet, three random feed samples were collected and analyzed for dry matter, CP and organic matter. The CP was determined by quantifying ammonium after digestion with sulphuric acid and hydrogen perioxide at 500^0C.

Table 3.2: Characteristics of experimental zeolite

Property	Unit	Value
Properties		
Silica/Alumina Ratio		5.8-6.4
Silica/Aluminum Ration		5.1-5.6
Bulk Density (dry solid)	kg/m^3	1394
Bulk Density (dry, loose mat'l)	kg/m^3	817-961
Hardness		5.1
Pore Size	angstorms	4
Pore Volume	%	15
Specific Surface Area	m^2/gm	40
Alkali Stability (pH)		7-13
Acid Stability (pH)		1-7
Thermal Stability	°C	700
Crushing Strength	kgs/cm^2	176
Wet Attrition (Avg.)	%	6-7
Heavy Metal Content		
As	mg/kg	27
Al	mg/kg	39 000
Cd	mg/kg	1.0
Cu	mg/kg	46
Co	mg/kg	4
Cr	mg/kg	5.4
Fe	mg/kg	4 300
Mo	mg/kg	10
Ni	mg/kg	2
Pb	mg/kg	64
Se	mg/kg	1
St	mg/kg	160
Zn	mg/kg	250

Table 3.3. Composition of Pig Rations

Ration		Ration 1	Ration 2	Ration 3	Ration 4
Property	**Unit**				
Crude Protein	%	17. 2	17. 2	15. 5	15. 5
Crude Fat	%	7	7	2	2
Crude Fiber	%	5	5	5	5
Sodium (Na)	%	0. 2	0. 2	0. 2	0. 2
Calcium (Ca)	%	0. 75	0. 75	0. 68	0. 68
Phosphorus (P)	%	0. 65	0. 65	0. 58	0. 58
Copper (Cu)	mg/kg	125	125	125	125
Zinc (Zn)	mg/kg	100	100	100	100
Vitamin A	I.U./kg	5400	5400	5400	5400
Vitamin D3	I.U./kg	1200	1200	1200	1200
Vitamin E	I.U./kg	40	40	40	40
Selenium	mg/kg	0. 3	0. 3	0. 3	0. 3
Zeolite	%	0	4	4	4
Energy	Kcal	3250	3250	2925	2760
Crude Protein	%	100	100	90	90
Energy	%	100	100	90	85

3.3.3 Methodology

3.3.3.1 Animals

The research protocol was approved by the Animal Care Committee of McGill University in accordance with the Canadian Council on Animal Care Guidelines. Each experimental pig was identified using an ear tag.

Two feeding experiments were performed. The first experiment was conducted using male subjects while the second experiment was conducted using female subjects. Three batches of the metabolic study were carried out. The first batch used pigs from Exp1, while the second and third batches used pigs from Exp 2.

Experiment 1. Twenty male crossbred (75% Landrace x 25% Yorkshire) growing pigs with average initial weight of 50.18 ± 2.95 kg were randomly divided into groups

of four and each group was placed in an individual pen in the growing/finishing room of the pig unit. Each group or pen was randomly assigned one of the four dietary treatments. The pigs were fed their respective diet for two weeks, and then weighed. Three pigs from each pen were randomly selected and immediately placed randomly into individual metabolic cage in the metabolic room where they remained for 8days. This trial was identified as Batch 1 (B1).

The average weight of the pigs at the onset of B1 was 66.53 ± 2.70 kg. After the 8-day metabolic test period, pigs were removed from the metabolic cages, weighed and returned to the previous pens in the growing/finishing room.

Experiment 2. Twenty-four female crossbred (75% Landrace x 25% Yorkshire) growing pigs with an average initial weight of 48.95 ± 5.25 kg were randomly assigned to four pens in the growing/finishing room, with six pigs per pen. Each one of the same four dietary treatments used for B1 was randomly assigned to a pen. The pigs were weighed after being fed their respective diet for two weeks. After weighing, three pigs from each pen were randomly selected and immediately placed randomly into individual metabolic cage for 8 days (3 cages/ treatment). This test was identified as Batch 2 (B2). The remaining pigs (three pigs per pen) continued to be housed in the previous pens and fed their respective experimental diets.

The average weight of the subjects at the onset of B2 was 63.13 ± 3.77 kg. After the 8-day metabolic experimental period, pigs in B2 were removed from the metabolic cages, weighed and returned to their previous pens in the growing/finishing room. The pigs not used in B2 were weighed and immediately placed randomly into individual metabolic cage (3 pigs/ treatment) to form Batch 3 (B3). The average weight of the pigs at the start of B3 was 62.86 ± 6.61 kg. After 8 days in the metabolic cages, all B3 pigs were weighed and returned to the growing/finishing room.

3.3.3.2 Management

3.3.3.2.1 Period of Pigs in Growing/finishing Pen

Pigs were allowed to have ad libitum access to feed and water for two to three weeks spent in the growing/finishing pens. Every day, the feed placed into the feeders was weighed. Pigs were weighed weekly, at which time the amount of feed left in the feeders were weighed. Whole feed intakes per pen were calculated by subtracting the leftover feeds from the initial amount of feed offered. Thus, average daily gain (ADG) for each individual pig, and pen average daily feed intake (ADFI) and feed conversion rate (FCR=Feed /Gain ratio) could be calculated.

3.3.3.2.2 Period of Pigs in Metabolic Cage

During each metabolic test period, pigs were housed in individual stainless steel metabolic cage in a mechanically ventilated room with the temperature maintained between 24 and 26^{0}C. Each metabolic cage measured 128.7cm x 43.7cm and contained a nipple waterer, and a single feeder. Pigs were allowed a 3-day acclimation period to adjust to the metabolic cages and dietary treatment. A bar inside and across the metabolic cage was adjusted to prevent pigs from turning around and to properly collect their feces and urine. Two clean independent trays were installed under each metabolic cage. The top tray was used to collect feces and perforated to allow for the drainage of urine into the bottom tray.

To eliminate the potential variability betwen the different bags of feed, four bags each of 25kg from each type of experimental ration were mixed together and stored in single containers, at the onset of the metabolic experiment. Three samples of feed were randomly taken from each container and tested for dry matter, CP, and organic matter content. During the adjustment and collection periods, pigs were continuously fed their respective experimental diet assigned once in the growing/finishing pen. The diets were offered once daily at 10:00 am every morning in a feed trough at the front of the cage. Before adding fresh and newly weighed feed, the feed not consumed from the previous

day was removed, weighed, and recorded to compute the net feed consumption. Pigs had ad libitum access to water throughout the trial, and the waterers were checked on a daily basis to ensure that they were working properly.

3.3.3.3 Feces and Urine Collection

Before the collection period, the metabolic cages and collection trays were thoroughly cleaned. Pigs were allowed 3 days (-2 d, -1 d, 0 d) to adjust to the metabolic cages prior to the 5 days (1 d, 2 d, 3 d, 4 d, 5d) of feces and urine collection.

3.3.3.3.1 Feces Collection

The feces generated by each pig while in the metabolic cages over 5-day collection period were collected daily following each feeding. The collected and weighed feces for each pig in B1 and B3 were placed into labeled plastic containers with caps and stored in -18^{0}C until further analyzed. The feces collected daily in B2 were composited, and stored in 12 labeled 20L containers at -18^{0}C. Prior to analysis, feces from each pig were thawed and thoroughly mixed to ensure uniform consistency of samples. Three subsamples for each pig in B2 or daily subsamples for each pig in B1 and B3 were taken for further analysis.

3.3.3.3.2 Urine Collection

Total urinary output was collected and recorded daily for each pig following feces collection. Thirty ml of formaldehyde (10%) were added initially to the urine collection trays in B2 and B3, at each collection, to prevent ammonia losses and limit microbial growth during the collection and storage of urine (Adeola et al., 1995). The collected urine were pooled by pig and stored in individual labeled capped container at -18^{0}C. Before analysis, all urine samples were completely thawed. Three subsamples of urine of each pig were taken for subsequent analysis after thorough mixing.

3.3.3.4 Dry Matter and Organic Matter Analysis

The dry matter or total solid content of the samples was determined by drying in an

oven at 103°C for 24h. Feces and feed were also tested for total organic matter content by burning in a furnace at 500°C for 4 hours after drying at 103°C for 24h. (Lefcourt and Meisinger, 2001; Thilsing-Hansen et al., 2002a).

3.4 Statistical Analysis

In both experiments, all data from individual batch were subjected to analysis of variance using the GLM procedure of the SAS system (1999) for a completely randomized design (CRD) model where pig was the experimental unit. This statistical model included the effects of dietary treatment. The data in combination of B2 and 3 were analyzed by the Randomised Complete Block Model (RCB), which model included the effects of dietary treatment and batch. All comparisons were adjusted for multiple comparisons by Bonferroni test and differences were considered significant if the P-value was < 0.05.

3.5 Results and Discussion

3.5.1 Effects of Zeolite on Pig Performance

During the whole course of the study, only one pig showed signs of diarrhoea while in the growing/finishing pen, and it recovered the second day after being placed in the metabolic cage. No other pigs showed signs of disease or excessive stress or died. The pigs appeared to be healthy and did not reject their diets throughout the entire collection period.

The average initial weight of the experimental pigs when placed on their respective diet in growing/finishing pens was 50.18 ± 2.95 kg and 48.95 ± 5.25 kg for Exp 1 and Exp 2, respectively. The performance (ADG, ADFI and FCR) of the pigs is listed in Table 3.4 and Table 3.5, for the two phases of the experiment, growing/finishing pen phase and metabolic cage phase. The growing/finishing pen data was insufficient to be statistically analysed since only one pen (experimental unit) for each dietary treatment. In term of ADG, the pigs fed the regular diet (R1) had the highest body weight gain in

the first week among four treatments, in both of two tests. However, in the second week, the pigs fed R3 (90 % CP and energy with 4 % zeolite) produced the highest body weight gain compared to other three diets, with of 0.085 and 0.3 kg/d exceeding those of the pigs fed the control diet for tes1 and test 2, respectively. Meanwhile, the ADFI for the pigs fed R3 was highest, whereas the ADFI for pigs fed R2 (100% CP and energy with 4% zeolite) was the least, amongst the four treatments for both of male and female pigs. In addition, the ADFI for the pigs fed R4 (90% CP and 85% energy with 4% zeolite) was slightly higher than that of the pigs fed the control diet. These preliminary data indicates that supplementation of zeolite to grower pigs' diet can reduce the ADFI when compared to the same CP and energy level diet.

Table3.4 Effects of dietary treatment on performances of experimental pigs in growing/finishing pens[1].

Treatment	Experiment 1							Experiment 2						
	IBW[2]		Week1		Week2		Ave.	IBW		Week1		Week2		Ave.
	Mean	SD	Mean	SD	Mean	SD		Mean	SD	Mean	SD	Mean	SD	
	Average Daily Body Weight Gain (kg/day)													
Ration1	50.04	3.26	1.231	0.09	1.103	0.15	1.167	48.90	4.88	0.819	0.11	0.931	0.51	0.875
Ration2	50.74	2.81	0.963	0.26	1.096	0.21	1.029	49.80	5.71	0.769	0.21	0.650	0.28	0.710
Ration3	50.06	2.93	1.163	0.13	1.188	0.15	1.176	48.87	5.19	0.514	0.09	1.231	0.23	0.873
Ration4	49.90	2.78	1.189	0.13	1.074	0.18	1.132	48.25	5.23	0.726	0.11	1.024	0.23	0.875
	Average Daily Feed Intake (kg/day)													
Ration1			2.157		2.417		2.287			1.671		1.924		1.798
Ration2			1.911		2.031		1.971			1.707		1.535		1.621
Ration3			2.457		2.791		2.624			1.757		2.526		2.142
Ration4			2.360		2.746		2.553			1.785		2.319		2.052
	Feed Conversion Rate (Feed/Gain)													
Ration1			1.760	0.14	2.223	0.28	1.992			2.077	0.31	2.268	1.66	2.173
Ration2			2.140	0.72	1.911	0.39	2.025			2.400	0.85	2.645	1.15	2.522
Ration3			2.136	0.25	2.380	0.30	2.258			4.066	0.93	2.12	0.43	3.093
Ration4			2.005	0.24	2.625	0.51	2.315			2.511	0.39	2.358	0.50	2.434

[1]Values are averaged within each pen. There is only one pen for each dietary treatment in each experiment. There were five pigs in each pen in experiment 1 and six pigs per pen in experiment 2.

[2]IBW means initial body weight of pigs, SD stands for standard deviation.

