6.0 Adverse Health Effects

6.1 Toxicology

Thomas L. Carson, DVM, PhD
Department of Veterinary Diagnostic and Production Animal Medicine
College of Veterinary Medicine
Iowa State University
Ames, Iowa

Gary D. Osweiler, DVM, PhD
Department of Veterinary Diagnostic and Production Animal Medicine
College of Veterinary Medicine
Iowa State University
Ames, Iowa

Peter S. Thorne, PhD
Department of Occupational and Environmental Health
Institute for Rural and Environmental Health
University of Iowa
Iowa City, Iowa
6.1 Abstract - Toxicology

Valid evaluation of the health effects of airborne substances released from animal production units should be based on the important and well-established toxicological principles of dosage and response. Dosage is the most important factor that determines response to poisons. Toxicity is the quantitative amount of toxicant required to produce a defined effect, but the hazard or risk of toxicosis depends not only on the inherent toxicity of the agent, but on the probability of exposure to the toxicant under conditions of use. Acute, subacute, and chronic toxicity are different chronological quantitations of chemical toxicity and are determined by relative dosage and time of exposure. Many factors can alter animal or human response to toxicants, including those inherent in the toxicant, the organism, the environment and the combinations of these major factors.

Toxicological evaluation depends heavily on determination of exposure and evidence for the contribution of interacting factors that can alter toxicity. Quantitative expressions of toxicity and exposure are essential for thorough toxicological evaluation and prognosis.

Response to exposure by airborne toxicants is likely to involve the respiratory system because it is a portal of entry. Study of CAFO issues suggests consideration of the mechanisms of injury by volatile agents and particulates, as well as understanding the potential effects of both acute and chronic exposure. Respiratory system effects are manifest in relatively limited ways (bronchoconstriction, pulmonary edema, asthma, carcinogenesis), and careful attention must be given to evidence for cause and effect from among a wide range of insults and levels of exposure. Similar considerations are important for systemic effects that are manifested in other parts of the body.

Laboratory animals are often as experimental models of human disease to help establish the mechanism of action and the correlation between exposure levels of airborne toxicants and clinical response. Clinical response to these pollutants depends not only on the concentration of the specific compound, but also the frequency and duration of exposure.

Studies of aerial ammonia in laboratory animals have demonstrated dose-effect and duration-effect patterns for damage to the respiratory tract similar to that observed in humans. Acute exposures to moderate concentrations of ammonia irritate the upper respiratory tract. Prolonged or repeated exposures to lower levels of ammonia produce inflammation and lesions of the respiratory tract. Exposures to high concentrations of ammonia result in severe damage to the upper and lower respiratory tract and alveolar capillaries.

Controlled studies with hydrogen sulfide in laboratory animals have shown that levels of 500 ppm or greater are likely to be lethal, similar to the response observed in humans. Exposure to sub-lethal levels of hydrogen sulfide have produced progressive effects ranging from increased respiratory rate, to pulmonary edema, to histopathological changes in the nasal cavity and lung tissue.

Endotoxins, glucans, and microorganisms maybe important components of bioaerosols associated with animal production units. Inhalation of these compounds have been shown to produce respiratory system effects including airway constriction and obstructive breathing pattern, inflammatory tissue responses, and overt infection of lung tissue.
6.1 Toxicology

6.10 Overview of Toxicology

Toxicology is the study of poisons, and their effects on living organisms. This includes an understanding of sources of poisons, circumstances of exposure, their effects, diagnosis and treatment and the application of management or educational strategies to prevent poisoning. More than many of the specialties in veterinary medicine, toxicology is based on the important principle of dose and response. Response is dependent not only on presence of a potential toxicant but on the amount of exposure as well. (Osweiler, 1996) With emphasis in this report on accountability of Concentrated Animal Feeding Operations (CAFOs) for substances released from animal production units, there is increasing need to be aware of and apply the dosage and response principle to best estimate the need for regulation or remediation.

