

ACUTE INTERSTITIAL PNEUMONIA IN FEEDLOT CATTLE

by

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## Abstract

Acute Interstitial Pneumonia (AIP) is a costly issue that affects feedlot cattle, especially during hot and dry summers. Research has yet to elucidate the exact etiology of AIP; therefore this study was conducted to determine possible factors that contribute to AIP in feedlot cattle. During the summer of 2011 in a 55,000 head feedyard in southwest Kansas, animals exhibiting clinical signs of AIP were selected for ante-mortem examination and data collection. The animal population within the feedlot consisted of 75% heifers and 25% steers. Approximately 50% of the animal population was black hided animals. Ante-mortem data consisted of rumen gas cap measurement for NH<sub>3</sub> and H<sub>2</sub>S, rumen pH, serum chemistry, rectal temperature, and body weight. Post-mortem cases with and without ante-mortem evaluations were also selected for an additional examination. Postmortem examination included similar data as ante-mortem examination with addition of visual and histological examination of lung tissue. There were 31 ante-mortem cases of clinical AIP with the following observations (mean ± SD): rectal temperature 105.3 ± 0.7 °F, weight 1098 ± 123 lbs., H<sub>2</sub>S 136 ± 133.3 ppm, and rumen pH 6.4 ± 0.5. Twenty-five healthy cohorts were selected from identical pens to serve as controls. Observations from control animals were: rectal temperature 103.7 ± 1.1 °F, weight 1113 ± 133.3lbs, H<sub>2</sub>S 269.8 ± 311.6 ppm and rumen pH 6.2 ± 0.6. A total of 61 post-mortem cases with a preliminary diagnosis of AIP were analyzed and displayed the following values: H<sub>2</sub>S 1279.7 ± 1569 ppm, and rumen pH 6.3 ± 0.36. Fifty-three of these postmortem cases had diffuse, focal and /or patchy AIP confirmed by histology.

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## **Dedication**

I dedicate this thesis to my family, who has always been there to support me as I attained an education. Accomplishment of this goal has made me the first person of all generations within both of my families to obtain both a bachelor's degree and a master's degree. I would also like to dedicate this to my special fiancé, Areli, who has been by my side through this entire journey. Areli, was one of the few people who believed in me after graduating from junior college and she did her share to convince me to pursue a higher education at Kansas State University.

# **Chapter 1 - Literature Review**

## **Introduction**

In Europe, atypical interstitial pneumonia has been observed for over 200 years and referred to by other informal descriptive terms such as dust pneumonia, fog fever, lungers, panthers and bovine asthma<sup>7</sup>. In 1962 the more relevant and descriptive name of “atypical interstitial pneumonia” was originally presented by D. C. Blood<sup>3</sup>. The term atypical was assigned to this disease for two reasons first, to better illustrate the unusual cellular reaction occurring within the lungs of the affected animals and secondly to take into account the poor response of treatments allocated to suspect or affected animals<sup>7</sup>. Currently the most creditable etiology of acute interstitial pneumonia (AIP) is that related cattle on pasture, although these findings are not accountable for the cause of AIP in feedlot cattle<sup>16, 27</sup>. Etiology of AIP in feedlot cattle still remains unknown and death is usually the result in affected cattle regardless of the types of treatment methods applied<sup>15, 25, 26</sup>.

## **Acute Interstitial Pneumonia**

Acute interstitial pneumonia has been known to be the cause of histologic lesions within cattle placed on feed<sup>1</sup>. The term AIP is also descriptive for the formation of hyaline membranes, emphysema and edema accumulation within the lungs of the affected animal<sup>1, 7, 14</sup>. Acute interstitial pneumonia has continued to cause tremendous issues within feedlots, especially during hot and dry summers. As bovine respiratory disease complex (BRDC) continues to be number one cause of morbidity and mortality in feedlot environments across the country, AIP has been categorized as the second most important disease in the feedlot industry<sup>1, 16, 25, 26</sup>. In the year 2000 the National Animal Health Monitoring System (NAHMS) distributed a survey to

all the feedlots within the nation to evaluate the industry's morbidity and mortality specifics.

Personnel of all participating feedlots reported in the survey that 3.1% of all cattle placed in a feedlot environment tend to develop acute interstitial pneumonia.

This particular type of pneumonia has been known to affect cattle placed on feed in a very sporadic manner<sup>2, 15</sup>. Clinical symptoms of AIP have been observed only days after animals experience any dietary changes<sup>7</sup>. Others studies have revealed that the effect of AIP in feedlot cattle is very rapid and in some occasions cattle may die within only a period of 24 hours after the onset of the disease<sup>3</sup>. Common clinical signs found in affected cattle may include dyspnea, excessive salivation (frothing at the mouth), swayback, elongated neck, bowed front legs, grunting and in possible increased body temperature<sup>3, 7</sup>. If familiar with auscultation, listening to the lungs carefully may also be a helpful method to identify affected animals. Listening to different types of sounds by approaching the animal from different sides can indicate certain lesions in the lungs. For example, loud bronchial tones from the ventral side are a sign of consolidation, inconsistency of sounds of inhalation from the dorsal side indicate severe and the sound of rubs and dry rales from the dorsal side indicate emphysema<sup>3, 6</sup>. An ante-mortem diagnosis of AIP may be challenging as it can be difficult to differentiate this particular disease from other respiratory issues if only these clinical signs are taken into account<sup>2</sup>. As mentioned before, the term "atypical" was selected to identify and/or describe the cellular reactions and lesions that appeared in AIP affected animals; therefore, the best way to determine an animal AIP positive is by gross and microscopic pathological findings at necropsy<sup>2, 7</sup>. During the post-mortem examination, findings may include heavy and enlarged lungs, pulmonary edema and emphysema and hyaline membranes<sup>2, 7, 26</sup>.

## **Factors Associated with AIP**

In the year 2000 a study performed by Ayroud et al demonstrated that in feedlot environments heifers on average were more likely to be affected by AIP than steers. Later Loneragan et al confirmed that certainly heifers did have a higher probability to be affected by this specific type of pneumonia. In fact during that exact study, Loneragan et al concluded that the number of affected animals was in a 3:1ratio heifers to steers respectively <sup>15</sup>.

#### *Melengestrol Acetate (MGA)*

These findings are one of the reasons why some researchers have questioned the use of melengestrol acetate (MGA) to be a contributing factor to the cause of AIP in animals placed under feedlot diets. A study performed in sheep showed that after an oral administration of 3-methylindole (3MI), MGA increased the probability of pulmonary edema in the animals <sup>21</sup>. Since this research was done on sheep further investigations are recommended to find any correlation between these findings and feedlot heifers. Stanford et al found that feeding MGA to feedlot heifers significantly increased the number of animals affected by AIP <sup>23</sup>. Animals were confirmed AIP positive by performing AIP-related emergency slaughter. According to their findings, Stanford et al claim that avoiding the involvement of MGA in the heifers' feed diets should be positive step for the feedlot industry <sup>23</sup>.

#### *Implants*

Implants and implant techniques have been questionable factors that are suspected to contribute to the cause or development of AIP in feedlot environments <sup>17</sup>. Even though these types of management factors have been proposed to be related with AIP, research currently lacks of further investigation and testing of this specific subject <sup>25</sup>. Evaluation of variables and strategies of this type may be difficult to accomplish since AIP affects cattle in a sporadic manner.

### *Late Feeding Periods*

Days on feed plays a tremendous role as a factor that is also involved in making this particular disease a more cost effective issue for the feedlot industry. Cattle on feed tend to become affected by AIP close to their market weight in most cases about 15 to 45 days from their projected harvest date <sup>2,27</sup>. In another study performed in western United States 14 participating feedlots presented data for 108 animals that had died consequently of AIP. These 108 animals averaged 127 days on feed before death had occurred <sup>16</sup>. Also Woolums et al reported that in one of their studies cattle subject of AIP had died after being placed on feed for at least 45 days <sup>27</sup>. In 2005 a nationwide survey to analyze management practices among feedlot was released. Only 65 feedlots responded with a completed survey. Managers from the responding feedlots reported that 80.5% of their deaths as a result of AIP occurred in cattle placed on feed for 60 days or more <sup>28</sup>.