When the experimental pigs were placed in the metabolic cages, all performance parameters (ADFI, ADG, FCR) were statistically analysed (Table 3.5) since individual pigs became the experimental unit. Only two significant differences ($P < 0.05$) appeared and they pertained to the ADFI and FCR between R2 and R4 in B1, indicating the male pigs significantly ($P < 0.05$) consumed higher amount of 4% zeolite diet daily (0.43 kg/d) when CP and energy concentration in diets was decreased from 100% to 90% and 85%, respectively. Consequently, the feed conversion rate of pigs fed R4 was significantly lower ($P < 0.05$) by 1.63 kg of feed/kg of body weight gain, compared to the pigs fed the 4% zeolite diet with 100% CP and energy (R2). In term of ADFI, no significant differences existed among R1, R2 and R3 in all three metabolic tests ($P > 0.05$). However, the ADFI of pigs receiving R2 (100% CP and energy with 4% zeolite) was the lowest of all four rations, with 0.11 kg/d and 0.14 kg/d less feed for male and female pigs respectively, when compared to that of pigs receiving R1, as well as 0.30 kg/d and 0.07 kg/d less feed for male and female pigs respectively when compared to that of pigs receiving R3.

Zeolite supplementation along with reduction of CP and energy in pig diets had no significant effect ($P > 0.05$) on pig body weight gain. However, the ADG of pigs receiving R2 and R3 was larger than that of pigs fed R1 and R4, with the ADG of pigs fed R2 being the highest and that fed R4 being the smallest (Table 3.5).

Diet supplementation of 4% zeolite had no significant ($P > 0.05$) effect on feed conversion but obviously increased the feed utilization efficiency. Pigs fed R2 had the best feed conversion rate, while pigs fed R4 had the lowest FCR out of all 4 rations. The addition of 4% zeolite in R2 decreased the FCR by 0.71 and 0.8 kg feed/kg body weight gain for male and female pigs respectively, compared to the control diet (R1), which indicates that the feed utilization efficiency was improved 22% for male pig and 23% for female pigs. Furthermore, the FCR of pigs fed R3 was also reduced by 0.07 and 0.19 kg feed/kg body weight gain for male and female pigs respectively, compared to that of

pigs receiving R1. Thus, the feed digestibility efficiency was improved by 2.3% for male and 5.5% for female pig, with zeolite supplementation, even if the protein and energy concentration in the diet was decreased by 10%, compared to NRC recommendations. In contrast, the FCR of pigs fed R4 (90% CP and 85% energy with 4% zeolite) was increased significantly ($P < 0.05$), when compared to that of pigs fed R2 (100% CP and energy with 4% zeolite) in B1. Therefore, the addition of 4% zeolite along with the 10 and 15% lower diet CP and energy, respectively, significantly ($P < 0.05$) reduced the feed utilization efficiency for male pigs. When compared to R1, R4 increased the FCR by 0.92 and 0.7 kg of feed/kg of body weight gain, for male and female pig respectively, suggesting that the feed utilization efficiency was decreased by 29% and 20% for male and female pigs, respectively. In conclusion, diets supplementation with 4% zeolite allowed the pigs to utilize feed more efficiently for growth, and the diet CP and energy content could be decreased to 90% of that recommended by NRC.

Similar conclusions can be reached by examining the protein and energy conversion (Table 3.6). Zeolite supplementation did not show any significant effect on protein and energy conversion rate ($P > 0.05$). However, pigs fed R2 had the best protein and energy conversion rate, and those on R4 had the worst, amongst all four rations. Zeolite supplement at 4% in R2 decreased the requirement by 23.45% and 22.67% for CP and energy per kg body weight gain respectively, suggesting that R2 demanded on the average 125g less protein and 2442 kcal less energy per kg of body weight gain, compared to the control R1. Moreover, the protein and energy conversion of pigs fed R3 was also improved by 14% (80g) and 13% (1450 kcal) respectively, compared to that of pigs fed R1, regardless of 10% reduced protein and energy concentration. More research is required to verify that the weight gain for R2 is muscle or fat.

In contrast, when the diet CP and energy was decreased to 90% and 85% respectively (R4), 11% (65g) more CP and 6% (620 kcal) more energy was required per kg body weight gain as compared to feeding R1, even with 4% zeolite supplementation

in R4.

Table 3.5 Influences of dietary treatment on performances of experimental pigs in metabolism cages[1].

Treatment[2]	Ration 1		Ration 2		Ration 3		Ration 4		SEM	P-Values
	LSM	SD	LSM	SD	LSM	SD	LSM	SD		
	Average Initial Body Weight (kg)									
Batch 1	65.93	3.85	65.13	2.35	68.76	3.49	66.30	0.20		
Batch 2	62.73	5.20	63.60	5.53	63.20	3.65	62.97	3.71		
Batch 3	67.10	5.85	58.77	7.04	63.60	5.69	61.97	9.60		
Batch2 +3	64.92	5.50	61.18	6.25	63.40	4.28	62.47	6.53		
	Average Daily Feed Intake (kg/d)									
Batch 1	2.05^{ab}	0.09	1.94^{a}	0.17	2.24^{ab}	0.22	2.37^{b}	0.05	0.12	0.03
Batch 2	1.90	0.12	1.62	0.07	1.75	0.07	1.62	0.28	0.24	0.39
Batch 3	2.20	0.18	2.20	0.23	2.21	0.15	2.37	0.37	0.24	0.55
Batch2 +3	2.05	0.25	1.91	0.35	1.98	0.48	2.00	0.50	0.18	0.76
	Average Daily Body Weight Gain (kg/d)									
Batch 1	0.65	0.04	0.80	0.07	0.74	0.13	0.59	0.11	0.08	0.14
Batch 2	0.67	0.26	0.69	0.18	0.56	0.06	0.37	0.20	0.19	0.27
Batch 3	0.60	0.04	0.78	0.09	0.70	0.37	0.59	0.15	0.21	0.43
Batch2 +3	0.54	0.31	0.74	0.14	0.63	0.31	0.48	0.20	0.15	0.25
	Feed Conversion Rate (Feed Intake/Gain)									
Batch 1	3.17^{ab}	0.38	2.46^{a}	0.41	3.10^{ab}	0.57	4.09^{b}	0.72	0.44	0.04
Batch 2	3.31	1.85	2.47	0.74	3.15	0.32	4.03	1.13	1.02	0.47
Batch 3	3.68	0.05	2.85	0.49	3.39	0.93	4.24	1.38	0.88	0.43
Batch2 +3	3.46	1.32	2.66	0.60	3.27	0.62	4.16	1.21	0.72	0.07

[1]Values are the least square mean of three replicates of pigs per treatment in each batch.

[2]All experimental pigs were placed in the metabolism cages for 8 days; Only male pigs in B1 and female pigs in B2 and 3.

LSM represents least square means, SD stands for standard deviation, SEM being standard error mean. P-values are equal to probability of differences between treatments, reported from ANOVA tables.

All data in each batch were analyzed by statistical CRD model, the data in combination of B2 and 3 was analyzed by the statistical RCB model. All comparisons were adjusted for multiple comparisons by Bonferroni test.

[a-c]Values within rows with no common superscripts are significantly different ($p< 0.05$).

Table 3.6 Effects of dietary treatment on protein and energy conversion of feeds with experimental pigs in metabolism cages[1].

Treatment[2]	Ration 1		Ration 2		Ration 3		Ration 4		SEM	P-Values
	LSM	SD	LSM	SD	LSM	SD	LSM	SD		
Feed Conversion Rate (Feed Intake/Gain)										
Batch 1	3.17^{ab}	0.38	2.46^{a}	0.41	3.10^{ab}	0.57	4.09^{b}	0.72	0.44	0.04
Batch 2	3.31	1.85	2.47	0.74	3.15	0.32	4.03	1.13	1.02	0.47
Batch 3	3.68	0.05	2.85	0.49	3.39	0.93	4.24	1.38	0.88	0.43
Batch2+3	3.46	1.32	2.66	0.60	3.27	0.62	4.16	1.21	0.72	0.07
Feed Crude Protein Conversion (kg Protein / kg Gain)										
Batch 1	0.55	0.07	0.42	0.07	0.48	0.09	0.63	0.11	0.07	0.08
Batch 2	0.57	0.32	0.43	0.13	0.49	0.08	0.63	0.21	0.19	0.74
Batch 3	0.63	0.01	0.49	0.09	0.53	0.07	0.66	0.22	0.12	0.54
Batch2+3	0.60	0.08	0.46	0.09	0.51	0.09	0.65	0.21	0.10	0.27
Feed Energy Conversion (Kcal / kg Gain)										
Batch 1	10285.93	1273	7998.14	1344	9071.11	1690	11290.54	2007	1305	0.14
Batch 2	10768.55	6018	8031.65	2428	9213.75	1496	11122.80	2109	3646	0.79
Batch 3	11950.41	####	9259.03	1616	9915.75	1655	11702.40	3829	2503	0.63
Batch2+3	11245.00	2813	8645.33	1792	9564.75	1561	11481.60	2411	1908	0.38

[1]Values are the least square mean of three replicates of pigs per treatment in each batch.

[2]All experimental pigs were placed in the metabolism cages for 8 days; Only male pigs in B1 and female pigs in B2 and 3.

LSM represents least square means, SD stands for standard deviation, SEM being standard error mean. P-values are equal to probability of differences between treatments, reported from ANOVA tables.

All data in each batch were analyzed by statistical CRD model, the data in combination of B2 and 3 was analyzed by the statistical RCB model. All comparisons were adjusted for multiple comparisons by Bonferroni test.

[a-c]Values within rows with no common superscripts are significantly different ($p< 0.05$).

3.5.2 Effects of Zeolite on Pig Metabolic Parameters

All data including total feed intake (TFI), total feces excretion (TFE), total feed retention (TFR), total dry feed retention (DFR), total organic matter retention (OMR) and total urine excretion (TUE) were daily collected for five days and statistically analyzed. Zeolite supplementation at 4% had no significant ($P > 0.05$) effect on all parameters (Table 3.7, Table 3.8 and Table 3.9). However, there were some significant differences ($P < 0.05$) when diet CP and energy was considered. Importantly, some beneficial trend appeared with zeolite supplementation.

In all three batches, TFE was significantly different ($P < 0.05$) between R4 and the other three diets only in B1, which demonstrate that the male pigs fed R4 produced significant more ($P < 0.05$) feces compared to other three treatments (Table 3.7). When the B2 and B3 data was combined and analysed, TFE was significantly different ($P < 0.05$) for R4 as compared to R2 and R3. Thus, feeding R4 (90% CP and 85% energy with 4% zeolite addition) had a significant negative impact on pig manure production, more feces were produced. Table 3.7 indicates that, in all three batches, pigs fed R2 produced the least feces while pigs receiving R4 produced the most feces out of four dietary treatments. Furthermore, the male and female pigs fed R2 produced 0.63 kg and 1.27 kg less feces within five days, respectively, than the pigs receiving R1, though this difference was not statistically significant.

Correspondingly, the same trend was observed with TFR (Table 3.7) and OMR (Table 3.8). Amongst the three metabolic tests, TRF and OMR were significantly different ($P < 0.05$) for R4 as compared to R2 in B1, and for R4 as compared to R1 and R2 in B2 and B2+B3. Thus, pigs fed R4 significantly ($P < 0.05$) retained less feed (Table 3.7) and OM (Table 3.8) in the gastrointestial tract (GIT), compared to R2 for male and R1 and R2 for female pigs. In fact, pigs receiving R4 retained the least feed and OM in the GIT, amongest four dietary treatments. This observation explains why the pigs fed R4 had the lowest feed conversion rate, and supports the conclusion that feeding R4 had a significant negative impact on pig manure production. In addition,

pigs fed R2 retained most feed and OM in GIT, amongst four diets, likely because zeolite slows down the passage of feed in the GIT by adsorbing water.

Table 3.7 Effects of dietary treatment on feed digestion of experimental pigs in metabolism cages[1].

Treatment[2]	Ration 1		Ration 2		Ration 3		Ration 4		SEM	P-Value
	LSM	SD	LSM	SD	LSM	SD	LSM	SD		
	Total Feed Intake (kg / 5 days)									
Batch 1	9.22	0.73	10.03	1.27	10.91	1.66	12.04	0.34	0.93	0.070
Batch 2	10.77	0.51	9.05	0.35	8.04	2.59	8.92	1.55	1.26	0.248
Batch 3	9.40	2.31	10.32	1.74	10.01	0.69	11.34	1.64	1.39	0.591
Batch2 + 3	10.09	1.67	9.58	1.38	9.02	2.01	10.13	1.95	0.99	0.625
	Total Feces Excretion (kg / 5 days)									
Batch 1	3.81^{a}	0.78	3.18^{a}	0.59	4.76^{a}	1.13	7.31^{b}	0.68	0.67	0.001
Batch 2	4.77	0.32	2.93	0.40	2.86	1.88	5.90	1.73	1.07	0.056
Batch 3	4.13	1.27	3.42	0.41	4.89	1.04	7.50	3.24	1.49	0.106
Batch2 + 3	4.45^{ab}	0.90	3.18^{a}	0.45	3.87^{a}	1.76	6.69^{b}	2.48	0.91	0.006
	Total Feed Retention (kg / 5 days)									
Batch 1	5.41^{ab}	0.51	6.86^{a}	0.71	6.15^{ab}	0.87	4.73^{b}	0.58	0.56	0.024
Batch 2	6.00^{a}	0.54	6.13^{a}	0.65	5.18^{ab}	1.16	3.02^{b}	1.20	0.77	0.015
Batch 3	5.27	1.13	6.90	1.34	5.11	0.92	3.84	1.60	1.04	0.103
Batch2 + 3	5.64^{a}	0.89	6.51^{a}	1.08	5.15^{ab}	0.94	3.43^{b}	1.34	0.64	0.001

[1]Values are the least square mean of three replicates of pigs per treatment in each batch.

