THE PENNSYLVANIA STATE UNIVERSITY
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DEPARTMENT OF ANIMAL SCIENCE

# EFFECT OF AGE ON CAUSES OF BOVINE DIARRHEA

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## ABSTRACT

Diarrhea in cattle of all types continues to hinder both production and welfare. This study examined 157 cases of bovine diarrhea submitted to the Animal Diagnostic Lab (State College, PA) between January 2014 and September 2015. It was hypothesized that causes of diarrhea in these cattle would differ between age groups. While trends indicated this, the findings were not statistically significant. It is speculated that this was much due to the retrospective nature of the study. However, this examination provided valuable framework for further examination. With improvements and a change to a prospective organization, these findings have the potential to become solidified information that would assists farmers in better understanding diarrhea's causes leading to more efficient control of the spread of these pathogenic agents.

# TABLE OF CONTENTS

## LIST OF FIGURES

# LIST OF TABLES

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## Chapter 1

## Introduction

Enteric diseases, particularly diarrhea are a common issue in bovine herds of all sizes and the morbidity and mortality that occurs can result in severe economic losses to the dairy producer. Understanding the role of causative agents and their pathogenesis may enable future prevention and treatment of such diseases. It is also important to understand these pathogens since many of them are zoonotic and have the potential to impact public health. Identification of peak age susceptibilities would enable better biosecurity practices. It is hypothesized that the primary causative agents associated with diarrhea will be influenced by the age group of the cattle. The study aims at identifying etiological agents associated with diarrhea in various age groups of cattle from cases submitted to the Penn State Animal Diagnostic Laboratory.

Enteric disease resulting in diarrhea (or scours) continues to have a severe economic impact in dairy and beef herds (Divers, 2013 and Meganck et al., 2015.) The adverse ramifications of diarrhea can be observed beginning in the neonatal stages and continuing into adulthood (Divers, 2013 and Bartels et al., 2010.) Lasting effects of disease can be felt for the remainder of the affected animal's life. Diarrhea is one of the two most important causes of health problems in calves under one year of age. In an attempt to develop optimally producing adults, it is best for calves to proceed through their adolescence without these growth hindering conditions. (Bartels et al., 2010.)

Causes of diarrhea in cattle are numerous. Often frequency of disease is dependent upon geographical location, season of the year, and other variables. Some of the main etiologic agents associated with diarrhea in cattle are Enterotoxigenic *Escherchia coli*, Rotavirus, Coronavirus *Cryptosporidia*, and *Salmonella spp*, *Clostridium perfringens*, Giardia, *Myobacterium avium subsp.*, Bovine Viral Diarrhea, and parasites such as Coccidia and Strongids (Helman, 2000, Sweeney, 1996 and Gruenberg, 2014.) Many agents are classically associated with particular ages of cattle (Figure 1.)

**Table 1 Age of Infection with common agents**

| Age of Infection with Common Agents | |
|---|---|
| ***Escherichia coli*** | 1-7 days |
| ***Clostridium perfringens*** | 1-14 days |
| **Rotavirus** | 5-14 days |
| **Coronavirus** | 5-30 days or adults (Winter Dysentery) |
| **Cryptosporidiosis** | 7-21 days |
| **Giardia** | 8-14 days |
| **Bovine Viral Diarrhea Virus** | 6-24 months |
| **Strongylosis** | <1 year |
| **Coccidiosis** | 2 months to 1 year |
| **Johne's Disease (Paratuberculosis)** | >1 year |
| **Salmonellosis** | All ages |

## Common Enteric Pathogens in Cattle

*Escherichia coli*

*Escherichia coli* is a rod-shaped, gram negative, facultative anaerobic bacterium. It is part of the normal flora of the lower intestine in warm-blooded animals. There are numerous strains of *Escherichia coli*, many of which are non-pathogenic. Exposure can occur via contaminated water, food or through direct and indirect contact with other animals. Pathogenic *E. coli* can infect inside the intestinal tract or outside in sterile locations like the urethra (CDC, 2015.)

Some varieties of *E. coli* are more pathogenic than others as they express virulence factors, these *E. coli* are known as Enterotoxigenic *Escherichia coli* (ETEC.) Enterotoxigenic *Escherichia coli* have the ability to attach and colonize intestinal villi with its fimbrae and pilli. Following attachment they produce enterotoxins, which interfere with normal physiology of the gut, causing diarrhea. While ETEC is among the most common pathogenic *E. coli* other types such Enterohaemorrhagic *E. coli* and Neurotoxigenic *E. coli* also have been known to cause diarrhea (Helman, 2000.)

*Rotavirus*

Rotavirus is a virus from the family Reoviridae. It often infects the juveniles of many livestock species as well as humans (Gruenberg, 2014 and Caserta, 2016.) There are eight different species of rotavirus identified as A, B, C, D, E, F, and G. Most infections in livestock are due to types A-E. The virus is transmitted via fecal-oral contact (Kirkwood, 2010.) It causes disease by infecting villus tip of epithelial cells in the proximal half of the small intestine, causing damage to the cells resulting in villus atrophy. This damage to the intestinal wall causes malabsorptive diarrhea that is often large in volume and contains mucous (Helman, 2000 and Gruenberg, 2014.)

