



Exposure assessment to airborne contaminants in the indoor environment of Swine Farms

February 2007

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Acknowledgements

The authors would like to acknowledge financial support of the National University of Ireland Galway, Millennium Fund and the Health and Safety Authority who funded this project.

The authors would like to thank Teagasc for their help with coordinating the field sampling and all the farmers who participated in this study, for their time, understanding and relentless cooperation.

Special thanks to the NUI Galway students, Michelle Feeney, Thomas Murphy, Elaine Browne and James McLynn for help with sample collection.

Executive Summary

Introduction:

The agriculture industry in Ireland is of major importance to both our economy and our way of life. Despite notable advances, an occupation in the farming sector inadvertently results in multiple exposures to a variety of hazards, including respiratory hazards. As a result, farmers tend to have higher rates of asthma and respiratory symptoms than other occupational groups. Data from the Teagasc National Farm Survey has shown that 9.9% of Irish farmers have reported work related illnesses (HSA, 2003). One third of the illnesses reported were respiratory in nature. However, there is currently no data on the extent to which Irish agricultural workers are exposed to various respiratory hazards in their working environments.

Aims:

The primary objective of this research project was to evaluate Irish swine farmers' occupational exposure to certain respiratory hazards, namely: carbon dioxide, ammonia, swine confinement dust, and bacterial endotoxin. Worker exposure levels were compared to the recommended health limits developed by Donham (2000) for the prevention of acute respiratory symptoms in swine workers

Method:

Five intensive pig farms (approximate size 500 - 2200 sows) at various locations throughout Ireland participated in the study. Similar animal house ventilation and manure collection systems were used on all farms. Workers participating in the study were classified into similar exposure groups (SEG's), based on the farm units in which they were working i.e. the farrowing unit, the dry sow unit, the weaner unit, the finishing unit and the farmer who worked throughout all units. Personal occupational exposure monitoring was carried out, involving obtaining samples of the air breathed in by the swine confinement

workers, in order to determine their exposure to the above respiratory hazards. Statistical analysis of exposure data allowed comparisons to be made within the SEG's and with recommended health limits for the prevention of acute respiratory symptoms.

Results:

Results from this research project show that swine confinement workers are potentially exposed to concentrations of workplace contaminants at levels above recommended health limits. For example, swine confinement dust exposure concentrations of up to three times in excess of recommended health limits were measured. Throughout the study a lack of both awareness and use of respiratory protection equipment amongst farm workers was noted. In addition none of the farm workers monitored in this study participated in an occupational health surveillance program.

Conclusions:

Exposure data collected in this research project indicate that swine confinement workers may be at an increased risk of developing respiratory disease from exposure to workplace hazards. There is a need for increased training and education to promote awareness of occupational health issues and the importance of implementing workplace exposure controls in the sector. A literature review completed as part of this study showed that there are available proven exposure reduction practices developed by international researchers which could be implemented in Ireland without much cost to the farmer. Exposure monitoring and health surveillance programs are recommended across the swine industry, particularly for vulnerable groups such as young people, pregnant workers, or those with existing respiratory diseases.

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Abbreviations

ACGIH = American Conference of Governmental Industrial Hygienists

ARDS = Acute Respiratory Distress Syndrome

CEN = Comité Européen de Normalisation

CFU = Colony forming units

CH₄ = Methane

CO = Carbon monoxide

CO₂ = Carbon dioxide

db = decibel

db(A) = A-weighted decibels

DNA = Deoxyribonucleic Acid

EN = European Norm

EU = Endotoxin Unit

FEF75 = Forced Expiratory Flow at 75% Vital Capacity

FEV₁ = Forced Expiratory Volume-In-One-Second

H₂S = Hydrogen sulphide

HSA = Health and Safety Authority

IgE = Immunoglobulin E

INAB = Irish National Accreditation Board

IOM = Institute of Occupational Medicine

IrDA = Infrared Data Association

ISO = International Standards Organisation

LAF = Laminar Airflow cabinet

LAL = *Limulus Amebocyte* Lysate

LPS = Lipopolysaccharide

MMI = Mucus Membrane Irritation

MSDS = Material Safety Data Sheet

NH₃ = Ammonia

NIHL = Noise Induced Hearing Loss

ODTS = Organic Dust Toxic Syndrome

OELV = Occupational Exposure Limit Values

p_{peak} = Peak Sound Pressure

PCR = Polymerase Chain Reaction

PPE = Personal Protection Equipment

ppm = Parts Per Million

rpm = Rotations Per Minute

SEG = Similar Exposure Group

SPSS = Statistical Package for Social Science

STEL = Short Term Exposure Limit

TWA = Time Weighted Average

USB = Universal Serial Bus

VRBA = Violet Red Bile Agar

1.0 Overview of Chapter

This research presents work aimed at determining the occupational exposure of workers in the swine industry to respiratory hazards. In this chapter an overview of the agriculture industry in Ireland is discussed, while focusing on the swine industry for the remainder of the research. The principal occupational hazards these workers are exposed to are identified, with particular emphasis on respiratory hazards, namely gases, swine confinement dust and endotoxin. The background to the study is also outlined and the overall objectives of the study are highlighted.

1.1 Agriculture Industry in Ireland

The agriculture industry in Ireland is of major importance to both our economy and the Irish way of life. Traditionally, most country families practiced some form of farming and currently farmers represent seven per cent of the Irish workforce (Teagasc, 2006). The relatively recent move towards intensive livestock production and larger confinement buildings has brought with it not only large increases in productivity per farm worker, but also has resulted in an increased potential exposure to physical, chemical and biological health hazards. Although these modern swine confinement buildings may appear 'cleaner', the air quality inside these units has become an issue for both workers and for the environment. Thus, despite notable advances in an occupation in the farming industry inadvertently results in multiple exposures to a variety of dusts, toxic gases and bioaerosols - many of which may contribute to respiratory symptoms and disease. Agricultural workers have higher rates of long-term sick leave associated with respiratory disease than any other workers (Hoppin *et al.*, 2002). Accordingly, second to chronic back pain (forty-nine per cent), respiratory problems account for thirty-five per cent of illnesses reported by Irish farmers (Health and Safety Authority, 2003).

1.2 Swine Production

Upon joining the European Union, farmers in Ireland began to specialise in different activities, resulting in a drastic trend from almost every farm keeping pigs outdoors to currently less than 600 commercial pig farms. Donham and co-workers (1977) were the first to observe the harmful effects of working with swine. Since then much research has been directed to the hazards encountered by the workers in these swine confinement buildings. It has been acknowledged that the increased frequency of symptoms of respiratory disease is related to the number of years and percentage of the day spent working with swine (Donham *et al.*, 1989). Hoppin and co-workers (2003), in a study aimed at investigating the role of animal exposures and wheeze, found that among European farmers, swine farmers had more work related symptoms and were fifty per-cent more likely to wheeze than cattle farmers.

1.3 Respiratory Hazards in the Swine Industry

The air of swine confinement buildings is very complex and contains many contaminants that are hazardous to human health. The respiratory hazards to which swine workers are exposed include gases, swine confinement dusts and microorganisms or their components that can become airborne and be inhaled. Gases are predominantly produced in swine production facilities either directly by animals and excreta or microbial degradation of manure (Lemay, 2002). Gases typically produced include carbon dioxide, carbon monoxide, ammonia, methane and hydrogen sulphide. Inhaled gases can act by being irritant, toxic or asphyxiating. Dust is an aerosol containing solid particles made airborne by mechanical disintegration of solid particles, ranging in size from less than 1 μm to greater than 100 μm . The dust generated within indoor swine buildings may contain many types of particles including: Animal dander; faecal material and urine of both pigs and rodents; feed components; bedding materials; absorbed gases and chemicals. Importantly, this dust also contains microorganisms such as viruses, bacteria, yeasts, moulds and their by-products (Kirychuk, 2002). Such dust is more appropriately referred to as 'bioaerosols', as it is primarily made up of particles of organic origin. Somewhere in the region of seventy to ninety per cent of swine confinement dust is thought to be biologically active in

its effects (Borg, 1999). Therefore this dust can serve as either an irritant or occasionally an allergen. Furthermore, it is relevant to note that in addition to adversely affecting human health, excessive dust affects the health of the swine, increases labour requirements for building and equipment maintenance, and interferes with the performance of ventilation systems.

Endotoxin is an additional respiratory hazard of concern regarding the health of workers in swine confinement buildings. They are a group of lipopolysaccharide (LPS) molecules making up the outer membrane of gram-negative bacteria, and dose-response relationships have been found between endotoxin in organic dusts and respiratory symptoms (Rylander, 2002; Beijer and Rylander, 2005). Endotoxins are ubiquitous in nature but livestock confinement units present one of the highest concentrations to be found anywhere (Thorne, 2004). Interestingly it has been suggested that endotoxins may be a more significant contributor than dust is to swine workers' problems with chronic cough and bronchitis (Hoppin, 2003) .

1.4 Background to Project

While the levels of the above hazards have been investigated in Europe (Simpson *et al.*, 1999), Asia (Chang *et al.*, 2001) and America (Cormior *et al.*, 1990), there is no published data on the extent to which they contaminate the air of Irish agricultural buildings. This lack of data is an important point as it not known to what extent variables such as the temperature and relative humidity of the various climates would affect the levels of the particular contaminants. The focus of the objectives of the Health and Safety Authority's (HSA) National Strategy for Workplace Well-being is on the health of employees in the workplace and on how this can be improved through well-defined and practical programs based on quality information . This initiative is in line with the relatively recent recognition of the importance of the health of workers, be it in the primary, secondary or tertiary sectors. As regards agriculture, we know that a significant target of the HSA Farm Safety Plan 2003 -2007 is to improve the health and safety of farm workers through engineering/workplace design, enforcement and education/training. This project aims to address this important data gap that exists in the Irish agricultural sector, specifically the actual exposure levels of workers in the swine industry to respiratory hazards.

1.5 Research Objectives

- Thus, the primary objective of this project is to evaluate Irish swine farmers' occupational exposure to respiratory hazards, namely gases, swine confinement dusts and endotoxins. The farms employed will take account of variables such as size, facilities, age and productivity. The workers will be identified using Similar Exposure Groups (SEGS), depending on the unit in which they work, either: Weaner unit, farrowing unit, dry sow unit, fattening unit and the general farmer who works in all units.
- The results are to be compared to both the Occupational Exposure Limit Values (OELV's) and the recommended health limits for the prevention of acute respiratory symptoms in the swine workers.
- In addition, while there have been notable advances in engineering controls, it is necessary to consider the various occupational exposure control measures and best - practices that exist within the swine industry and their possible implications on the health of swine workers.

2.0 Overview of Chapter

In this chapter an overview of the relevant research and literature is discussed. The nature of swine production in Ireland is outlined. A summary of the occupational hazards encountered in swine confinement buildings is given, namely gases, swine confinement dust, microorganisms, endotoxin, confined spaces, chemical hazards and noise exposures. In addition, occupational diseases such as acute bronchitis, sinusitis, organic dust toxic syndrome, occupational asthma and zoonotic infections experienced by swine confinement workers are identified. Furthermore, control measures and best practices that exist within the industry are documented in this chapter and their possible implications on the health of the workers are discussed.

2.1 Swine Production in Ireland

Traditionally every farm in Ireland had at least one pig kept outdoors; however, the current trend is towards more intensive swine confinement buildings. There are an estimated 1.7 million pigs in Ireland, with more than half of all pigs being found in just four counties, namely Cork, Cavan, Tipperary and Waterford (Teagasc, 2006). The average breeding herd has 355 sows with the pigs housed indoors, in specialised confinement buildings. The current swine density in Ireland is 40 pigs/km², which is similar to countries like the United Kingdom, Spain and Germany (Anderson, 2001). Most pig production units in Ireland are integrated units, where the entire production cycle takes place in one location. The designs of these swine confinement buildings are aimed at protecting animal welfare and simplifying management, while allowing one person to care for approximately 150 sows and their offspring through to slaughter weight (Teagasc, 2006). The life cycle of such pigs consists of: Dry sows, farrowers, piglets (birth to 6.5 kg), weaners (1st stage 6.5 - 15kg; and 2nd stage 15 - 35 kg), and fatteners (35 - 93 kg). The feed used for swine in Ireland is mainly meal and water. This can be delivered to the pigs separately, in a dry feeding system, or pre-mixed in a wet feeding system. Most farms employ automatic systems for the distribution of the feed. As with other industries, the viability of the swine industry is influenced by variables such as the environment, consumer demands/trends competitive ability and the economy

2.2 Occupational Hazards found in Swine Confinement Buildings

2.2.1 Gases

Although approximately 160 different gases have been identified in the ambient air of swine confinement buildings, many of these gases are present only in trace amounts and are not linked to occupational respiratory illnesses. However, many of these gases produced in small quantities, such as volatile acids, amines and mercaptans contribute to the characteristic odours in swine facilities (von Essen, 2001). Gases of concern typically produced in swine confinement buildings include: Carbon dioxide (CO₂), carbon monoxide (CO), ammonia (NH₃), methane (CH₄) and hydrogen sulphide (H₂S).

OELV exist to protect workers from excessive exposures to toxic chemicals in the workplace. In addition to this 8-hour reference period, the following terms, which are used to quantify the environment of the worker, shall be referred to:

- The Time Weighted Average (TWA) is the employee's average airborne exposure in any 8-hour work shift of a 40 hour working week. Concentrations are set at levels to which nearly all workers may be repeatedly exposed without adverse effects.
- The Short Term Exposure Limit (STEL) is the employee's 15-minute time weighted average exposure that shall not be exceeded at any time during a working day. These are set at concentrations to which workers can be exposed continuously for a short period of time without suffering from adverse effects.

2.2.1.1 Carbon dioxide (CO₂)

Carbon dioxide occurs primarily as a normal by-product of pig respiration. Exposures to high levels of carbon dioxide (20,000 ppm) can result in deep rapid breathing (Doss *et al.*, 2002). If there is a ventilation failure (or lack of) in a fully occupied, completely enclosed fattening unit the carbon dioxide level can rise rapidly, and in addition to depletion of oxygen, create an asphyxiate atmosphere in as little as 6 hours (Donham, 2000). Carbon dioxide levels in swine confinement buildings are used as a measure of the units' air quality, and the adequacy of its ventilation system. The reason for this is that the rate of carbon dioxide production per animal is known, and if concentrations are kept below 0.5 per cent then other gases do not usually cause problems (Pearson, 1988). It is important to note that there is a seasonal variation in the levels of this gas; while it may be over 4000 ppm in winter it is often under 1000 ppm in summer (Lemay, 2002). This can be explained by the increased ventilation rates during the summer. Chang and co-workers (2001) reported mean concentrations between 600 and 895 ppm carbon dioxide. As per the 2002 Code of Practice to the Safety, Health & Welfare (Chemical Agents) Regulations, 2001:

- 8 hour OELV - 5000 ppm
- 15 min STEL - 15,000 ppm

Donham's (2000) recommended health limit for swine confinement workers' exposure to carbon dioxide is 1,540 ppm, as it was found that concentrations in excess of this were associated with a higher proportion of ill health in workers.