### *Moldy Sweet Potatoes*

In previous studies it has been reported that the feeding of moldy sweet potatoes has been suspected to cause acute interstitial pneumonia. In one particular study conducted in Tift County, GA there were a total of 69 of 275 Hereford cattle died as result of fed moldy sweet potatoes <sup>20</sup>. The heard was on a 6 to 8 inch tall Bermuda grass pasture with the option of feeding cull sweet potatoes provided from a curing shed. Mortality of the 69 animals occurred during the months of April, May and June. During the month of April 6 cow died and but only one animal was submitted to examination. In early May 2 more cows were found dead and both submitted to examinations, at this time the rest of the heard had no sign of sickness. On June 5<sup>th</sup> about 200 bushels of damaged moldy sweet potatoes were placed within the pasture for the cattle to consume. On the day of June 8<sup>th</sup> a total of 18 animals were found and other animals expressed

signs of illness. Later within the next 4 day period an additional 30 animals were found dead to equal a total of 61 animals dead during the same week. Of all 69 deads only four cattle were submitted for further evaluation. Findings at the gross level confirmed firm and large lungs with present interstitial and interlobular emphysema on all the lobes of the lungs on the three of the four animals. Microscopic evaluation of two animals revealed the presence of edema, formation of hyaline membranes, and interstitial emphysema. Samples of the sweet potatoes that were fed to the cattle were submitted for evaluation and discovered more than 150 fungal isolants present. Among all the isolants three different types of Fusarium were recognized. The researchers tried to reproduce the disease by performing different experimental designs with what was thought health animals, but did not any comparable results. Finally, a batch of sweet potatoes was purposely damaged following steps that were thought to be similar to those followed with the 200 bushels what caused the 61 deaths. The purposely damaged batch was also infested with Fusarium Solani. The sweet potatoes were then fed to 16 cattle and resulted the death of three animals. Gross and microscopic lesions found in these animals were similar to those found on the previous four animals sampled from the 61 deads. In the reproduction of the disease through different experimental methods the morphological species Fusarium Solani became the main suspect to cause the disease.

In a study conducted in Northeastern Brazil about 400kg of sweet potatoes were fed to 18 milking cows, 1 steer and 1 bull.

#### *Time of Occurrence*

Acute interstitial pneumonia has been found to affect more animals during hot and dry summer and fall seasons <sup>2, 16</sup>. In a survey study conducted by Woolums et al managers from the responding feedlots reported that 62% of their AIP deaths occurred during the summer season.

The percentage of feedlots affected by the hot temperatures of the summer may fluctuate depending on the location of the feedlot. Woolums et al also reported that feedlots located in the northern part of the United States had a lower possibility to report morbidity and mortality as a result of AIP<sup>28</sup>. Considering the ambient temperature and the location of the feedlots may have a correlation with the number of cattle affected by AIP during this time of the year as typically the northern part of the nation may present lower temperatures in comparison to the southern states.

### *3-Methylindole*

Acute interstitial pneumonia is known to be associated with the movement of grazing cattle from poor quality pastures to lush pastures. When a change in pasture of this type occurs L-tryptophan is metabolized to 3-methylindole (3MI) by lactobacillus microbes in the rumen<sup>29</sup>. L-tryptophan is an essential amino acid regularly found in feed diets. 3-mythelindole becomes absorbed through the ruminal and intestinal walls to enter the body's circulation system<sup>23</sup>. Once in the lungs, this specific agent becomes activated by Clara cells found in the bronchioles leading to the cause of AIP like lesions and pulmonary edema and emphysema in cattle<sup>4,9</sup>. Further observations of this agent have been performed to discover that a metabolite derivative of 3MI known as 3-methylindolenine (3MEIN) is the responsible compound causative of certain pulmonary lesions<sup>24</sup>.

In a study conducted in southern Alberta feedlots, 3-MEIN concentrations in the plasma and 3-MI concentrations in the urine were compared among AIP suspected cattle and a control group<sup>2</sup>. The AIP suspect group consisted of a total of 38 animals. All 38 animals were submitted for emergency slaughter due to their health state<sup>2</sup>. Thirty-one of the 38 animals resulted AIP positive by confirmation upon gross and histological evaluations of lung pathology.

All animals that tested positive for AIP were heifers. The control group consisted of the 7 remaining animals that tested negative for AIP and 17 additional pen mates. After evaluating the results of the samples collected, researchers concluded that animals with AIP had higher 3-MEIN plasma concentrations than animals without AIP. Animals with AIP also had lower 3-MI concentrations excreted in the urine compared to animals not affected by AIP. However, 3-MI concentrations in lung tissue did not differ between the AIP and the control groups. In another study 3-MEIN adducts concentrations in lung tissues within the AIP and the control groups were detected and recorded <sup>15</sup>. In this case animals that were confirmed AIP positive upon pulmonary tissue histology had higher 3-MEIN adduct concentrations in lung tissues than animals that had died of others causes. Researchers also found that cattle affected by AIP also had higher 3-MEIN adduct concentrations in the blood in comparison to their healthy pen mates.

#### *Bovine Respiratory Syncytial Virus*

Bovine respiratory syncytial virus (BRSV) is another factor that is thought to contribute to the cause of AIP. A particular study was performed to compare the proportions of BRSV infections present in the lungs of animals that had died of AIP and animals that had died of other causes non-related to AIP. In this study the researchers concluded that animals affected by AIP had greater proportions of pulmonary BRSV infections present than the animals that had died of other causes <sup>5</sup>.

#### *Bronchopneumonia*

Bronchopneumonia (BP) is another respiratory condition known to be strongly implicated with AIP in feedlot settings. In two previous studies focused in the clinical management and the concurrent lesions of respiratory disease in feedlot animals found that a significant amount of animals with AIP also had BP present. During the first study of the total 149 animals affected by

AIP, 144 animals had BP present confirmed by histopathology of pulmonary tissue <sup>10</sup>. This means that about 97% of the cattle were affected by both diseases which maybe encouraging to conduct further research to find if there are any possible correlations between AIP and BP. It would be interesting to find which of these two diseases is more prone to be present first within the cows pulmonary system and under what circumstances. Possibly BP could lead to or influence the cause of AIP or vice versa in feedlot cattle. During the second study investigators reported that out of 28 animals that had died from AIP 21 of them also had BP lesions present <sup>22</sup>. Once again the majority of the animals within a study of this type had both AIP and BP which I insist that there may be possible correlations between diseases.

### *Management Practices*

### **Economics**

As mentioned before acute interstitial pneumonia stands as the second leading cause of morbidity and mortality of cattle in the feedlot industry. Most costs attributed to this disease are direct and indirect. Costs of affected animals can relate to many different things such as: cost of animal when bought from producer, shipping costs, vaccination/conditioning, feed and treatment possibly applied. Once in the feedlot the value of the animals becomes amplified on a daily basis. Therefore, when an animal dies an investment by the feedlot is completely lost. In the past investigators have referred to the number of mortalities as a result of AIP as a percentage of all cattle placed. The percentage of AIP mortality ranged from 0.03 to 0.15% in different studies <sup>10</sup>. In a particular study, participating feedlots reported that about 10.4% of their mortalities were caused by AIP <sup>25</sup>. Taking into consideration this percentage of mortalities, these feedlots are facing significant losses. In the year 2000 the National Animal Health Monitoring System (NAHMS) distributed a survey to all the feedlots within the nation to evaluate the industry's

morbidity and mortality causes. Personnel of all participating feedlots reported in the survey that 3.1% of all cattle placed in a feedlot environment tend to develop acute interstitial pneumonia<sup>18</sup>.

Occasionally, producers resort to emergency slaughter of affected cattle to avoid complete loss. Producers who face the problems of AIP within their production are very familiar with the costs that may affect their budgets. Therefore, emergency slaughter may conserve part of the investment attributed to the affected animals. In some cases emergency slaughter at the farm or production facility other than a slaughter house can still have associated losses of up to \$500 per animal<sup>8</sup>. If affected animals are detected in an early stage of AIP they can still be sent to slaughter following the standard guidelines<sup>23</sup>. Early detection of these animals can possibly reduce costs related to onsite emergency slaughter.