Rotavirus infections and its effects on the health and growth rate of mostly neonatal calves make it one of the most economically detrimental neonatal bovine diseases (Vega et al., 2015.) It is characteristically found in suckling calves between the ages of 5 and 14 days old. Though it has been found in calves as old as 8 weeks (Gruenberg, 2014 and Helman, 2000.) Generally, the younger the animal is when it contracts the virus, the more severe the infection (Vega et al., 2015.)

*Coronavirus*

Coronavirus is part of the family Coronaviridae. It affects the respiratory and gastrointestinal tracts of both mammals and birds (Cavanagh, 2005, Hogue et al, 1984 and Helman, 2000.) In cattle, the enteric variety is transmitted via the fecal-oral route (Helman, 2000.) The virus replicates in intestinal enterocytes causing lesions similar to those of rotavirus. Coronavirus infects both the small and large intestine and causes atrophy of colonic ridges and affects a larger portion of each villus tip than does rotavirus explaining why the malabsorptive diarrhea produced is generally more severe than that caused by rotavirus. (Gruenberg, 2014, and Helman 2000.)

Coronavirus generally affects calves between 5 and 30 days of age but has also been identified as a potential causative agent of winter dysentery in adult cattle (Helman, 2000 and Jeong et al., 2005.) Winter dysentery causes hemorrhagic diarrhea. It is more commonly found in cattle housed in enclosed spaces, making it more frequent in dairy cattle (Helman, 2000.) This is significant since winter dysentery can cause decreased milk production, and cause economic losses to the dairy producer (Jeong et al., 2005.)

*Cryptosporidia*

Cryptosporidiosis is another common etiologic agent causing diarrhea in juvenile ruminants and is most commonly found in calves from 1-3 weeks of age. It is caused by the protozoa *Cryptosporidium parvum* (Helman, 2000.) This pathogen is contracted through fecal-oral contact with oocysts (Araque, 2006.) Cryptosporidiosis can sporulate internally and infect the rest of gut. This ability for autoinfection enables rapid development of severe disease that can be maintained for a long period of time, making relapses quite common (Helman, 2000.)

It infects by attacking the apical surface of enterocytes in the distal small intestine and proximal colon. Here it forms a parasitoporous vacuole, a blister-like structure made by some protozoa to protect themselves while developing inside the host cell. Infection from Cryptosporidium results in atrophy and necrosis of intestinal villi and decreased activity of mucosal enzymes. This causes the intestinal tract to be compromised in such a way that its absorptive ability is hindered and fermentation of nutrients is decreased, resulting in diarrhea (Araque, 2006.)

*Salmonella*

Salmonellosis is caused by infection with *Salmonella* species. *Salmonella spp.* are gram-negative, facultatively anaerobic bacilli with flagella (Giannella, 1996.) There are more than 2000 known serotypes each considered a separate species (Giannella, 1996 and Helman, 2000.) All 2000 serotypes have the ability to cause gastrointestinal upset and diarrhea in calves, most often affecting cattle over one week old (Helman, 2000, McGurk and Peek, 2003.) The various serotypes do have the ability to be transmitted between different species including birds, cats, dogs, cattle, people, flies and pigs. They can be transmitted through fecal-oral contact, contact with saliva and nasal secretions through shared water or in aerosol form (especially in confinement facilities) and though milk and colostrum (McGurk and Peek, 2003.) It is important to note that the infection can cause an asymptomatic, chronically infected carrier state in all ages. The carrier state in cattle is most often caused by the host-adapted, *Salmonella dublin* strain (McGurk and Peek, 2003.)

When ingested, the bacteria can survive the gastric acid barrier and then occupy mucosa of the small and large intestines. Once it invades the mucosa it produces enterotoxins that stimulate the host's body to release proinflammatory cytokines triggering severe inflammation. This is what causes the characteristic diarrhea. It also has the potential to cause ulceration and destruction of the intestinal mucosa (Giannella, 1996.) Enteric Salmonellosis can manifest as fever, diarrhea and abdominal cramps with the most common form being uncomplicated, self-limited gastroenteritis.

*Clostridium perfringens*

*Clostridium perfringens* is one of multiple pathogens in the genus *Clostridium,* family Clostridiaceae. It is an anaerobic, gram-positive, spore-forming bacilli common in the environment and in healthy intestinal flora of both humans and other mammals (Giannella, 1996.) Because of this, it is considered an opportunistic pathogen that can become harmful when an animal's immune system is compromised (Petit et al., 1999.) *C. perfringens* not only is the causative agent many enterotoxemic diseases of lambs and calves but also of gas gangrene and food poisoning in humans (Giannella, 1996.) As a pathogen, it is most commonly found in calves less than two weeks old. *C. perfringens* has five types, A, B, C, D and E. All have the potential to cause diarrhea in calves but type C is most common (Daly and Rotert, 2007 and Helman, 2000.)