Determinants of exposure that affect dosage may be more than simply the gross amount of material with access to animals or man. Rather, the effective dosage at a susceptible receptor site determines the ultimate response. Thus, environmental factors that influence exposure, species differences in organisms within an exposure area, vehicle differences that affect absorption, specific drug or chemical interactions that potentiate response, and organ dysfunction that limits elimination may all be factors which influence the ultimate dosage and the outcome of exposure. (Osweiler, 1996)

Toxicological Principles Of Evaluation For Cafo Issues

A poison or toxicant is any natural or synthetic solid, liquid or gas that when introduced into or applied to the body can interfere with homeostasis of the organism or life processes of cells of the organism by its own inherent qualities, without acting mechanically and irrespective of temperature. For CAFOs, toxicants considered are natural products that would normally be handled by ecological assimilation, but may be locally in unnatural or excessive concentration. Knowledge of the chemical nature and specific effects of toxicants and their combinations is the only certain way to assess hazard from such exposure. Suggestions about potential adverse effects of natural products from livestock waste may be gained from comparative experimental studies, from know effects of substances at high concentrations within CAFOs, and from well-controlled and properly interpreted epidemiological studies. This chapter will review the known biological effects of compounds identified in CAFOs, and will also present evidence gained from epidemiological studies.

Toxicological conventions should be followed in assessment of risk to different populations. Toxicity is the quantitative amount or dosage of a poison that will produce a defined effect. For example, the acute lethal dosage of hydrogen sulfide to swine could be described as a concentration in air, e.g. 1,000 parts-per-million or as the equivalent amount on a body weight basis. Toxicity values do not describe the biological effects, but only the quantitative amount (dosage) required to produce a defined effect (e.g. death, respiratory distress, immune suppression, etc). Dosage is the correct terminology for toxicity expressed as amount of toxicant per unit of body weight. Commonly accepted dosage units are mg/kg body weight or moles or micromoles of agent/per kg body weight. In comparative toxicology, relative effects in large and small animals relate dosage to the body surface area, which is approximately equal to (body weight)\(^{2/3}\). This relationship, and others relevant to interspecies comparisons, should always be considered when comparing laboratory or farm animal toxicity data against risk for humans. Generally, as animals increase in weight, the body surface area increases proportionally less, and this may affect the rate of metabolism, excretion and receptor interaction with toxicants. For many toxicants, larger animals
will be poisoned by relatively lower body weight dosages than are smaller mammals. (Eaton and Klaassen, 2001; Osweiler, 1996)

From a public health and diagnostic toxicology perspective it is essential to know what exposure level will not cause any adverse health effect. This level is usually referred to as the "no observed adverse effect level" (NOAEL). (Eaton and Klaassen, 2001) Usually a NOAEL in laboratory animals is based on chronic exposures ranging from ninety days to two or more years depending on the species. The inhalation toxicity for gases or aerosols, including particulates, is often expressed as the concentration of material (i.e. the weight of compound per volume or weight of air). The no-effect level is the largest dosage or concentration that does not result in detrimental effects. In industrial hygiene, the concept of protecting human health from exposure is quantified to an assumed normal work day exposure and given a value called the Threshold Limit Value (TLV), which includes a safety factor between exposure allowed and concentrations where adverse effects may be expected.

**Response to Toxicants**

Toxicant evaluation is usually classified according to chronological scale that accounts for both dosage and response. **Acute toxicity** refers to effects of a single dose or multiple doses measured during a twenty-four-hour period. Toxic effects apparent over a period of several days or weeks are classified as **subacute**. **Subchronic** toxicity refers to toxic effects that occur between 30 days and ninety days exposure. **Chronic** effects are those produced by prolonged exposures of three months to a lifetime. Chronic effects are affected by the cumulative tendencies of the toxicant. The ratio of the acute to chronic LD\(_{50}\) dosage is called the **chronicity index**. (Eaton and Klaassen, 2001) Compounds with strong cumulative properties have larger chronicity index. The potential for individual products from CAFOs to cause cumulative effects should include evaluation of their cumulative potential or chronicity index. Conversely, organisms may develop **tolerance** for a compound such that repeated exposure increases the size of the dose required to produce lethality. For example, the single dose LD\(_{50}\) of potassium cyanide in rats is 10 mg/kg, while rats given potassium cyanide for ninety days are able to tolerate a dosage of 250 mg/kg without lethality.