2.2.1.2 Carbon monoxide (CO)

Carbon monoxide is produced from incomplete combustion of organic matter. It occurs in exhaust fumes from improperly maintained or malfunctioning engines and direct burning heaters where there is inadequate ventilation. Acute exposure to carbon monoxide has an insidious onset with giddiness, headache, chest tightness and nausea; unconsciousness rapidly supervenes at concentrations in excess of 3500 ppm (Harrington *et al.*, 1998). As per the 2002 Code of Practice to the Safety, Health & Welfare (Chemical Agents) Regulations, 2001:

- 8 hour OELV - 20 ppm
- 15 min STEL - 100 ppm

2.2.1.3 Ammonia (NH₃)

Ammonia is released into the air from the breakdown of urea in the urine of the animals. In addition to the storage of liquid manure, ammonia gas is produced from the drying of manure and urine on the solid floor surfaces of the pig houses. As it is water-soluble, ammonia is rapidly absorbed in the upper airways, with the result of damaging the upper airway epithelia and impeding lung cilia from clearing dust particles (Merchant, 2002). Ammonia has a low odour threshold of less than 5 ppm, meaning that its presence is readily detectable above this concentration. Eye irritation and respiratory problems occur around 6 to 20 ppm and above; while at 40 to 200 ppm headaches, nausea, reduced appetite, irritation to airways, nose and throat occurs (Doss *et al.*, 2002). There is much speculation in the literature as to the possibility of ammonia gas particles adhering to respirable dust particles, and consequently being carried deep into the lungs of exposed individuals, thus adding to their potential toxicological effects.

The release of ammonia from urea is a slow process governed by factors such as ammonia concentration, pH, temperature, air velocity and emitting surface area. Accordingly, as with carbon dioxide levels, ammonia levels can vary with the time of year in question; ammonia concentrations can be between 20 and 30 ppm under winter conditions but are often much lower in the summer, often due to increased ventilation rates (Lemay, 2002). In a study aimed at determining the temporal variation of indoor air quality in enclosed swine confinement buildings ammonia levels, O'Shaughnessy and co-workers (2002) reported ammonia to average at only 3.6 ppm in their well-maintained study site. Chang and co-workers (2001) reported mean concentrations of less than 5 ppm ammonia, while Wathes and co-workers (1998) reported peaks of 18 ppm. As per the 2002 Code of Practice to the Safety, Health & Welfare (Chemical Agents) Regulations, 2001:

- 8-hour OELV - 20 ppm
- 15 min STEL - 35 ppm

Donham's (2000) recommended health limit for swine confinement workers' exposure to ammonia is 7 ppm.

2.2.1.4 Methane (CH₄)

Methane is the colourless, odourless, flammable gas present in 'natural gas', which is continuously produced by anaerobic decomposition of slurry. Methane is rarely a problem in swine buildings, however at high concentrations (500,000 ppm) can cause headaches and even asphyxiation (Doss *et al.*, 2002). If it accumulates in slurry stores and is present at the correct concentration methane poses as an explosion hazard. Deep pit units are more likely to promote methane accumulation and it is imperative that they are properly ventilated.

2.2.1.5 Hydrogen sulphide (H₂S)

Hydrogen sulphide is a colourless gas that smells like ‘rotten eggs’ and it is both an irritant and an asphyxiant. It is produced during the decomposition of manure -slurry or sewage and remains there until movement causes its release. This gas is of serious concern in both the cattle and swine industries. Chronic low-level exposure to hydrogen sulphide is associated with anosmia, the loss of ability to detect odors. This lack of ability to smell hydrogen sulphide results in no warning of concentrations that can result in loss of consciousness. At higher levels, hydrogen sulphide exposure causes hydrogen sulphide poisoning, pulmonary edema, Acute Respiratory Distress Syndrome (ARDS), coma and death. The ambient level of hydrogen sulphide in a well-ventilated swine unit will be less than 3 ppm. However, when manure is being agitated hydrogen sulphide can rapidly reach levels that can cause immediate unconsciousness and death in just a few seconds; with hydrogen sulphide levels above 500 ppm, unconsciousness may result in just a few breaths. This gas is the principle hazard in confined swine areas. Reported mean concentrations in the literature range from 0.2 to 10 ppm hydrogen sulphide (Chang *et al.*, 2001; Donham *et al.*, 1977). As per the 2002 Code of Practice to the Safety, Health & Welfare (Chemical Agents) Regulations, 2001:

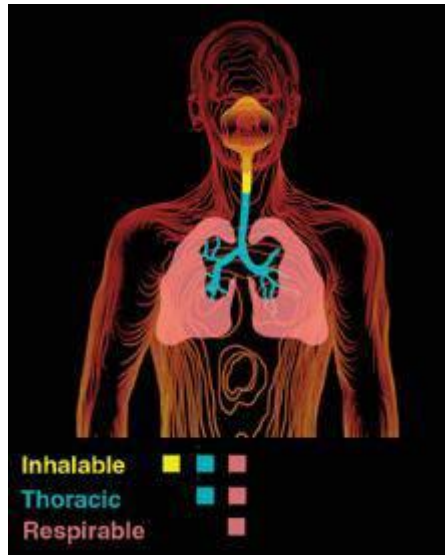
- 8-hour OELV - 10 ppm
- 15 min STEL - 15 ppm

2.2.2 Swine Confinement Dust

When dust measurements are being taken in relation to health effects, the sampling convention relates to the penetration of the aerosol to regions of the respiratory tract and its aerodynamic diameter. The definition of these conventions has been agreed between the Comité Européen de Normalisation (CEN), the International Standards Organisation (ISO) and the American Conference of Governmental Industrial Hygienists (ACGIH).

- The **inhalable fraction**, which includes the thoracic and respirable fractions, is defined as the mass fraction of the total airborne particles that are inhaled through the nose and/or mouth. Inhalable fractions have no median aerodynamic diameter but are generally less than 100 μm . Some of these airborne particulates are trapped in the mucous of the nose and pharynx and are prevented from travelling deeper into the lungs.
- The **thoracic fraction**, which includes the respirable fraction, is defined as the mass fraction that penetrates the respiratory system beyond the larynx. These particles have a mean aerodynamic diameter of 11.64 μm .
- The **respirable fraction** is defined as the mass fraction that penetrates to the unciliated airways of the lung, known as the alveolar region, where gaseous exchange takes place. The respirable fraction has a mean aerodynamic diameter of 4.25 μm (Ashton and Gill, 2000).

Plate 1: The Inhalable, Thoracic and Respirable Dust Fractions



[SKC website] <http://www.skinc.com> [Accessed 19th July 2006]

The amount of dust in the air of livestock buildings is correlated to environmental factors such as ventilation, feeding practices, bedding materials, dung and slurry handling, and animal activity (Takai and Pedersen, 2000). Wathes and co-workers (1998) found that the inhalable dust emissions from pig buildings were forty per cent higher in summer than winter, while respirable dust emissions were not affected greatly by the season. In addition, the risk of health effects from swine confinement dust depends on a combination of not only the size and shape of the dust particles, but also the duration the worker spends in the dusty area, what is contained in the dust and if a respirator is worn or if engineering controls are implemented during working activities (Kirychuk, 2002).

Typical standards for inhalable and respirable dust do not take into consideration the biologically active nature of the dust found in swine confinement buildings, hence reduced exposure levels have been recommended. Donham (1995) suggests the following threshold values for swine workers exposure to swine confinement dust:

- Total dust - 2.4 mg/m^3
- Respirable dust - 0.23 mg/m^3

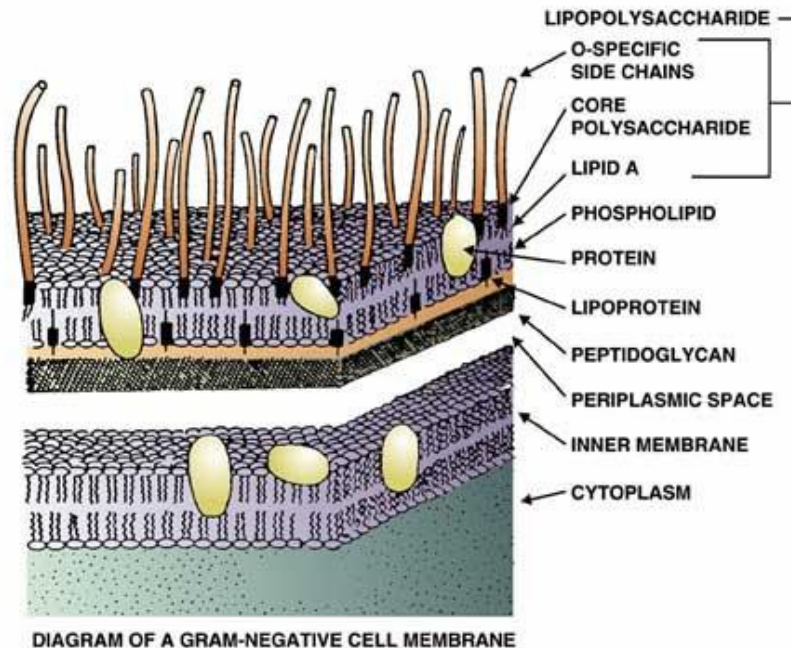
As with carbon dioxide and ammonia, exposures to concentrations in excess of the above values were found to be associated with higher levels of ill health in workers. In addition Donham and co-workers (2002) highlighted that the resultant adverse health effects of ammonia and particulates in combination was greater than the additive effect of ammonia and particulates by a factor of fifty three to one hundred and fifty-six per cent. These authors concluded that maximum exposure concentrations must be less than the individual exposure limits as workers are exposed to both substances simultaneously.

2.2.3 Microorganisms

One must acknowledge the vastness of microbial diversity and the fact that it is a result of successful evolutionary events that have conferred survival value on the microorganisms in existence today (Madigan *et al.*, 2000). A brief understanding of what constitutes this microbial diversity is desirable. The major groups of living organisms are Bacteria, Archae and Eukarya. Several evolutionary branches occur within the Bacteria, which include all known pathogenic prokaryotes and most of the bacteria found in the soil, water, animal digestive tracts, and many other environments. Conversely, most Archae are anaerobes; cells incapable of living in air and inhabiting what humans consider as being extreme environments. The microbial Eukarya include the algae, fungi and protozoa. Three major groups of fungi are recognised: moulds, yeasts and mushrooms.

These microorganisms and their products are easily accumulated and aerosolised in the densely populated and enclosed areas of swine confinement buildings. It has been shown that swine workers are highly exposed to microbes present in dust at their workplaces; Cormior and co-workers (1990) found that the air of swine confinement buildings is highly contaminated with bacteria, yeast and moulds at a level up to 1200 – fold greater than so called “normal air”. The bacteria are gram-positive and gram-negative and largely of fecal origin (Donham, 2000). Importantly dust in swine buildings contains far more than purely viable organisms; microbial products with health implications include antigens, glucans, and endotoxins

Plate 2: Structure and Components of Gram -negative Bacteria Cell Membrane



[The Horseshoe Crab website] <http://www.horseshoecrab.org> [Accessed 2nd August 2006]

As endotoxins are a major cell wall component of gram -negative bacteria, these gram -negative species are of particular interest. Examples of common gram -negative bacteria are the species *Aeromonas*, *Citrobacter*, *Enterobacter*, *Escherichia* and *Pseudomonas*. Gram -positive organisms found in swine buildings include *Enterococcus*, *Streptococcus*, *Bacillus*, *Aerococcus*, and *Micrococcus*. Gram -positive microorganisms represent the majority of bacteria while gram -negative organisms are generally less than twenty -five per cent of the viable bacteria (Thorne, 2002). Exposure assessments have shown that airborne bacteria in swine confinement buildings reach levels of up to 10^7 colony forming units/ m^3 (CFU/ m^3), with *Micrococcus* and *Staphylococcus* being predominant (Chang *et al.*, 2001). Several studies have found that fattening units contain the highest airborne levels of culturable bacteria and gram -negative bacteria, probably due to the fact that they are more densely stocked and the larger body size of the growing pigs (Cormior *et al.*, 1990; Chang *et al.*, 2001).

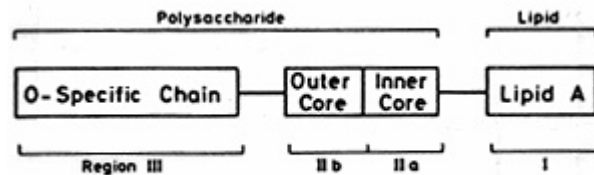
Yeasts and moulds are also very concentrated in the air of swine confinement buildings. As with bacteria, they appear to be present at similar portions to what might be found outdoors, but are concentrated by a factor of 100 to 10,000 (Donham, 2000). Although many of these fungi are parasitic on animals, including humans, they are generally less significant pathogens than are bacteria and viruses. Nevertheless, the role of more than eighty airborne fungi in the development of respiratory allergy and asthma has been established (Adhikari *et al.*, 2004). Prevalent airborne fungi associated with swine buildings include *Cladosporium*, Ascospores, smut spores, and Basidiospores. Fungal products or components of note include conidia and microconidia, hyphal fragments, mycotoxins and glucans (Thorne, 2002). Some fungi such as *Aspergillus* and *Penicillium* species are known to produce extrinsic allergic alveolitis, a clinical entity found in swine workers (Cormior *et al.*, 1990).

It is necessary to acknowledge the restrictions of the pure culture approach for the culturing and enumeration of microorganisms. The term "the great plate count anomaly" was coined by Staley and Konopka in 1985 to describe the difference in orders of magnitude between the numbers of cells from natural environments that form colonies on agar media and the numbers countable by microscopic examination using methods such as DNA staining (Connon and Giovannoni, 2002). The concentrations of non-culturable aerobic and anaerobic organisms in the particulate matter in swine confinement buildings is known to be 10 to 100 -fold higher than the culturable organisms. Nonetheless culture is still one of the most popular methods of bioaerosol sampling as it allows determinations of microbial composition and concentration simultaneously (Chang *et al.*, 2001). However, since many of the airborne organisms are not culturable, it is often necessary to employ non-culture based methods for a more precise identification. These molecular biological techniques include use of direct count methods with deoxyribonucleic acid (DNA) staining and epifluorescence microscopy, fluorescent in situ hybridization, and Polymerase Chain Reaction (PCR) techniques (Thorne, 1992). For the purpose of determining exposure in the current study, the endotoxin content of the air may be a more reliable measure of biological exposure than bioaerosols, as it is independent of the ability to culture the microorganism.

2.2.3.1 Endotoxin

Endotoxins are a major cell wall component of gram-negative bacteria. They are highly inflammatory substances and are believed to be a major agent in initiating respiratory disease in swine confinement workers and animals. Endotoxins are lipopolysaccharide (LPS) molecules that consist of three distinct regions: O-specific polysaccharide, core polysaccharide (outer and inner cores), and lipid A.