### **Treatments**

Currently, there is no official or conventional set of treatments or prevention protocols to be utilized among affected animals. The discovery of treatments or preventions for AIP has been difficult to find due to the complexity of the disease<sup>23</sup>. Since there are multiple pathways and factors that lead to the cause of AIP, the difficulty level of finding or creating treatment or prevention protocols is affected significantly.

## **Chapter 2 - Acute Interstitial Pneumonia in Feedlot Cattle**

### **Introduction**

#### ***Nature of Problem***

Acute Interstitial Pneumonia (AIP) is a respiratory disease that affects cattle. Acute interstitial pneumonia is also known as atypical interstitial pneumonia, fog fever, and dust pneumonia<sup>7</sup>. Cattle affected by the disease present dyspnea, panting, and death. At necropsy, lungs grossly display edema and emphysema<sup>7</sup>. AIP has been speculated to be the second most cost effective respiratory disease among cattle on feed<sup>1</sup>. In 2000 the National Animal Health Monitoring System reported that 78.4% of all feedlots had at least one placement develop AIP<sup>19</sup>. Acute interstitial pneumonia occurs sporadically in feedlot cattle, especially during dry and hot weather patterns<sup>25</sup>. This type of pneumonia is known to exist among cattle that are near to their market weight which results in large financial losses<sup>16</sup>. A survey of U.S. feedlot personnel estimated that 3.1% of the total cattle placed on feed develop AIP<sup>18</sup>. Research has yet to elucidate the exact etiology of AIP; therefore this study was conducted to determine possible factors that lead to the cause of AIP in feedlot cattle.

### **Materials and Methods**

#### ***Study Design***

A case-control study was performed to evaluate possible factors contributing to AIP in feedlot production settings. The study was approved by the Institutional Animal Care and Use Committee at Kansas State University. Evaluations included measurement of concentrations of ammonia and hydrogen sulfide in the rumen gas cap, and serum chemistry analysis focusing on

amylase and lipase values. Lung tissue samples were obtained for histological evaluation from animals with clinical and/or post-mortem findings suggestive of AIP.

Variation in feed consumption was compared in a case control approach among pens with at least one case of AIP vs. selected control pens with no reported cases of AIP within the last 15 days. A second comparison in feed variation among AIP vs. selected control pens was made using a 5-day rolling average compared to the 6<sup>th</sup> day feed consumption. For example the average of days 10-14 vs. day 15, days 9-13 vs. day 14, days 8-12 vs. day 13 and so forth.

The ambient temperature at the feedlot location was automatically recorded by a central station and a weather censor located on site. Collection of samples for the study was conducted from June 10 to August 10, 2011. At the time of sample collection, a demographic form was completed which included the date of sample collection, suspected cause of death, sex, lot and tag number, and hide color. All sample collections were performed by a Kansas State University graduate student and properly trained personnel from the hospital crew.

### ***Study Population***

The study was conducted in a feedlot with a capacity of approximately 55,000 head in southwest Kansas. Animal population demographics consisted of 75% heifers and 50% black-hided animals. Average body weight at arrival of cattle within the participating feedlot was 778.5 lbs. At initiation of the study the cattle ranged from 3 to 166 days on feed with an average of 64.3 DOF. There were 3 diet changes during the time of the study (Table 3). Changes in diet maintained the same ingredients but at different percentages.

### ***Selection Criteria and Sample Collection***

#### ***Ante-Mortem***

Animals exhibiting clinical signs of AIP were removed from their pen and taken to the hospital facilities for examination. In some cases samples were collected in the home pen if the animal was unable to rise. Clinical signs suggestive of AIP included extension of the neck, panting, excessive salivation, breathing through the mouth and refusal to travel<sup>3,7</sup>. Affected animals also may express aggressive attitudes when being approached for handling<sup>13</sup>. Animals diagnosed with AIP (case) were paired with a healthy control animal of comparable physical characteristics from the same pen. All ante-mortem samples were obtained from both the AIP suspect and control animal sequentially.

Rumen gas-cap samples were collected by a rumen puncturing technique<sup>12</sup>. The animal was restrained in a squeeze chute and the left paralumbar fossa region was cleaned with a solution of 2% chlorhexidine. Before puncturing the rumen, the needle was attached to an air-tight, clear, rubber tube which was connected to the appropriate glass measurement tube (Gastec detector tubes)<sup>A</sup> which was in turn attached to a 100 ml capacity Gastec pump. A 12-gauge needle was introduced into the rumen. A total of 200 ml of rumen gas was collected for measurement of NH<sub>3</sub> and H<sub>2</sub>S (100 ml per gas measured).

Rumen ammonia and rumen hydrogen sulfide levels were determined using two Gastec detector tubes with different sizes in scale range were used for each gas. For the ammonia samples, the first determination was taken using a Gastec tube <sup>A</sup>NO.3L which measured the presence of NH<sub>3</sub> in a scale range of 0.5 – 30 ppm. If the amount of NH<sub>3</sub> present surpassed the scale range of the <sup>A</sup>NO.3L tube, a second gas sample of 100 ml was taken substituting the <sup>A</sup>NO.3L tube with a <sup>B</sup>NO.3M tube that measured NH<sub>3</sub> amounts of 50 – 500 ppm. The same procedure was followed for the hydrogen sulfide samples. H<sub>2</sub>S samples were initiated using a <sup>C</sup>NO.4H tube which had a measurement scale range of 100 – 2,000 ppm. For occasions in which

the amounts of H<sub>2</sub>S present surpassed the scale range of the <sup>C</sup>NO.4H tube, a second 100 ml sample was taken using a <sup>D</sup>NO.4HH tube of 0.1 – 2.0% scale range.

Rumen fluid was obtained following a rumen sampling procedure similar to the technique used to collect the rumen gas samples. A 12-gauge needle was introduced into the rumen on the left side of the animal just caudal to the 13<sup>th</sup> rib at the level of the point of the elbow. At least 6 mL of rumen fluid was obtained to measure the rumen pH using an Eco tester ph2 meter.

Approximately 12 mL of blood was obtained from the jugular vein and divided into two aliquots of 6 mL each and deposited into serum separator tubes (Kendall Healthcare Monoject) and refrigerated immediately. The samples were centrifuged on site and continuously refrigerated before taken to the clinic in charge of performing the analysis for amylase and lipase concentrations. An amylase and lipase panel was utilized for evaluation using IDEXX equipment (IDEXX Westbrook, Maine).

Rectal temperature was determined using an electronic digital thermometer (GLA M700). Body weight was recorded in the hospital chute for those animals receiving an ante-mortem examination. The weights for the animals receiving a post-mortem examination but without an ante-mortem examination that died in their home pen were estimated using their home pen's weight average.

#### ***Post-Mortem***

Throughout the study, necropsies were performed by experienced personnel trained by the feedyard's consulting veterinarian. Only those animals suspected of AIP that died early in the morning and showed no signs of physical decomposition were examined. Post-mortem examination was performed on suspected AIP mortalities regardless of whether they had received an ante-mortem examination and were entered into the study population.

Rumen gas-cap samples were obtained at necropsy by following the same technique performed during the ante-mortem examination except that the rumen was exposed. Rumen pH was collected by making a small incision in the rumen wall to insert the pH meter.

### ***Tissue Samples***

Lung tissue samples were obtained by only one individual for consistency. Samples were obtained from different lobes of each side (four total for each animal). If consolidated tissue was present, samples were collected preferably from those lobes with lines of demarcation between consolidated and non-consolidated tissue. Tissues were sectioned in blocks of similar dimensions (approximately 2.50 in. length x 0.75 in. width x 0.75 in. depth). Each sample was individually placed in a Whirl-Pak® bag containing formalin, labeled with the lot number, animal ID and lobe location. All four bags containing the samples were then placed in a larger re-sealable zipper storage bag to keep them grouped by animal. Formalin fixed-samples were stored in a room with no sunlight and at a temperature of approximately 70° F. Samples were shipped on a weekly basis to the Arizona Veterinary Diagnostic Laboratory at the University of Arizona for histopathology analysis.