**Toxicity and Risk**

The concept of risk or hazard is important to toxicology. While toxicity defines the amount of a toxicant that produces specific effects at a known dosage, hazard or risk is the probability of poisoning under the conditions of expected exposure or usage. Compounds of high toxicity may still present low hazard or risk if exposure to the toxicant is limited. CAFO risk evaluation should include estimation of dosage at remote or off site locations, and measurement or estimation of exposure at such locations is essential. Factors discussed in previous chapters relating to dispersion and dilution in the environment are essential in estimating the risk for a compound, even if it is of high inherent toxicity. Moreover, binding of toxicant gases to particulates may either reduce or increase their toxic properties so that risk is a function of all factors and interactions.

**Factors That Affect Response To Toxicants**

Many factors inherent in the toxicant, the animal or the environment can alter a toxicity value determined under defined experimental conditions. The toxicity of a compound may vary with the route of exposure. Usual routes of exposure to environmental agents are oral, dermal and inhalation. Gases are absorbed directly through pulmonary membranes, but aerosols including dusts may be deposited in lower airways or lungs if they are in a range between 0.1 and 5.0 um. Systemic retention occurs when macrophages laden with particles gain access to the pulmonary lymphatic
Retention of inhaled particles in the gastrointestinal tract can occur when large particles trapped by cilia and mucus in the nasopharynx and trachea are swallowed. (Eaton and Klaassen, 2001)

Many environmental and physiological factors can influence the toxicity of compounds, and such factors, or others possibly unknown, can substantially influence response to toxicants. Accurate evaluation of CAFO risk to both on-site and off-site persons must consider multiple factors and their interactions to properly support regulatory and remedial activity. Some examples of factors that alter response to toxicants are presented in Table 1

<table>
<thead>
<tr>
<th>Alteration or Change</th>
<th>Mechanism or Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Changes in chemical composition or salts of inorganic agents</td>
<td>Toxicity of metals may be altered by valence state. Sodium salts are more water soluble than parent compounds, promoting absorption.</td>
</tr>
<tr>
<td>Instability or decomposition of chemical</td>
<td>Volatile compounds can decompose or change to more toxic form upon exposure to sunlight, as with nitrogen and nitrogen oxides.</td>
</tr>
<tr>
<td>Ionization</td>
<td>Generally, compounds that are highly ionized are poorly absorbed and thus less toxic. The pH of the source of pit gases may influence ionization of some products.</td>
</tr>
<tr>
<td>Vehicle effects</td>
<td>Non-polar and lipid soluble vehicles usually increase toxicity of toxicants by promoting absorption and membrane penetration.</td>
</tr>
<tr>
<td>Protein binding</td>
<td>Binding to serum albumin is common for many drugs and toxicants, limiting the bioavailability of the agent and reducing toxicity.</td>
</tr>
<tr>
<td>Chemical or drug interactions</td>
<td>Chemicals may directly bind, inactivate or potentiate another. One chemical may also induce microsomal enzymes to influence the metabolism of another.</td>
</tr>
<tr>
<td>Biotransformation</td>
<td>Prior exposure to the same or similar chemical may induce increased metabolic activity of microsomal mixed function oxidases (MFOs). Foreign compounds activated by MFOs can then be conjugated by Phase II metabolism and excreted. If toxicants are activated by MFO activity, then toxicity may be increased. Liver disease, very young or very old animals, and specific breeds or strains of animal can alter ability of MFO to begin metabolism followed by Phase II</td>
</tr>
</tbody>
</table>
Liver disease

Reduced synthesis of conjugating or binding agents (glutathione, metallothionein), essential proteins and coagulation factors may alter response to absorbed chemicals.

Nutrition and diet

Vitamin C and vitamin E can aid in scavenging of free radicals and repair of cellular protective mechanisms.

**Respiratory System Response to Injury**

Response of airways and lung to injury is dose dependent and expressed in chronological terms as acute, subacute or chronic. Response of the respiratory tract to toxicants is manifest in relatively few ways in response to many different chemicals, and a few specific mechanisms of injury are known. (Haschek and Rousseaux, 1998; Witschi, 2001)

**Mechanisms of Respiratory System Injury**

Respiratory damage depends on relatively few recognized molecular and cellular mechanisms that account for a wide variety of toxicant exposures. Many recognized effects are related to the oxidative burden imposed on the respiratory tract. (Witschi, 1997) This includes generation of unstable and reactive free radicals that lead to oxidative chain reactions and subsequent cellular damage or destruction. Cellular injury then results in release of microsomes and flavoproteins, neutrophils, monocytes and macrophages that can sustain the conversion of molecular oxygen to reactive oxygen metabolites. Many of these effects are an excessive response to what is a normal respiratory defense mechanism against microorganisms and low- or high-molecular-weight antigenic materials. Immunologic consequences are triggered when foreign materials in the respiratory tract sensitize the lung or airways to further exposure of the same material. (Witschi, 2001). Further consequences of oxidative damage or covalent binding in the pulmonary systems can result from damage and cross linking of DNA with potential subsequent development of carcinogenesis. The consequences of these mechanisms can be acute or chronic respiratory damage and the physiological dysfunction that accompanies each.