Plate 3: General Architecture of Lipopolysaccharide



[Todar's Online Textbook of Bacteriology website] www.textbookofbacteriology.net

[Accessed 2nd August 2006]

The lipid A region of LPS exhibits little variation across genera and imparts the toxicity to endotoxin, while immunogenicity is associated with the polysaccharide components. Subsequent to LPS biosynthesis in the cytoplasmic membrane, LPS molecules are transported to the surface and function as the principle surface antigens of gram-negative bacteria. It is the lipid A region that causes harmful health effects in humans after exposure to endotoxin. Humans are able to produce antibodies to endotoxins after exposure, but these are generally directed at the polysaccharide chain, and do not protect against a wide variety of endotoxins. The term LPS, although not totally accurate, is often used interchangeably with endotoxin; this term was adopted before the discovery of the toxic lipid A moiety.

It has been shown that multiple exposures to endotoxin-containing swine confinement building air induces airway hyper-responsiveness, increases mucus-containing airway epithelial cells, lung inflammation, increased asthma severity, mucous membrane irritation (MMI), chronic bronchitis, byssinosis, toxic pneumonitis, and hypersensitivity pneumonitis (Charavaryamath *et*

al., 2005). The alterations in pulmonary function are characterised most typically by a decline in forced expiratory volume-in-one-second (FEV₁); and this was found to be in a dose dependent way (Donham *et al.*, 1989). Conversely, it has been reported that low-level endotoxin exposure induces tolerance to subsequent endotoxin challenges by challenging the immune system (von Essen, 2001).

Similarly conflicting evidence exists in relation to endotoxin exposure and asthma. Some studies indicate an increased risk of asthma after endotoxin exposure, while others indicate that endotoxin exposure protects from asthma. Radon, (2006) postulates that these differences can be explained when different asthma phenotypes are considered and the possibility that not all asthma is associated with allergic sensitisation. Atopic asthma is caused, in susceptible individuals, by sensitisation to and subsequent inhalation of allergens (Merchant, 2002). The risk of atopic asthma, mainly dominated by eosinophilic response, is decreased in those exposed to endotoxins. In contrast, the risk of nonatopic asthma, often referred to as asthma - like syndrome and characterised by neutrophilic response, is enhanced in subjects with higher endotoxin exposure.

While numerous studies have been undertaken in the area of endotoxin and its health effects, the exact connection between endotoxin exposure and respiratory illness is still poorly understood (Portengen *et al.*, 2005). This problem is a result of many factors, including inconsistent associations between exposure and adverse health affects, underdeveloped sampling collection techniques, limitations on the accuracy of current endotoxin analysis techniques, and only very limited information on the relationship between concentrations of endotoxin in the air and settled dust (Pheatt, 2001). Furthermore, exposure to ammonia and endotoxin-rich dust has been shown to act synergistically to adversely affect respiratory health in both animals and humans (Sigurdarson *et al.*, 2004).

Despite the fact that there is a clear recognition of endotoxin being a respiratory hazard there are no occupational exposure limit values. Rylander (1985) calculated an endotoxin threshold of 33 ng/m³ based on spirometric data; Castellán (1987) recommended an endotoxin exposure

limit of 9 ng/m³ based on a cotton dust study; and Palchak (1988) suggested an endotoxin threshold of 30 ng/m³ in the pharmaceutical industry to trigger medical surveillance (White, 2002). Much of the endotoxin literature reports results in weight units (usually nanograms). However it is important to note the inaccuracy in comparing data from studies that report results in weight units of endotoxin, since different endotoxin preparations do not necessarily have equivalent potencies. To address this concern the Endotoxin Unit (EU) was implemented as a measure of activity or potency of endotoxin, as opposed to gravimetric methods (approximately 10 EU equals 1 ng i.e. 300 EU = 30 ng).

Recommended exposure limit thresholds for endotoxin have been suggested ranging from 50 to 2000 EU/m³ (Radon, 2002). There is a drastic range in the levels of endotoxin detected in swine confinement buildings. Chang and co-workers (2001) reported low levels of endotoxin exposure for swine workers. These authors reported average concentrations of airborne total endotoxin between 37 and 298 EU/m³, while respirable endotoxin was between 14 and 129 EU/m³. It is relevant to note however, that this particular exposure assessment was carried out in open style swine houses where there would invariably be more air circulation from the outdoors. Conversely, Simpson and co-workers (1999) reported high levels of endotoxin exposure for swine workers in enclosed confinement buildings in the region of 149,230 EU/m³.

2.2.3.2 Endotoxin Sampling, Extraction & Analysis

An area of considerable uncertainty is that of endotoxin sampling and extraction of samples in the environmental setting. Filter media and extraction solutions are potential sources of variation in the overall procedure. Commonly used filters include glass fibre, cellulose esters, polyvinyl chloride and polycarbonate membrane. Laitinen (1999) found that samples collected on glass fibre filters showed highest amounts of detectable endotoxin after collection. It was also determined that the best efficiency was attained by extraction with non-pyrogenic water within 8 hours after sampling, followed by storage of the extracts at 4°C until they were analysed.

The effect of preservation method on detectable concentrations of endotoxin is an issue of great importance. It has been recommended that the extracted samples should be stored in a refrigerator rather than in a deep-freezer because freezing and thawing decreases the concentration of detectable endotoxin in the LAL assay. A twenty-five per cent reduction in endotoxin activity per freeze-thaw cycle has been reported in many studies, while storage of samples for a period of less than 1 year at 7°C had no effect (Douwes *et al.*, 1995). According to Laitinen (1999) the air samples extracted on the day of collection showed larger amounts of endotoxin than those stored at 4°C without extraction. Furthermore, the difference was found to increase with prolonged preservation time.

Concentrations of endotoxin can be quantified by several modifications of the *Limulus Amebocyte* Lysate (LAL) assay. This is an *in vitro* test utilising lysate of blood cells of the horseshoe crab, *Limulus Polyphemus*, which enzymatically interacts with endotoxins (Ziljstra *et al.*, 1997). The LAL assay measures LPS potency, which is dependent on factors such as the fatty acid content of the Lipid A portion, the polysaccharide content and LPS aggregational properties (White, 2002). There are several variations of the LAL assay available for quantification of endotoxins from different environmental settings. The LAL endpoint assay, generates ranks or levels at which the endotoxin is present or not, while the kinetic LAL assay is capable of determining exact levels of endotoxin present in a sample.

2.2.4 Confined Spaces

The Safety, Health and Welfare at Work (Confined Space) Regulations 2001 are applicable to all workplaces involving work in confined spaces. Examples of confined spaces in swine confinement facilities include enclosed tanks, manure pits and grain bins. According to Kirychuk (2002) there are four main dangers in such confined spaces:

1. Oxygen deficiency and oxygen enrichment
2. Fire and/or explosion
3. Build up of harmful levels of gases, vapours or particles resulting in potential health hazards and immediately dangerous to life and health
4. Drowning in liquids and/or entrapment in free-flowing solids

2.2.5 Chemical Hazards in Swine Confinement Buildings

In addition to the previously discussed respiratory hazards, swine workers encounter many chemical hazards during their typical working day. These include solvents, veterinary drugs and cleaners. The material safety data sheet (MSDS) for every chemical indicates how the worker can be exposed to the chemicals by identifying the routes of entry. The risks of working with such chemicals must be assessed - identifying how often, for how long and under what conditions exposure occurs (Kirychuk, 2002).

2.2.6 Noise Exposure in Swine Confinement Buildings

Farmers are exposed to loud noises from both animals and equipment on the farm. Exposure to high levels of noise over an extended period, or intense noise for a short period can damage one's hearing. Permanent damage known as Noise Induced Hearing Loss (NIHL) cannot be repaired because of damage to the cilia in the cochlea of the ear, which sense and transmit sound messages to the brain. The noise levels within a swine confinement building will vary greatly throughout the day, with the levels increasing greatly during feeding, due to both the increase in animal activity and the dry feeding systems themselves. Automatic feeding systems allow the swine workers to avoid the areas during feeding. In addition, automatic feeding systems that feed all animals at the same time reduce noise from pigs waiting to be fed.

Engineering controls to reduce noise are difficult and often impractical in swine confinement buildings as the animals are the primary source of high noise levels. Adsorptive materials and baffles are not an option in these settings due to the requirements for stringent cleaning with high-pressure spraying equipment. Thus, personal protection equipment is a viable option in the swine industry for prevention of noise induced hearing loss, providing noise attenuation in excess of 30 decibel (db) at frequencies most common in swine confinement buildings (Donham, 2000).

The Safety, Health and Welfare at Work (Control of Noise at Work) Regulations 2006 contain strict noise exposure limits and responsibilities for employers. The Regulations contains noise 'Exposure Limit Values' and Exposure Action Levels', which are based on ambient noise levels and trigger different degrees of protective measures. In each case a daily or weekly average noise exposure value in A-weighted decibels (dB(A)) is accompanied by a peak sound pressure (p_{peak}) to take account of high instantaneous noise levels. As per the Regulations, the limits are as follows:

- Exposure Limit Value of 87 dB(A) and a p_{peak} of 200 Pa
- Upper Exposure Action Value of 85 dB(A) and a p_{peak} of 200 Pa
- Lower Exposure Action Value of 80 dB(A) and a p_{peak} of 112 Pa

Where noise exposure exceeds the Upper Exposure Action Value, workers are obliged to use the individual hearing protectors, which the employer must make available to them when the Lower Exposure Action Value is exceeded.

2.3 Workers in the Swine Industry

It has been well documented that workers in the swine industry often leave this environment because of respiratory symptoms within weeks or months of commencing employment. Accordingly, swine workers are considered a 'survivor population', comprising of those who can tolerate exposure to the various levels of hazards. However, there are several groups of workers that deserve mentioning. As with many other industries in Ireland, foreign workers constitute a noteworthy proportion of work force in the swine industry. This directly results in many social and language barriers, which have implications for their training, health and health care. Language barriers can impede following safety instructions on labels and training in proper work practices.

Also of interest is the fact that pregnant workers are more susceptible to carbon monoxide poisoning and hormonal drugs, such as prostaglandin and oxytocin, which are used in the swine industry. Carbon monoxide is occasionally present in levels of 50 to 150 ppm that could harm the human fetus but may not be acutely toxic to adults (Donham, 1995). It has been reported that the swine environments and cigarette smoke exposures are likely to be additive in terms of the risk of developing bronchitis (von Essen, 2001). Smokers have also been associated with a reduced baseline pulmonary functioning and a greater decline in pulmonary functioning during a work shift (Donham, 1995). All susceptible workers, including those predisposed to asthma, should be accommodated for, monitored accordingly and special attention given to their specific needs.

2.4 Occupational Diseases in Swine Workers

There are several illnesses habitually experienced by those working in the swine farming industry:

2.4.1 Acute bronchitis

Acute bronchitis is the most common complaint among swine workers, affecting as many as seventy per cent of swine workers. This is an irritant-induced inflammatory condition of the airways. The symptoms of bronchitis are cough and sputum production, which occur for usually less than a year and typically dissipate within a year with decreased exposure (Donham, 2000). However it may lead to chronic bronchitis, with cough and sputum production for two or more years being characteristic of this disorder (Merchant *et al.*, 2002).

2.4.2 Sinusitis

Sinusitis is often chronic among swine workers who may complain of a continual or frequent cold “they just cannot shake,” and symptoms such as a stuffy head, difficulty in breathing through the nose, headache, and/or “popping ears” (Merchant *et al.*, 2002). These symptoms are often accompanied by an irritant rhinitis and pharyngitis (Donham, 2000). Sinusitis and rhinitis have been collectively referred to as mucus membrane irritation (MMI), which has been attributed mainly to exposure to the combination of bioaerosol, endotoxin and ammonia.

2.4.3 Organic Dust Toxic Syndrome (ODTS)

Organic Dust Toxic Syndrome is a febrile illness characterised by a spectrum of symptoms including malaise, joint and muscle pain, chest tightness, headache and nausea after exposure to large amounts of organic dust contaminated with microorganisms (von Essen, 2001). Symptoms appear four to eight hours after exposure occurs and can last for several days. ODTS results from unspecific stimulation of the immune system by a high concentration of bioactive substances. These mostly consist of microorganisms and their products such as endotoxins, to which swine workers are constantly exposed. It is likely that endotoxin exposure is the cause of the signs and symptoms of ODTS because they can be reproduced by experimental exposure (Rylander *et al.*, 1989; von Essen, 2001). ODTS has been frequently mistaken for “farmers’ lung”, as they have the same acute symptoms; however this condition is seen mainly in dairy farmers, as opposed to swine farmers. In a European farmers’ study the lifetime prevalence of ODTS in pig farmers was found to be twenty-three per cent (Radon, 2006).

2.4.4 Occupational asthma

Occupational asthma includes periodic airway obstruction, chest tightness, wheezing, and dyspnea; this does not occur on first exposure but may develop after weeks to months. Swine workers with pre-existent asthma typically experience severe asthma upon first exposure to animal confinement facilities and select themselves out of these jobs. Hence, reference to the “survivor population”. Occupational asthma may result from repeated exposure to the work environment. It has two basic mechanisms: 1) immunologically mediated or allergic [Immunoglobulin E (IgE)], or 2) chronic irritation. Allergic occupational asthma is not a common cause of swine farmer illness, as these are the workers who commonly self-select themselves out of employment. Non-allergic occupational asthma or asthma-like syndrome, on the other-hand, has been found to affect up to twenty per cent of swine confinement workers (Merchant *et al.*, 2002)

2.4.5 Zoonotic infections

A zoonotic infection is one that can be transmitted between animals and humans. Several swine infections can be transmitted to humans, some with potentially serious outcomes. These include brucellosis, erysipeloid, streptococcus suis meningitis, asc ariasis, swine influenza, scabies, ringworm, leptospirosis, toxoplasmosis and salmonellosis. Toxoplasmosis is a risk for the fetus of pregnant workers, and streptococcus suis meningitis may be fatal and permanent hearing loss has been reported in survivors. A concern that has been raised in relation to microbes in the swine industry is antibiotic resistance. Swine are often fed low -levels of antibiotics for growth promotion or more frequently for the treatment of infectious diseases. Humans may acquire resistant zoonotic pathogens directly or may be infected with a nonpathogenic, resistant organism that may then transfer the resistant gene to a pathogen in the gut of the individual (Donham, 2000). However, many zoonotic infections can be prevented by control ling the diseases in the animal and through the following of good occupational hygiene principles. These occupational hygiene principles fall into three categories: Abiding by safe work practices; using good personal hygiene methods; and wearing personal p rotection equipment (Health Services Executive, 2005). Myers and co -workers (2006) conclude that swine workers should be included in pandemic surveillance, and in antiviral and immunisation strategies.