Animals were classified AIP positive by presenting lesions confirmed by histology in at least one lobe. Presence of bronchopneumonia, bronchiolitis, and interstitial pneumonia lesions were also evaluated during the histopathology exams. Histology confirmed the presence of AIP, bronchiolitis and bronchopneumonia in post-mortem cases. Presence of AIP was classified as three different patterns; diffuse, focal and patchy. Bronchiolitis lesions were also identified as chronic or acute. Histopathological lesions were defined as focal AIP (less than 10% of lobule affected), patchy AIP (more than 10% but less than 50% of the lobule affected), diffuse AIP (more than 50% of the lobule affected), bronchopneumonia (focal or diffuse acute/chronic

bronchopneumonia), chronic bronchopneumonia (bronchopneumonia plus evidence of chronicity such as fibrosis or atelectasis), interstitial pneumonia (interstitial pneumonia lacking lesions described for AIP), and AIP (interstitial pneumonia with alveolar fibrin, hyaline membrane formation, and pneumocyte hyperplasia).

### ***Feed Consumption***

Feed consumption data were obtained from the feedlot's software program (Cattle Management Software (CMS)), which were recorded daily by the feed mill personnel. Feed intake variations were calculated based on the dry matter consumption per animal and compared on a pen-to-pen basis. Pens with at least one post-mortem case diagnosed with AIP (n=44) were paired with a pen that had no recorded AIP cases. The control pen was selected based on sex, days on feed, average weight, number of cattle in the pen, and location within the feedlot. Dry matter intake for case and control pens (n=88) were evaluated during the 14 days prior to an AIP incident. For a second comparison, dry matter intakes for the five days prior to an incident were compared individually to the rolling average of the previous five days. For example, the dry matter intake two days before the incident was compared to the average of days 3, 4, 5, 6 and 7 before the incident. For statistical analysis, dry matter intake variations were categorized as the number of pens varying from the previous day or the rolling average by  $\geq 0.25$ , 0.50, or 0.75 lbs.

### ***Temperature and Humidity***

Temperature and humidity were recorded through the duration of the study using a personalized weather station (PWS, model: Davis Vantage PRO 2). High and low daily temperatures were gathered from the weather station for a graphical analysis in comparison with the daily incidence of AIP in cattle.

### ***Statistical analysis***

A  $P$  value  $\leq 0.05$  was selected to establish statistical significance among evaluations.

Probability values  $> 0.05$  but  $\leq 0.1$  were described as values approaching statistical significance.

Ante-mortem observations of rectal temperature, weight, rumen H<sub>2</sub>S, and rumen pH were analyzed using a mixed-effects model analysis of variance with ante-mortem classification as a fixed effect and with sex and pairing as random effects. Analysis was conducted using the Proc Mixed procedure in SAS. This certain analysis was focused on 23 pairs composed of AIP affected animals and controls.

Ante-mortem observations of amylase and lipase demonstrated values with biphasic distributions of zero and positive values. Therefore, amylase and lipase values were evaluated using a Wilcoxon Ranked Sum procedure in SAS in which all values were utilized.

Post-mortem observations of H<sub>2</sub>S and rumen pH were recorded for AIP suspected animals and non-AIP animals. The number of cattle in the various ante and post-mortem examination groups varied. Due to the large differences in numbers and the lack of histological confirmation of control animal lung pathology, these data were reported but not statistically evaluated.

Feed consumption changes were analyzed using the Proc Mixed procedure to compare the pens with AIP cases to control pens without AIP cases. For these comparisons, classification of the pen as having or not having an AIP case was a fixed effect and no random effects were included in the model.

## Results

A total of 31 animals were diagnosed with AIP and received an ante-mortem examination. Eighty-seven percent of these were heifers as compared to an estimated heifer

percentage in the feedlot of 75%. Proportions of hide color for the 31 animals were 74% black, 16% red and 10% white.

Seventeen of the 31 animals presented for ante-mortem examination also received a post-mortem examination. The remaining 14 animals were in an advanced state of decomposition by the time necropsies were performed. In addition, 44 possible AIP cases were only examined post-mortem due to mortality occurring prior to the opportunity for an ante-mortem exam. Twenty-three case controls were examined ante-mortem and 11 post-mortem controls were selected at the time of necropsy, these animals had to be suspected of other types of death causes and not AIP. Post-mortem AIP suspects and controls were paired by the best match of the animal's demographic information (sex, weight, home pen).

### ***Ante-mortem Results***

There were no significant ( $P = 0.99$ ) differences in amylase or lipase concentrations between control animals and AIP suspects. Rumen pH values among the control and the AIP suspect groups approached significance ( $P = 0.06$ , Table 1). Cattle suspected of AIP ( $H_2S 110 \pm 67.7$ ) had numerically lower hydrogen sulfide concentration average than the control group ( $H_2S 272 \pm 59.1$ ) with an approaching significance of  $P = 0.09$ . At approximately day 30 of the study, collection of ammonia samples from the rumen gas cap were ended due to large proportion of non-detectable measurements by the equipment employed in this study. Cattle suspected of AIP had significantly ( $P < 0.0001$ ) greater rectal temperature values ( $105.4 \pm 0.3^\circ\text{F}$ ) compared with the animals classified as controls ( $103.5 \pm 0.03^\circ\text{F}$ ). There were no significant differences in body weight values between the control cattle and the AIP suspect cattle ( $P = 0.22$ ).

### ***Post-mortem Results***

Rumen pH values obtained from the 60 AIP ( $6.3 \pm 0.4$ ) animals were significantly greater ( $P = 0.01$ ) than the values obtained from 11 control ( $5.7 \pm 0.6$ ) animals. Statistical significance was not calculated for the rumen pH between the AIP and control groups due to the lack of lung histology of the control group. Sample acquisition of ammonia was excluded from the post-mortem phase for the same reasons mentioned previously in the ante-mortem section.

### ***Histology***

From the 16 animals with both ante-mortem and post-mortem examinations, 13 (81%) had diffuse, focal and patchy AIP confirmed by histology. Twelve (92%) of the 13 animals with AIP also presented acute and chronic bronchiolitis lesions. The other four animals with no histological AIP lesions also displayed bronchiolitis lesions. From these 16 animals, a total of 10 (63%) had some form of bronchopneumonia with seven also being AIP positive confirmed by histology.

Lung samples from a total of 60 animals were submitted to the laboratory. Diffuse, focal and patchy AIP lesions were present in 45 of the 60 animals. Bronchopneumonia was confirmed to be present in 34 of the 60 animals. Fifty-five of the 60 had lesions consistent with bronchiolitis. Nineteen of the 60 animals had presented interstitial pneumonia lesions.

### **Other results**

#### ***Weather patterns compared to AIP incidence***

High and low daily temperatures were gathered from the weather station for a graphical analysis in comparison with the daily incidence of AIP in cattle. After graphing the temperature and incidence values and reviewing the patterns there was no apparent association evident between temperature changes and the incidence of AIP (Figure 1).

#### ***Feed consumption variation***

Comparisons of feed variations between the pens with at least one AIP incident during the time of the study and their assigned control pen were made. Feed variations were statistically evaluated between the AIP and control groups in  $\geq .25$ , .50 and .75 lbs. daily change. During both 14 (compared to the previous day) and 5 day (compared to the five day rolling average) periods prior to the AIP incidence, the number of changes of  $\geq 0.25$ , 0.5, and 0.75 lbs. per head per day did not differ between pens with and without AIP cases ( $P = .13$  to  $.88$ ). The results are illustrated in Figures 2-7, with statistical results reported in the Figures.

## **Discussion**

Unfortunately, the exact etiology of acute interstitial pneumonia (AIP) in feedlots still remains unknown. Following bovine respiratory disease (BRD), AIP is the second most costly disease confronted by feedlots<sup>1</sup>. In other studies AIP has been observed to be a prevalent cause of death in cattle that are in feeding periods close to slaughter<sup>11, 13</sup>. In one particular study the cattle under observation presented AIP 24 days on average before the expected slaughter date<sup>2</sup>. As described in table 2.1, cattle developed AIP at an average body weight of 1111 lbs. which can be considered close to market weight. A current definite economic effect remains unpredicted. Most of the economic cost comprises the value of feed and treatments invested in the cattle. Cattle that display AIP-like symptoms tend to respond poorly to treatments applied<sup>28</sup>.

Cattle reported as AIP suspects developed clinical signs such as extension of the neck to facilitate breathing, excessive salivation, grunting, panting, breathing through mouth and refusal to travel. These same clinical signs have been described as symptoms of cattle affected by AIP in other studies<sup>3, 7</sup>.