**Acute Respiratory Injury**

Acute airway damage in the transport passages (nasopharynx, trachea, bronchi, bronchioles) is reflected as bronchoconstriction and/or excess or reduced mucus and ciliary function. (Haschek and Rousseaux, 1998; Witschi, 2001). Response to irritants in nasal passages can cause acute or chronic rhinitis or, at higher concentrations, pause in respiration which develops as a reflex protective mechanism. Autonomic nervous system response to irritants is associated with acute reflex contraction of trachea and bronchi, resulting in decreased airway diameter and increased resistance to air flow. This results in wheezing, coughing, dyspnea and reduced exercise tolerance. This response is most likely triggered by irritant gases with moderate water solubility. Effects of short-term exposure resolve quickly when the irritant gas is no longer present and if no permanent cellular damage has occurred; long-term exposure may lead to chronic effects.

Acute lung damage can result in two major effects on lung tissue. Toxic pulmonary edema, which is characterized by alveolar or interstitial fluid accumulation and a thickened alveolar-capillary interface results in reduced oxygen and carbon dioxide exchange. Highly water-soluble irritant gases, including ammonia and hydrogen sulfide, which reach the lung parenchyma can damage cellular membranes.
and allow fluid leakage leading to pulmonary edema. Inflammatory response and cellular accumulation may accompany the edema and, if severe, result in prolonged changes including fibrogenesis. Acute alveolar endothelial damage and necrosis stimulates Alveolar Type II cell proliferation. These cells are physically thicker than Type I cells, and as immature replacements of Type I cells (alveolar endothelium) markedly reduce oxygen and carbon dioxide exchange (Witschi, 2001).

**Chronic Respiratory Injury**

Chronic response to injury may come from excessive and prolonged acute injury or from low-level or subclinical damage. In either event, manifestation is commonly as fibrosis or other chronic inflammatory change, emphysema, asthma or carcinogenesis.

Fibrosis is the result of excessive production of collagen in lung parenchyma and can occur at the alveolar, alveolar duct and bronchiolar levels. Type I and III collagen constitute approximately 90 percent of lung collagen. Increases in collagen, especially Type I, increase stiffness of the lung and reduce compliance, with severe fibrosis resulting in reduced vital capacity and reduced exercise tolerance.

Emphysema is characterized by “abnormal enlargement of the airspaces distal to the terminal bronchiole, accompanied by destruction of the walls, without obvious fibrosis”. (Snider et al, 1985) Emphysema arises from interference with or lack of alpha$_1$-antiprotease, leading to loss of pulmonary elastin and subsequent alveolar wall breakdown. This leads to reduced alveolar surface and hyperinflation of alveoli and lungs with excessive compliance.

Asthma is characterized by increased airway activity with excessive contraction of large airways in response to irritants. Effects may be initiated by exposure to antigens or by chemicals that serve as haptens, with contributing influences by inflammatory cells and cytokines (Barnes et al 1998). Effects are mild to severe dyspnea, which can be acute, recurring and influenced by inhalation of a variety of pollutants (Witschi, 2001).

Respiratory carcinogenesis, especially lung cancer in humans is common and associated with environmental, industrial and personal exposures to a variety of chemicals. For most lung cancers, there is likely a dose-response relationship but clinical disease is often manifested later in life after long-term exposure. Animal studies are helpful in definition of mechanisms and in selected dose-response considerations. However, animal studies are important to interpret carefully in the context of significant differences in laboratory animal susceptibility and for the dosages used in experimental studies compared to ambient exposures of human populations (Hahn, 1997; Malkinson, 1998).