When an animal is exposed to *C. perfringens* in the environment, the bacteria survives the gastric barrier of the stomach and proliferates in the intestine. This transition is made easier in calves since *C. perfringens* survives by ingesting starches and sugars. In adults, starches are rapidly digested in the rumen, often starving the bacteria, but since the rumen is not fully developed in the calf, the bacteria are able to survive more readily on the available starches and sugars. Small amounts of *C. perfringens* are tolerable and often normal to a cow's gut flora. However if an animal is stressed, has a change in diet or is ill, the bacteria can proliferate and produce toxins that will damage the intestinal lining, causing diarrhea. High levels of these toxins can allow them to enter the blood stream causing systemic inflammation, shock and cardiac arrest that can lead to death (Daly and Rotert, 2007.) It has been documented that rapid proliferation of the bacteria can lead to sudden death before any clinical signs surface in particularly young or weak cattle (Helman, 2000.) More recently, *C. perfringens* type A has been linked to cases of jejunal hemorrhage syndrome, causing bouts of hemorrhagic diarrhea and enteritis. This seems to be more common in high-producing dairy cattle early in their lactation. It is thought that improving nutritional plans and ensuring that diet changes are made gradually may help prevent this malady (Stampfli, 2014.)

*Giardia*

Giradia is a protozoan parasite that may infect many mammalian and non-mammalian species. The species that is responsible for infecting both humans and cattle is *Giardia intestinals,* and is spread through contact with contaminated water and food sources (Zhang et al., 2016.) Giardia is a non-invasive parasite that colonizes the small intestine by attaching to the apical surface of the epithelium in the duodenum and proximal jejunum (Helman, 2000 and Hodges and Gill, 2010.) It has been isolated in cattle between the ages of two and four weeks and is accepted as a cause of diarrhea though the pathogenic mechanisms by which it causes it are unknown (Helman, 2000.) However, it is may be present in juvenile and adult cattle that have no symptoms (O'Handley et al., 2001.) Thus, Giardia's significance as a primary pathogen is still subject of discussion.

*Johne's Disease (Paratuberculosis)*

*Myobacterium avium subsp.* Paratuberculosis (MAP) is better known as Johne's Disease. MAP is a gram positive acid-fast bacillus, mycobactin-dependent, slow growing bacteria. It is also an obligate pathogen, as it is unable to multiply without the host. It affects cattle and small ruminants such as sheep and goats. Most often it is transmitted via the fecal-oral route but can also be transferred through colostrum, milk or in-utero. Most often animals are infected as calves, though clinical signs may not arise until later in life (Sweeney, 1996.)

MAP lodges in the ilium where specialized absorptive cells called M cells phagocytose the bacteria. The M cells lay over the Peyer's Patches (aggregates of lymphoid tissue.) The function of Peyer's Patches is to sample luminal contents to take back to macrophages and lymphocytes to educate the immune system. When MAP is brought back to the macrophages and phagocytized, instead of being destroyed like other antigens MAP cells proliferates and thrives within the cell. Eventually the macrophage will die, allowing MAP to, spread throughout the ilium. The immune system of the animal will employ more macrophages and lymphocytes to try to kill the bacteria, creating the characteristic thickening of the intestinal wall that compromises nutrient absorption and causes diarrhea (Sweeney, 1996.)

The disease has four distinct stages. In the first stage of the disease, there are no clinical signs of the disease, however the animal may shed the organism at a low level and thus can still transmit the disease. These animals are generally younger than two years old. In the second stage, the disease manifests itself in a subclinical form. Adult animals with no overt clinical signs however they may have low fertility, mastitis or be prone to other infections. In the third stage of the disease, the animals shows clinical signs characterized by gradual weight loss, decreased appetite and diarrhea. This usually occurs after an incubation period between two and eight years. In the fourth stage of the disease, the clinical signs of this stage are lethargy, emaciation, bottle jaw (intermandibular edema) due to hypoproteinemia (low protein levels in blood), cachexia (weakness and bodily wasting) and pipe stream diarrhea (Sweeney, 1996.)

*Bovine Viral Diarrhea*

Bovine viral diarrhea virus (BVDV) is the causative agent of Bovine Viral Diarrhea (BVD). BVDV is a member of the genus *Pestivirus*. Many strains of BVDV exist and the differences found in subgenotypes are enough that vaccinated animals can still contract another type of the virus if exposed to a novel subtype. The subtypes are grouped into either genotype I or II. Both genotypes can be found worldwide but specific strains tend to be more regional. Each type of BVDV is divided into noncytopathic and cytopathic biotypes, based on its ability to cause overt cytopathic change and cell death in cultures. A cytopathic effect is a structural change in the host cell resulting in lysis or cell death without lysis due to an inability to reproduce. Cytopathic varieties can develop in animals persistently infected with non-cytopathic varieties. This is often a result of a mutation due to recombination of non-cytopathic viral RNA with itself, heterologous viral RNA or host cell RNA (Helman, 2000.) BVDV is transmitted through oral contact with an infected animal's biological products, biting insects, by breeding with infected semen or before birth from an infected mother to her fetus (APHIS, 2007.)

A fetus will contract BVD through the placental blood if the dam is infected. When a calf or cow comes in contact with the virus, it will replicate in epithelial cells of the mucous membrane where it was first introduced. The virus is spread to the rest of the body two ways, either phagocytes will ingest the cells infected with the virus and it will be transferred to lymphoid tissues or it can travel in the blood stream. Either way, viremia results within two to four days post exposure (Fray *et al*. 1998.) When the virus reaches the mesenteric and submucosal ganglia of the gastrointestinal tract, the virus can interfere with normal function of the gastrointestinal system and diarrhea can result. The systemic viremia also will cause the animal to become immunocompromised, opening them up for other infections to take over and resulting in a vast assortment of secondary symptoms (Lanyon *et al*., 2014.)