At necropsy observations of gross lesions were made and animals' suspects of AIP presented pulmonary characteristics such as enlarged lungs, dark red in color, rubbery texture

and interstitial edema and emphysema. In comparison with studies performed by Curtis et al, Woolums et al and Doster, animals suspected of being affected with AIP did present similar gross lesions to those reported in the studies previously mentioned<sup>6, 7, 28</sup>.

Histological evaluation of samples submitted demonstrated that bronchiolitis lesions were consistent among more than 90% of the cattle affected with AIP. The presence of bronchopneumonia was also recorded. About 47% of the cattle with AIP confirmed by histology also bronchopneumonia lesions present. The percentage of AIP cattle with bronchopneumonia was relatively lower than those reported in previous studies. In a study of feedlot-associated AIP 97% of the cattle affected with AIP (n=149) also presented evidence of bronchopneumonia (n=144)<sup>10</sup>. In another study 75% of the cattle that had resulted with AIP (n=28) also became affected with bronchopneumonia (n=21)<sup>22</sup>.

Hydrogen sulfide ( $\text{H}_2\text{S}$ ) concentrations were expected to be different between cattle affected by AIP and control animals. Numerically AIP animals seemed to have lower concentrations of  $\text{H}_2\text{S}$  than the control animals. However, the statistical results demonstrated that there was no difference in  $\text{H}_2\text{S}$  concentrations between the two groups.

In previous studies researches have claimed that heifers are more prone to become affected by AIP than steers placed in feedlot environments. In a particular study conducted in southern Alberta feedyards, Ayroud et al confirmed that all the animals suspect of AIP were heifers even though the heifer to steer ratio during the time of the study was 80,000 to 10,000 respectively<sup>2</sup>. Woolums et al also report that in some occasions heifers had died of AIP at higher ratios than steers<sup>27</sup>. During the ante-mortem phase 87% of the animals suspect of AIP were heifers (n=27) and during the post-mortem phase 96% of the cattle with AIP confirmed on histology were heifers. According to our high percentages of heifers affected by the disease we

can say that numerically heifer can become affected by AIP at higher numbers than steers. Though, throughout the time of the study about 75% of the entire population at the feedlot was heifers. According to the differences in populations between heifers and steers we cannot statistically conclude that heifers are more prone to become affected than steers.

AIP has been known to affect cattle more commonly during hot and dry summers and spring time <sup>2, 25</sup>. Others have claimed for AIP to be very common in the fall season as well <sup>9</sup>. This study in particular was only conducted for about 60 days during the months of June, July and August at only one location. Therefore, due to the lack of data for the rest of the months of the year a conclusion cannot be made to agree with previous studies in accordance to AIP being a disease more commonly occurring during the spring, summer and fall seasons.

In a nationwide survey conducted to evaluate the association between management practices and the risk of AIP in feedlot environments managers reported that about 80.5% of all the AIP deaths were from cattle that had been on feed for 60 days or more <sup>28</sup>. In some studies AIP has been reported to affect cattle placed on feed as early as 45 days and continues to affect cattle up to 15 to 45 days prior to expected slaughter date <sup>23, 27</sup>. During the time of the study the days on feed of the cattle affected ranged from 24 to 131 days on feed. A simple graph was made using the affected animals home pen's days on feed. Refer to Figure 2.8 for days on feed of AIP affected pens.

**Table 2.1** – Statistical analysis of amylase and lipase between AIP and Control groups.

Ante-Mortem Examination						Amylase	Lipase	
		Rectal Temp (°F)	Weight (lbs)	Rumen H2S (ppm)	Rumen pH	N of Serum Chemistry	Amylase U/L	Lipase U/L
AIP (30)	Mean	105.4	1111.1	110.0	6.2	30	8.9	11.1
	SEM	0.30	57.5	67.7	0.3		2.3	5.8
Control (25)	Mean	103.5	1148.0	272.6	5.9	25	8.9	11.2
	SEM	0.30	57.5	59.1	0.3		2.4	5.8
P-Value		<0.0001	0.2233	0.0921	0.0588		0.99	0.99

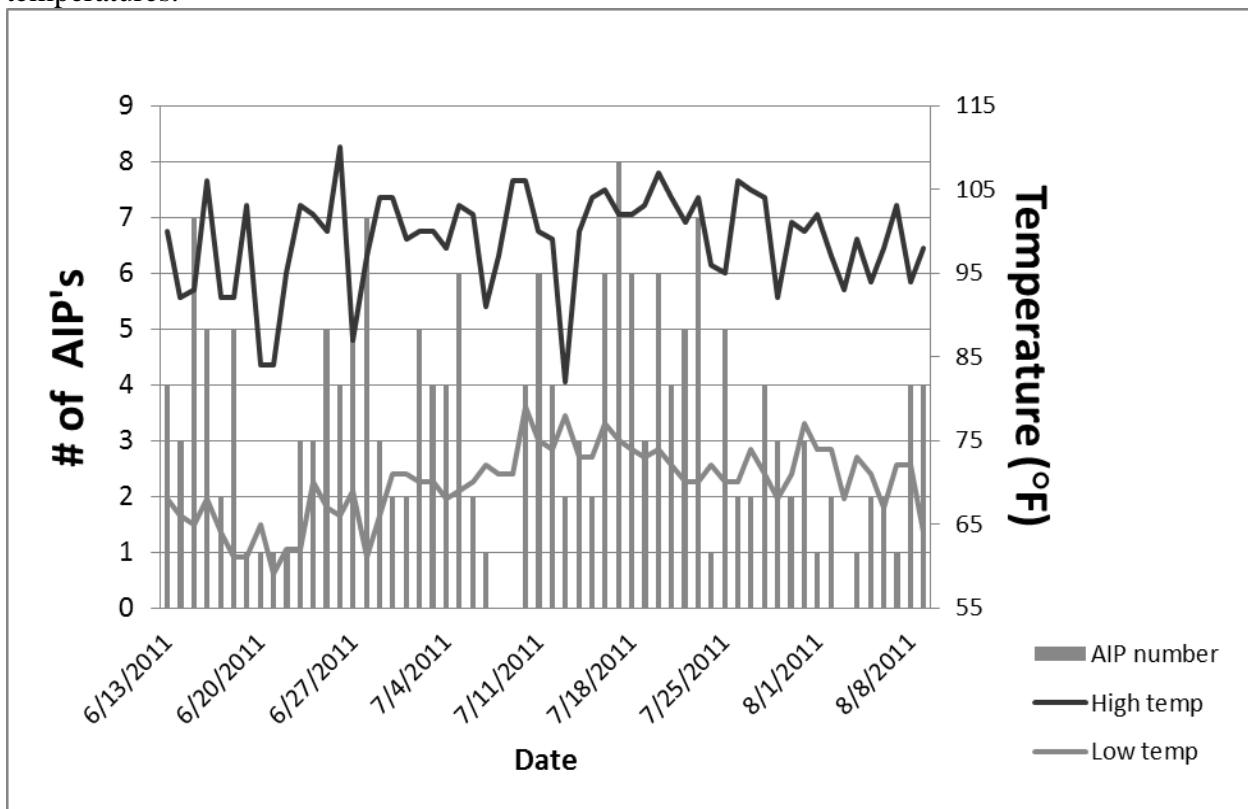
**Table 2.2** – Numerical evaluation of H<sub>2</sub>S and rumen pH between AIP and Control groups.