[2]All experimental pigs were placed in the metabolism cages for 8 days; Only male pigs in B1 and female pigs in B2 and 3.

LSM represents least square means, SD stands for standard deviation, SEM being standard error mean. P-values are equal to probability of differences between treatments, reported from ANOVA tables.

All data in each batch were analyzed by statistical CRD model, the data in combination of B2 and 3 was analyzed by the statistical RCB model. All comparisons were adjusted for multiple comparisons by Bonferroni test.

[a-c]Values within rows with no common superscripts are significantly different ($p < 0.05$).

Table 3.8 Influences of dietary treatment on digestion of dry feed and organic matter with experimental pigs in metabolism cages[1].

Treatment[2]	Ration 1		Ration 2		Ration 3		Ration 4		SEM	P-Value
	LSM	SD	LSM	SD	LSM	SD	LSM	SD		
	Total Feed Intake (kg / 5 days)									
Batch 1	9.22	0.73	10.03	1.27	10.91	1.65	12.03	0.34	1.83	0.070
Batch 2	10.77	0.51	9.05	0.35	8.04	2.59	8.92	1.55	1.26	0.248
Batch 3	9.40	2.31	10.32	1.74	10.01	0.69	11.34	1.64	1.39	0.591
Batch2 + 3	10.09	1.67	9.58	1.38	9.02	2.01	10.13	1.95	0.99	0.625
	Total Dry Feed Retention (kg / 5 days)									
Batch 2	7.96	0.41	6.47	0.36	5.94	1.69	5.91	1.06	0.84	0.120
Batch 3	6.78	1.75	7.54	1.35	6.90	0.51	7.57	0.57	0.95	0.770
Batch2 + 3	7.37	1.31	7.01	1.06	6.42	1.23	6.74	1.19	0.69	0.580
	Total Organic Matter Retention (kg / 5days)									
Batch 1	3.79^{ab}	0.48	5.21^{a}	0.48	4.22^{ab}	0.65	2.76^{b}	0.56	0.45	0.004
Batch 2	4.12^{a}	0.48	4.47^{a}	0.56	3.69^{ab}	0.86	1.65^{b}	1.06	0.63	0.008
Batch 3	3.65	0.76	5.19	1.03	3.41	0.78	1.90	1.70	0.95	0.051
Batch2 + 3	3.89^{a}	0.62	4.83^{a}	0.84	3.55^{ab}	0.75	1.78^{b}	1.31	0.53	<0.001

[1]Values are the least square mean of three replicates of pigs per treatment in each batch.

[2]All experimental pigs were placed in the metabolism cages for 8 days; Only male pigs in B1 and female pigs in B2 and 3.

LSM represents least square means, SD stands for standard deviation, SEM being standard error mean. P-values are equal to probability of differences between treatments, reported from ANOVA tables.

All data in each batch were analyzed by statistical CRD model, the data in combination of B2 and 3 was analyzed by the statistical RCB model. All comparisons were adjusted for multiple comparisons by Bonferroni test.

[a-c]Values within rows with no common superscripts are significantly different ($p < 0.05$).

Male and female grower pigs fed R2 retained 290g and 174g of more feed daily (Table 3.7), and 284g and 188g of more OM daily (Table 3.8), in the GIT, respectively, as compared to the pigs fed the zeolite-free diet (R1). Pigs fed R3 showed inconsistent results for TFE, TFR and OMR, probably due to the varibility of the subject.

However, 4% zeolite supplementation had no effect ($P > 0.05$) on TUE, but had a significant ($P < 0.05$) effect on the urine total solid content between female pigs fed R1 and those fed R4 (Table 3.9). In general, the pigs fed R1 had the highest urine total solid content while those fed R4 had the lowest, amongst all four dietary treatments.

Table 3.9 Influences of dietary treatment on urine excretion of experimental pigs in metabolic cages[1].

Treatment[2]	Ration 1		Ration 2		Ration 3		Ration 4		SEM	P-Value
	LSM	SD	LSM	SD	LSM	SD	LSM	SD		
	Total Urine Excretion (L)									
Batch 2	10.96	2.94	6.33	1.68	7.03	3.87	4.82	1.98	2.25	0.115
Batch 3	9.79	6.61	9.67	1.19	5.81	2.76	6.21	3.44	3.28	0.500
Batch2 + 3	10.38	4.62	8.00	2.24	6.42	3.08	5.52	2.62	1.93	0.096
	Total Solid in Urine (G)									
Batch 2	262.43	32.93	192.33	20.00	194.14	71.73	179.57	19.82	34.21	0.140
Batch 3	231.03	28.34	230.49	61.69	227.13	6.99	172.89	24.83	29.65	0.220
Batch2 + 3	246.73[a]	32.42	211.41[ab]	46.04	210.64[ab]	49.03	176.23[b]	20.43	22.35	0.041

[1]Values are the least square mean of three replicates of pigs per treatment in B2 & 3.

LSM represents least square mean, SD stands for standard deviation, SEM being standrd error mean. P-values are equal to probability of differences between treatments, reported from ANOVA tables.

All data in each batch were analyzed by statistical CRD model, the data in combination of B2 and 3 was analyzed by the statistical RCB model. All comparisons were adjusted for multiple comparisons by Bonferroni test.

[ab]Values within rows with no common superscripts are significantly different ($p< 0.05$).

3.6 Conclusions

While in the metabolic cages, the grower pigs fed a ration supplemented with 4% zeolite did not perform significantly better than those on the control ration ($P > 0.05$). However, consistant beneficial trends, some of which were significantly different ($P < 0.05$) occurred when the diet CP and energy concentration were changed along with the supplementation of 4% zeolite.

Our results demonstrate that 4% zeolite supplementation to the diet of grower pigs promotes better feed utilization when the ration CP and energy was decreased to 90% of that recommended by NRC.

3.7 Acknowledgement

This project was financed by the Fédération des producteurs de porcs du Québec, the Natural Science and Engineering Research Council of Canada (NSERC), Promix (Upton Québec) and KMI Zeolites.

CHAPTER IV

General Conclusion

The environmental impact of the pig production is closely related to the amount of manure produced and its nutrient contents. Reducing the amount of manure produced and its nutrient content can reduce the negative environmantal impact of the industry on resources such as soil, water and air. The most logical solution to this problem is to find an economical and viable way of improving feed digestion. Zeolite, and especially clinoptilolite, is said to improve the digestion of feed by adsorbing water and slowing down the passage of food throught the GIT, and by temporarily adsorbing excess nitrogen.

The present study demonstrated that 4% zeolite (90%+ clinoptilolite) supplementation in grower pig diets improved feed utilization efficiency. It was observed that 4% zeolite added to the regular diet of grower pigs decreased the average daily consumption of feed and the amount of feces produced, increased feed and organic matter retention in the GIT, improved feed and protein as well as energy conversion, and finally increased body weight gain, compared to a regular diet without zeolite and 4% zeolite diets with low CP and energy. Nevertheless, 4% zeolite supplementation, combined with the 10% reduction in CP and energy, in grower pig diets, also improved feed and protein as well as energy conversion, and increased body weight gain, when compared to those pigs fed the regular diet. In general, 4% zeolite supplementation to the regular diet, improved the feed utilization efficiency by 23% and decreased the amount of feces produced (190g/day), thereby, reducing the environmental impact of pig manures. If the diet CP and energy concentrations are reduced by 10%, along with addition of 4% zeolite, the negative impact of pig manure

on environment can also be decreased to a lesser extent. The further study needs to look at the quality of the carcasses to relate the weight gain to that of either muscle or fat.

However, reducing the diet CP and energy by 10% and 15% respectively, along with 4% zeolite supplementaiton had significantly negative impacts on pig performance and manure production. Such feed significantly ($P < 0.05$) increased feed consumption and the amount of feces produced, and decreased feed and OM retention in the GIT as well as feed conversion rate.

4.1 Research to be Continued

The present research has studied the effect of zeolite supplement on pig performance and on part of the metabolic products. The potential to reduce the environmental impact of pig manure was also examined. Nevertheless, further research needs to certify this potential effect, such as:

1. The changes in mineral concentration in the collected samples such as feces, urine and blood plasma.

2. The efffect of zeolite supplementation on the retention time of feed through the GIT of pigs.

3. The impact of zeolite addition on the total feed digestibility and the apparent digestibility of individual minerals in feed.

4. The effect of zeolite supplementation on the manure nitrogen volatilization during storage and manure handling.

References

Adeola, O., Lawrence, B.V., Sutton, A.L. and Cline, T.R. 1995. Phytase-induced changes in mineral utilization in zinc-supplemented diets for pigs. Journal of Animal Science 73:3384-3391.

Airoldi, G., Balsari, P. and Chiabrando, R. 1993. Odor control in pig houses by the use of natural zeolites: First approach to the problem. In: Livestock Environment IV, Fourth International Symposium, ASAE. St-Joseph, Michigan, USA, 701-708.

AI-Kanani, T., Akochi, E., Mackenzie, A.F., Alli, I. and Barrington, S.F. 1992. Odour emission from liquid hog manure: Effect of added amendments and aeration on odour presence and odour offensiveness. Journal of Environmental Quality 21:704-708.

Baidoo, S.K. 1996. Manure modifications through dietary manipulation: Dietary effect on volume, content and odour. In: Manure Management Symposium Proceedings. Winnipeg, Canada, 161-168.

Barrington, S.F. 2001. Technologies to address environmental issues. London Pig Conference - The Pork Industry and Public Issues: 5-6.

Barrington, S.F. 1996. Practical methods for controlling odors. In: Manure Management Symposium Proceedings. Winnipeg, Canada, 149-158.

Barrington, S.F. and EI Moueddeb, K. 1995. Zeolite as pig manure additive to control odours and conserve N. CASE paper no. 95-510, Saskatoon, Canada.

Bartko, H., Seidel, H. and Kovac, G. 1993. In: Zeolite '93. Fourth International Conference on the Occurrence, Properties and Utilization of Natural Zeolites. International Committee on Natural Zeolites, Department of the Earth Sciences, SUNY-College at Brockport, Brockport, New York 1442, USA, 43-44.

Berg, W. and Hornig, G. 1997. Emission reduction by acidification of slurry—Investigations and assessment. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkerloord, The Netherlands, 459-466.

Bernall, M.P. and Lopez-Real, J.M. 1993. Natural zeolites and sepiolite as ammonium and ammonia adsorbent materials. Bioresearch Technology 43:27–33.

Betcher, R.N., Rudolph D.L. and Nicholson, R. 1996. Impact of agricultural activities on groundwater quality. In: Manure Management Symposium Proceedings. Winnipeg, Canada, 53-62.

Bundy, D.S., Li, X.W. and Hoff, S.J. 1997. Malodour abatement by covering materials. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 101-110.

Burnett, W.E. and Dundero, N.C. 1970. Control of odors from animal wastes. Transaction of the ASAE 13:221-224.

Burton, C.H. 1992. A review of the strategies in the aerobic treatment of pig slurry: Purpose, Theory and Method. Journal of Agricultural Engineering Research 53:249-272.

Canadian Pork Council. 1996. Literature review on the impacts of hog production on public health. http://www.cpc-ccp.com/.

Canh, T.T., Verstegen, M.W.A., Aarnink, A.J.A. and Schrama, J.W. 1997. Influence of dietary factors on nitrogen partitioning and composition of urine and feces of fattening pigs. Journal of Animal Science 75:700-706.

Carlton Trail Regional Hog Information. 2005.http://collections.ic.gc.ca/hog/heart.html

Carpenter, G.A. 1986. Dust in livestock buildings—A review of some aspects. Journal of Agricultural Engineering Research 33:227-241.

Carter, S.D., Cromwell, G.L., Lindemann, M.D., Turner, L.W. and Bridges, T.C. 1996. Reducing N and P excretion by dietary manipulation in growing and finishing pigs. Journal of Animal Science 74 (Suppl. 1):59.

Cefali, E.A., Nolan, J.C., McConnell, W.R. and Walters, D.L.1995. Pharmacokinteic study of Zeolite A, sodium aluminosilicate, magnesium silicate and aluminum hydroxide in dogs. Pharmaceutical Research 12:270–274.

Chiba, L.I., Peo, E.R.Jr., Lewis, A.J., Brumm, M.C., Fritschen, R.D.and Crenshaw, J.D. 1985. Effect of dietary fat on pig performance and dust levels in modified open front and environmentally regulated confinement building. Journal of Animal Science 64:763-781.

Chiba, L.I., Peo, E.R.Jr. and Lewis, A.J. 1987. Use of dietary fat to reduce dust, aerial ammonia and bacterial colony forming particle concentrations in pig confinement buildings. Transactions of the ASAE 30:464-468.