2.5 Occupational Exposure Control Measures in the Swine Industry

In relation to control measures it is imperative to refer to Schedule 3 of the Safety, Health and Welfare at Work Act, 2005, which contains the General Principles of Prevention. As with any kind of contaminated working environment, swine workers health and safety can be addressed using different approaches that can be grouped as: 1) engineering control; 2) administrative control; and 3) personal protection equipment (PPE). In the hierarchy of safety controls, engineering solutions to health and safety problems are to be given priority. Administrative controls such as limiting the amount of time that workers spend in the contaminated areas are not expected to be effective in the relatively uncontrolled livestock industry (Barber *et al.*, 1999). Personal protection equipment should be considered as an invaluable means for protecting ones health and for susceptible workers or tasks with longer than normal exposure.

It is important that these controls are implemented with worker health i n mind, and not just with pig production as the principal incentive. Upon consideration and implementation of the various controls, swine workers health can be protected through a comprehensive program of environmental monitoring and control, through the use of efficient management and work practices, education/awareness training and health surveillance. Little attention has been given to exposure monitoring and health assessments in the swine confinement industry in Ireland. Improved health surveillance with baseline spirometry and ongoing screening for respiratory disease is important and should be implemented for swine confinement workers.

It is imperative that while considering the following measures, to remember that no single technique will present a universal solution due to the complexity of the sources of contamination in swine confinement buildings. More appropriately a combination of engineering controls and good practices will be necessary to provide an integrated solution.

2.5.1 Dust Control

As dust particles can carry gases and odours, control of dust improves the farmers' exposure and helps significantly in odour reduction. Factors determining the amount of dust include cleanliness of the buildings, animal activity, temperature, relative humidity, ventilation rate, stocking density and feeding method. Strategies that can greatly reduce or control the amount of dust in swine buildings include the following measures:

2.5.1.1 Clean interior building surfaces

The intervals of cleaning the swine units vary significantly from one farm to the next. However, most farms seem to adhere to an "All in, All out" policy. This entails all of the animals of a particular age or reproductive stage being housed in the same room, and being moved to different facilities or marketed at the same time. Accordingly, the farrowing units would be cleaned every 4 weeks, as would the 1st and 2nd stage weaner units. Depending on the weight gain of the fatteners, approximately 1 kg per day, the fattening unit would be cleaned every 8-10 weeks. However the dry sow unit would not follow the same clear-cut cycle as not all sows farrow at the same time; hence this unit may be only partially cleaned from time to time. The time between animal groups is used to empty pits, pressure wash, and disinfect all of the interior surfaces. Strict adherence to this practice would invariably help to reduce dust levels .

2.5.1.2 Reduce dust from feed

Feed is one of the main sources of dust and has accordingly been targeted as one of the most common methods for dust reduction. Addition of oil to dry swine rations significantly reduces the amount of dust in a building. Gestation rations are often mixed with water, which also greatly reduces dust (Chastain, 2000). Addition of lignin to straw has been shown to aid in reducing the dust released from straw. One particular Danish study found that addition of a thirty-nine per cent solution of lignin to shredded straw provided a ninety per cent reduction in the tendency to release dust and a reduction in the endotoxin content and fungal spores released by seventy and eight per cent respectively (The National Committee for Pig Production, 1999).

Proper and timely maintenance of feeders, augers, and other feed handling equipment is required for proper dust control (Chastain, 2000). The covering of feeders in a weaner house and levels of dust in the air was examined in a Dutch trial. There was no reduction of dust and checking and cleaning of feeders was made more difficult for the workers; thus it was not recommended to cover the mouth of feeders for dry feed (Roelofs and Binnendijk, 2000). However, this suggestion has not been reflected on in other literature and the practice remains in many farms of covering the mouths of the feeders.

2.5.1.3 Spraying with vegetable oil

A rational way of reducing airborne dust concentrations is to make the dust particles more adhesive so that they are not dispersed by animal activity. Sprinkling vegetable oil, such as soybean or canola oil, in very small amounts inside swine buildings has been shown to control dust as well as odour and some gases (Zhang, 1996; Lormior *et al.*, 2002). This control method has received a lot of attention in recent years, and in combination with good management practices appears to be very effective. Soybean oil can be stored in a bulk storage tank either inside or outside the production building and transferred to a smaller reservoir inside the building as needed. This may involve manual application with a hand sprayer, or it can be

distributed automatically to minimise labour. The delivery system should be configured to allow spraying of oil to a pair of pens, primarily in the sleeping/laying areas of the pen and avoiding the walkways. This will adsorb dust particles in these areas and allow the pigs to distribute the oil throughout the rest of the pen with their activities (Schmidt and Heber, 2005).

Once a day sprinkling at 0.5 ml/ft^2 has been shown to reduce dust by forty to fifty per cent, odour up to sixty per cent and hydrogen sulphide up to sixty per cent (Jacobsen *et al.*, 1999; Lormior *et al.*, 2002). This technique also resulted in ammonia emission rates being reduced by nineteen per cent (Schmidt and Heber, 2005). Lemay and co-workers (2000) designed an oil sprinkling system using undiluted crude canola oil to control dust that achieved a reduction in the inhalable and respirable dust particle counts by ninety and eighty-six per cent respectively. Water or a surfactant or emulsifier can be added to the oil for better distribution and convenience of cleaning the units between groups of pigs, and for reducing the incidence of clogged nozzles. It must be noted that this practice requires additional labour and techniques for cleaning of the units. Nonetheless this technology has been credited for being cost-effective for swine producers and remains one of the most hopeful options for reducing dust levels in swine houses.

The pigs' level of activity is an important factor in determining the dust particle concentration in swine confinement buildings. In the course of half an hour the concentration of airborne dust in a house with ad libitum feeding can be drastically affected due to a change in the level of activity. Therefore, it is recommended that work in the houses should be done at a calm pace to avoid exciting the pigs. In a comprehensive comparative study of different dust control methods in swine buildings, Takai and Pedersen (2000) found that a combined method of spraying an oil-water mixture controlled by an animal activity sensor and animal fat to the dry feed reduced the airborne dust concentrations and dust exposures of the swine workers by eighty and eighty-five per cent respectively.

2.5.2 Air cleaning

The use of electrostatic air cleaners and vacuum cleaning has been investigated in swine confinement buildings. These methods have shown little improvements in reduction of dust and contaminants from the air of the buildings and are thus are not thought to be feasible relative to the cost and labour input involved in their use (Gustafsson, 1999). Research into wet scrubbers and dry filters has found them to be technically difficult and impractical. Thus, as is recommended by Wathes and co-workers (1998) further research is needed for cleaning of the air of these swine units *in situ*. The most practical methods available for air cleaning are ventilation systems.

2.5.2.1 Ventilation system

There are multiple demands on the ventilation systems in swine confinement buildings. However, the main purposes of any swine facility ventilation system are to: (1) maintain an adequate supply of fresh air for the animals, (2) remove excess moisture during cold weather, (3) remove combustion gases from heaters, (4) provide adequate temperature control during mild weather, and (5) limit the temperature rise during hot weather. A well -designed and managed ventilation system will control the levels of gases, dusts and vapours, and is an important factor in controlling odours from swine confinement buildings (Chastain, 2000).

Mechanical Exhaust Ventilation is the most common type of system used in modern swine facilities. It takes three basic components: Properly sized fans, properly sized and distributed fresh air inlets, and controls. The fans and inlets must be designed to provide at least three stages of ventilation. A proper setting of the minimum ventilation rate is one of the first things to look at to maintain acceptable ammonia and carbon dioxide concentrations in a swine facility. Considering that the swine confinements buildings' ventilation system is not controlled by the room ammonia concentration, it cannot react to an increase in ammonia release (Lemay *et al.*, 2002) A minimum, continuous ventilation rate for winter, a mild weather rate for

temperature control during the autumn and spring, and a maximum rate to control the temperature rise of the building in summer are required (Chastain, 2000). Ideally the system would be connected to an alarm in the case of a failure and a back up system should be in place. Furthermore, it has been recommended that more thorough mixing of the stagnant indoor air of well-insulated buildings may reduce the effects of clouds of pollutants containing gases, dusts and endotoxins (Pickrell, 1991).

2.5.3 Diet formulation

The first method to reduce ammonia emission caused by excessive nitrogen is reducing the nitrogen content in the swine diets. About seventy per cent of the protein ingested by swine normally ends up in the manure as urea, ammonia and other compounds. Bacterial action on the manure will result in production of several foul smelling compounds, which create unfavorable working conditions (Lynch, 2000). Reduction of dietary protein combined with supplementation of synthetic amino acids such as lysine in swine diets has been shown to reduce total nitrogen excretion by twenty-five to forty per cent. Reduction of dietary protein by twenty nine per cent has resulted directly in a reduction of ammonia emission by fifty-two per cent (Kay and Lee, 1997; Lemay *et al.*, 2002). Moreover, concentrations of other major odour components responsible for swine odour are significantly lower in slurry from swine fed low crude protein diets compared to a control diet (Hobbs *et al.*, 1996; Lemay *et al.*, 2002). Thus, diet formulation can have significant effects on ammonia emissions and on the level to which workers are exposed.

2.5.4 Manure Handling and Storage

Hydrogen sulphide produced during agitation of manure can have lethal consequences. Teagasc (1999) have published concise guidelines for manure management in intensive agricultural enterprises, which is based on the following framework elements: Manure quantity and quality; reducing manure nutrient content; operational procedures; and quality assurance. A systematic approach encompassing the above elements should be formulated and strictly adhered to for manure handling in every swine farm. Ventilation should be operating at maximum capacity during the operation and afterwards for a sufficient period before anyone commences work in the building. It is also recommended to always have two people that are properly equipped working together as protective equipment can, and does fail. Meyer (1997) recommended that ammonia-emission from livestock housing can be reduced by drying the manure quickly, minimising the period of time during which the manure is in contact with the air, and importantly minimising the contact surface area of the manure with the air. In addition, Meyer (1997) demonstrated that emission from manure storage could be reduced by at least eighty per cent provided that the right kind of covering is used. Also commercial manure additives are available for mitigation of odour production, reduction of ammonia and hydrogen sulphide emissions, and breaking down of solids.

2.5.5 Personal Protection Equipment (PPE)

As with all occupations, PPE can provide an invaluable means for protecting the health of swine confinement workers. However, it is imperative that PPE be used as part of a supervised respirator program and as an adjunct to management practices and engineering controls. Voluntary use of this equipment is hindered by issues such as: Discomfort and difficulty in communicating; lack of awareness or acceptance of the real risks of working unprotected in hazardous areas; and the costs associated with their use (Barber *et al.*, 1999).

The use of personal respiratory protection while working with swine is still not common. However frequent use of dust masks may prevent respiratory illness from occurring. To be effective, disposable masks must be of correct fit and the worker must be willing to wear the device for the duration of the entire working shift. It is also imperative that reusable masks are maintained and stored correctly. Dust masks are particularly appropriate for tasks that result in higher than normal exposure or those that have proven to result in undesirable health affects in a particular worker. The workers must use the correct type of dust mask, which can be decided on by looking at the code of the mask. The European Norm (EN) 149 Type P2 is ideal. However EN of FFP1, which is often used on swine farms, offers inadequate protection (Lawlor, 2002). Dust masks provide no protection against toxic gases, thus an alternative range of equipment is required. Only a self-contained breathing apparatus should be worn when entering a manure pit or other confined space on the farm. In a study comparing the effectiveness of two strategies for reducing dust exposures, Barber and co-workers (1999) found that wearing a dust mask was a more effective means of reducing the effects on pulmonary function than the oil spraying treatment; the reduction in dust was approximately ninety-five per cent for a properly fitted mask.

3.0 Overview of Chapter

This chapter contains an overview of the project sampling strategy and a detailed description of the methodologies used for personal sampling of ammonia, carbon dioxide, swine confinement dust and endotoxin. Details on the microbiological analysis of the bioburden of the air in the swine confinement buildings through use of settle plates are specified. In addition, the statistical tests that were used for analysing the results are noted in this chapter.

3.1 Sampling Strategy

Several farms were involved in the sampling for this study, which was carried out during the months of June and October 2006. The farms involved varied in size (ranging from 200 to 2,200 sows), the number of employees (ranging from 1 to 15 employees), and in the age of the facilities (ranging from 4 to 40 years old). The smaller farms were of older design, while the larger farms had more modern designs and facilities, and an increased number of employees.

The swine farm workers are identified depending on the unit on the farm in which they work, and were thus classified as Similar Exposure Groups (SEGs). The SEGs are as follows: Farrowing unit worker, weaner unit worker, fattening unit worker, dry sow unit worker, and farmer. The first four SEGs would spend the majority of their working day in the relevant units, while the farmers would spend their day working throughout all of the swine confinement units.

The **farrowing unit workers'** duties include: Inducing sows for birth; assisting in farrowing, both physically and in the administration of oxytocin; clipping tails and teeth of piglets at birth; administration of injections to piglets, (iron and vaccines); recording all births and deaths of piglets; assessing house temperature, and piglet and sow welfare on a daily basis; and introducing piglets to creep feed as soon as possible.

The **weaner 1st stage unit workers'** duties include: Grouping piglets in homogenous groups usually consisting of a set number; total switch over to solid creep feed; ensuring high

temperature in the unit for first week, usually 28°C with a reduction of 2°C each week to a constant of 22°C; inject weaners as appropriate; move onto a link feed after a week or so; and maintain good clean feeders and drinkers. The main duty of these workers is to assess piglet welfare, as this is a very critical period and animals are very susceptible. Poorly thriving weaners need a lot of attention at this time. In the **weaner 2nd stage** unit the workers main duty is general observation for poorly thriving pigs. The swine undergo their biggest diet change in this stage, moving on to a weaner ration. The weaners require a lot less supervision at this point and there is no house heating. They are not mixed any more and they remain in the same groups as in weaner stage-one through to leaving the fattening unit for slaughter.

The **fattening unit worker**: At this stage the pigs are moved onto a fattening ration. Observation is important as when the pigs move onto wet feed, their feed quantities must be constantly amended and it is important to ensure that all pigs are feeding properly. An important issue at this stage is the change of floor type, which can cause lameness in the fatteners. The general welfare of the swine is constantly assessed.

The **dry sow unit worker**: Not as much attention is needed with pregnant sows as they are housed in stalls. The worker must walk through all the stalls at feeding times to ensure that the sows are feeding properly. These workers are also responsible for serving of weaned sows and gilts; conducting daily boar walks to see if sows are in heat (boar effect); and keeping records of all sows served and repeats. Dry sow unit workers are responsible for the general welfare of the sows, including vaccines, recording parities for culling, and tagging sows with lost tags.

All workers are responsible for cleaning and washing of their respective units, as described in Section 2.5.1.1, of Chapter 2.

The **farmer** is most commonly found on the smaller farms, where one worker may be responsible for all of the above duties and running of the entire farm on a daily basis.