Post-Mortem Examination		H2S	Rumen pH
AIP (60)	Mean	1279.7	6.30
	SEM	232.3	0.05
Control (11)	Mean	1841.7	5.70
	SEM	363.9	0.17
<i>P</i> -Values		0.57	<0.01

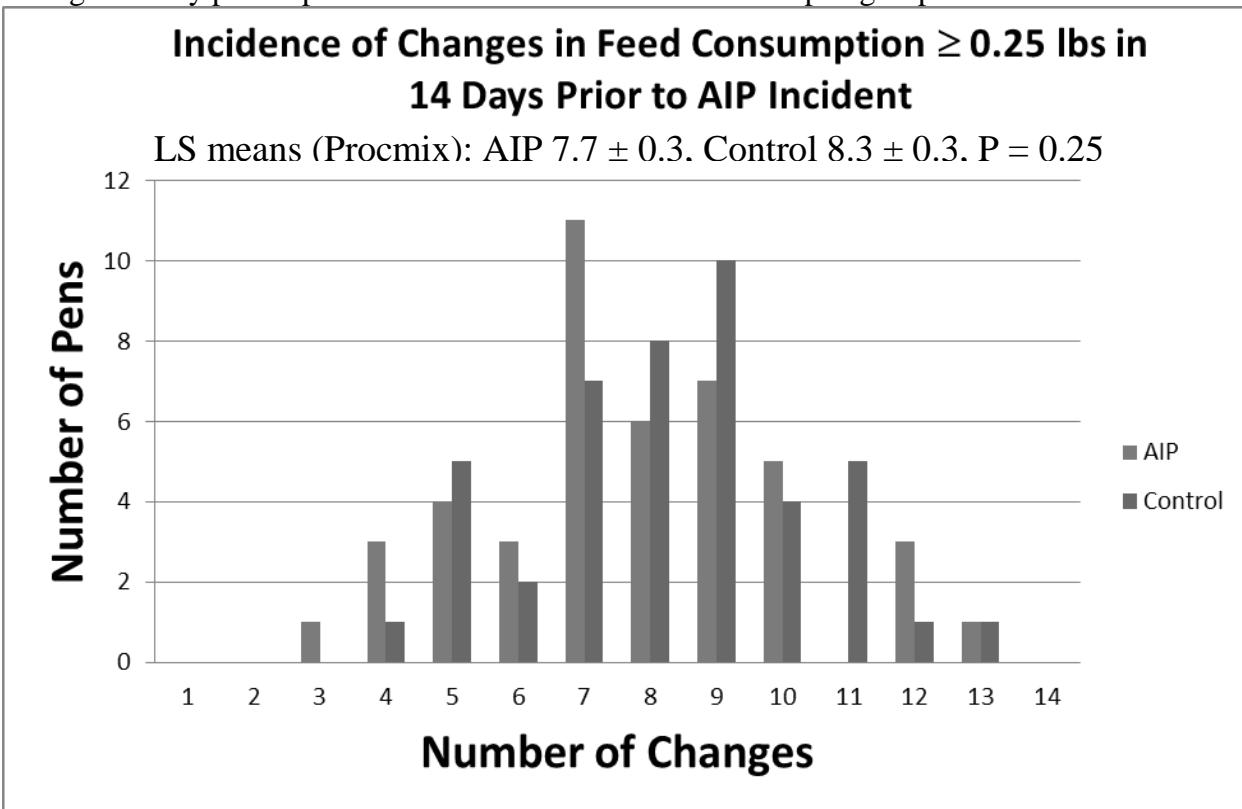
**Table 2.3** - Ingredients of diet ration changes during the time of the study.

	<b>5/1/2011</b>	<b>7/14/2011</b>	<b>7/23/2011</b>
<b>Flaked Corn</b>	44.3%	43.9%	43.5%
<b>High Moisture Corn</b>	20.0%	20.0%	20.0%
<b>Wet Distillers Grain</b>	22.0%	22.0%	22.0%
<b>Dried Distillers Grain</b>	3.7%	3.7%	3.7%
<b>Liquid Supplement</b>	5.3%	5.3%	5.3%
<b>Corn Stalks</b>	4.7%	5.1%	5.5%

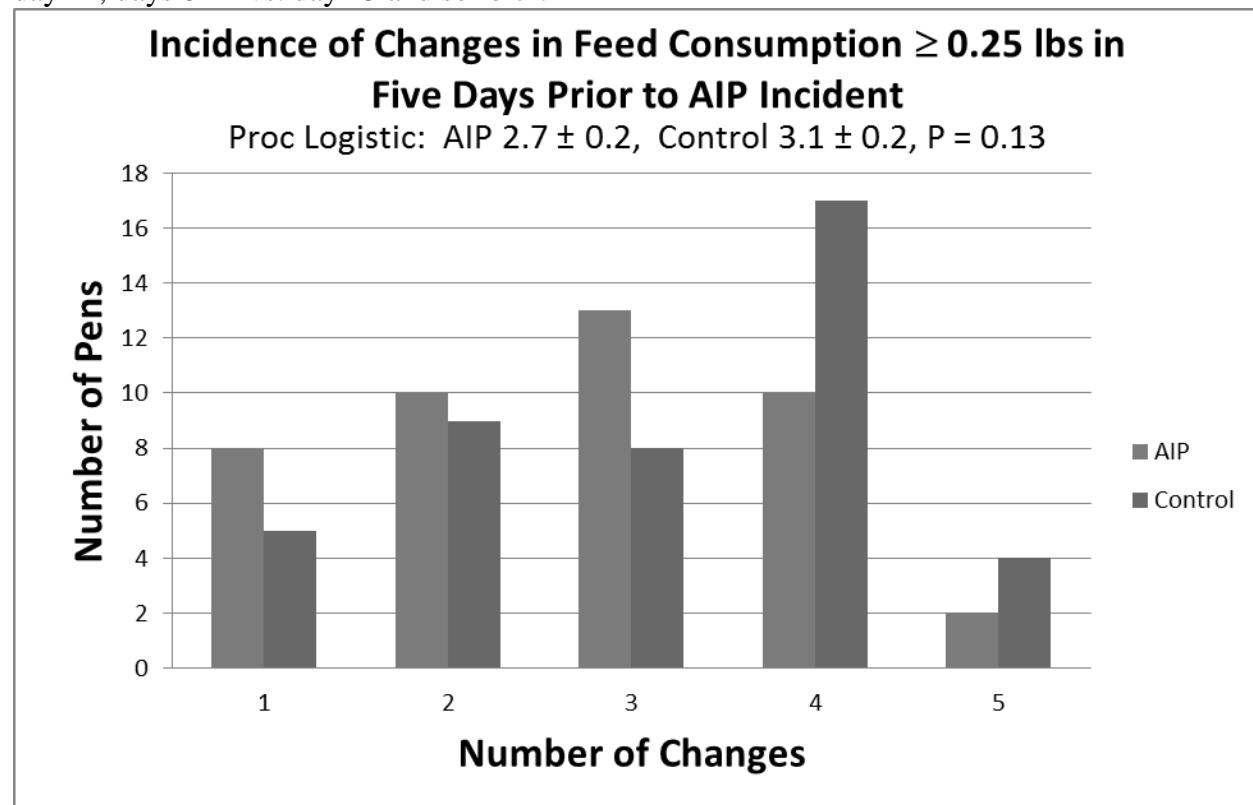
**Figure 2.1** – Graphical comparison between daily AIP incidence vs. daily high and low temperatures.