Choiniere, Y., Marquis, B. and Gingras, G. 1997. Ammonia and contaminant concentrations with conventional versus pit ventilation in finishing pig units. In: Proceedings of the International Symposium on Ammonia and Odour Control from

Animal Production Facility. Vinkeloord, The Netherlands, 365-372.

Cook, T.E., Cilley, W.A., Savitsky, A.C. and Wiers, B.H. 1982. Zeolite A hydrolysis and degradation. Environmental Science and Technology 16:344-350.

Cromwell, G.L., Stahly, T.S., Coffey, R.D., Monegue, H.J. and Randolph, J.H. 1993. Efficacy of phytase in improving the bioavailability of phosphorus in soybean meal and corn-soybean meal diets for pigs. Journal of Animal Science 71:1831-1840.

Daniel, T.C., Sharpley, A.N. and Lemunyon, J.L. 1998. Agricultural phosphorus and eutrophications: A symposium overview. Journal of Environmental Quality 27:251-257.

Denis, C. 1999. Zeolite in pig diet: Effect on growth performance and air quality. M.Sc. Thesis, Department of Agricultural and Biosystems Engineering, McGill University, Montreal, Canada.

Dong, L., Heber, A.J., Patterson, J.A., Strobel, B.R., Jones, D.D. and Sutton, A.L. 1997. Bioscrubber for removing ammonia from pig house exhaust air. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 529-532.

Elliott, M.A. and Edwards, H.M. 1991. Comparison of the effects of synthetic and natural zeolite on laying hens and broiler chicken performance. Poultry Science 70:2115-2130.

Enemark, J.M.D., Frandsen, A.M.S., Hansen, T.T. and Jorgensen, R.J. 2003a. Aspects of physiological effects of sodium zeolite A supplementation in dry, non-pregnant dairy cows fed grass silage. Acta Veterinaria Scandinavica Supplementum 97:97-117.

Enemark, J.M.D., Kirketerp-Moller, C.N. and Jorgensen, R.J. 2003b. Effect of prepartum zeolite A supplementation on renal calcium excretion in dairy cows around calving and evaluation of a field test kit for monitoring it. Acta Veterinaria Scandinavica Supplementum 97:119-136.

Environment Canada. 2005. Canadian Water Quality Guidelines. http://www.ec.gc.ca.

Fethiere, R., Miles, R.D. and Harms, R.H. 1994. The utilisation of sodium in Na zeolite A by broilers. Poultry Science 73:118-121.

Garcia Moreno, M. 1993. Sphagnum moss for pig manure nitrogen conservation. M.Sc. Thesis, Department of Civil Engineering and Applied Mechanics, McGill University, Montreal, Canada.

Gore, A.M., Kornegay, E.T. and Viet, H.P. 1986. The effects of soybean oil on nursery air quality and performance of weaning pig. Journal of Animal Science 63:1-7.

Guingand, N., Granier, R. and Massabie, P. 1997. Characterization of air extracted from pig housing: effect of the presence of slurry and ventilation rate. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 49-55.

Hobbs, P.J., Misselbrook, T.H. and Pain, B.F. 1997. Characterisation of odorous compounds and emissions from slurries produced from weaner pigs fed dry feed and liquid diets. Journal of the Science of Food and Agriculture 73:43-45.

Hobbs, P.J., Pain, B.F., Kay, R.M. and Lee, P.A. 1996. Reduction of odorous compounds in fresh pig slurry by dietary control of crude protein. Journal of the Science of Food and Agriculture 71:508-514.

Hoff, S.J., Bundy, D.S. and Li, X.W. 1997. Dust effects on odor and odor compounds. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 101-110.

Holthaus, D.L., Richardson, C.R., Swift, S.M., Gibson, A.S. and Bunger, F.L. 1996. Effect of zeolite materials on rumen fermentation characteristics when compared to sodium carbonate. Journal of Animal Science 74 (supplement):284.

Janzen, H.H., Desjardins, R.L., Asselin, J.M.R. and Grace, B. 2003. The Health of Our Air: Toward sustainable agriculture in Canada. http://www.agr.gc.ca/.

John, M. and Wilson, P. 2003. Pig production: Biological principles and applications. USA.

Johnson, M.A., Sweeney, T.F. and Muller, L.D. 1988. Effects of feeding synthetic zeolite A and sodium bicarbonate on milk production nutrient digestion and rate of digest passage in daily cows. Journal of Dairy Science 71:946-953.

Jongbloed, A.W., Everts, H. and Kemme, P.A. 1991. Phosphorus availability and requirements in pigs. Recent Advances in Animal Nutrition. In: W. Haresign and D.J.A. Cole (Editors), Butterworths, London.

Jongbloed, A.W. and Lenis, N.P. 1992. Alteration of nutrition as means to reduce environmental pollution by pigs. Livestock Production Science 31:74-95.

Jongbloed, A.W. and Lenis, N.P. 1998. Environmental concerns about animal manure. Journal of Animal Science 76:2641-8.

Jorgensen, R.J., Hansen, T., Jensen, M.L. and Thilsing-Hansen, T. 2001. Effect of oral drenching with zinc oxide or synthetic zeolite A on total blood calcium in dairy cows. Journal of Dairy Science 84:609-613.

Kay, R.M.and Lee, P.A. 1997. Ammonia emission from pig buildings and characteristics of slurry produced by pigs offered low crude protein diets. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 253-260.

Kerr, B.J., McKeith, F.K., and Easter, R.A. 1995. Effect on performance and carcass characteristics of nursery to finisher pigs fed reduced crude protein amino acid-supplemented diets. Journal of Animal Science 73:433–440.

Kornegay, E.T., and Verstegen, M.W.A. 2001. Pig nutrition and environmental pollution and odor control. Pages 609–603 in Pig Nutrition. 2nd ed. E. T. Kornegay and M. W. A. Verstegen, ed. CRC Press LLC, Boca Raton, FL.

Kyriakis, S.C., Papaioannou, D.S., Alexopoulos, C., Polizopoulou, Z. and Tzika, E.D. 2002. Experimental studies on safety and efficacy of the dietary use of a clinoptilolite-rich tuff in sows: a review of recent research in Greece. Microporous and Mesoporous Materials 51:65-74.

Lau, A.K., Vizcarra, A.T., Lo, K.V. and Luymes, J. 1996. Recirculation of filtered air in pig barns. Canadian Agricultural Engineering 38:297-304.

Lavoie, J., Marchand, G. and Gingras, G. 1997. Pit ventilation in pig-housing facilities. Canadian Agricultural Engineering 39:317-326.

Lee, P.A. and Kay, R.M. 1997. Economic implications of reduced crude protein diets for pigs to reduce ammonia emissions. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 699-706.

Lefcourt, A.M. and Meisinger, J.J. 2001. Effect of adding alum or zeolite to dairy slurry on ammonia volatilization and chemical composition. Journal of Dairy Science 84:1814-21.

Liberti, L., Lopez, A., Amicarelli, V. and Boghetich, G. 1995. In Natural Zeolites '93: Occurrence, Properties, Use, Editors: Ming, D.W. and Mumpton, F.A. (Int. Comm. Nat. Zeolites, Brockport, NY), pp. 351-362.

Liu, J., Bollinger, D.W., Ledoux, D.R. and Veum, T.L. 1998. Lowering the dietary calcium to total phosphorus ratio increases phosphorus utilisation in low phosphorus corn-soybean meal diets supplemented with microbial phytase for growing-finishing

pigs. Journal of Animal Science 76:808-813.

Ma, C.S., Tzeng, C.M., Lai, M.K. and Tsai, A.H. 1979. The feeding effect of zeolite on the litter size at birth of pig. Science Agriculture 33:203-209.

Ma, C.S., Yang, S.K., Tzeng, C.M. and Wu, M.K. 1984. Effects of feeding clinoptilolite on embryo survival in pig. In: Zeo-Agriculture. Use of natural zeolites in agriculture and aquaculture. International Committee on Natural Zeolite, Editors: W.G. Pond and F.A. Mumpton. Westview Press Inc., Boulder, Colorado. 155-160.

Masse, D.I., Droste, R.L., Kennedy, K.J., Patni, N.K. and Munroe, J.A. 1997. Potential for the psychrophilic anaerobic treatment of pig manure using a sequencing batch reactor. Canadian Agricultural Engineering 30:25-33.

Meyer, A.M. 1997. Ammonia Control in Agriculture. In: Proceedings of the International Symposium on Ammonia and Odour Control from Animal Production Facility. Vinkeloord, The Netherlands, 575-577.

Milán, Z., Sánchez, E., Weiland, P., Borja, R., Martín, A. and Ilangovan, K. 2001. Influence of different natural zeolite concentrations on the anaerobic digestion of piggery waste. Bioresource Technology 80:37-43.

Miner, J.R. 1999. Alternatives to minimize the environmental impact of large pig production units. Journal of Animal Science 77:440-444.

Ministere de I'Environnement et de la Faune du Quebec. 1996. Document de reflexion sur la capacite des sols du territoire quebecois a supporter les elevages. Unpublished document of "la table de concertation sur le projet de reglement sur la reduction de la pollution d'origine agricole". Quebec, Canada.

Morken, J. and Sakshaug, S. 1997. New injection technique—Direct Ground Injection (DGI). In: Proceedings of the International Symposium on Ammonia and Odor Control from Animal Production Facility. Vinkeloord, The Netherlands, 585-590.

Mumpton, F.A. 1984. Natural zeolites. In: Zeo-Agriculture. Use of natural zeolites in agriculture and aquaculture. International Committee on Natural Zeolite, Editors: W.G. Pond and F.A. Mumpton. Westview Press Inc., Boulder, Colorado. 33-34.

Mumpton, F.A. 1999. Uses of natural zeolites in agriculture and industry. Proceeding of the National Academy of Sciences 96:3463–3470.

Mumpton, F.A. and Fishman, P.H. 1977. The application of natural zeolite in animal science and aquaculture. Journal of Animal Science 45:1188-1202.

National Research Council (NRC). 1998. Nutrients requirements of pig. 10th ed., National Academy Press, Washington, DC.

Nestrov, N. 1984. Possible applications of natural zeolites in animal husbandry. In: Zeo-Agriculture. Use of natural zeolites in agriculture and aquaculture. International Committee on Natural Zeolite, Editors: W.G. Pond and F.A. Mumpton. Westview Press Inc., Boulder, Colorado. 167-174.

Nicholson, R.J., Webb, J. and Moore, A. 2002. A review of the environmental effects of different livestock manure storage systems, and a suggested procedure for assigning environmental ratings. Biosystems Engineering 81:363-377.

Nielsen, V.C. 1987. Manure management and environmental effects. Pig housing and the environment: occasional publication No.11—Brirish society of animal production. 109-116.

Olver, M.D. 1997. Effect of feeding clinoptilolite (zeolite) on the performance of three strains of laying hens. British Poultry Science 38:220–222.

O'Neill, D.H. and Phillips, V.R. 1991. A review of the control of odor nuisance from livestock buildings: part 1, influence of the techniques for managing waste within the building. Journal of Agricultural Engineering Research 50:1-10.

O'Neill, D.H. and Phillips, V.R. 1992a. A review of the control of odor nuisance from livestock buildings: Part 3, Properties of the odorous substances which have been identified in livestock wastes or in the air around them. Journal of Agricultural Engineering Research 53:23-50.

O'Neill, D.H., Stewart, W.I. and Phillips, V.R. 1992b. A review of the control of odor nuisance from livestock buildings: Part 2, The cost of odour abatement systems as predicted from ventilation requirements. Journal of Agricultural Engineering Research 53:23-50.

Ortatatli, M. and Oguz, H. 2001. Ameliorative effects of dietary clinoptilolite on pathological changes in broiler chickens during aflatoxicosis. Research in Veterinary Science 71:59-66.

Papaioannou, D.S., Kyriakis, S.C., Papasteriadis, A., Roumbies, N., Yannakopoulos, A. and Alexopoulos, C. 2002. Effect of in-feed inclusion of a natural zeolite (clinoptilolite) on certain vitamin, macro and trace element concentrations in the blood, liver and kidney tissues of sows. Reseach in Veterinary Science 72:61-68.

Pecover, S.R. 1987. A review of the formation and geology of natural zeolite. In: Natural zeolite, New South Wales Geological Survey, Report GS 1987/145, Department

of Mineral resources, Australia.

Perkins, S.L. and Feddes, J.J.R. 1996. The effect of timing of floor application of mineral oil on dust concentrations in a pig farrowing unit. Canadian Agricultural Engineering 38:123-127.

Pond, W.G. 1984. Response of growing lambs to clinoptilolite or zeolite Na added to corn, corn-fish meal and corn-soybean meal diets. Journal of Animal Science 59:1320-1327.

Pond, W.G. 1989. Effects of dietary protein level and clinoptilolite on the weight gain and liver mineral response of growing lambs to copper supplementation. Journal of Animal Science 67:2772-2781.