All swine confinement workers' personal exposure to ammonia, carbon dioxide and swine confinement dust was investigated. After review of the literature it was decided to focus on

determining the endotoxin exposures of the weaner unit worker, fattening unit worker and farmer. According to Chang and co-workers (2001) these workers were exposed to the highest levels of endotoxin during their working day. Sampling involved the workers wearing the relevant monitoring devices for a minimum of seventy five per cent of an eight-hour working day. Also, settle plates were used for microbiological analysis of the air in the swine confinement buildings. The typical working shift varied, but generally commenced at 08.30 hours and finished at 17.30 hours. A 30-minute break was taken both in the morning and in the afternoons, with an hour break taken for lunch. The workers had their breaks either outside in the fresh air or in canteens located on the farms. At the end of each working day, a monitoring record sheet was compiled, which contained all the relevant data for that day's sampling (Refer to Appendix).

Work has previously been carried out here in the National University of Ireland, Galway on determining and assessing the occupational exposure of swine confinement workers to gases, swine confinement dust and noise. Thanks to sponsorship from the HSA, it was possible to undertake the current study in order to continue this effort and also commence carrying out microbiological and endotoxin analysis. The carbon dioxide, ammonia and swine confinement dust results obtained were incorporated into a pre-existing database, resulting from these previous studies. This data, along with the endotoxin data was analysed using Statistical Package for Social Science (SPSS). The microbiological data from the settle plates was analysed using GraphPad InStat 3.

3.2 Gas Sampling in Swine Confinement Units

3.2.1 Ammonia Sampling

The electrochemical ToxiPro Single Gas Detector was used to continuously monitor the ammonia levels of the workers in the SEGs in the swine confinement buildings. This device is small and light weight which enabled the workers to continue with their daily tasks safely and at ease. The ToxiPro was clipped to the workers' collar in their breathing zone, for the duration of their working day, and upon removal the device automatically saves the measurements data in memory. The readings were downloaded to the computer via a universal serial bus (USB) - infrared beam. From the data output generated it was possible to determine the start time, end time, duration, peak and average of the ammonia exposure readings.

Plate 4: Personal Ammonia Sampler



Picture taken by Author (2006)

3.2.2 Carbon Dioxide Sampling

Carbon dioxide was measured using the Anagas CD 98 infrared analyser. This compact and lightweight device measures in the range of 0 -10,000 ppm CO₂ or zero to sixty per cent CO₂. On-site this device was clipped onto the workers belt for a personal exposure measurement. The Anagas CD 98 has a data storage facility and data is downloaded via an Infrared Data Association (IrDA) communications link.

Plate 5: Carbon Dioxide Sampler



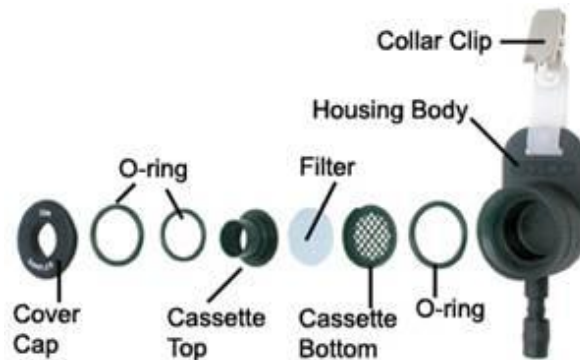
Picture taken by Author (2006)

3.3 Collection of Swine Confinement Dust

3.3.1 Equipment Used

- IOM Inhalable Dust Sampler (Figure 4)
- 25 mm glass fibre filters
- Polyurethane foams (PUF)
- Tygon tubing
- SKC suction pump
- The Gilibrator Primary Flow Calibrator
- Sartorius ME5-F Microbalance
- Flat tipped tweezers for handling filters

Plate 6: Exploded view of the IOM Inhalable Dust Sampler



[SKC website] <http://www.skcinc.com> [Accessed 19th July 2006]

3.3.2 Preparation of sampling equipment

1. The IOM Inhalable Samplers were cleaned before use. This involved the units being disassembled, soaked in laboratory detergent solution, rinsed thoroughly with water, wiped with adsorptive tissue and allowed to dry thoroughly before reassembly .
2. All filters and foams were pre-conditioned in the weighing-room over night before weighing. The temperature and relative humidity were recorded.
3. The filters were loaded into clean and dry sampling heads using flame sterilised flat tipped tweezers.
4. The weight of the filter and cassette was firstly recorded, which was used to determine the respirable fraction of the swine confinement dust.
5. The weight of the entire unit with the foam in place was then recorded, which was used to determine the inhalable fraction.
6. Each loaded sampling head was connected to a sampling pump using Tygon tubing, ensuing that no leaks occurred.
7. The pump was switched on and the calibrated flow meter was attached to the sampling head so that it measured the flow through the sampler inlet orifice. The flow rate was set to 2.2 l/min with an accuracy of plus or minus five per cent.

Plate 7: IOM Inhalable Sampler



Picture taken by Author (2006)

3.3.3 Sampling Train

1. The sampling head was fixed to the worker on their lapel and as close to the mouth and nose as possible.
2. The sampling pump was then either placed in a convenient pocket or attached to the worker in a manner that caused minimum inconvenience, such as to a belt around the waist. The pump was switched on and note was taken of the time at the start of the sampling period.
3. The workers and the equipment were observed periodically throughout the sampling period in order to ensure that the pump was operating as desired. At the end of the sampling period the sampling pump was switched off and record was taken of the time.
4. The sampler was disconnected from the sampling head and stored in its cassette.
5. On return to the lab the flow rate of the pump was measured with the calibrated flow-meter using the actual sample to ensure that over-loading of the filter did not occur. The sample identity and all relevant sampling data were carefully recorded in the monitoring record sheet.
6. The mean flow rate was calculated by averaging the flow rate measurements from before and after the sampling period and calculating the volume of air sampled, in litres, by multiplying the flow rate in litres per minute by the sampling time in minutes.

3.3.4 Gravimetric Analysis

1. The samples were allowed to re-condition overnight in the balance room. Again note was taken of the temperature and relative humidity.
2. Prior to weighing they were placed in front of a static eliminator for 3 minutes to eliminate any static build up.
3. The whole unit was weighed and recorded (inhalable fraction).
4. The foam and cassette front were removed and the filter and cassette rear were weighed and recorded (respirable fraction).

3.4 Microbiological Analysis of the air of Swine Confinement Buildings

3.4.1 Media Preparation

All media were supplied from LAB M[®]: UK.

3.4.1.1 Malt Extract Agar (Yeast and Moulds)

Malt extract agar is a selective media for the enumeration of yeasts and moulds. This media was prepared as follows:

1. 25 g of media was weighed and added to 500 ml of de ionised water in a Schott bottle (500 ml) and mixed vigorously.
2. The mixture was autoclave at 115°C for 10 minutes at 103.5 kPa.

3.4.1.2 Nutrient Agar (Total Colony Forming Units)

Nutrient agar is a general-purpose agar for the culture of non-fastidious organisms and is used to determine the total colony forming units. This media was prepared as follows:

1. 14 g of media was weighed and added to 500 ml of deionised water in a Schott bottle (500 ml) and mixed vigorously.
2. The mixture was autoclaved at 121°C for 15 min at 103.5 kPa.

3.4.1.3 Violet Red Bile Agar (VRBA) (Coliforms)

VRBA is a medium for the general enumeration of coliform organisms. This media was prepared as follows:

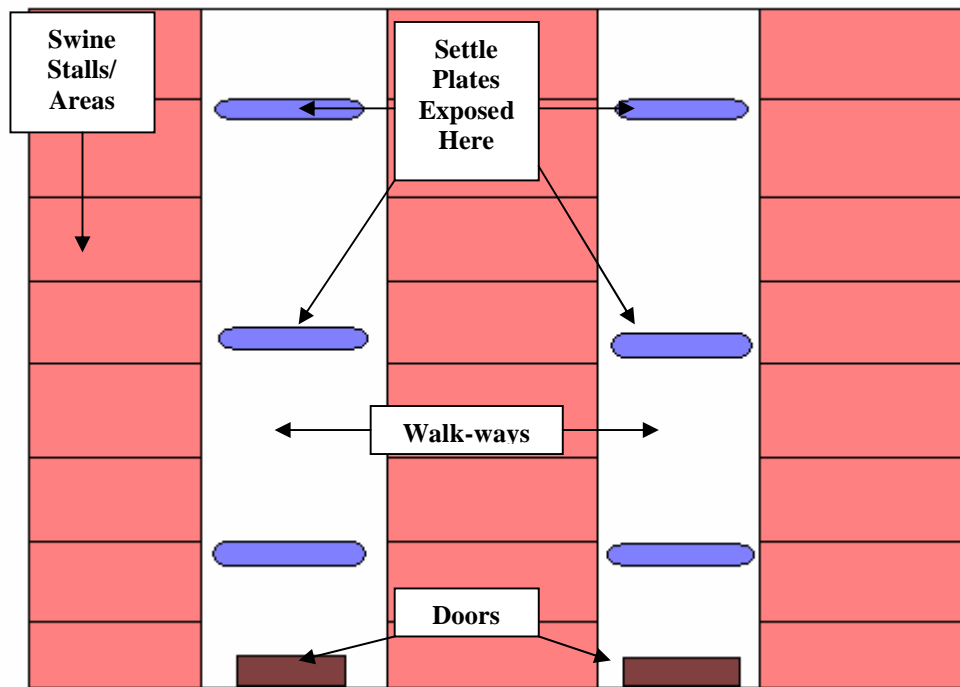
1. 19.25 g of media was weighed and added to 500 ml of deionised water in a glass conical flask (1 l) and mixed vigorously.
2. The flask was plugged with cotton wool and the solution was brought to the boil with frequent stirring over a Bunsen burner.
3. The solution was then allowed to cool to 47°C.

Using aseptic techniques the media was poured into petri dishes. The dishes were stacked so as to avoid condensation and left at room temperature until solidified. If necessary the dishes were dried by being exposed for ten minutes in a laminar airflow (LAF) cabinet. All media were stored at 4°C until required.

3.4.2 Settle Plates for the Culture & Enumeration of Microorganisms

Settle plates were exposed at various locations in the pig farms for a specified time to determine the microbial bioburden of the air. The locations were chosen in an attempt to account for possible variability in the units. Figure 3 is an example of locations that were chosen for exposure of the settle plates in one particular swine confinement unit. For the first sampling session an exposure time of 10 minutes was employed. Subsequently, an exposure time of 2 minutes was used in an attempt to get the colony count in the region of 30 to 300 colony-forming units per plate.

Plate 8: Sample Plan of Locations where Settle Plates were exposed in a Swine Confinement Unit



3.4.3 Media Incubation Periods

All plates were inverted and incubated as follows:

- Malt Extract Agar: 20°C for 5 days
- Nutrient Agar: 30°C for 72 hours.
- VRBA: 37°C for 24 hours

3.5 Endotoxin Sampling

The protocol that was used for endotoxin sampling is similar to that used for the collection of swine confinement dust, described in Section 3.3, of Chapter 3.

3.5.1 Equipment used

The equipment used for endotoxin sampling is similar to that described in Section 3.3.1 of Chapter 3.

3.5.2 Preparation of sampling equipment

Preparation of sampling equipment was carried in a similar manner to that described in Section 3.3.2, of Chapter 3. However, foams were not available for endotoxin sampling and it was not necessary to weigh the units. Thus, preparation was carried out as follows:

1. The IOM Inhalable samplers were cleaned before use. This involved the units being disassembled, soaked in laboratory detergent solution, rinsed thoroughly with water, wiped with adsorptive tissue and allowed to dry thoroughly before reassembly .
2. The 25 mm glass fibre filters were individually wrapped in tin foil and placed in a glassware oven at 180°C for 4 hours.
3. The filters were loaded into clean and dry sampling heads using flame sterilised flat tipped tweezers.
4. Each loaded sampling head was connected to a sampling pump using Tygon tubing, ensuing that no leaks occurred.
5. The pump was switched on and the calibrated flow meter was attached to the sampling head so that it measured the flow through the sampler inlet orifice. The flow rate was set to 2 l/min with an accuracy of plus or minus five per cent.

3.5.3 Sampling Train

The IOM Inhalable Sampler was used for collection of the inhalable endotoxin samples. The equipment was placed on the worker in the same manner as described Section 3.3.3, of Chapter 3.

3.5.4 Endotoxin Extraction

1. Samples were extracted by placing the filters in 10 ml of sterile pyrogen -free water (Water for injections: B| Braun: Germany) in 30 ml sterilins.
2. The samples were placed on a shaking table at 90 rotations per minute (rpm) at room temperature for 1 hour.
3. Samples were subsequently centrifuged at 4000 rpm for 10 min.
4. The solutions were then decanted into fresh sterilins in a LAF.

3.5.5 Effect of Freezing during Storage on Endotoxin Sample

As mentioned previously (Section 2.2.3.2 of Chapter 2) there is much debate in the literature as to the effect of freezing samples during storage on the endotoxin levels that are subsequently detected. In order to address this uncertainty, one particular extracted sample was divided into two separate sterilins. Sample 1 was stored at 4°C while the other sample (Sample 2) was frozen. The LAL assay determined endotoxin levels to be as follows:

- Sample 1 (4°C) = 21 EU/m³ air
- Sample 2 (frozen) = 4.2 EU/m³ air

Freezing of the sample during storage was found to result in an eighty per cent reduction in endotoxin levels, as opposed to storage of the same sample at 4°C. Thus, in accordance with Douwes and co-workers (1995) it was decided to store the samples at 4°C until being transported for analysis

3.5.6 Endotoxin Analysis

Microchem Laboratories carried out endotoxin analysis. This is an Irish National Accreditation Board (INAB) accredited laboratory. Analysis was carried out using the *Limulus Amebocyte* Lysate Endosafe Assay (Refer to Appendix). This gel-clot LAL test method is conducted as follows:

1. Equal parts of **Endosafe**[®] LAL reagent and the endotoxin sample are mixed together.
2. The mixture is then promptly incubated undisturbed for 60 minutes at 37 °C.
3. A positive response on the gel clot indicates that there is an amount of endotoxin in the sample that equals or exceeds the reagents' labelled sensitivity.

Thus, due to the nature of the endpoint assay, i.e. the gel either clots or not at a certain dilution of the sample, the results are generated as cut-off or break points of EU/ml. These results are then converted to EU/m³ air, as described in Section 4.4, of Chapter 4.

3.6 Data Analysis

Raw data for the swine confinement dust (mg/m³), ammonia (ppm), carbon dioxide (ppm) and endotoxin (EU/m³) sampling was analysed using SPSS version 14.0 for Windows, which is a data management and analysis software. Data was analysed using a combination of descriptive statistics, parametric, non-parametric and post-hoc tests.

Results from the microbiological analysis of the air (CFU/m²) were analysed using GraphPad Instat version 3.06, 32 bit for Windows, GraphPad Software, San Diego California, United States of America. Analyses were mainly carried out using Mann-Whitney U Test and Kruskal-Wallis Tests. In all cases, p values of > .05 were considered as significant.