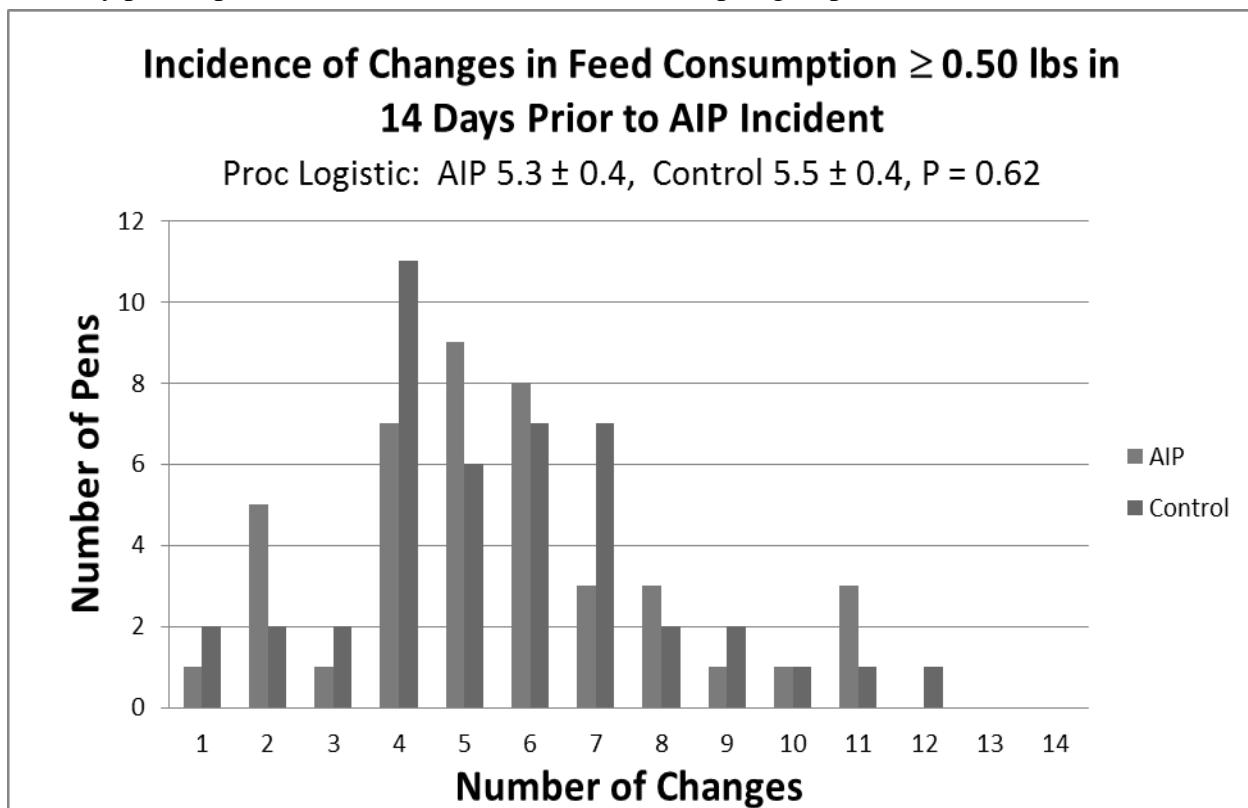
**Figure 2.2** – Statistical evaluation of incidence of changes in feed consumption  $\geq 0.25$  lbs. during a 14-day period prior to an AIP incident in the affected pen group.



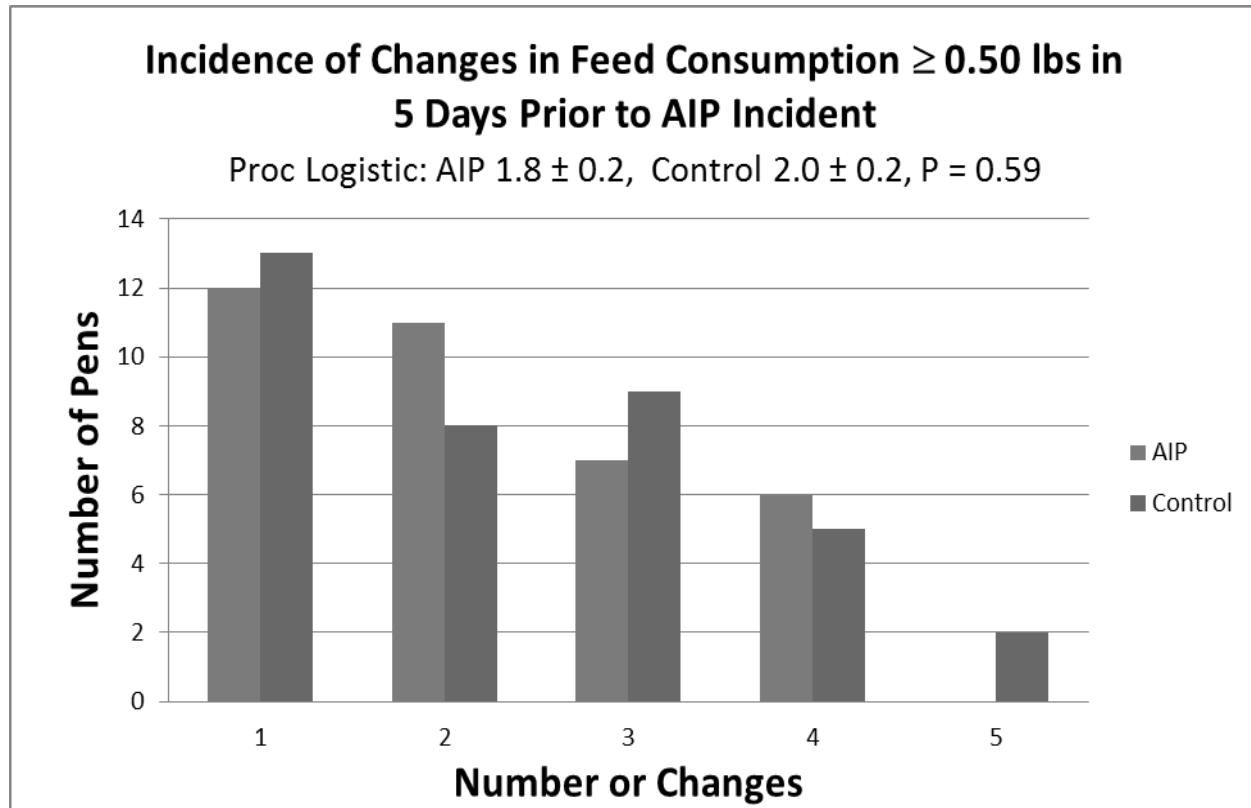
**Figure 2.3** - Statistical evaluation of incidence of changes in feed consumption  $\geq 0.25$  lbs. during a 5-day period compared to the 5-day rolling average prior to an AIP incident in the affected pen group. For example the average of days 10-14 compared to the average of day 15, days 9-13 vs. day 14, days 8-12 vs. day 13 and so forth.



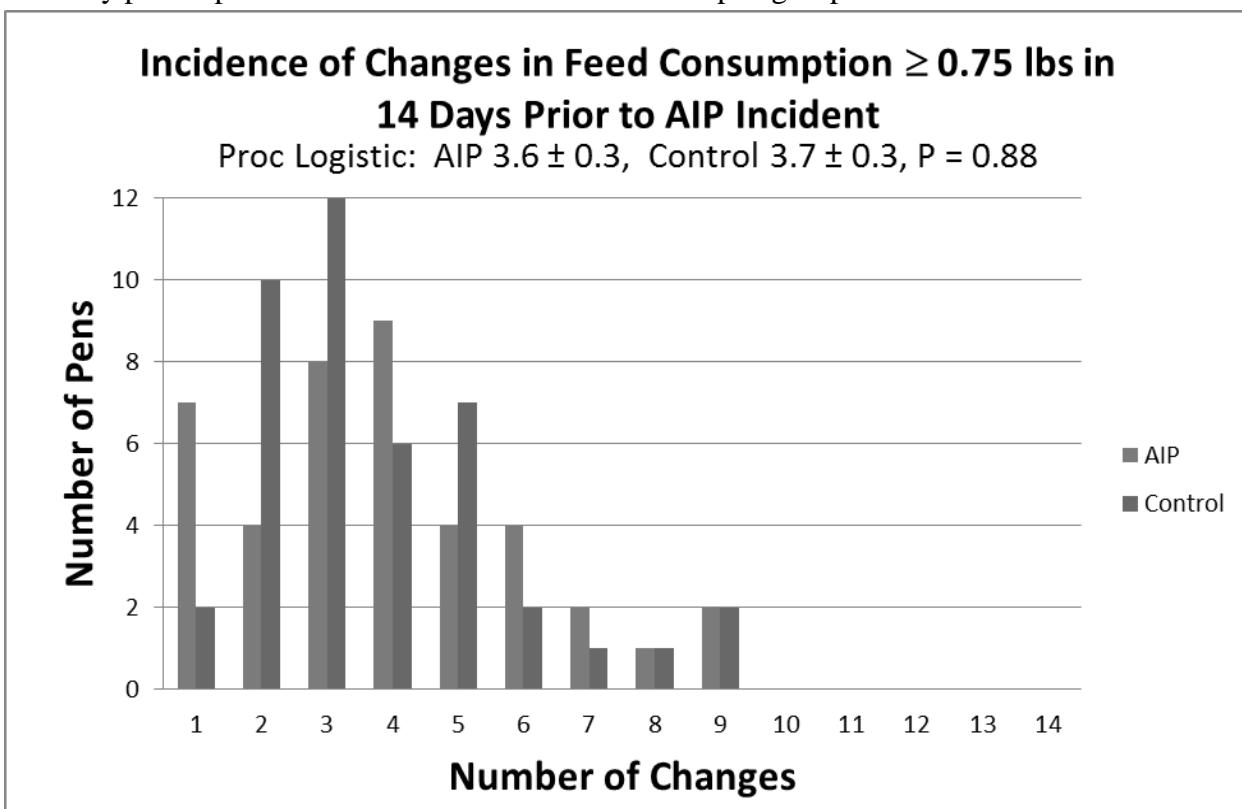
**Figure 2.4** - Statistical evaluation of incidence of changes in feed consumption  $\geq 0.50$  lbs. during a 14-day period prior to an AIP incident in the affected pen group.