Pond, W.G., Jen, J.T. and Crouse, J.T. 1989. Tissue mineral element content in pig fed clinoptilolite. Bull. Reviews of Environmental Contaminationa and Toxicology 42:735-742.

Pond, W.G. and Yen, J.T. 1982. Response of growing pig to dietary clinoptilolite from two geographic sources. Nutrition Reports International 25(5):837-848.

Pond, W.G., Yen, J.T. and Varel, V.H. 1988. Response of growing pig to dietary copper and clinoptilolite supplementation. Nutrition Reports International 37:795-803.

Poulsen, H.D. and Oksbjerg, N. 1995. Effects of dietary inclusion of a zeolite (clinoptilolite) on performance and protein metabolism of young growing pigs. Animal Feed Science and Technology 53:297-303.

Reynolds, S.J., Donham, K.J., Whitten, P., Merchand, J.A., Burmeister, L.F. and Popendorf, W.J. 1996. Longitudinal evaluation of dose-response relationships for environmental exposures and pulmonary function in pig producer workers. American Journal of Industrial Medicine 29:33-40.

Ritter, W.F. 1989. Odor control of livestock wastes: State-of-the-art in North America. Journal of Agricultural Engineering Research 42:51-62.

Roderick, I.M., Peter, G.S. and Vincent, H. V. 1998. Biochemical identification and biological origin of key odor components in livestock waste. Journal of Animal Science 76:1331–1342.

Rolland, D.A., Laurent, S.M. and Orloff, H.D. 1985. Shell quality as influenced by zeolite with high ion-exchange capacity. Poultry Science 64:1177-1187.

Ruurd, T.Z., Matt, A.O., Symeon, Z. and David, E. 2001. Diet Manipulation to Reduce

Nutrient Content in Pig Manure. University of Saskatchewan, Saskatoon, Canada.

SAS Institute, 1999. SAS/STAT User's Guide, Version 8. SAS Institute, Inc., Cary, NC.

Schiffman, S.S. 1998. Livestock odors: implications for human health and well-being. Journal of Animal Science 76:1343-1355.

Schulte, D.D. 1997. Critical parameters for emissions. In: Proceedings of the international Symposium on Ammonia and Odor Control from Animal Prodcution Facility. Vinkeloord, The Netherlands, 23-32.

Shurson, G.C., Ku, P.K., Miller, E.R. and Yokoyama, M.T. 1984. Effects of zeolite A or clinoptilolite in diets of growing pig. Journal of Animal Science 59:1536-1545.

Siemers, V. and Van den Weghe, H. 1997. Biofilteer/ wetscrubber combination for the reduction of ammonia, odour and dust emissions of pig fattening houses. In: Proceedings of the International Symposium on Ammonia and Odor Control from Animal Production Facility. Vinkeloord, The Netherlands, 537-544.

Simard, R.R., Cluis, D., Gangbazo, G. and Beauchemin, S. 1995. Phosphorus status of forest and agricultural soils from a wastershed of high animal density. Journal of Environmental Quality 24:420-425.

Smith, D.R., Moore, P.A. Jr., Maxwell, C.V., Haggard, B.E.and Daniel, T.C. 2004. Reducing phosphorus runoff from pig manure with dietary phytase and aluminum chloride. Journal of Environmental Quality 33:1048-54.

Sneath, R.W., Burton, C.H. and Williams, A.G. 1992. Continuous aerobic treatment of piggery slurry for odor control scaled up to a farm-size unit. Journal of Agricultural Engineering Research 53:81-92.

Statistics Canada. 1996. Agricultural Financial Statistics, Catalog no.21-205-XPB. Statistic Canada, Ottawa, Canada.

Statistics Canada. 1997. Agricultural Profile of Quebec, Catalog no.95-176-XPB. Statistic Canada, Ottawa, Canada.

Statistics Canada. 2001. Hogs Marketed in Canada by Province,1984-2004.

Statistics Canada. 2003. Total Pigs on Farms by Province (Inventory),1986 – 2004.

Sutton, A.L., Kephart, K.B., Patterson, J.A., Mumma, R., Kelly, D.T., Bogus, E., Jones, D.D. and Heber, A. 1996. Manipulating pig diets to reduce ammonia and odor

emissions. In Proc. 1st Int. Conf. Air Pollution from Agric. Operations, Kansas City, MO. 445–452

Sutton, A.L., Kephart, K.B., Patterson, J.A., Mumma, R., Kelly, D.T., Bogus, E., Jones, D.D. and Heber, A. 1997. Dietary manipulation to reduce ammonia and odorous compounds in excreta and anaerobic manure storage. In: Proceedings of the International Symposium on Ammonia and Odor Control from Animal Production Facility. Vinkeloord, The Netherlands, 245-252.

Sutton, A.L., Kepphart, K.B., Verstegen, M.W.A., Canh, T.T. and Hobbs, P.J. 1999. Potential for reduction of odorous compounds in pig manure through diet modification. Journal of Animal Science 77:430-439.

Sweeny, T.F., Cervantes, A., Bull, L.S. and Hemken, R.W. 1984. Effect of dietary clinoptilolite on digestion and rumen fermentation in steers. In: Zeo-Agriculture. Use of natural zeolites in agriculture and aquaculture. International Committee on Natural Zeolite, Editors: W.G. Pond and F.A. Mumpton. Westview Press Inc., Boulder, Colorado. 183-193.

Swine Odor Task Force. 1995. Option for managing odor. Report from the North Carolina Pig Odor Task Force. Agricultural Research Service, North Carolina State University. (Internet document, 47pp).

Tabbara, H. 2003. Phosphorus loss to runoff water twenty-four hours after application of liquid pig manure or fertilizer. Journal of Environmental Quality 32:1044-52.

Takai, H., Moller, F., Iversen, M., Jorsal, S.E. and Bille-Hasen, V. 1993. Dust control in pig buildings by spraying of rapeseed oil. In: International Livestock Environment Symposium, St-Joseph, MI, USA: ASAE, 726-733.

Tamminga, S. 1996. A review on environmental impacts of nutritional strategies in ruminants. Journal of Animal Science 74:3112-3124.

Thilsing-Hansen, T., Jorgensen. R.J., Enemark, J.M. and Larsen, T. 2002a. The effect of zeolite A supplementation in the dry period on periparturient calcium, phosphorus, and magnesium homeostasis. Journal of Dairy Science 85:1855-1862.

Thilsing-Hansen, T., Jorgensen, R.J., Enemark, J.M.D., Zelvyte, R. and Sederevicius, A. 2002b. The effect of zeolite A supplementation in the dry period on blood mineral status around calfing. Acta Veterinaria Scandinavica Supplementum 97:87-95.

Tuitoek, K., Young, L.G., de Lange, C.F.M. and Kerr, B.J. 1997. The effect of reducing excess dietary amino acids on growing-finishing pig performance: An evaluation of the ideal protein concept. Journal of Animal Science 75:1575–1583.

Turner, L.W., Cromwell, G.L., Bridges, T.C., Carter, S. and Gates, R.S. 1996. Ammonia (NH_3) emission from pig waste as influenced by diet manipulation. in Proc. 1st Int. Conf. Air Pollution from Agric. Operations, Kansas City, MO. 453–458.

USDA, Agricultural Research Service. 2003. Air quality: malodorous compounds. http://www.ars.usda.gov

Valencia, Z. 1996. Phytase and lignin in the diet of piglets. Part 1: Nutritional implications of microbial phytase supplementation in the diet of early weaned piglets. MSc. Thesis. McGill University, Quebec, Canada.

Vrzgula, L. and Bartko, P. 1984. Effects of clinoptilolite on weight gain and some physiological parameters of pig. In: Zeo-Agriculture. Use of natural zeolites in agriculture and aquaculture. International Committee on Natural Zeolite, Editors: W.G. Pond and F.A. Mumpton. Westview Press Inc., Boulder, Colorado. 161-166.

Ward, T.L., Watkins, K.L. and Southern, L.L. 1993. Research Note: Interactive effects of sodium zeolite A and Eimea acervulina infection on growth and tissue minerals in chicks. Poultry Science 72:2172–2175.

Ward, T.L., Watkins, K.L., Southern, L.L., Hoyt, P.G. and French, D.D. 1991. Interactive effects of SZA and Cu in growing pig: growth and bone and tissue mineral concentrations. Journal of Animal Science 69:726-733.

Watkins, K.L., Vagnoni, D.B. and Southern, L.L. 1989. Effect of dietary SZA and excess Ca on growth and tibia Ca and P concentration in uninfected and Eimea acervulina-infected chicks. Poultry Science 68:1236-1240.

Williams, P.E.V. and Kelly, J.M. 1994. Animal production and pollution problems. In: Livestock Production for 21st Century. Priorities and research needs ed. P.A. Thacker. University of Saskatchewan Press, Saskatoon, Canada, 159-186.

Williams, A.G. and Nigro, E. 1997. Covering slurry stores and effects on emissions of ammonia and methane. In: Proceedings of the International Symposium on Ammonnia and Odour Control from Animal Prodcution Facility. Vinkeloord, The Netherlands, 421-428.

Williams, A.G., Shaw, M. and Adams, S. J. 1984. The biological stability of aerobically-treated piggery slurry during storage. Journal of Agricultural Engineering Research 29:231-239.

Williams, A.G., Shaw, M., Selviah, C.M. and Cumby, R.J. 1989. The oxygen requirements for deodorising and stabilising pig slurry by aerobic treatment. Journal of

Agricultural Engineering Research 43:291-311.

Young, J.S., Classen, J.J., Bottcher, R.W. and Westerman, P.W. 1997. Biofiltration system for testing the reduction of odor from pig building. In: Proceedings of the International Symposium on Ammonia and Odor Control from Animal Production Facility. Vinkeloord, The Netherlands, 521-528.

Zhang, Y., Nijssen, L., Barber, E.M., Feddes, J.J.R. and Sheridan, M. 1994. Applying mineral oil to reduce dust in pig buildings. Transactions of ASAE 100:1043-1050.

Zhu, J., Bundy, D.S., Li, X. and Rashid, N. 1997a. A procedure and its application in evaluating pit additives for odor control. Canadian Agricultural Engineering 39:207-214.

Zhu, J., Bundy, D.S., Li, X.W. and Rashid, N. 1997b. The hindrance in the development of pit additive products for pig manure odor control-A review. Journal of Environmental Science and Health A32:2429-2448.

Zhu, J. 2000. A review of microbiology in pig manure odor control. Agriculture, Ecosystems and Environment 78:93-106.

Guidelines for completing the form are available at www.mcgill.ca/rgo/animal/

McGill University
Animal Use Protocol – Research

Protocol #: 4461
Investigator #: 939
Approval End Date: MAY 31, 2005
Facility Committee: ACR

tle: Zeolite as natural swine feed additive to reduce the environmental impact of manure
ust match the title of the funding source application)

New Application ☒ **Renewal of Protocol #** 4461 ☐ **Pilot** **Category** *(see section 11)*: B

Investigator Data:

ncipal Investigator: Suzelle Barrington **Phone #:** 514 -398-7776
it/Department: Department of Bioresource Engineering **Fax#:** 514 -398-8387
dress: Macdonald Stewart MS1024, Macdonald Campus **Email:** suzelle.barrington@mcgill.ca

Emergency Contacts: Two people must be designated to handle emergencies

me: Suzelle Barrington **Work #:** 514-398-7776 **Emergency #:** (450) 825-2530
me: Denis Hatcher **Work #:** 514-398-8644 **Emergency #:** (514) 457-9276)

Funding Source:

ternal ☒
urce (s): NSERC and FPPQ
er Reviewed: ☒ YES ☐ NO**
atus : ☒ Awarded ☐ Pending
nding period: until summer 2005

Internal ☐
Source (s): _____
Peer Reviewed: ☐ YES ☐ NO**
Status: ☐ Awarded ☐ Pending
Funding period: _____

For Office Use Only:

ACTION	✓	DATE
CCs		
DB	✓	May 7/04
APPROVED		

All projects that have not been peer reviewed for scientific merit by the funding source require 2 Peer Review Forms to be npleted e.g. Projects funded from industrial sources. Peer Review Forms are available at www.mcgill.ca/rgo/animal

oposed Start Date of Animal Use (d/m/y): June 1st 2004 **or ongoing** ☒
pected Date of Completion of Animal Use (d/m/y): June 1st 2005 **or ongoing** ☐

vestigator's Statement: The information in this application is exact and complete. I assure that all care and use of animals in this oposal will be in accordance with the guidelines and policies of the Canadian Council on Animal Care and those of McGill University. I shall uest the Animal Care Committee's approval prior to any deviations from this protocol as approved. I understand that this approval is valid one year and must be approved on an annual basis.

incipal Investigator's signature:		**Date:** March 24th/2004
iair, Facility Animal Care Committee:		**Date:** 30 March 2004
iiversity Veterinarian:		**Date:**
iair, Ethics Subcommittee (as per UACC policy):		**Date:**
pproved Animal Use	**Beginning:** June 1, 2004	**Ending:** MAY 31, 2005

] This protocol has been approved with the modifications noted in Section 13.