Software provided with both the ammonia and carbon dioxide personal samplers were used for downloading the information and generating preliminary tables and graphs. Microsoft Excel was employed for producing tables and figures used for the presentation of results.

4.0 Overview of Chapter

Swine confinement dust, ammonia, carbon dioxide and endotoxin samples were collected during the period of May-July 2006. Table 1 is an overview of the number of personal samples that were collected throughout this period.

Table 1: Previous Project Database: Number of Personal Samples Collected

<i>SEG</i>	Inhalable Dust	Respirable Dust	NH₃	CO₂	Inhalable Endotoxin
Weaner	1	1	4	1	10
Fattening	1	1	1	0	8
Farrowing	5	5	2	0	NS
Dry Sow	5	5	1	1	NS
Farmer	2	2	2	1	19
Total	14	14	10	3	37

NS denotes: Not Sampled

For the purpose of data-analysis this data has been incorporated with a pre-existing dataset (explained in Section 3.1, of Chapter 3) resulting in a larger dataset, presented in Table 2. This is the dataset that was analysed for the purpose of the current project.

Table 2: Incorporated Database: Total Number of Personal Samples

SEG	Inhalable Dust	Respirable Dust	NH₃	CO₂	<i>Inhalable Endotoxin</i>
Weaner	12	12	8	5	10
Fattening	6	6	14	5	8
Farrowing	10	12	6	4	NS
Dry Sow	11	11	6	7	NS
Farmer	8	7	5	2	19
Total	47	48	37	23	37

NS denotes: Not Sampled

Results from the above data were analysed using SPSS version 14.0 for Windows. Settle plates were employed for the culture and enumeration of the microorganisms present in the air of the various units in the swine confinement buildings. Results from this research were analysed using GraphPad InStat version 3.06. In this chapter the major findings of the above research are highlighted and their significance acknowledged where applicable.

4.1 Gas Exposure of Swine Confinement Workers

4.1.2 Carbon dioxide

Table 3 illustrates that the mean values, ranges and standard deviations of carbon dioxide peak (ppm) exposures experienced by the weaner, fattening, farrowing, dry sow and swine farmer worker groups.

Table 3: Carbon dioxide peak exposures (ppm) of the Various SEGs

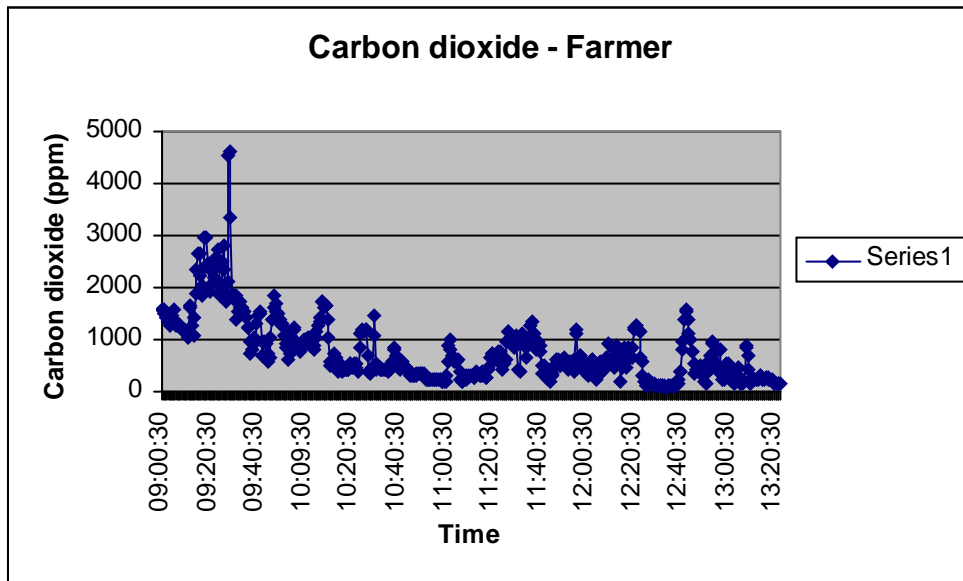
SEG	Mean (ppm)	Std. Deviation (ppm)	Number of Samples
Weaner	1722 (430-2970)	955	5
Fattening	2090 (1190-3480)	991	5
Farrowing	1488 (1151-1690)	243	4
Dry sow	2041 (1390-2680)	511	7
Farmer	4700 (4620-4780)	113	2

A one-way analysis of variance revealed significant differences between the SEGs ($F(4,18) = 7.950, p < .001$). Post-hoc comparison showed that the farmer SEG (Mean = 4700, S.D. = 113) experienced significantly more exposure to carbon dioxide than any other of the SEGs; weaner (Mean = 1722; $p < .001$); fattening (Mean = 2090; $p < .004$); farrowing (Mean = 1488; $p < .001$); dry sow (Mean = 2041 $p < .002$).

The most pertinent point that emerges from consideration of the carbon dioxide results is that the swine workers are frequently exposed to levels of carbon dioxide in excess of the recommended 1540 ppm carbon dioxide. Carbon dioxide levels in excess of this are considered to reflect poor air quality in the swine confinement buildings and greater potential risk of respiratory disease for the swine confinement workers.

Plot 1 shows an example of a farmers' daily exposure to carbon dioxide. A peak reading of 4700 ppm carbon dioxide is evident at 09.30 hours, which may have occurred when the farmer entered a building for the first time that day. From this time onwards, the carbon dioxide peaks appear to be cyclical, which may be a result of the farmer entering and leaving various units throughout the day.

Figure 1: Example of Farmers Carbon Dioxide Exposure



4.1.3 Ammonia

Table 4 presents the mean values, ranges and standard deviations of ammonia exposures of the weaner, fattening, farrowing, dry sow and swine farmer worker groups.

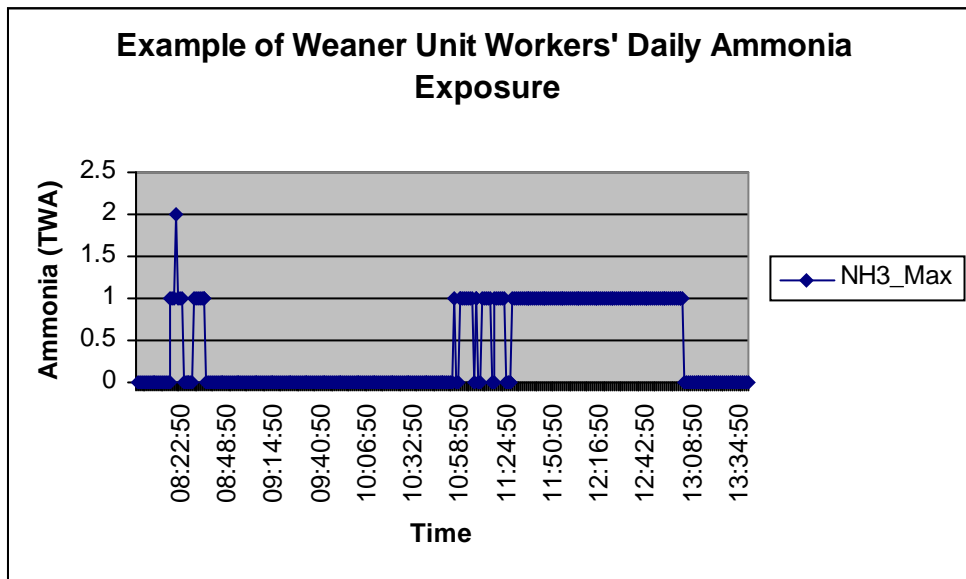
Table 4: Ammonia Exposures of the Various SEGs

SEG	Mean (TWA)	Std. Deviation (TWA)	Number of Samples
Weaner	0.41 (0.03-1.0)	0.44	8
Fattening	1.24 (0.09-2.9)	0.84	14
Farrowing	0.72 (.09-2.0)	0.71	6
Dry sow	1.47 (0.02-3.0)	1.07	6
Farmer	0.64 (0.01-2.0)	0.87	5

The highest ammonia TWA exposures were found in the dry sow (Mean = 1.5; S.D = 1.07) and the fattening (Mean = 1.2; S.D = 0.88) SEGs. However, a one-way analysis of variance showed that there were no significant differences between the ammonia exposures experienced by the various workers in the swine confinement buildings.

Plot 2 shows an example of a weaner unit workers' typical exposure to ammonia. On this particular day the worker commenced work at 08.00 hours and he/she entered the weaner unit at approximately 08.20 hours, as is evident from the initial peak value of 2 ppm ammonia. The worker then spent the early part of the morning doing tasks outside of the actual weaner unit, as is visible from the plot. The worker then returned, at approximately 11.00 hours to carrying out tasks inside of the weaner unit.

Figure 2: Example of Weaner Unit Workers Daily Exposure to Ammonia



4.2 Swine Confinement Dust

4.2.1 Inhalable Swine Confinement Dust

Table 5 shows the mean values, ranges and standard deviations of the inhalable dust fraction exposures experienced by the different swine confinement workers.

Table 5: Inhalable Dust Exposures of the Various SEGs

SEG	Mean (mg/m³)	Std. Deviation (mg/m³)	Number of Samples
Weaner	4.33 (0.25-7.6)	2.30	12
Fattening	2.75 (1.9-5.0)	1.16	6
Farrowing	1.94 (0.29-4.4)	1.51	10
Dry sow	1.36 (0.25-3.5)	0.79	11
Farmer	3.01 (1.1-5.6)	1.49	8

From Table 5 it is apparent that the weaner unit worker has the highest inhalable dust exposure (Mean = 4.3 mg/m³; S.D. = 2.3). A one-way analysis of variance was carried out to determine if this exposure was significantly different from the other SEGs. This analysis showed that the weaner unit worker had significantly higher exposure to inhalable swine confinement dust than other workers ($F(4,42) = 5.717, p < .001$). Post-hoc comparison showed that the weaner unit workers' exposure was significantly higher than both the farrowing (Mean = 1.9; $p < .011$) and the dry sow workers (Mean = 1.4; $p < .001$).

4.2.2 Respirable Swine Confinement Dust

Table 6 shows the mean values, ranges and standard deviations of the respirable dust fraction exposures experienced by the different SEGs.

Table 6: Respirable Dust Exposures of the Various SEGs

SEG	Mean (mg/m³)	Std. Deviation (mg/m³)	Number of Samples
Weaner	0.25 (0.03-0.63)	0.19	12
Fattening	0.16 (0.01-0.3)	0.09	6
Farrowing	0.46 (0.01-3.4)	0.95	12
Dry sow	0.10 (0.01-0.31)	0.11	11
Farmer	0.25 (0.09-0.63)	0.24	7

From Table 6, it is apparent that the farrowing unit workers have the highest exposure to respirable dust (Mean = 0.46; S.D. 0.19). Upon carrying out a one-way analysis of variance it is clear that this is not a significantly higher exposure than that experienced by any of the other SEGs.

4.3 Microbiological Analysis

4.3.1 Settle Plates for the Culture & Enumeration of Microorganisms

Settle plates were used for analysis of the bioburden of the air in the swine confinement buildings. The term bioburden is used to refer to the number of microorganisms with which an object is contaminated. Coliforms, total colony forming units, yeasts and moulds were collected using settle plates from the air of the swine confinement units and enumerated. This was carried out in both a relatively modern building with several workers and an older swine farm with less personnel working on-site. For the purpose of data analysis the more modern unit will be referred to as “Farm A”. Farm A has units varying in age from 4 to 10 years, with 13 employees working in the units. The older farm will be referred to as “Farm B”, with units being on average 40 years old and with 2 employees working in the units. The following data refers to total colony forming units per exposed plate (surface area = 63.6 cm²). The results were analysed using GraphPad InStat 3.

4.3.2 Comparison of the Microbial Bioburden in Farms A and B

Firstly the total numbers of each group of microbes present in Farm A and Farm B were compared. The Mann-Whitney U Test was used to carry out non-parametric analysis of the unpaired groups. Table 7 presents the results of this analysis

Table 7: Comparison of the Microbial Bioburden in Farms A and B

Microorganisms	Farm A (CFU/plate)	Farm B (CFU/plate)
Coliforms	Mean = 21 S.D. = 34 (n=15)	Mean = 6 S.D. = 6 (n=19)
Total colony forming units	Mean = 530 S.D. = 325 (n=16)	Mean = 639 S.D. = 375 (n=14)
Yeasts	Mean = 49 S.D. = 57 (n=14)	Mean = 101 S.D. = 67 (n=18)
Moulds	Mean = 19 S.D. = 12 (n=14)	Mean = 6 S.D. = 2 (n=18)

This analysis revealed that neither the number of coliforms nor the total colony forming units varied significantly between the two farms. The moulds were found to be present at significantly different levels in the two farms ($p < .0001$). Farm A was found to have higher levels of moulds (Mean = 19.4; S.D. = 12.1) than Farm B (Mean = 5.8; S.D. = 2.3). Conversely, the yeasts were found to be present at a significantly higher level in Farm B than they were in Farm A; Farm A (Mean = 49.3; S.D. = 56.6); Farm B (Mean = 100.6; S.D. = 66.8).

4.3.3 Comparison of the Microbial Bioburden in Different Units between the Two Farms

Both Farm A and Farm B have farrowing, weaner and dry sow units - Farm B does not have a fattening unit. Accordingly coliforms, total colony forming units, yeasts and moulds in these found in the air of these 3 areas were compared in order to determine if there were any significant differences. Table 8 presents the total numbers of microbes that were found to be present in the different units in Farm A and Farm B.

Table 8: The number of Microbes present in Different Units in Farm A and Farm B

Microorganisms	Farm A				Farm B		
	Dry sow Unit (CFU/plate)	Weaner Unit (CFU/plate)	Farrowing Unit (CFU/plate)	Fattening Unit (CFU/plate)	Dry sow Unit (CFU/plate)	Weaner Unit (CFU/plate)	Farrowing Unit (CFU/plate)
Coliforms	Mean=14 S.D. = 12 (n=4)	Mean=19 S.D. = 31 (n=3)	Mean = 5 S.D. = 4 (n=5)	Mean=59 S.D. = 63 (n=3)	Mean=3 S.D. = 3 (n=6)	Mean=1 S.D. = 2 (n=3)	Mean=9 S.D. = 7 (n=10)
Total colony forming units	Mean=597 S.D. = 562 (n=4)	Mean=493 S.D. = 214 (n=3)	Mean=577 S.D. = 483 (n=5)	Mean=548 S.D. = 190 (n=4)	Mean=438 S.D. = 291 (n=6)	Mean=1148 S.D. = 350 (n=3)	Mean=702 S.D. = 458 (n=10)
Yeasts	Mean=64 S.D. = 84 (n=8)	Mean=24 S.D. = 19 (n=3)	Mean=63 S.D. = 64 (n=4)	Mean=37 S.D. = 7 (n=3)	Mean=55 S.D. = 37 (n=8)	Mean=117 S.D. = 17 (n=4)	Mean=112 S.D. = 81 (n=10)
Moulds	Mean=20 S.D. = 19 (n=4)	Mean=13 S.D. = 11 (n=3)	Mean=20 S.D. = 4 (n=4)	Mean=27 S.D. = 5 (n=3)	Mean=6 S.D. = 2 (n=4)	Mean=4 S.D. = 2 (n=4)	Mean=6 S.D. = 3 (n=10)

Results from the Mann-Whitney U Test analysis revealed that the number of moulds in the dry sow and farrowing units were the only groups to differ significantly between Farm A and Farm B; at levels of ($p < .0427$) and ($p < .0020$) respectively. Moulds were found to be present in the dry sow unit of Farm A (Mean = 20; S.D. = 19) at a significantly higher level than in the dry sow unit of Farm B (Mean = 6; S.D. = 2). Also moulds were found to be present in the farrowing units of Farm A (Mean = 20; S.D. = 4) at significantly higher level than in the farrowing unit of Farm B (Mean = 6; S.D. = 3).