**Systemic Effects of Airborne Toxicants**

Airborne toxicants can affect systems other than or in addition to the respiratory tract. Lung is an efficient absorption organ and readily transports volatile compounds to the systemic circulation. Neurological and immune system consequences may occur secondary to inhalation exposure. A limited amount of xenobiotic metabolism is possible in lung, so that some bioactivation of toxicants can occur upon first pass pulmonary absorption. Effects of absorbed volatile agents will depend on the eventual target organs and susceptible receptors. These specific effects in target tissues and organs will be discussed in detail in subsequent sections of this chapter.
6.1.1 Toxicology of ammonia

Experimental studies indicate that the concentration of aerial ammonia which is acutely lethal to laboratory animals is dependent on the duration of the exposure. The lethal concentration of ammonia in rats and mice increases 5-10 times as the duration of exposure decreases from 16 hours to several minutes (Hilado et al. 1977; Kapeghian et al. 1982; Weedon et al. 1940). Exposure frequency also appears to be an important factor in determining lethality. Continuous exposure to 653 ppm of ammonia for 25 days resulted in nearly 64% lethality in rats, whereas intermittent exposure to nearly twice this concentration was tolerated for 42 days (Coon et al. 1970). It also appears that male rats are more sensitive than female rats to the lethal effects of aerial ammonia (Appelman et al. 1982).

Studies in laboratory animals have demonstrated dose-effect and duration-effect patterns for damage to the respiratory tract similar to that observed in humans. Acute exposures to moderate concentrations of ammonia (≤1000 ppm) irritate the upper respiratory tract, whereas exposures to high concentrations (>4000 ppm) result in severe damage to the upper and lower respiratory tract and alveolar capillaries (Coon et al. 1970; Kapeghian et al. 1982; Mayan and Merilan 1972; Richard et al. 1978a,b; Schaerdel et al. 1983). Prolonged or repeated exposures to lower levels of ammonia (>150 ppm) produce inflammation and lesions of the respiratory tract (Broderson et al. 1976; Coon et al. 1970).

No overt symptoms of neurological disorders were reported in guinea pigs or monkeys that were exposed to up to 1105 ppm ammonia for 6 weeks (Coon et al. 1970). However, acute exposure to low levels of ammonia (100 ppm) has been shown to depress free-access wheel running behavior in rodents (Tepper et al. 1985). This may represent avoidance of sensory or upper airway irritation, but these same effects can be seen after injection of ammonium salts.

6.1.2 Toxicology of hydrogen sulfide

Controlled studies using dogs, rats, mice, and rabbits exposed acutely to high concentrations of hydrogen sulfide gas for various periods of time have shown that levels of 500 ppm or greater are likely to be lethal, similar to the response observed in humans exposed to high levels (Beck, 1979; Elovaara, 1978; Higuchi and Fukamachi, 1977; Haggard, 1922; Lopez, 1987, 1988a, 1988b, 1989; Kage, 1992; Khan, 1990; Prior, 1988, 1990; Savolainen, 1980; Smith and Gosselin, 1964; Tansy, 1981).

In addition to an increase in respiration rate that was noted in rats exposed to 100-200 ppm hydrogen sulfide for 1 hour (Higachi and Fukamachi, 1977), a number of histological and biochemical changes were noted in the respiratory tissues and fluids of rats acutely exposed to 200, 300 or 400 ppm hydrogen sulfide for 4 hours (Lopez, 1987; Green, 1991). Histopathological changes were reported in the nasal cavity of rats exposed to greater than 200 ppm hydrogen sulfide for 4 hours (Lopez, 1988b). Moderate-to-massive pulmonary edema was evident in rats exposed to 375 ppm hydrogen sulfide for 4 hours (Prior, 1990), and slight pulmonary congestion was found in rats exposed to 75 ppm hydrogen sulfide for 1 hour (Kohno, 1991). Significant decreases in numbers of viable pulmonary alveolar macrophages were noted in the lung lavage fluid of rats exposed for 4 hours to 400 ppm hydrogen sulfide (Khan, 1991).