It is possible for an animal to become persistently infected with BVDV. When the virus is spread to the rest of the body it can be maintained asymptomatically in areas of the body that are immunoprivileged such as testicular tissue, ovarian follicles, white blood cells and the central nervous system. Chronically infected animals are still contagious (Goyal and Ridpath, 2008) Cattle infected in-utero before 150 to 180 days gestation may be aborted, after this period the fetus will generally survive. However, they may be born with a persistent infection (Lanyon *et al*., 2014.) BVD contracted after birth is most common in cattle between the ages of 6 and 24 months. Clinically, the presentation can vary in severity from inapparent to severe enteric, fatal mucosal disease complex. The most characteristic symptom of infection is diarrhea. The virus becomes deadly when it reaches the point of profuse enteritis (Helman, 2000.)

*Coccidia*

Coccidiosis is caused by protozoa in the genera *Eimeria* and *Isospora*. Cattle are affected by *Eimeria spp.* Many mammals and birds can get coccidiosis, however it is host-specific so there is no interspecies transfer reported thus far. Coccidia oocysts enter the environment when they are shed in an infected animal's feces and healthy animals will become infected when they ingest sporulated oocysts. Coccidia are considered opportunistic pathogens, they thrive and become more prevalent when animals are malnourished, the environment has poor sanitation and is overcrowded (Constable, 2015b.)

Not all oocysts shed in feces are sporulated, so not all oocysts shed are infective. When oocysts sporulate, amorphous protoplasm develops into sporozoites (small bodies.) Once ingested the sporozoites leave the oocyst and attack the intestinal mucosa. When inside the cells of the mucosa the sporozoites develop into multinucleated schizonts. Each nucleus of the schizonts becomes a merozoite, which is an infective body that can enter a new cell and repeat the process, enabling spread of the infection. Eventually the merozites will undergo sexual reproduction by developing into macrogametocytes and microgametocytes, which are male and female counterparts. These bodies will either create a few microgametes or one macrogamete. When a macrogamete is fertilized by a microgamete it will develop into an un-sporulated oocyst that can then be deposited in feces. This development and disruption in the cells of the intestine causes destruction of not only the intestinal epithelium but often connective tissue beneath it. This damage results in the commonly observed clinical signs of hemorrhagic diarrhea, tenesmus, and dehydration (Constable, 2015b.)

Coccidia infections are commonly found in calves between the ages of two months and one year. The incubation period is between 16 days and 4 weeks depending on the species. Severity of symptoms are varied, generally calves may appear weak or thin and often have watery diarrhea. In more severe cases, diarrhea may contain blood, mucous and sloughed epithelium and calves may develop a fever and weight loss. Enteritis is generally contained in the large intestine but weakness derived from the infection can lead to development of concurrent infections (Constable, 2015a.)

It is thought that coccidiosis could have a nervous form characterized by neurological symptoms like muscle tremors, nystagmus, and hyperesthesia, animals who progress to this stage generally die within one day. However, there is little evidence supporting this set of symptoms' linkage to coccidia and no proven mechanism as to how these signs develop (Constable, 2015a.)

*Strongyles*

Strongylosis is caused by an infestation of *Strongyloides spp.* Strongyles are parasitic nematodes. There are many different species of strongyles that affect different types of livestock, though their mechanisms of pathogenesis are similar. Some of the most common varieties that affect cattle are found in the genus *Nematodirus*. *N. helvetianus,* otherwise known as "the thread-necked strongyle," is most notable for often causing noticeable clinical symptoms, especially in younger calves. Strongyles are transferred through the fecal-oral route. Ova are passed through the feces of infected animals and develop into invective larvae on pasture (Melancon, 1999.) They have also been known to be transmitted through colostrum (Fox, 2014.)

This first larval stage (L1) is not infective. Under favorable conditions, the larvae will develop in the egg until it gets to the L3 stage. If it the environment is too cold or dry the larvae in the L1 or L2 stage can halt development until conditions become more favorable, allowing them to persist in pasture for long periods of time. Once the larvae reach the L3 stage, they will hatch from the egg and now have the ability to infect an animal, but they are now susceptible to drying. Once an animal ingests a L3 larvae, the larva will molt in the abomasum giving rise to the L4. The L4 travels to the intestine and will burrow into the intestinal wall, eventually emerging as an adult. Females then lay eggs that are passed through the feces to repeat the cycle (Melancon, 1999.)

The penetration into the intestinal wall by the L4 is particularly irritating and can cause inflammation and damage the mucosa. If present in significant enough numbers, this will lead to the clinical signs of diarrhea, anorexia and dehydration (Melancon, 1999.) While strongyles are present in many asymptomatic cattle, most clinical infections occur in calves under six months old (Fox, 2014.) Generally, cattle become immune to strongyles sometime after six months of age from both exposure and general maturation (Melancon, 1999.)

*Non-Pathogenic Causes*

In the case of neonatal calves, if there is no evidence of a pathogenic cause for diarrhea the most likely cause is nutritional such as changing milk replacer or an environmental stressor like transport or vaccinations. (Kehoe and Heinrichs, 2005.) A sudden increase in milk production of the dam or increased quantity of milk replacer fed at one time with more time in between feedings can cause diarrhea. Calves with a primary lactase deficiency may also develop diarrhea. Generally, nutritionally related diarrhea is not severe and will resolve with little to no treatment (Helman, 2000.)