4.3.4 Comparison of the Microbial Bioburden in Different Units in each Individual Farm

A further analysis was carried out for both of Farms A and B. This involved comparing the different groups of microorganisms found in the air of each individual farm. The Kruskal - Wallis Test is a nonparametric test to compare three or more unpaired groups. Using this test to analyse the data it appears that there are no significant differences in the numbers of coliforms, total colony forming units, yeasts and moulds found in the four units of Farm A. The same non-parametric analysis of variance test of the three locations in Farm B revealed that the only significant difference was in the coliform numbers found between the weaner and farrowing units ($p < .0241$) of Farm B. Coliforms were found to be present in the weaner unit (Mean = 1; S.D. = 2) at a significantly lower number to that found in the farrowing unit (Mean = 9; S.D. = 7). Thus it appears that with the exception of this one significant difference, the levels of total colony forming units, coliforms, yeasts and moulds are the same within the units of the two farms.

For the purpose of comparison with the literature, the above results were extrapolated to number of CFU/m² as follows:

$$27 \text{ CFU/plate (63.6 cm}^2\text{)} = 43 \text{ CFU/100 cm}^2 \Rightarrow$$
$$43 \text{ CFU/100 cm}^2 = 4300 \text{ CFU/m}^2 \Rightarrow 4.3 \times 10^3 \text{ CFU/m}^2$$

Mean levels of microorganisms found in the swine confinement buildings were as follows:

- Coliforms (Mean = 2×10^3 CFU/m²; S.D. = 3.8×10^3 CFU/m²)
- Total colony forming units (Mean = 1.2×10^5 CFU/m²; S.D. = 1.6×10^5 CFU/m²)
- Yeasts (Mean = 1.3×10^4 CFU/m²; S.D. = 1.1×10^4 CFU/m²)
- Moulds (Mean = 4.4×10^3 CFU/m²; S.D. = 1.0×10^4 CFU/m²)

4.4 Endotoxin Exposures of Swine Confinement Workers

The sampling parameters for endotoxin sampling were as follows:

- 5 hours (300 minutes) at 2 l/min = 600 l = 0.60 m³ air.
- Each filter was extracted in 10 ml of pyrogen -free water.

Due to the nature of the LAL end-point assay results are generated as break-point values and are reported as EU/ml. In order to convert this to EU/m³ air the following calculation was applied:

- 1000 EU/ml = 10,000 EU/filter (each filter was extracted in 10 ml of pyrogen free water)
- 10,000 EU/filter
0.60 m³ air = 16,667 EU/ m³ air

Table 9 presents the values that were generated from samples from the weaner, fattening and farmer worker groups.

Table 9: Endotoxin Results (EU/m³ air) for Weaner, Fattening and Farmer Workers

SEG	Inhalable Endotoxin			
	<16,667 EU/m ³ air	16,667 EU/m ³ air	166,667 EU/m ³ air	>166,667 EU/m ³ air
Weaner (n=10)	4	4	2	0
Fattening (n=8)	2	3	2	1
Farmer (n=19)	5	4	10	0

It is apparent from the results in Table 9 that the workers are exposed to high levels of endotoxin in the swine confinement buildings. These results were anticipated due to the high levels of bacteria that were also found to be present in the various units (Section 4.3.1, of Chapter 4).

Endotoxin results consist of ordinal scale data. As this involves ranking of the endotoxin level it is necessary to carry out non-parametric analysis of variance. For this purpose a test called the Kruskal-Wallis one-way analysis of variance by rank was used. This analysis revealed that there were no significant differences between the levels of endotoxin exposure experienced by the different workers. Results for each of the worker groups contain endotoxin levels up to 166,667 EU/m³ air. However on one occasion (fattening worker) a level in excess of 166, 667 EU/m³ was detected.

5.0 Overview of Chapter

The primary objective of this study was to evaluate Irish swine farmers' occupational exposure to respiratory hazards, namely gases, swine confinement dust and endotoxins. In this chapter the main findings of the study in relation to the research objectives are discussed. The results of this research are considered and compared to other studies and research carried out in similar fields. The significance of this research and its results is considered while limitations of the current study are acknowledged. In addition recommendations that resulted from this discussion are offered and moreover, suggestions for future research are proposed.

5.1 Gas Exposure of Swine Confinement Workers

Carbon dioxide is produced by manure decomposition, animal respiration and heating systems. In the current study carbon dioxide exposures of swine confinement workers ranged from 430 to 4780 ppm. The farmer who works throughout all the units in the swine farm experienced significantly higher peak exposures of carbon dioxide than any of the other swine workers, with a mean peak exposure of 4700 ppm carbon dioxide. On both days where high carbon dioxide exposures were experienced by the farmer SEG worker (4620 and 4780 ppm), the worker was cleaning out non-slatted units, which would have a build up of manure on its surface, thus accounting for the high carbon dioxide exposure experienced by the workers. It is important to note that very high peak levels of carbon dioxide exposure were also experienced by the fattening unit (2090 ppm) and the dry sow unit workers (2041 ppm). While the OELV for carbon dioxide is 5000 ppm, concentrations in excess of 1540 ppm are considered to reflect poor air quality and greater potential risk for the development of respiratory disease (Donham, 2000). Although the 8-hour OELV was not exceeded, carbon dioxide levels detected in this study frequently reached concentrations greater than 1540 ppm. Mean carbon dioxide peak exposures in excess of this value were observed for all workers, except for the farrowing unit worker, who were exposed to just slightly lower levels of 1488 ppm carbon dioxide. Consequently, these high exposures indicate that swine workers are at an increased risk of

experiencing headaches and dizziness during their typical working day and potentially developing respiratory disease. Studies have shown a significant correlation between carbon dioxide and lung function as related to forced expiratory flow at 75% vital capacity (FEF75) (Donham *et al.* 1988) and between exposure to carbon dioxide and phlegm production (Zejda *et al.*, 1994).

Significant amounts of ammonia are released from manure and urine on the floors of the swine confinement buildings or from manure storage pits. In the current study, ammonia TWA exposures of the swine confinement workers ranged from 0.01 to 3.0 ppm and there were no statistically significant differences in the ammonia exposures experienced by the various worker groups. However, the dry sow and fattening unit SEG workers had the highest exposures, with mean ammonia TWA values of 1.5 and 1.2 ppm respectively. The high ammonia and carbon dioxide exposures in the fattening units are possibly attributable to the presence of swine excrement on non-slatted floors and high stocking density of the fattening pigs. The 8-hour OELV for ammonia is 20 ppm, while Donham (2000) recommended an exposure health limit of 7 ppm ammonia. While neither of these limits was exceeded in the current study, short-term peak concentrations of 16 ppm ammonia were observed during cleaning activities in fattening units which had non slatted floors. Such exposures reported in the current study have been associated with irritation of the mucous membrane of the eyes, noses and throats of the swine confinement workers. However due to the irritating nature of ammonia, serious health problems are rarely seen because workers remove themselves from high exposures (Preller *et al.*, 1995). As high ammonia exposures are associated with activities such as cleaning of non-slatted floors, it is especially important to ensure that the units are adequately ventilated during these periods.

Carbon dioxide exposure levels in the current study range from 430 to 4780 ppm, while ammonia TWA exposures range from 0.01 to 3 ppm. Chang and co-workers (2001) reported levels of 5 ppm ammonia and 600 to 895 ppm carbon dioxide in their study carried out in open style swine houses. However, the levels reported by these authors are well below most of the published data, which involve studies carried out in enclosed swine buildings. Average

ammonia levels and carbon dioxide personal exposures reported for workers in swine confinement buildings range from 5 to 34 ppm and from 1640 to 2632 ppm, respectively (Attwood *et al.*, 1987; Donham *et al.*, 1989; Malcom *et al.*, 2005; Zejda *et al.*, 1984). Ammonia levels reported in the current study appear to be quite low. However, an important point to remember is that ammonia levels are affected by seasonal variations, which may account for some of the variation in reported results. Ammonia concentrations are generally found to increase from summer to winter in response to a decreased ventilation rate to maintain internal temperatures in the colder winter temperatures (Crook *et al.*, 1991). While the farmer SEG mean carbon dioxide exposures appear to be quite high as discussed previously, the exposures of the other SEG worker groups' appear to be within the range of that reported in the literature.

5.2 Exposure of Swine Workers to Swine Confinement Dust

When considering the levels of exposure experienced by the workers to swine confinement dust, it is important to remember that Donham (1995) suggested an exposure threshold value of 2.4 mg/m^3 total dust and 0.23 mg/m^3 respirable dust. These values were suggested as repeated exposures to concentrations in excess of these were found to be associated with higher levels of ill health in swine confinement workers. Levels of personal inhalable swine confinement dust exposures found in the current study range from 0.25 to 7.6 mg/m^3 , while the respirable fraction range from 0.01 to 3.4 mg/m^3 .

The weaner unit worker (4.33 mg/m^3) was found to have the highest level of personal inhalable dust exposure across the different SEG worker groups. All of the weaner unit (4.33 mg/m^3), fattening unit (2.75 mg/m^3) and farmer (3.01 mg/m^3) SEG workers were found to be exposed to levels of swine confinement dust above the suggested threshold value of 2.4 mg/m^3 . The farrowing unit (1.94 mg/m^3) and dry sow unit (1.36 mg/m^3) SEG workers were not exposed to concentrations of swine confinement dust above this recommended limit. In relation to respirable swine confinement dust, the farrowing unit worker (0.46 mg/m^3) was found to have the highest personal exposure levels. The weaner unit (0.25 mg/m^3), farrowing unit (0.46 mg/m^3) and farmer (0.25 mg/m^3) workers exceed the recommended threshold limit of 0.23 mg/m^3 respirable dust. However, the fattening unit (0.16 mg/m^3) and dry sow unit (0.10 mg/m^3) workers were below the exposure threshold limit of 0.23 mg/m^3 respirable dust. There are thought to be a number of influencing factors contributing to the personal exposure levels experienced by the swine confinement workers, including: Duration the swine worker spends in the unit; level of activity of animals in the units; stocking density of the units; feeding systems and type of feed; surface area of the units; types of tasks done by the workers and manner in which they are carried out; and interval of cleaning of the various units.

As mentioned previously, the highest personal inhalable particulate exposure levels were experienced by the weaner unit SEG worker (4.33 mg/m^3). This high exposure may be attributable to the greater activity of the younger swine and the higher pig density in the weaner

units. A further possible influencing factor is the provision of dry based ration to swine in the weaner unit, whereas wet feeding systems are to be found in the other units. The weaner unit SEG workers duties involve close interaction with the young swine, such as grouping them into homogenous groups, administering injections and general observation of their welfare during this critical stage. The high swine confinement dust exposures of the farmer workers (3.01 mg/m^3) are possibly a result of the varied nature of the tasks carried out and the numerous units entered by these workers. As mentioned previously the farmer is responsible for the general running of the entire farm and thus must enter all the swine units several times throughout their typical working day. In addition to carrying out all the required tasks in the different swine units, the farmer is responsible for the moving of all the swine from each stage to the next, which may be responsible for generating high dust levels. In addition to caring for the swine, the farmer may be involved with working with other animals and general farming activities such as building maintenance. As with the weaner unit worker (4.33 mg/m^3), the high dust levels experienced by workers in the fattening unit (2.75 mg/m^3) may be a result of the high stocking density found in these units. The low levels of personal dust exposure of those working in the dry sow unit may be explained by the fact that these units are generally not as densely stocked and that the sows are often pregnant and relatively inactive.

Also of relevance are the cleaning intervals in the different units. Both the farrowing units and the weaner units are cleaned approximately every four weeks, in conjunction with the “All in, All out” policy. However, the fattening unit is cleaned approximately every eight to ten weeks, depending on the duration of time involved for the fatteners to reach their target weight of approximately 93 kg. Thus these units may not be as ‘clean’ as the farrowing and weaner units. This may be a possible influencing factor for the relatively higher exposures of the fattening unit worker (2.75 mg/m^3).

It has been suggested that the respirable fraction of swine confinement dust constitutes a significant proportion of the total inhalable fraction. Donham (1986) reported that seven per cent of the total weight of the dust was respirable (Gustafsson, 1999). With the exception of the farrowing unit worker, results reported in the study appear to concur with this statement. The

portion of total dust that is respirable in the farrowing unit appears to be exceptionally high, constituting approximately twenty four per cent of the total inhalable dust. This is perhaps due to the fact that the farrowing unit workers' tasks include a significant amount of interaction with the small piglets, such as clipping their tails and teeth. During such tasks the piglets are extremely active and may result in dispersing higher amounts of respirable dusts.

Both inhalable and respirable swine confinement dust levels recorded in the current study appear to be similar to those currently published in the literature. Inhalable dust exposures determined in the present study, ranged between 0.25 and 7.61 mg/m³ across all units, were similar to those of other published studies, which ranged from 1.32 to 8.8 mg/m³ (Attwood *et al.*, 1987, Cormior *et al.*, 1990, Donham *et al.*, 1986; Mackiewicz, 1998; Malcom *et al.*, 2005; Preller *et al.*, 1995; Simpson *et al.*, 1999). These concentrations are approximately ten-fold greater than total dust levels reported by Chang and co-workers (2001), who found average personal exposures between 0.15 and 0.34 mg/m³. Respirable dust exposures determined in the present study, ranged between 0.01 and 3.4 mg/m³ across all units, were also comparable to other published studies; means of respirable airborne dust exposures in swine confinement buildings have been reported between 0.13 and 2.5 mg/m³ (Donham *et al.*, 1986; Zejda *et al.*, 1994). Again the mean respirable exposures dust reported by Chang and co-workers (2001) were on the lower end of the scale, ranging from 0.08 to 0.24 mg/m³.