Guidelines for completing the form are available at www.mcgill.ca/rgo/animal/

McGill University
Animal Use Protocol – Research

tle: Zeolite as natural swine feed additive to reduce the environmental impact of manure
ust match the title of the funding source application)

New Application ☒ **Renewal of Protocol #** 4461 ☐ **Pilot** **Category** *(see section 11)*: B

Investigator Data

incipal Investigator: Suzelle Barrington **Phone #:** 514 -398-7776

it/Department: Department of Bioresource Engineering **Fax#:** 514 -398-8387

ldress: Macdonald Stewart MS1024, Macdonald Campus **Email:** suzelle.barrington@mcgill.ca

Emergency Contacts

me: Suzelle Barrington **Work #:** 514-398-7776 **Emergency #:** (450) 825-2530

me: Denis Hatcher **Work #:** 514-398-8644 **Emergency #:** (514) 457-9276)

Funding Source(s)

cternal ☒	**Internal** ☐
ource (s): NSERC and FPPQ	**Source (s):**
er Reviewed: ☒ YES ☐ NO**	**Peer Reviewed:** ☐ YES ☐ NO**
atus : ☒ Awarded ☐ Pending	**Status:** ☐ Awarded ☐ Pending
nding period: until summer 2005	**Funding period:**

All projects that have not been peer reviewed for scientific merit by the funding source require 2 Peer Review Forms to be mpleted e.g. Projects funded from industrial sources. Peer Review Forms are available at www.mcgill.ca/rgo/animal

oposed Start Date of Animal Use (d/m/y): June 1st 2004 **or ongoing** ☒

pected Date of Completion of Animal Use (d/m/y): June 1st 2005 **or ongoing** ☐

Investigator's Statement

rincipal Investigator's signature:		**Date:**

Approved by:

hair, Facility Animal Care Committee:		**Date:**
niversity Veterinarian:		**Date:** May 5, 2004
hair, Ethics Subcommittee (as per UACC policy):		**Date:**
pproved Animal Use		**Ending:**

☐ This protocol has been approved with the modifications noted in Section 15.

0 7 MAI 2004

Research Personnel and Qualifications

the duties of the Principal Investigator and of all individuals who will be in contact with animals in this study and their employment classification (Investigator, technician, research assistant, undergraduate, graduate student, fellow). If undergraduate students are involved, the role of the student and the supervision received must be described. Training is mandatory for all personnel listed here. Refer to *www.animalcare.mcgill.ca* for details. Each person listed in this section must sign to indicate that s/he has read this protocol. *(Space will expand as needed)*

Name	Classification	Animal Related Training Information	Occupational Health & Safety Program *	Signature
elle Barrington	Project coordinator		On-line AC. training done	
asha Lapointe	M.Sc. student	Trained by D. Hatcher	To take on-line course	
nghong Wan	M. Sc. student	Being trained by D. Hatcher	Took AC course given for handling animals	
nis Hatcher	Animal Science Coordinator			
Pika	Technician		On-line AC. training done	
X. Zhao	Co-supervisor		On-line AC. training done	

students working with the animals are applying for the occupational health program.

**ndicate for each person, if participating in the local Occupational Health & Safety Program, see *www*
ails.**

Summary (in language that will be understood by members of the general public)

5 a) AIMS AND BENEFITS: Describe, in a short paragraph, the overall aim of the study and its potential benefit to human/animal health or to the advancement of scientific knowledge.

ducing the impact of manure on soil, air and water; improving the digestion efficiency of hogs; improving carcass quality and at value with no side effect on food safety. Demonstrating the mechanism by which zeolite can improve feed digestion.

5 b) SPECIFIC OBJECTIVES OF THE STUDY: Summarize in point form the primary objectives of this study.

tablish optimum protein to energy ratio in feed supplement with 4% zeolite for lean carcasses; demonstrate the potential for olite supplemented feed to improve piggery air quality; measure the impact of zeolite on manure mass, nutrient content, N ses and odour produced, as well as herd productivity. Demonstrate mechanism by which zeolite can improve feed digestion.

5 c) Indicate if and how the current goals differ from those in last year's application.

om July 2003 to November 2003, 192 pigs were used to identify the level of zeolite inclusion in the feed and the feed protein d energy dilution value, to obtain the best feed conversion and carcass quality. The results showed at 4% was the best zeolite lusion level and that the feed energy digestion was improved by 5%, as compared to 3% for the feed protein. The zeolite had impact on muscles, live and kidneys heavy metal content, when fed up to a level of 6%. We now need to show over-all nefits of zeolite (gain, room air and carcass quality) by a trial using a different room for hogs fed with and without zeolite. We o need to conduct test showing lower manure mineral content and less manure odours and zeolite digestion mechanisms (only tabolic cages allow us to collect individual hog manure samples). We also need to refine protein to energy ratio in the ration lean carcasses.

5 d) List the section / subsection numbers where significant changes have been made

is is an ongoing research project and the proposed test sequence is respected.

5 e) KEYWORDS: Using keywords only, list the procedures used on animals (e.g. anaesthesia, breeding colony, injection IP, gavage, drug administration, major survival surgery, euthanasia by exsanguination, behavioural studies). For a more complete list of suggested keywords refer to Appendix 1 of the Guidelines (www.mcgill.ca/rgo/animal/).

ite digestion mechanism, improved feed conversion, manure environmental impact.

Animals Use data for CCAC

6 a) Purpose of Animal Use (Check most appropriate one):

1. ☒ **Studies of a fundamental nature/basic research**
2. ☐ **Studies for medical purposes relating to human/animal diseases/disorders**
3. ☐ **Regulatory testing**
4. ☐ **Development of products/appliances for human/veterinary medicine**
5. **If for Teaching, use the Animal Use Protocol form for Teaching (www.mcgill.ca/rgo/animal)**

6 b) Will field studies be conducted? NO ☒ YES ☐ If yes, complete "Field Study Form"
Will the project involve the genetically altering animals? NO ☒ YES ☐ If yes, complete SOP #5 or #6
Will the project involve breeding animals? NO ☒ YES ☐ If breeding transgenics or knockouts, complete SOP#4

Animal Data

7 a) Please justify the need for live animals versus alternate methods (e.g. tissue culture, computer simulation)

The experiment is designed to test the effect of feeding zeolite on nutrient digestion, carcass quality, manure nutrient level, building air quality. The number of test hogs has been examined statistically, and is required to insure the significance of the ılts.

7 b) Describe the characteristics of the animal species selected that justifies its use in the proposed study (consider racteristics such as body size, species, strain, data from previous studies or unique anatomic/physiological features)

grower hogs weighing from 25 to 105kg.

7 c) Description of animals

Disease Control Assurance: To prevent introduction of infectious diseases into animal facilities, a health status report or veterinary inspection certificate may be required prior to receiving animals from all non-commercial sources or from commercial sources whose animal health status is unknown or questionable. Quarantine and further testing may be required for these animals.

If more than 6 columns are needed, please attach another page

	Sp/strain 1	Sp/strain 2	Sp/strain 3	Sp/strain 4	Sp/strain 5	Sp/strain 6
ecies	hogs					
pplier/Source	Haybay genetics					
·ain	Duroc x Landrace x Yorkshire					
ĸ	both					
ge/Wt	25-105kg					
To be purchased	408hogs					
Produced by in-use breeding						
Other g.field studies)						

eded at one e	204hogs					
er cage	1(metabolic) 6 (grower room)					
TAL# /YEAR	**408 hogs**					

7 d) Justification of Animal Usage: BASED ON THE EXPERIMENTAL OBJECTIVES OF THE PROJECT, cribe the number of animals required for one year. Include information on experimental and control groups, # per up, and failure rates. For breeding, specify how many adults are used, number of offspring produced, and how many pring are used in experimental procedures. Use the table below when applicable. The arithmetic explaining how the l of animals for each column in the table above is calculated should be made clear. *(Space will expand as needed)*

show the global effect of zeolite in the feed, we need to grow one group of hogs in a separate room and compare their formance to another group grown in another room. The treatment needs to be reversed per room, to show no room interaction. ce each room holds 96hogs, then each test requires 192hogs, repeated twice for 384hogs.
the metabolic tests, we need to compare three feeds to test for C.P. (protein) to energy ratio along with the effect of 4% lite in the feed. We need to test 8 hogs per treatement, to lower variance within treatments. The floor of the metabolic cages is lt of a plastic-covered wire mesh, which is more confortable than the concrete slabs, used in the piggery grower rooms. erefore, foot lesion is not at risk. The animals will be kep in metabolic cages for 10 days, maximum: 3 to 5 days to get them d to the cages (if a hog refuses to eat during this period, it is replaced) and 5 days on test.

table) The following table may help you explain the animal numbers listed in the 7c table:

ble will expand as needed)	Sp/strain 1	Sp/strain 2	Sp/strain 3	Sp/strain 4	Sp/strain 5	Sp/strain 6
t agents or procedures	4% zeolite	4% zeolite				
f animals per group	96 grower hogs	12 grower hogs				
sage / route of administration	feed	feed				
f endpoints	2	2				
her variables (sex,genotypes…)						
tal number of animals per year	**384**	**24**				

Animal Husbandry and Care

8 a) If projects involves non-standard cages, diet and/or handling, please specify

Standard metabolic cages are required and hogs are kept in these cages for a maximum period of 10 days.

8 b) Is there any component to the proposed procedures which will result in immunosuppression or decreased mune function (e.g. stress, radiation, steroids, chemotherapeutics, genetic modification of the immune system)?

NO ☒ YES ☐ **if yes, specify:**

8 c) Indicate area(s) where animal use procedures will be conducted:

Building: **Room:**

Indicate area(s) all facilities where animals will be housed:

Building: Piggery **Room:** Grower rooms and metabolic room

If animal housing and animal use are in different locations, briefly describe procedures for transporting animals:

Standard Operating Procedures (SOPs)

nplete this section if you plan to use any of the UACC SOPs listed below. IT IS UACC POLICY THAT THESE SOPS USED WHEN APPLICABLE. Any proposed variation of the SOPs must be described and justified. The Standard erating Procedures can be found at the UACC website at www.mcgill.ca/rgo/animal . The completed and signed SOP n must be attached to the protocol.

ck all SOPs that will be used:

od Collection UACC#1	☐	**Collection of Amphibian Oocytes UACC#9**	☐
esthesia in rodents UACC#2	☐	**Rodent Survival Surgery UACC#10**	☐
lgesia in rodents UACC#3	☐	**Anaesthesia & Analgesia Neonatal Rodents UACC#11**	☐
eding transgenics/knockouts UACC#4	☐	**Stereotaxic Survival Surgery in Rodents UACC#12**	☐
nsgenic Generation UACC#5	☐	**Field Studies Form**	☐
ockout/in Generation UACC#6	☐	**Phenotype Disclosure Form**	☐
duction of Monoclonal Antibodies UACC#7	☐	**Other, specify:**	☐
duction of Polyclonal Antibodies UACC#8	☐		☐

Description of Procedures

), IF A PROCEDURE IS COVERED BY AN SOP, WRITE "AS PER SOP", NO FURTHER DETAIL IS QUIRED.