Nutritional issues can also cause diarrhea in adult cattle. Copper and cobalt deficiency have been linked to diarrhea in adult cattle. Copper deficiency may be induced when there is elevated molybdenum in the diet. Cattle who graze on a pasture that is predominately fescue or pastures that are treated with a fertilizer high in nitrogen such as chicken manure may be susceptible to abdominal fat necrosis which can lead to diarrhea. It is important to note however that any stress or dietary changes can cause transient diarrhea that most often resolves with little to no treatment (Helman, 2000.)

# Chapter 2

## Materials and Methods

The Laboratory Information Management System (LIMS) was queried for cases submitted between January 2014 and September 2015 in which the primary test requested was "Diarrhea Panel" and Purpose of Test was Pathology. This subset was further refined by excluding submissions for animals other than cattle. For each individual case, tests performed were selected at the discretion of the pathologist on duty when the cases were submitted. A total of 157 cases were identified and were grouped by age for evaluation. The age groups into which the cattle were divided for this study were 1-7 days, 8-14 days, 15-30 days, 31-90 days, 91-180 days, 181-365 days, and greater than 365 days. These age divisions provided for analysis of a significant sample number while still separating animals in developmentally different stages. All specimens were tested using commercially available testing kits and techniques outlined in the standard operating procedures for the laboratory.

## Statistical Analysis

A total of 157 records from the LIMS database were tabulated in a Microsoft Excel sheet. The records were categorized into two groups; group one included animals < 90 days of age, while group two comprised of animals > 90 days of age. The average number of counts for each of the two groups were compared using the Tukey-Kramer (equal variance) or Dunnett's T3 (unequal variance) procedures. These two procedures were used due to unequal sample sizes observed in the two categories of a given count. The Tukey-Kramer procedure performs all pair-wise comparisons, testing whether the two means are significantly different. $P < 0.05$ was considered significant. The data was analyzed using OpenStat, advanced statistical software for data analysis (http://openstat.en.softonic.com/).

# Chapter 3

## Results

From January 2014 to September 2015, 157 total bovine samples were submitted to ADL with a primary complaint of diarrhea.

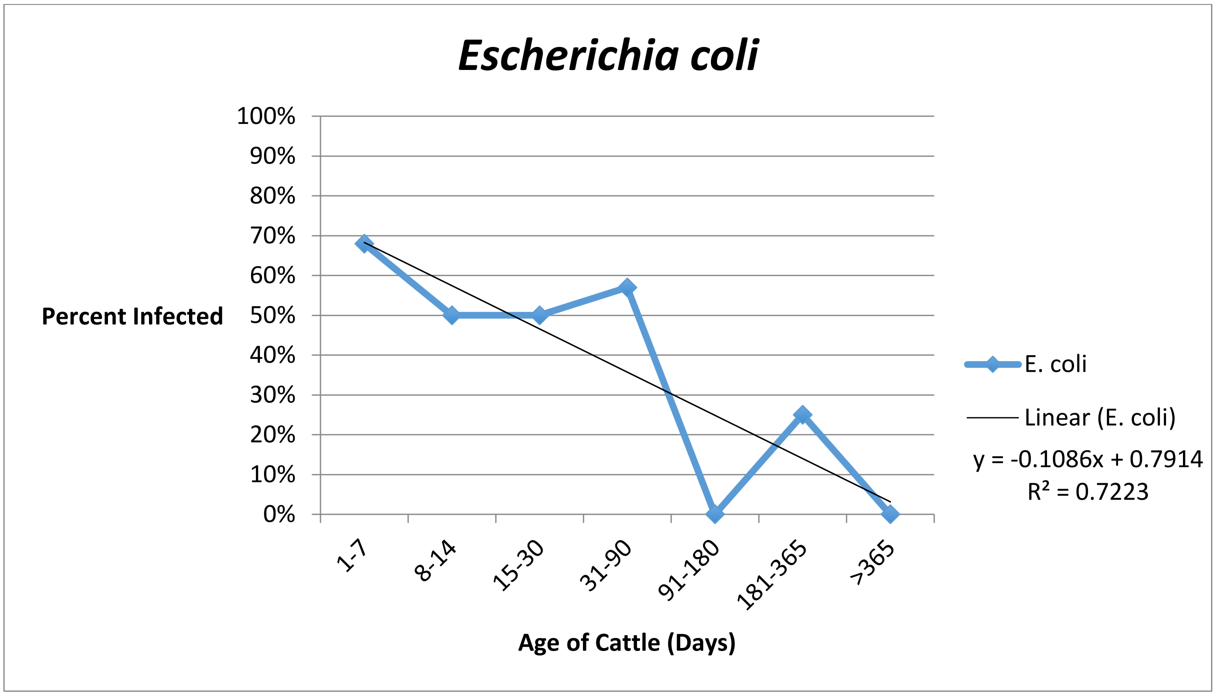
Forty one samples were from calves between 1 and 7 days of age, 28 were from calves between 8 and 14 days old, 22 were from calves 15 to 30 days old, 14 samples were from calves 31 to 90 days, 3 were from animals 91 to 180 days old, 4 were obtained from animals between the ages of 181 and 365 days old and 13 were from animals older than 356 days (Table 2.)

Raw data that depicts the animals that tested positive for each identified pathogen within these specified age groups is depicted in a table in the appendix.