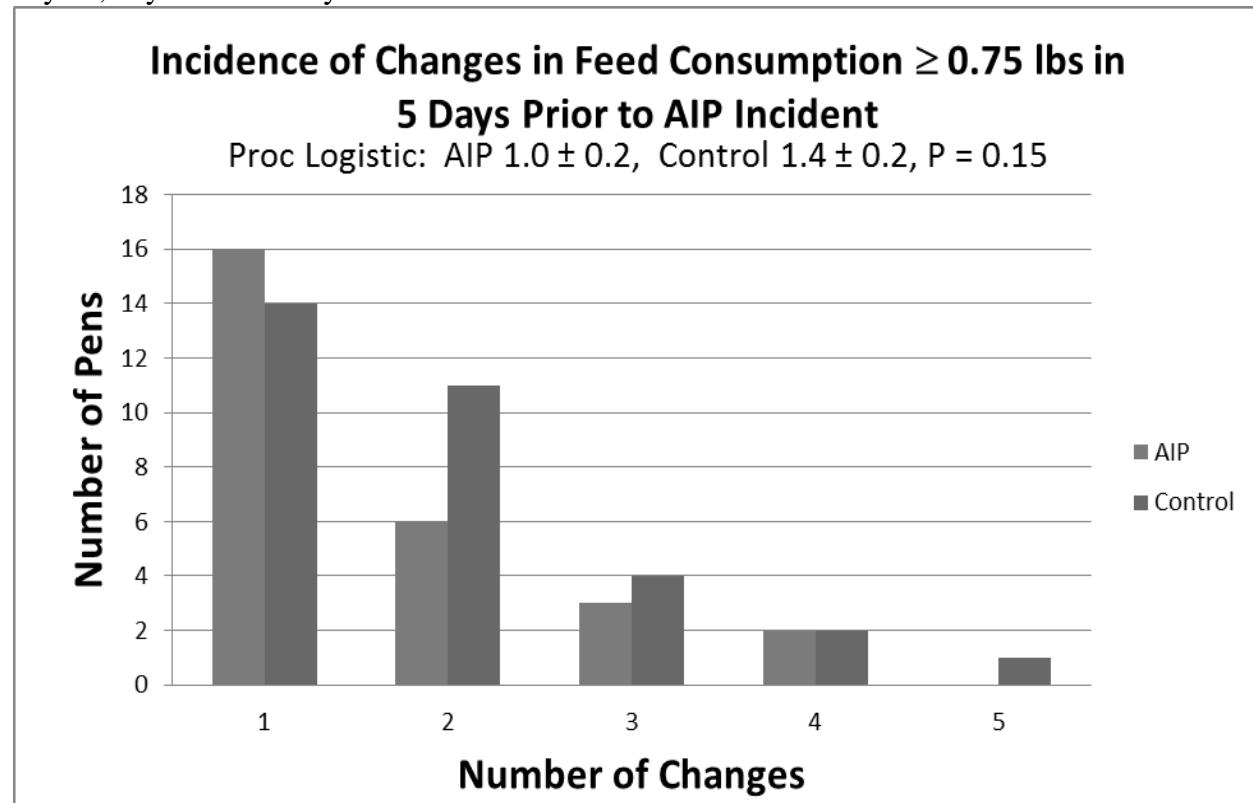
**Figure 2.5** - Statistical evaluation of incidence of changes in feed consumption  $\geq 0.50$  lbs. during a 5-day period compared to the 5-day rolling average prior to an AIP incident in the affected pen group. For example the average of days 10-14 compared to the average of day 15, days 9-13 vs. day 14, days 8-12 vs. day 13 and so forth.



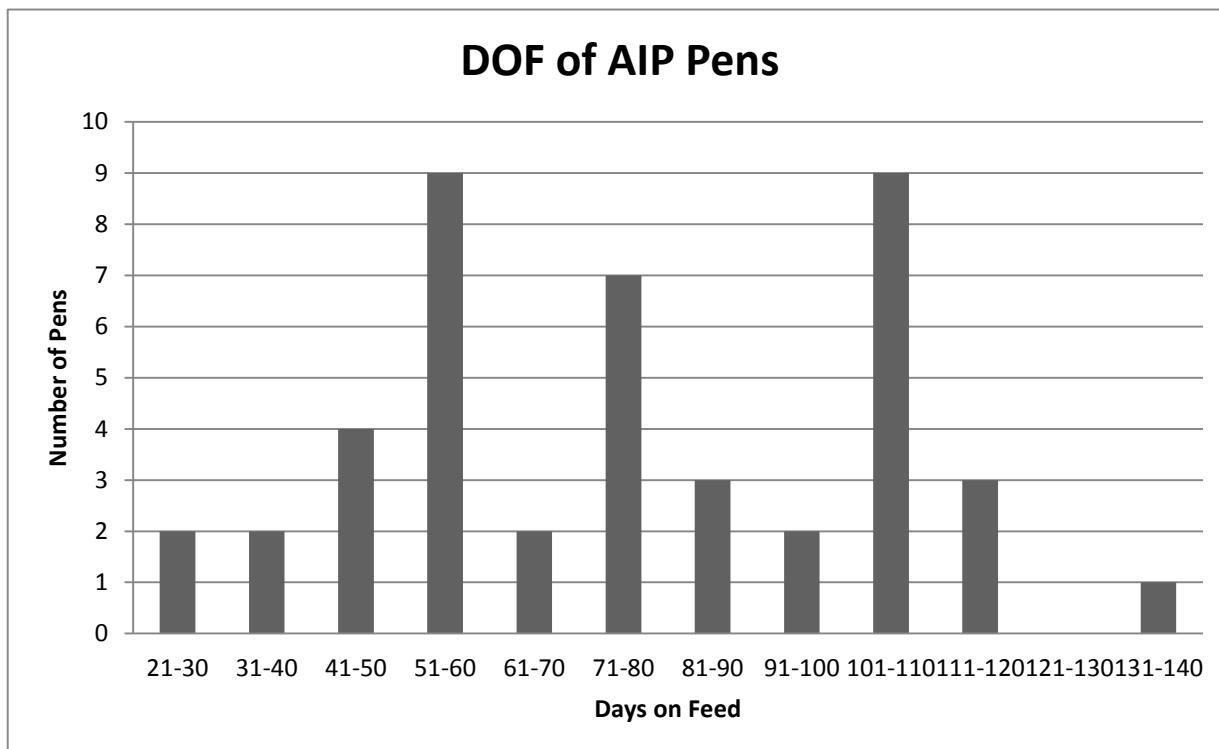
**Figure 2.6** - Statistical evaluation of incidence of changes in feed consumption  $\geq 0.75$  lbs. during a 14-day period prior to an AIP incident in the affected pen group.



**Figure 2.7** - Statistical evaluation of incidence of changes in feed consumption  $\geq 0.75$  lbs. during a 5-day period compared to the 5-day rolling average prior to an AIP incident in the affected pen group. For example the average of days 10-14 compared to the average of day 15, days 9-13 vs. day 14, days 8-12 vs. day 13 and so forth.



**Figure 2. 8** – Graphical identification of AIP affected pens by the number of days on feed.



#### Footnotes

Gastec Detector Tubes Models:

<sup>A</sup>NO.3L Ammonia (NH<sub>3</sub>) 1-30ppm scale range;

<sup>B</sup>NO.3M Ammonia (NH<sub>3</sub>) 50-500ppm scale range;

<sup>C</sup>NO.4H Hydrogen Sulfide (H<sub>2</sub>S) 100-2,000ppm scale range;

<sup>D</sup>NO.4HH Hydrogen Sulfide (H<sub>2</sub>S) 0.1-2.0% scale range.

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