R EACH EXPERIMENTAL GROUP, DESCRIBE ALL PROCEDURES AND TECHNIQUES, WHICH ARE NOT RT OF THE SOPs, IN THE ORDER IN WHICH THEY WILL BE PERFORMED – surgical procedures, nunizations, behavioural tests, immobilization and restraint, food/water deprivation, requirements for post-operative e, sample collection, substance administration, special monitoring, etc Appendix 2 of the Guidelines ww.mcgill.ca/rgo/animal/) provides a sample list of points that should be addressed in this section.

o types of tests are planned: 1) a test in the two grower rooms of the pigegry were 3 feeds will be tested : with 4% zeolite and % C.P. and energy; with 4% zeolite and 90% C.P. and 85% energy; and no zeolite with 100% C.P. and energy. The 100% els of C.P. and energy are those recommended for grower hogs (NRC), and will be adjusted as they grow. One grower room house 2 x 48 hogs, each two groups of 48 hogs being fed 4% zeolite and one of the two C.P./energy ratio. The other 96 hogs ised in the other room will be on the feed without zeolite and 100% C.P. and energy. The objective of this test is to compare m air quality for animals on feed with and without zeolite; for those hogs on zeolite feed within the same room, the objective o compare the effect of feed C.P. to energy ratio on carcass leaness. It was earlier found that zeolite is more efficient at roving energy than C.P. digestion. The test will be repeated twice, to switch room treatment and eliminate room effect. For test, hog rate of weight gain, feed consumption and feed conversion will be compared among treatments. The room air lity will be monitored and compared. The hogs will be supplied by Haybay genetics, the regular supplyer of the Macdonald npus piggery, and these hogs will be grown from 25 to 105kg. The carcass quality of these hogs will be examined at slaughter ise while cutting the carcasses.

a test using metabolic cages where 3 feed treatments will be applied to 8 hogs each weighing 60kg (24 in total). Because there only 12 cages, the metabolic procedure will be repeated twice, using 4 hogs per treatment, each time. The feeds to be tested : without zeolite with 100% C.P. and energy; with 4% zeolite and 90% C.P. and energy; 4% zeolite with 90% C.P. and 85% rgy. The 100% C.P. and energy levels are those recommended for 60kg grower hogs (NRC). For all three feeds, faeces and ne will be collect for mineral mass balance and for manure odour emissions. The faeces and urine will be collected separately compare the effect of zeolite on manure properties and feed digestion. Males will be used for this test, to eliminate the use of heters to collect urines separatly from the feces, to impose less stress on the hogs. Blood samples will be collected three times fore testing, on first day of testing and on last day of testing) to measure the effect of zeolite on blood mineral content and test hypothesis that zeolite regulates protein digestion. The feed will contain 1% iron oxide and 0.5% chromium oxide to measure d retention time and feed digestibility, respectively, to test for zeolite feed digestion mechanism. This is a standard procedure d by scientists. Animals other than those on test (1) above will be used as not to disturb the statistical design of test (1).

e blood samples will be taken through the ear vein (marginal vein) using a sterilized needle. Mr. Jan Pika will take the blood nples. Animals showing any sign of sickness are placed alone in a pen and treated as required. If not improving, the farm uses hanasia with a dead bolt. Euthanasia is performed on animals that were injured accidently and cannot stand or has not been ing for a few days or is unable to drink.

b) Experimental endpoint – for each experimental group indicate survival time

e experimental hogs conducted in the grower rooms will be sent for slaughter at 105kg. Carcass quality is measured by ghter house when the carcass are being cut (fat level, loin mass, etc.). The hogs used for the metabolic tests will be returned

) Clinical endpoint – describe the conditions, complications, and criteria (e.g. >20% weight loss, maximum tumour , vocalizing, lack of grooming) that would lead to euthanasia of an animal before the expected completion of the eriment (specify per species and project if multiple projects involved)

quency of monitoring: Zeolite is a beneficial additive and does not cause any complications. The animals will be fed up to ket weight and then sent to a commercial slaughter house.

) Specify person(s) who will be responsible for animal monitoring and post-procedural care *(must also be listed in ion 4)*

ne: Animal monitoring: Denis Hatcher and Natasha ointe	**Phone #:** 514-398-8644

) Pre-Anesthetic/Anaesthetic/Analgesic Agents: List all drugs that will be used to minimize pain, distress or omfort. If covered in an SOP, write "*As per SOP*", no further details is required. *(Table will expand as needed)*

Species	Agent	Dosage (mg/kg)	Total volume(ml) per administration	Route	Frequency/Duration

) Administration of ALL other substances: List all non-anaesthetic agents under study in the experimental ponent of the protocol, including but not limited to drugs, infectious agents, viruses. If covered in an SOP, write "*As SOP*", no further details is required. *(Table will expand as needed)*

Species	Agent	Dosage (mg/kg)	Total volume(ml) per administration	Route	Frequency/Duration

lite fed in feed at a level of 4%. The 2003 research data on carcass quality indicated no significant heavy metal accumulation zeolite being fed up to 6%.

) Method of Euthanasia

cify Species

	☐ **Anaesthetic overdose, list agent/dose/route:**
	☐ **Exsanguination with anaesthesia, list agent/dose/route:**
	☐ **Decapitation without anaesthesia *** ☐ **Decapitation with anesthesia, list agent/dose/route (including CO_2):**
	☐ **Cervical dislocation without anaesthesia *** ☐ **Cervical dislocation with anaesthesia, list agent/dose/route (including CO_2):**
	☐ **CO_2 chamber only**
	☐ **Other, specify:**
	☒ **Not applicable, explain:** animals will be sent to commercial slaughter house at 105kg.

or physical method of euthanasia without anaesthesia, please justify:

Category of Invasiveness B ☒ C ☐ D ☐ E ☐

egories of Invasiveness (from the CCAC *Categories of Invasiveness in Animal Experiments*). Please refer to this document for a e detailed description of categories.

egory A: Studies or experiments on most invertebrates or no entire living material.

egory B: Studies or experiments causing little or no discomfort or stress. *These might include holding animals captive, injection, utaneous blood sampling, accepted euthanasia for tissue harvest, acute non-survival experiments in which the animals are completely esthetized.*

egory C: Studies or experiments involving minor stress or pain of short duration. *These might include cannulation or eterizations of blood vessels or body cavities under anaesthesia, minor surgery under anaesthesia, such as biopsy; short periods of raint, overnight food and/or water deprivation which exceed periods of abstinence in nature; behavioural experiments on conscious mals that involve short-term stressful restraint.*

egory D: Studies or experiments that involve moderate to severe distress or discomfort. *These might include major surgery under esthesia with subsequent recovery, prolonged (several hours or more) periods of physical restraint; induction of behavioural stresses,*

rdance with University policy).
gory E: **Procedures that involve inflicting severe pain, near, at or above the pain threshold of unanaesthetized, conscious animals.** *confined to but may include exposure to noxious stimuli or agents whose effects are unknown; exposure to drugs or chemicals at levels (may) markedly impair physiological systems and which cause death, severe pain or extreme distress or physical trauma on aesthetized animals.* **According to University policy, E level studies are not permitted.**

Potential Hazards to Personnel and Animals It is the responsibility of the investigator to obtain the necessary hazard and/or Radiation Safety permits before this protocol is submitted for review. opy of these certificates must be attached, if applicable.

lazardous materials will be used in this study: ☒

) Indicate which of the following will be used in animals:

☐ **Toxic chemicals** ☐ **Radioisotopes** ☐ **Carcinogens**
☐ **Infectious agents (includes vectors)** ☐ **Transplantable tumours**

) Complete the following table for each agent to be used (use additional page as required):

t name			
ge			
e of administration			
uency of administration			
tion of administration			
ber of animals involved			
ival time after administration			

) After administration the animals will be housed in:

☐ **the animal care facility** ☐ **laboratory under supervision of laboratory personnel**

Please note that cages must be appropriately labeled at all times.

) Describe potential health risk (s) to humans or animals:

) Describe measures that will be used to reduce risk to the environment and all project and animal facility personnel:

Review/Modifications (to be completed by ACC only): The Animal Care Committee has made the following modification(s) to this animal use procedure protocol during the review process. Please make these changes to your copy to comply with the recommended changes as a condition of approval.

Canadian Food Inspection Agency | Agence canadienne d'inspection des aliments
Animal Health & Production Division
Ottawa, Ontario
K1A 0Y9
Fax: (613) 228-6614

October 16, 2003 JCG.SIF

Mr. J.C. Guilmain
J.C. Guilmain, Inc.
1034 Rang 20
Upton, Québec
J0H 2E0

Re: Application for Temporary Feed Registration

Dear Mr. Guilmain:

This letter is to inform you that a temporary registration (Registration No. T990700) is being granted for KMI zeolite (Registration No. T990700) to authorize the disposal of swine from this research trial that have been fed diets containing 4% and 6% KMI zeolite for slaughter. **This temporary registration expires on March 31, 2005**.

If you wish to register KMI zeolite in the future at levels greater than 2% in livestock feed, then the following information will be required:

1. Tissues from the current study should be held and analysed for heavy metals (arsenic, cadmium, chromium and lead) for liver, kidney and muscle from three pigs fed diets containing 4% and 6% KMI zeolite; and

2. Histopathology (as discussed previously) will be required for muscle, kidney and liver for four pigs at levels feds.

Please note that this ingredient has only been evaluated for safety and not for efficacy. Therefore, currently KMI zeolite is only approved as a flowing/anti-caking agent not to exceed to 2% in finished feed.

You have been charged fees in the amount of CAN$304.95 for the consideration of this application and this fee has been paid in full. If you have any questions, please do not hesitate to contact me at (613) 225-2342 ext. 4140.

Yours sincerely,

Paul Loeven, Toxicologist,
Feed Section.

c.c. Jacques Fafard, CFIA Québec Area Office
Catherine Italiano, CFIA Headquarters
Dr. Suzelle Barrington, McGill University

P:\New Ingredients\Zeolite Ore\temporary reg for KMI zeolite 6% Oct. 16, 2003.wpd

KMI Zeolite, Inc.

HCR37 Box 52

Sandy Valley, Nv. 89019

Material Safety Data Sheet	U.S. Department of Labor
	OMB No. 1218-0072
IDENTITY (as Used on Label and List)	
CLINOPTILOLITE / CLINO	
Section I	
KMI ZEOLITE, INC.	Emergency Telephone Number 1-800-878-0989 702-722-5725
HCR 37 Box 52	Telephone Number for Information 1-702-723-8415
Sandy Valley, Nv 89019	
	Date Prepared 06-18-2003
	Signature of Preparer (optional)

Section II-Hazardous Ingredients/Identity Information

Hazardous Components (Specific Chemical Identity, Common Name(s))	OSHA PEL	ACGIH TLV	%
(optional)			
N/A			

Section III Physical/Chemical Characteristics

Boiling Point	N/A	Specific Gravity (H_2O = 1)	1.5 - 1.7
Vapor Pressure (mm Hg)	N/A	Melting Point	N/A
Vapor Density (Air = 1)	N/A	Evaporation Rate (Butyl Acetate = 1)	N/A
Solubility in Water			
0 (Zero)			

Appearance and Odor

Crystalline sand-like appearance with earth-like odor.

Section IV-Fire and Explosion Hazard Data

Flash Point (Method Used)	Flammable Limits	LEL	UEL
N/A	N/A	N/A	N/A

Extinguishing Media N/A

Special Fire Fighting Procedures None Known/Applicable

Unusual Fire and Explosion Hazards None

(Reproduce locally) OSHA 174 Sept. 1985

Section V-Reactivity Data

Stability	Unstable		Conditions to Avoid
	Stable	X	N/A

Incompatibility (Materials to Avoid)

None

Hazardous Decomposition or Byproducts

None

Hazardous Polymerization	May Occur		Conditions to Avoid
N/A	Will Not Occur	X	N/A

Section VI Health Hazard Data

Route(s) of Entry	Inhalation?	Skin?	Ingestion?
Inhalation	Possible	N/A	Possible / Harmful

Health Hazards (Acute and Chronic)

None

Carcinogenicity	NTP?	IARC Monographs?	OSHA Regulated?
Non-Carcinogenic	N/A	N/A	For dust control only

Signs and Symptoms of Exposure

None

Medical Conditions Generally Aggravated by Exposure

None

Emergency and First Aid Procedures

Flush eyes to remove particles-wash skin with mild soapy water

Section VII-Precautions for Safe Handling and Use

Steps to Be Taken in Case Material Is Released or Spilled

It is recommended, to ensure compliance with various state regulations, that any major

quantities spilled in surface water be reported to appropriate state officials.

Waste Disposal Method

Spills on ground should be cleaned up in a manner that does not generate dust and disposed of in a landfill area.

Precautions to be Taken in Handling and Storing

N/A

Other Precautions

N/A

Section VII-Control Measures

Respiratory Protection (Specify Type)

OSHA approved dust respirators where dust is a problem.

Ventilation	Local Exhaust	Special
N/A	N/A	N/A
	Mechanical (General)	Other
	N/A	N/A

Protective Gloves	Eye Protection
N/A	Safety glasses or goggles to prevent dust from entering the eyes

Other Protective Clothing or Equipment

N/A

Work/Hygienic Practices

Keep work areas clean and dust free as much as possible. Use misters if required.

CORE LABORATORIES

QUALITY: A CONTINUING COMMITMENT AT CORE LABORATORIES

FAX COVER SHEET

TO:
COMPANY McGill University
ATTN: Suzelle Barrington
FAX NO.: (514) 398-8387

June 9, 2003

FROM:
COMPANY: CORE LABORATORIES CANADA LTD.
NAME: Trevor Finlayson
PHONE #: (403) 250-4037
PAGE: 3 (Including this page)

Dear Ms. Barrington,

Please find attached the results of one X-ray diffraction analysis performed on the submitted zeolite sample. This sample was analyzed for bulk mineralogy, and then compared to the standard sample (also attached).

Thank you for the opportunity of providing this service. Should you have any further questions or comments, please feel free to contact us.