5.3 Microbiological Analysis of the Air of Swine Confinement Buildings

Settle plates were used for the collection and enumeration of the microbial bioburden of the air of swine confinement buildings. Coliforms, total colony forming units, yeasts and moulds were collected and enumerated. As described in Section 4.4.1 of Chapter 4, samples were collected in two farms, one being a modern building (Farm A), while the other is an older swine farm (Farm B). Older farms, often up to 40 years old, tend to be smaller operations (200 sows), employing possibly 2 or 3 employees. The modern facilities, as new as 4 years old, tend to be larger (2,200) and more intensive production units, employing up to 15 employees. Hence, these units often have more resources at their disposal and are in a position to ensure that more stringent cleaning practices are adhered to. The purpose of this analysis was as follows: Firstly to investigate if there were any differences in the microbial bioburden of the air between the older and the more modern swine facilities; secondly to investigate if there were any differences in the microbial bioburden of the air between each of the different units; and finally if there is a possible predictive value of observed cleanliness of the units in relation to the microbial bioburden.

Firstly the overall microbial bioburden in the air was compared between each of the 2 farms. Analysis of the settle plate results found that there were very few statistically significant differences in the numbers of microorganisms found between the two farms. There was a significant difference in the numbers of yeasts and moulds that were found between the two farms. Farm A was found to have higher numbers of moulds than Farm B, while Farm B was found to be contaminated with higher numbers of yeasts than Farm A. Secondly the microbial bioburden of the air of the farrowing, weaner and dry sow unit were then compared between Farm A and Farm B. In this case it was found that the only statistically significant difference between the two farms was in the number of the moulds in two of the units. Farm A was found to have higher numbers of moulds in the dry sow and farrowing units than did Farm B. Overall these results imply that while there are differences in the numbers of moulds in units between the two farms, there are little differences in the levels of coliforms, total colony forming units and yeasts found between the modern and older swine confinement buildings.

Interestingly, Attwood and co-workers (1987) showed that there is a significant positive correlation between airborne levels of culturable bacteria and the interval of stall cleaning. However, the findings of the current study seem to be in concurrence with those of Duchaine and co-workers (2000) who reported that observed dirtiness of the swine confinement buildings has a poor predictive value concerning air quality. Thus, while the interior of the units in Farm A may appear to be cleaner, it is not possible to presume that the air of such apparently 'cleaner' swine confinement building will automatically have a lower microbial bioburden.

In addition the microbial bioburden in each of the units was compared within both of the individual farms. From this comparison it appears that there is no difference in the numbers of microbes found in the weaner unit, farrowing unit, dry sow unit and fattening unit of Farm A. As regards Farm B, only one statistically significant difference was found, which was that the farrowing unit was found to have higher numbers of coliforms than the weaner unit. Thus the numbers of total colony forming units, yeasts and moulds appear to be similar across all the units within both Farms A and B. Several other studies have found the fattening unit to be contaminated with the highest airborne levels of culturable bacteria and gram -negative bacteria, both in open-air swine houses and enclosed swine buildings (Chang *et al.*, 2001; Cormior *et al.*, 1990; Crook *et al.*, 1991). This finding appears to explain and be in concurrence with the fact that fattening units have also been commonly cited as having one of the highest levels of airborne endotoxin, which is the component of the cell wall of gram -negative bacteria that is associated with the development of respiratory disease. Along with additional endotoxin samples, it would be desirable to collect enough dust samples in each of the swine units of both Farms A and B in order to determine if there is a correlation between the dust levels, endotoxin levels and the microbial bioburden in the various units in these particular farms.

Furthermore it would be interesting to investigate the levels of bacteria emanating from both modern and older swine confinement buildings. Green and co-workers (2006), in a study aimed at evaluating the levels of bacteria in the plume emanating from swine confinement buildings, found that there was a marked increase in bacterial levels inside the facility (average of 18,132 CFU/m³) versus upwind (average of 63 CFU/m³) and a steady downwind decrease in bacterial levels out to approximately 150 m from the facility. Thus, in relation to the current study, it

would be desirable to carry out the sampling on a larger scale and across the different units before any conclusions are drawn as to the effect of the facilities on the microbial bioburden of the air both inside of the various units and in the air emanating from such swine confinement buildings. In addition it would be beneficial to account for variables such as the duration of cleaning, number and density of swine in the units, ventilation systems and ventilation rates, temperature and relative humidity. Both temperature and relative humidity have been documented as two important factors relating to the survival of bacteria in dust and the number of airborne microorganisms (Chang *et al.*, 2001).

Overall, the numbers of coliforms in the air of the swine confinement buildings ranged from 0 to 5.6×10^3 cfu/m², numbers of total colony forming units ranged from 3.1×10^2 to 9.5×10^5 cfu/m², numbers of yeasts ranged from 1.1×10^3 to 3.5×10^4 cfu/m² and the numbers of moulds ranged from 7.9×10^2 to 8.1×10^3 cfu/m². Undoubtedly settle plates are an invaluable microbiological method for assessing the likely number of microorganisms that are naturally deposited from the air onto a surface in a given time. However after extensive analysis of the literature, it is apparent that this method has not been widely applied across various industries and thus it is not possible to compare the numbers of microorganisms found in the current study with those found in other studies. Several researchers have used air samplers, such as the Anderson microbial sampler (Cormior *et al.*, 1990; Chang *et al.*, 2001), which use forced impaction of a certain amount of litres of air per minute onto the agar media. Such results are reported as cfu/m³, as opposed to cfu/m², which is reported in the current study. It would be desirable in future studies to employ microbial air samplers in order to make comparisons with the microbial bioburden reported in both swine farms and other industries. Swine confinement buildings have previously been placed among the working environments with the highest bioaerosol pollution, along with grain stores, seed stores, animal feed factories, poultry farms, herb processing plants and waste composting facilities (Chang *et al.*, 2001; Adhikari *et al.*, 2004; Fishwick *et al.*, 2001; Dutkiewicz *et al.*, 2001). As of yet there are no internationally accepted OELV for bioaerosols and microbial bioburdens in working environments; however several authors have suggested recommended values, which have been reviewed by Dutkiewicz and co-workers (2001).

5.4 Endotoxin Exposures of Swine Confinement Workers

As mentioned earlier there are several modifications of the *Limulus Amebocyte* Lysate assay available for the quantification of endotoxin from environmental settings. In the current study the LAL endpoint assay was used to determine the levels of endotoxin that were present in the samples from the various swine worker groups.

The LAL endpoint assay generates results as break-point values; break-point values found in the current study were as follows: <16,667 EU/m³ air; 16,667 EU/m³ air; 166,667 EU/m³ air; and >166,667 EU/m³ air. Results from this assay indicate that all swine confinement workers are exposed to endotoxin levels at concentrations 166,667 EU/m³ air and in one particular case (fattening unit worker) was exposed to a level in excess of 166,667 EU/m³. There are numerous suggestions in the literature for an exposure standard ranging from 50 –2000 EU/m³ (Radon, 2002). The Dutch Expert Committee on Occupational Standards of the National Health Council has proposed a limit of 50 EU/m³ over an 8-hour exposure period (Heederik and Douwes, 1997), while the International Commission on Occupational Health have proposed an occupational exposure limit of 125 EU/m³. Several authors have offered exposure limit recommendations for endotoxin of 90 EU/m³, 330 EU/m³ and 800 EU/m³ (Castellan 1987; Rylander, 1985; Donham and Cumro, 1999). Even accepting the higher recommended threshold limit of 800 EU/m³ results found in this study are 200 times greater than suggested, indicating that there is potential for development of acute respiratory effects in swine confinement workers.

However, exposures to high levels of endotoxin are not uncommon across various industries. Zock and co-workers (1995) in a study aimed at determining exposure in the potato processing industry reported that twenty three per cent of the workers had a mean exposure above 1000 EU/m³. Kullmann and co-workers reported levels of 34,800 EU/m³ in the air of dairy farms. According to Fishwick and co-workers (2001), there have been reported maximum concentrations of endotoxin in the region of 16,970 and 66,000 EU/m³ in the cotton mill industry (Gokani *et al.*, 1987; Christiani *et al.*, 1993). Simpson and co-workers (1999) carried

out a comparative study of dust and endotoxin exposure across nine different occupations. These authors found poultry workers to be the most highly exposed to endotoxin levels of 719,950 EU/m³ with swine workers exposed to 149,230 EU/m³. These exposures appear to be comparable to exposures of 166,667 EU/m³ found in the current study. In order to comprehend the significance of such high endotoxin exposures, it is only necessary to point out that low exposures of 70 EU/m³ have been reported for occupations such as mushroom handling (Simpson *et al.*, 1999).

Statistical analysis revealed that there were no significant differences between the levels of endotoxin exposure experienced by the different SEG workers. While the weaner unit SEG worker was exposed to the highest level of inhalable dust, the workers in this unit were not exposed to the highest level of endotoxin. The highest level of endotoxin exposure (>166,667 EU/m³) was a sample collected on a worker in the fattening unit. Interestingly, Chang and coworkers (2001) also found that the highest respirable endotoxin levels in the fattening unit were greater than the inhalable exposures found in any of the other swine units. These authors explained that this inconsistency might be due to the variation in the amount of endotoxin and dust found to be in the respirable fraction across the different swine units. The difference in the duration of time that the workers spent in the units was also thought to be relevant. The fattening units tend to be larger units of greater surface area than other units, and the workers often need to spend longer periods of time in these units than they do in perhaps the weaner unit.

As endotoxin is a component of the cell wall of gram-negative bacteria and their release is linked with bacterial death, these high levels of endotoxin appear to be in concurrence with the high levels of bacteria that were detected in the air of the swine confinement buildings. Theoretically, quantification of the load of gram-negative bacteria in the air via endotoxin detection and *vice versa* should be possible. Laitinen and co-workers (1999) have shown that endotoxin levels in the air of wastewater treatment plants can be estimated from bacterial counts, by use of a selective medium for gram-negative bacteria. These authors concluded that the LAL assay could be used to estimate the concentration of viable gram-negative bacteria in

the air. However Seedorf and co-workers (1998) did not confirm these conclusions in their study, finding that there was no significant correlation between the concentrations of gram-negative bacteria and aerial endotoxin. Due to the nature of the break-point value results generated by the LAL endpoint assay, it is not possible to attempt to draw any conclusions from the endotoxin results and bacterial counts reported in the current study. Thus no definitive conclusions can be drawn from this study as to the possibility of quantifying the load of gram-negative bacteria in the air via endotoxin detection and *vice versa*. However, it is evident from these results that swine confinement workers are exposed to high levels of endotoxin, which have previously been associated with the initiation of respiratory disease.

5.5 Conclusions

While neither the ammonia nor carbon dioxide personal exposures sampled in the current study are above the recommended 8-hour OELV, the carbon dioxide exposure data frequently exceeds the recommended limit for the prevention of acute respiratory symptoms in healthy swine confinement workers. In addition, swine workers are frequently exposed to high levels of inhalable and respirable swine confinement dust at concentrations above recommended health threshold limits. Where contaminant concentrations are below the 8-hour OELV, it is important to remember that this is often because the workers spend a significant proportion of the working day in the ambient air. It is apparent from this study also that the air of swine confinement buildings is contaminated with high levels of microorganisms. Also endotoxin levels detected in this study are 200-fold greater than suggested exposure limits for swine confinement workers (Donham and Cumro, 1999). Thus results from this research show that swine confinement workers are potentially exposed to levels of contaminants at hazardous levels above recommended maximum values for human health, which previous studies have associated with adverse health effects in these workers, and in particular with the development of respiratory disease.

An important concept that emerges from this study is that the hallmark of agricultural exposures, such as those experienced by swine workers, is their enormous diversity in type, extent and duration. Importantly, there is a stark lack of awareness of the affects an occupation in the swine industry has on the health of the workers. Swine workers health can be protected through implementation of a comprehensive program incorporating environmental monitoring and control through the use of engineering controls, management practices, worker education, health surveillance and use of personal protection equipment. By ensuring that swine workers receive adequate education and training, and follow specified health and safety policies, employers can reduce the risk of illness, injury and mortality in their workforce.

5.6 Limitations of the Study

It is necessary to acknowledge the following limitation of the current study:

- As results from settle plates are generated as cfu/m² and there is a lack of such published results, it is not possible to compare the microbial bioburden of the air of the swine confinement units with other workplaces. In order to address this it would be possible to use microbial air samplers, which have been widely used for determining the microbial bioburden of the air across various industries.
- While the results generated from the LAL end-point assay indicate that workers are potentially exposed to endotoxin levels in excess of recommended health limits, they do not specify the exact levels (EU/m³) of air. The kinetic LAL assay is a modification of this assay that may be availed of in order to determine the exact level of endotoxin exposure of the swine confinement workers.
- After analysis of the literature it was decided to concentrate on determining the endotoxin exposures of the weaner, fattening and farmer SEG workers, who were previously reported as being exposed to the highest levels of endotoxin. However, in addition it would be desirable to determine the levels of exposure experienced by the farrowing and dry sow SEG workers.

5.7 Future Research

In order to further our understanding in this area and address current knowledge gaps, further studies are warranted to investigate the following areas:

- Duchaine and co-workers (2000) found that there is a decrease in some contaminants found in swine confinement buildings during the summer months. However the effect of seasonal variations on the levels of the various contaminants has not been considered in the current study and should be investigated in future studies, taking into account variables such as temperature, relative humidity and wind velocity.
- While the occupational exposure of swine workers to respiratory hazards was the primary concern of the current study, they are not the only workforce at issue in the agricultural sector. Those individuals working with poultry and cattle are also exposed to similar contaminants and it would be interesting to carry out an analogous study across different workforces in the agricultural sector.
- Given these high exposure levels a major objective of this study was to document the various control measures and best practices in existence in the swine industry (Refer to Section 2.5, of Chapter 2). While the current study has documented many of the control measures and best practices future research is needed into the practicality and effects of such measures in protecting the health of swine confinement workers.

5.8 Study Recommendations

On the basis of the present study's findings, the following recommendations are offered:

- Exposure monitoring and health surveillance are required across the swine industry for protecting the health of swine confinement workers, particularly for vulnerable groups such as pregnant workers or those with respiratory disease.
- Attention needs to be directed to protecting the health of swine workers through: Comprehensive programs of environmental monitoring and control; the implementation of engineering controls; use of efficient management and best practices; and education/awareness training.
- The HSA have issued a 'Draft Code of Practice for Preventing Injury and Occupational Ill Health in Agriculture', which is intended to provide guidance and to improve the level of safety and health among all people engaged in the agriculture sector. Such practical education is imperative in order to make workers aware of the risks to which they are exposed and the measures that may be taken, by them and their employers to protect their health and well-being.

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Appendix A: Monitoring Record Sheet

<u>Monitoring Record Sheet</u>								
Author		<u>GASES</u>						
Farm		NH ₃ SEG		CO ₂ SEG				
Date		Result	TWA ppm	Result	TWA ppm			
Number of Workers being Monitored								
DUST/ENDO		Pre-calibration	Filter & Foam weight (pre)	Time On	Sample	Time Off	1. Post-cal 2. Average flow rate	Filter & Foam weight (post)
<u>Worker Name</u>	<u>Pump</u>							
Additional Notes								

Appendix B: Details on *Limulus Amebocyte* Lysate Assay