The effects of intermediate-duration exposures to hydrogen sulfide have been examined in rats, mice, and pigs. Respiratory effects were not observed in two strains of rats exposed to hydrogen sulfide at concentrations up to 80 ppm 6 hours/day, 5 days/week, for 90 days (CIIT 1983b, CIIT
In contrast to rats, inflammation of the nasal mucosa described as minimal to mild was observed in mice exposed to hydrogen sulfide at 80 ppm (CIIT 1983a). Respiratory effects were not observed at 30.5 ppm. No mortality was noted during 90-day studies in which rats and mice were exposed for 6 hours/day, 5 days/week, to up to 80 ppm hydrogen sulfide (CIIT 1983b, 1983c). (CIIT 1983a).

Guinea pigs exposed daily to 20 ppm of hydrogen sulfide for 11 days developed fatigue, somnolence, and dizziness (Haider, 1980). Neurochemical analyses revealed decreased cerebral hemisphere and brain stem total lipids and phospholipids. Lethargy was observed in rats following exposure to 400 ppm of hydrogen sulfide for 4 hours (Lopez, 1988b).

Rats were exposed to average concentrations of 100-200, 200-300, 300-400, or 400-500 ppm hydrogen sulfide; at 200-300 ppm, a decreased response rate in a discriminated avoidance task was observed (Higuchi and Fukamachi, 1977). Except at the highest concentrations tested, the response rates and percent avoidances recovered rapidly when ventilation with clean air was provided, although even at 400-500 ppm, they were almost normal the following day. When these same animals were tested for Sidman-type conditioned avoidance response at response-shock intervals of 10 or 30 seconds, an inverse relationship between hydrogen sulfide concentration and response rate was noted; this effect dissipated when exposure stopped (Higuchi and Fukamachi 1977). Excitement was observed when mice were exposed to 100 ppm of hydrogen sulfide for 2 hours at 4-day intervals (Savolainen, 1980). Exposure also resulted in decreased cerebral ribonucleic acid (RNA), decreased orotic acid incorporation into the RNA fraction, and inhibition of cytochrome oxidase. An increase in the glial enzyme marker, 2',3'-cyclic nucleotide-3'-phosphohydrolase, was seen. Neurochemical effects have been reported in other studies. Decreased leucine uptake and acid proteinase activity in the brain were observed in mice exposed to 100 ppm hydrogen sulfide for 2 hours (Elovaara, 1978). Inhibition of brain cytochrome oxidase and a decrease in orotic acid uptake were observed in mice exposed to 100 ppm hydrogen sulfide for up to 4 days (Savolainen, 1980).

The intermediate-duration effects of hydrogen sulfide on neurological function were examined by the measurement of motor and sensory nerve conduction velocities of the tail nerve or morphology of the sciatic nerve but, no neurotoxic effects were observed in rats exposed to 50 ppm hydrogen sulfide for 5 days a week, for 25 weeks (Gagnaire, 1986).

Neurologic function and neuropathology were evaluated in rats exposed to 0, 10.1, 30.5, or 80.0 ppm hydrogen sulfide for 6 hours/day, 5 days/week, for 90 days (CIIT, 1983c). Although absolute brain weights were decreased (5%) in rats exposed to 80 ppm hydrogen sulfide in this study, there were no treatment-related effects on neurological function or neuropathology. In addition, no signs of neurotoxicity were noted in a similar study in which mice and rats were exposed to 0, 10.1, 30.5, or 80.0 ppm hydrogen sulfide for 90 days (CIIT, 1983a, CIIT, 1983b).

**6.1.3 Toxicology of bioaerosols**

**Endotoxin**

The bioaerosol constituent present in swine barns that has been most studied is endotoxin. Endotoxin is a lipopolysaccharide (LPS) component of the outer cell wall of Gram negative (Gm-) bacteria. Endotoxin has been shown in both humans (Schwartz et al 1995, Jagielo et al 1996, Deetz
et al 1997) and animals (Schwartz et al 1994, Jagielo et al 1996, Thorne et al 1998, Thorne 2000) to be a potent pro-inflammatory agent through its ability to activate the innate immune system. Endotoxin is an amphipathic molecule consisting of a phospholipid fraction, called lipid A, bound to a polysaccharide. The polysaccharide has two components: the O-antigen and the core polysaccharide (Rietschel et al 1996). In swine CAFOs, endotoxin most likely includes pieces of other membrane materials in association with LPS. The biological activity of endotoxin rests largely with the lipid A fraction. Once inhaled, endotoxin will interact with macrophages or soluble CD14 inducing signal transduction via the TLR-4 receptor (Medzhitov et al 1997, Faure et al 2000, Gao et al 1998). Through multiple transcription factors (Gao et al 1998), the initiation of transcription of several genes coding for inflammatory mediators can trigger the production of pro-inflammatory cytokines. The cytokines most associated with inhalation of endotoxin are Interleukin (IL)-1, tumor necrosis factor (TNF)a, IL-6, IL-8 (humans), and MIP-2 (mice) (Thorne et al 1998, Deetz et al 1997). Recent evidence suggests a regulatory role for IL-10, IL-12 (Shnyra et al 1998), and interferon ? (IFN?) (Kline et al 1998). An aggressive response to endotoxin exposure results in a cascade of events producing airway narrowing and an obstructive breathing pattern (Pauwels et al 1990). Chronic inhalation exposure in mice has been shown to induce airway remodeling and collagen formation (George et al 2001).