**Table 2 Sample Set Details**

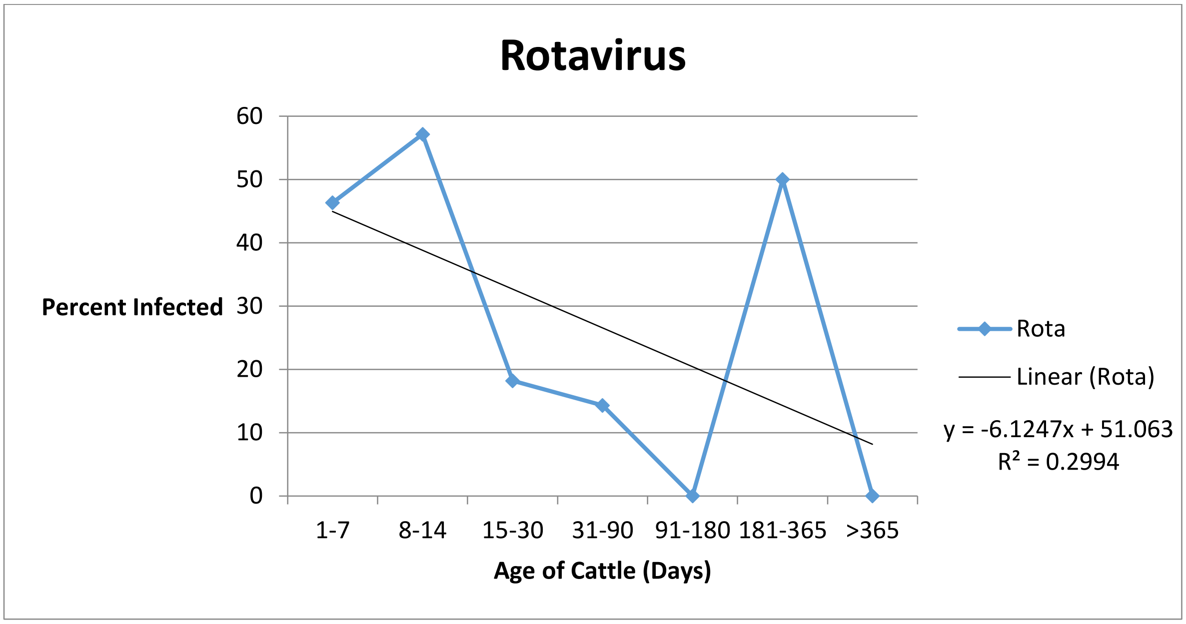
| Age Group | Number of Cases |
|---|---|
| **1-7 days** | 41 |
| **8-14 days** | 28 |
| **15-30 days** | 22 |
| **31-90 days** | 14 |
| **91-180 days** | 3 |
| **181-365 days** | 4 |
| **>365 days** | 13 |
| **Total** | 157 |