Best Regards,

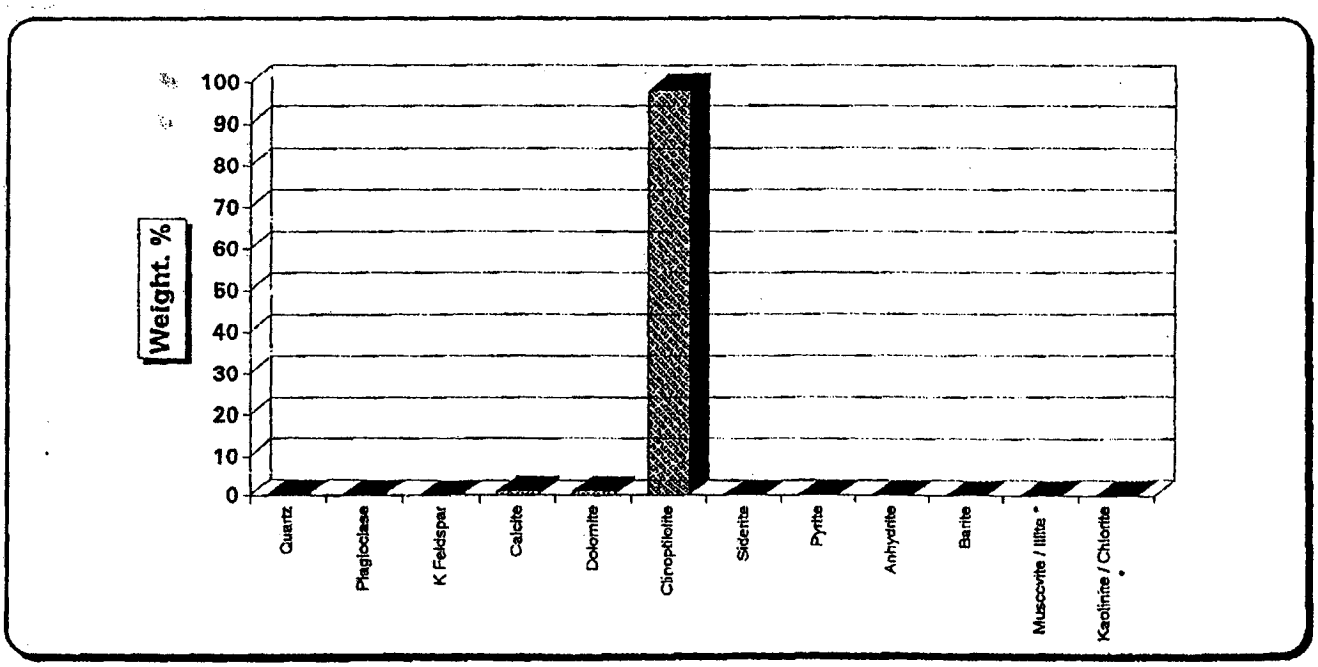
Trevor Finlayson
Lab Coordinator
Core Laboratories Canada Ltd.

PLEASE CONTACT HONG SHI MESSAGE IS NOT CLEARLY RECEIVED AT TEL: (403) 250-4005, FAX: (403) 250-4012

COMPANY: McGill University
WELL / LOCATION: Faculty of Agricultural and Environmental Sciences
SAMPLE: Zeolite Sample
DEPTH:

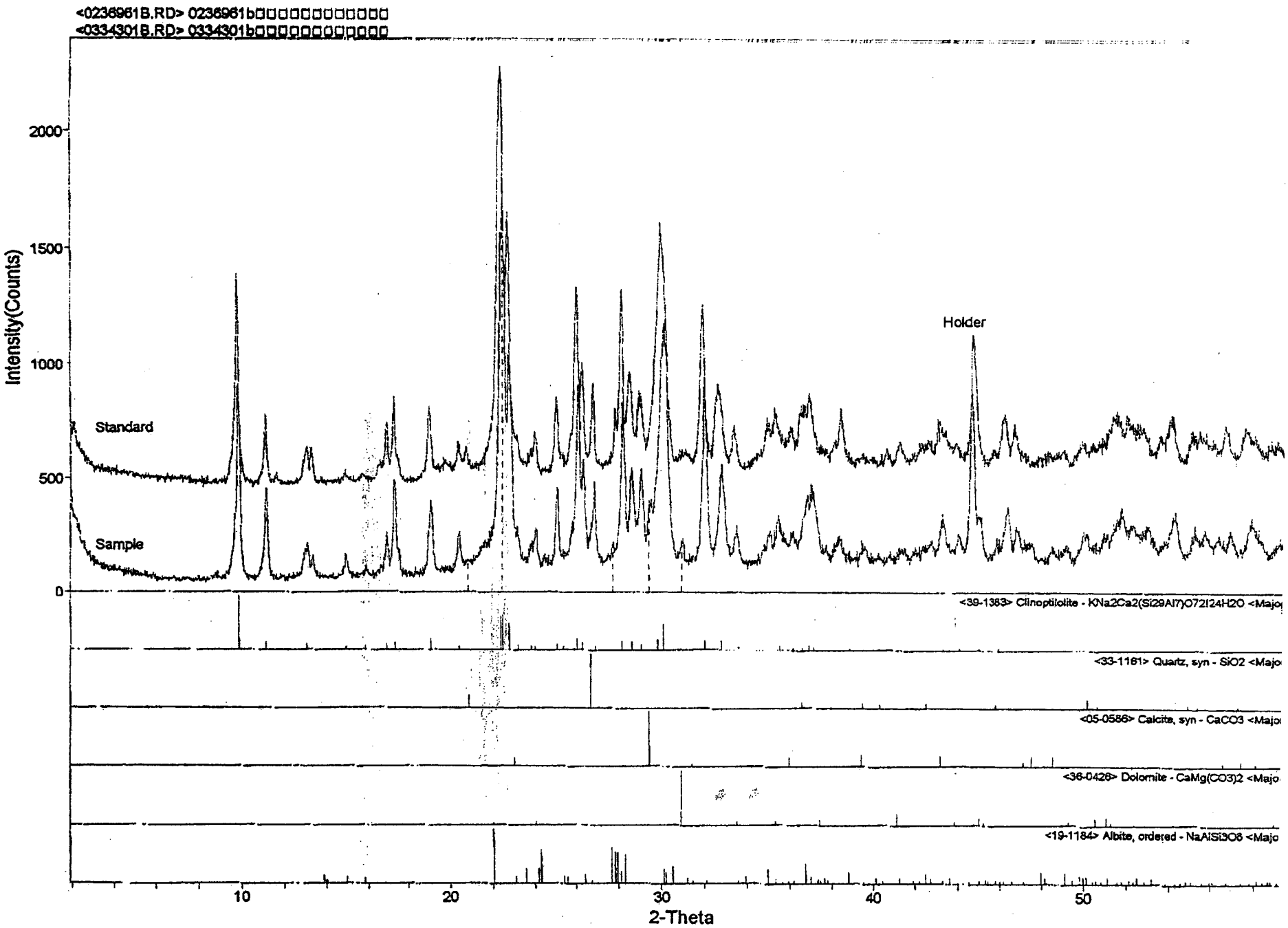
File #: 52135-03-3430

		BULK COMPOSITION (WEIGHT %)
Quartz	(SiO_2)	Trace
Plagioclase	($NaAlSi_3O_8$ - $CaAl_2Si_2O_8$)	Trace
K Feldspar	($KAlSi_3O_8$)	0
Calcite	($CaCO_3$)	1
Dolomite	($[CaMg]CO_3$)	1
Clinoptilolite	($KNa_2Ca_2(Si_{29}Al_7)O_{72}.24H_2O$)	98
Siderite	($FeCO_3$)	0
Pyrite	(FeS_2)	0
Anhydrite	($CaSO_4$)	0
Barite	($BaSO_4$)	0
Muscovite / Illite *	($KAl_2[AlSi_3O_{10}][OH]_2$)	0
Kaolinite / Chlorite *	($Al_2Si_2O_5[OH]_4$)	0



Due to inherent limitations in X-ray diffraction quantification, results must be considered semi-quantitative.

X-RAY DIFFRACTION ANALYSIS


<0236961B.RD> 0236961b
<0334301B.RD> 0334301b
Intensity(Counts)
2000
1500
1000
500
0
Standard
Sample
Holder
<39-1383> Clinoptilolite - KNa2Ca2(Si29Al7)O72I24H2O <Majo
<33-1161> Quartz, syn - SiO2 <Majo
<05-0586> Calcite, syn - CaCO3 <Majo
<36-0426> Dolomite - CaMg(CO3)2 <Majo
<19-1184> Albite, ordered - NaAlSi3O8 <Majo
10
20
30
40
50
2-Theta

Section 2

ICP Analysis of KMI zeolite

KMI Total Heavy Metal Analysis

Procedure: ICP by Bodycote, Essais de Matériaux Canada Inc. (an accredited laboratory).

Element	KMI zeolite mg/kg					
Sample	**1 January****	**2 February 13th**	**3**	**4 June 17th***	**5 July 23rd***	**Average**
As	11	29	36	33	-	27 (11)
Al					39 000	39 000
Cd	1.0	1.0	1.0	<1.0	<1.0	1.0
Cu	45	110	24	4	5	46 (46)
Co	<17	6	4	4.1	<1.0	4 (0.93)
Cr					<2.0	5.4 (0.68)
Fe					4 300	4 300
Mo	22	6	6	4.7	5	10 (8.2)
Ni	<45	2	2	<2	<2.0	2
Pb	<10	90	120	35	30	64 (50)
Se	1	<0.5	<0.5	1.5		1
St	240	170	110	120	-	160 (59)
Zn	<58	270	350	390	180	250 (134)

* random composite samples taken from zeolite used in feed experiment.

** report analysis not included since you already have this report in your files, as well as those of February 2003.

KMI

ZEOLITE, INC.

TECHNICAL DATA SHEET - 101

(Cream-Colored Zeolite)

TOTAL CATION EXCHANGE CAPACITY (CEC).....1.6 to 2.1 + meq/gm
(Total CEC determined for Ammonia. Values vary with Cation involved.)

Clinoptilolite Content (Based on XRD Results From The Mineral Lab, Inc.)....90% (+/- 5%)

Chemical Analysis (%) - The Mineral Lab, Inc.

	(XRF Results)	*(Normalized to 100%)*
SiO_2	66.5	76.7
Al_2O_3	10.6	12.2
CaO	1.16	1.34
MgO	0.53	0.61
Na_2O	3.12	3.60
K_2O	3.86	4.45
Fe_2O_3	0.92	1.06
MnO	0.04	0.05

Major Exchangeable Cations (USBM)
(In order of selectivity for exchange)
$Cs^+ > Rb^+ > K^+ > Ca^{+2} > NH_4^+ > Sr^+ > Na^+ > Li^+$

$NH_4^+ >> Na^+$

Heavy Metal Cations (USBM)
(In order of selectivity for exchange)
$Pb^{+2} > K^+ > Ca^{+2} > NH_4^+ > Cd^{+2} > Cu^{+2} > Co^{+2} > Zn^{+2} > Ni^{+2} > Hg^{+2}$

Primary Adsorbed Gases
NH_3 CO CO_2 SO_4 H_2S H_2O N_2
Freon, Formaldehyde, Mercaptans, Benzene, Methanol

Anaconda Research Lab And The Mineral Lab, Inc. Test Result Summary

Silica/Alumina Ratio	5.8 - 6.4
Silicon/Aluminum Ratio	5.1 - 5.6
Bulk Density (dry solid)	87 lbs/ft^3 (1394 kg/m^3)
Bulk Density (dry, loose mat'l)	51 - 60 lbs/ft^3 (817 - 961 kg/m^3)
Mohs Hardness	5.1
Pore Size	4 angstroms
Pore Volume	15%
Specific Surface Area	1357 yd^2/oz (40 m^2/gm)
Alkali Stability (pH)	7 - 13
Acid Stability (pH)	1 - 7
Thermal Stability	1292° F (700° C)
Crushing Strength	2500 lbs/in^2 (176 kgs/cm^2)
Wet Attrition (Avg.)	6 - 7%

McGill

University Animal Care Committee

McGill University
James Administration Building
845 Sherbrooke Street West
Room 429
Montreal, Quebec, Canada
H3A 2T5

Comité universitaire de protection des animaux

Université McGill
Pavillon James de l'administration
845, rue Sherbrooke Ouest
Bureau 429
Montréal, Québec, Canada
H3A 2T5

Tel.: (514) 398-2837
Fax: (514) 398-4853
www.mcgill.ca/rgo/animal

January 14, 2004

The McGill University Animal Care Committee certifies that

Yonghong Wan has successfully completed the

Advanced Level
of the
Theory Training Course on Animal Use for Research and Teaching
on
January 14, 2004.

The training includes the following topics:

- **Basic Level:** Regulations & Procedures, Ethics, Basic Animal Care, Occupational Health & Safety
- **Advanced Level:** Anesthesia, Analgesia, Euthanasia, Categories, Influencing Factors and Environmental Enrichment

Please note that this certificate does NOT include practical training, which is obtained by successfully completing an Animal Methodology Workshop where another certificate is issued.

Certification is valid for 5 years, starting on the date indicated above.

Deanna Collin
Animal Care Training Coordinator, animalcare@mcgill.ca

(Confirmation of training can be obtained by request to the above email address)

Note: Trainee must keep this certificate as other institutions may request it as evidence of training

This certificate is awarded to

Yonghong Wan

In recognition of attendance, participation and having passed an examination in the following course at

Macdonald Campus Farm of McGill University, Ste Anne de Bellevue.

Livestock Behavior and Handling

Dairy Swine Poultry

Session Date: March 20th, 2004

certified by:

Philippe Lavoie BSc
Farm Manager

May 20th 2004
Date

LITHO. IN U.S.A.