**Glucans**

Studies of the past five years have provided evidence that glucans may also be important immunomodulators (Rylander 1999, Fogelmark et al 1997). ß(1 ? 3)-glucans are glucose polymers with variable molecular weight and degree of branching that may appear in triple helix, single helix or random coil structures (Williams 1994). ß(1 ? 3)-glucans originate from a variety of sources, including fungi, bacteria, and plants (Stone and Clarke 1992). They are water insoluble structural cell wall components of these organisms, but may also be found in extracellular secretions of microbial origin. Glucans may account for up to 60% of the dry weight of the cell wall of fungi, of which the major part is ß(1 ? 3)-glucan (Klis 1994). Recently it has been suggested that ß(1 ? 3)-glucans play a role in bioaerosol induced inflammatory responses and resulting respiratory symptoms (Williams 1994, Rylander et al 1992, Fogelmark et al 1994).

**Microorganisms**

Infectious microorganisms may present an occupational hazard when inhaled (Thorne 2001, Douwes et al 2002). Fortunately, airborne transmission of zoonotic pathogens at sufficient doses to cause disease appears to be uncommon in CAFOs. The most notable infectious bioaerosol in agricultural occupational environments is *Mycobacterium tuberculosis* (Schenker et al. 1998). However, this arises from transmission from person-to-person. Tuberculosis occurs with high prevalence among immigrant farm laborers. More germane to CAFOs in Iowa is concern over the emergence of antibiotic resistant pathogenic organisms that may arise under the influence of antibiotics added to feed.

Non-infectious microorganisms are a more significant problem in CAFOs by virtue of the enormously high concentrations at which they occur. There has been limited study of the effects of inhaled bacteria and fungi in laboratory animal models of human disease. Most of the studies in the literature have used a lung infection model to study host defense against lung pathogens or to assess the efficacy of antimicrobial therapies. However, a few studies are informative. McCray et al (1999) demonstrated severe inflammation with neutrophilic infiltration to the lungs of mice following 4 hr inhalation exposure to *Pseudomonas aeruginosa* at a concentration of 3.3 x 10^8 CFU/m³. This study
used bacterial lung burdens that resemble those attainable in CAFOs. The bacteria were cleared from the lungs within 24 hours and the inflammation resolved by 72 hours after exposure. Thorne and Gassman studied the relative potency of inhaled Gram-negative organisms and Gram-positive organisms for lung inflammation in mice (Gassman et al 2000). This study demonstrated that the Gram-negative bacteria: *Enterobacter agglomerans* and *Pseudomonas aeruginosa* were orders of magnitude more potent that the Gram-positive organisms: *Bacillus magaterium* and *Micrococcus luteus* at initiating inflammation. In this study, markers of inflammation included influx of neutrophils to the lung and increased concentration of interleukin-6 (IL-6) and tumor necrosis factor (TNFa). It was concluded that the endotoxin derived from the Gram-negative organisms was the cell component primarily responsible for the inflammation.

Fungi and fungal conidia are also found airborne in CAFOs. Fungi have been studied primarily as allergens and as sources of mycotoxins. There is no reported evidence of animal or human health problems due to mycotoxin delivery arising from inhalation of fungal spores for the common fungi found in CAFOs. Studies of allergen potency for fungi found in CAFOs have focused on human studies rather than on animal models.

### References - Overview (Osweiler)


### References - Toxicology of ammonia (Carson)


References -Toxicology of hydrogen sulfide (Carson)

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