**Figure 1 *Escherichia coli* isolated obtained from cattle submitted to the ADL**



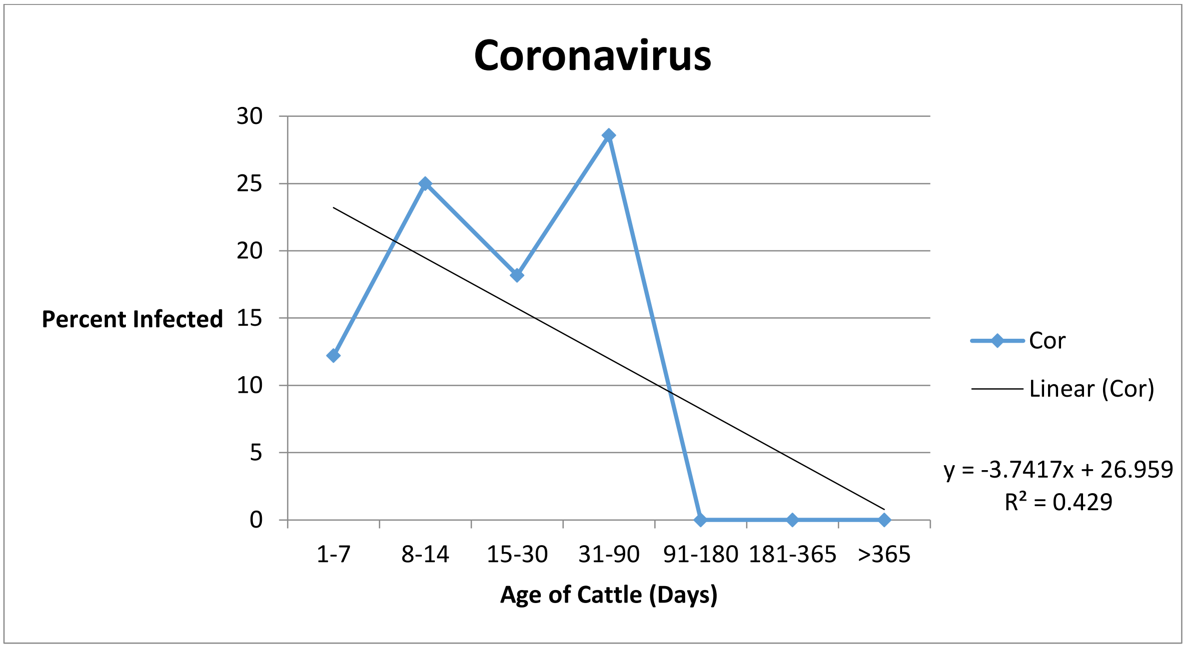

*E. coli* was isolated from 28 of the 41 (68%) calves aged 1 to 7 days. In calves 8 to 14 days 14 of the 28 (50%) yielded a positive result. In the 15 to 30 day age group 11 of the 22 animals were positive (50%.) In animals aged 31 to 90 days 8 of 14 (57%) cases yielded a positive result. *E. coli* was not detected in any animals between the ages of 91 and 180 days. In the 181 to 365 day old group 1 of the 4 (25%) had growth. There was no *E. coli* found in animals older than one year. A linear trend line was created to determine if the number of reported cases with E. coli increased or decreased with each age group. It was observed that as the age increased the number of animals with E. coli declined. A 72% correlation was observed with increasing age and the number of cases with *E. coli* (Figure 1.)

**Figure 2 Rotavirus isolated obtained from cattle submitted to the ADL**



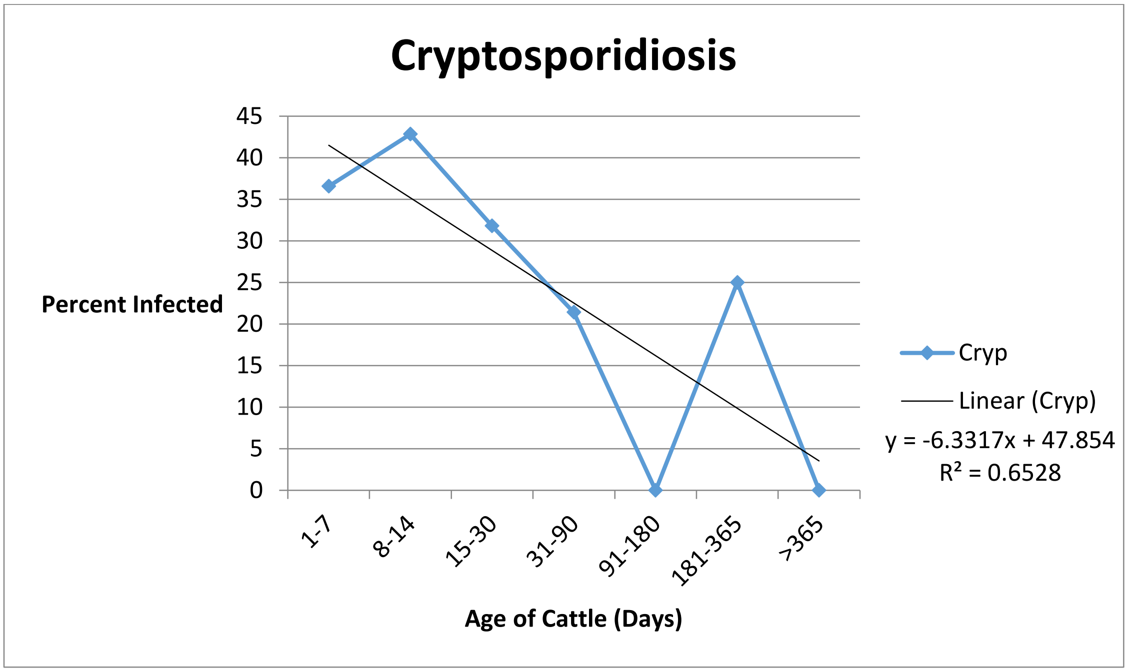

Of the 41 diarrhea cases seen in calves between the ages of 1 and 7 days, 19 tested positive for rotavirus (46%.) In calves between the ages of 8 to 14 days, 16 of the 28 tested positive (57%.) In the 15 to 30 day age range, 4 of the 22 (18%) calves tested positive. In animals between the ages of 31-90 days 2 of the 14 (14%) animals tested positive. There were no rotavirus positive animals between the ages of 91 to 180 days. Rotavirus reemerged in the 181 to 365 days age group with 2 of the 4 (50%) animals in this age bracket testing positive. Rotavirus was not detected in cattle greater than one year of age. The linear trend line created indicated that as the age increased the number of animals with rotavirus declined. A 30% correlation was observed with decreasing age and the number of cases with rotavirus (Figure 2.)

**Figure 3 Coronavirus isolated obtained from cattle submitted to the ADL**



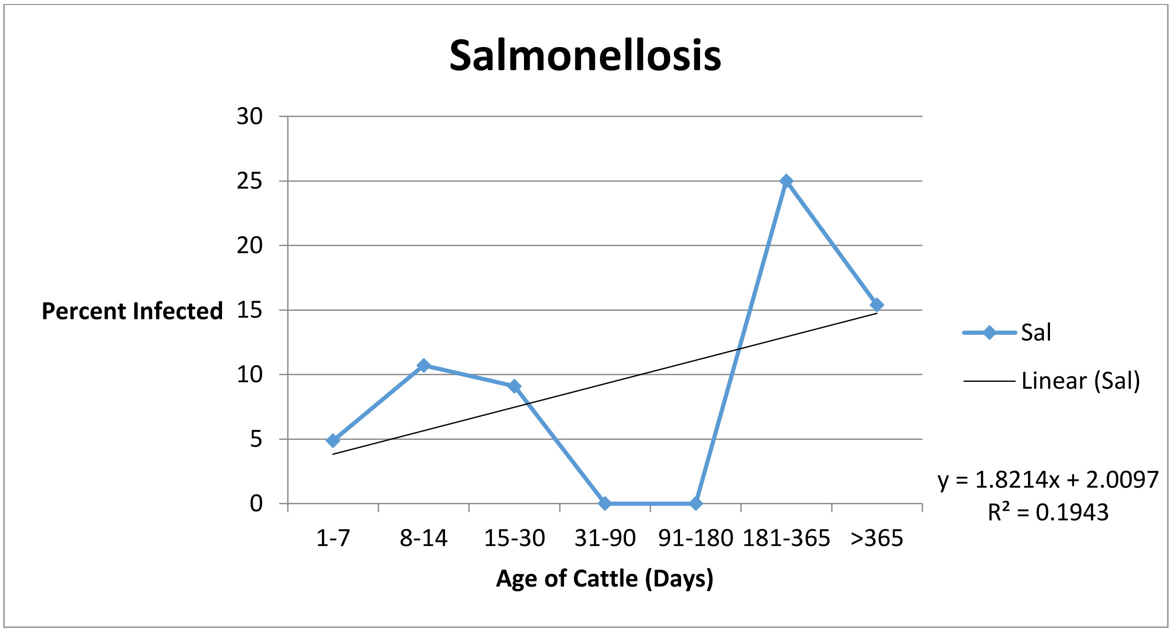

In calves one to seven days of age, 5 of 41 (12%) were positive for coronavirus. Of the 28 animals in the 8 to 14 day age group 7 tested positive (25%.) There were 22 calves between the ages of 15 and 30 days, 4 were positive for coronavirus (18%.) Finally, out of 14 between 31 and 90 days old, 4 were positive (29%.) No animals older than 90 days tested positive for coronavirus. As indicated by the linear trend line, the number of animals with coronavirus declined with age with a 43% correlation between decreasing age and animals diagnosed with coronavirus (Figure 3.)

**Figure 4 Cryptosporidiosis isolated obtained from cattle submitted to the ADL**



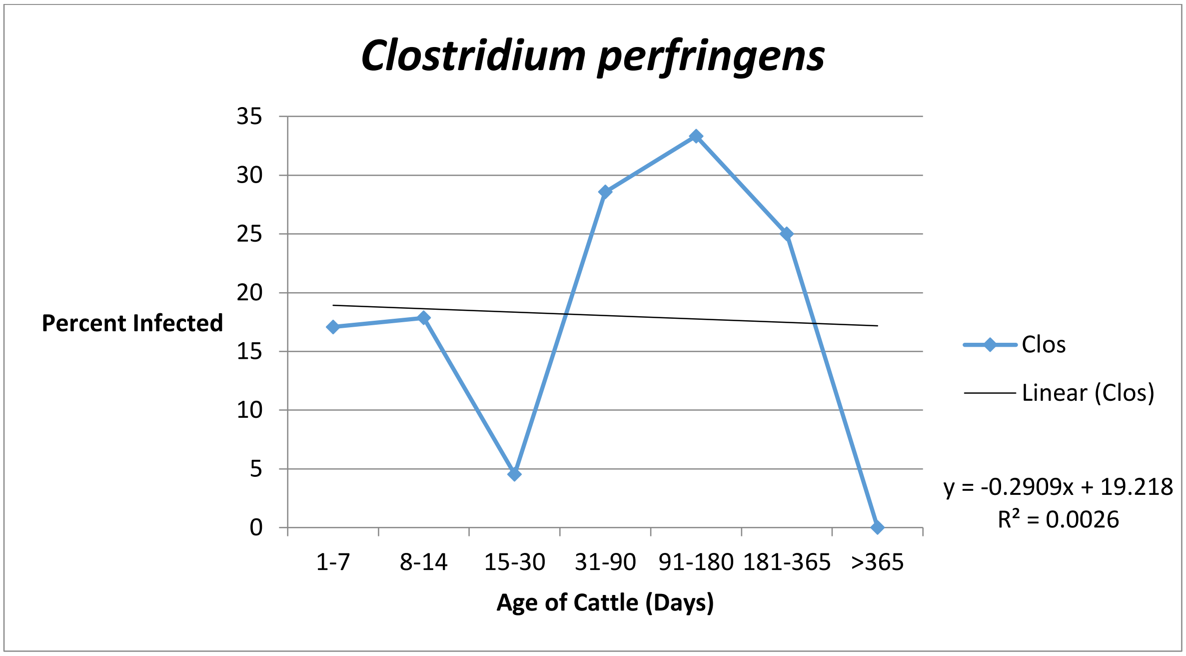

Of calves ages 1 to 7 days, 15 out of 41 tested positive (37%.) In the 8 to 14 day age group 12 of the 28 (43%) received a positive diagnosis. For animals 15 to 30 days old, 7 of 22 (32%) tested positive. In ages 31-90 days 3 of the 14 calves were positive (21%.) No animals in the 91 to 180 day range or any over one year old received a diagnosis of cryptosporidiosis. However, 1 of the 4 animals (25%) between the ages of 91 and 180 days did test positive. The linear trend line created showed that the number of reported cases with Cryptosporidiosis decreased with each age group. A 65% correlation was observed with decreasing age and the number of cases with Cryptosporidiosis (Figure 4.)

**Figure 5 Salmonella spp. isolated obtained from cattle submitted to the ADL**



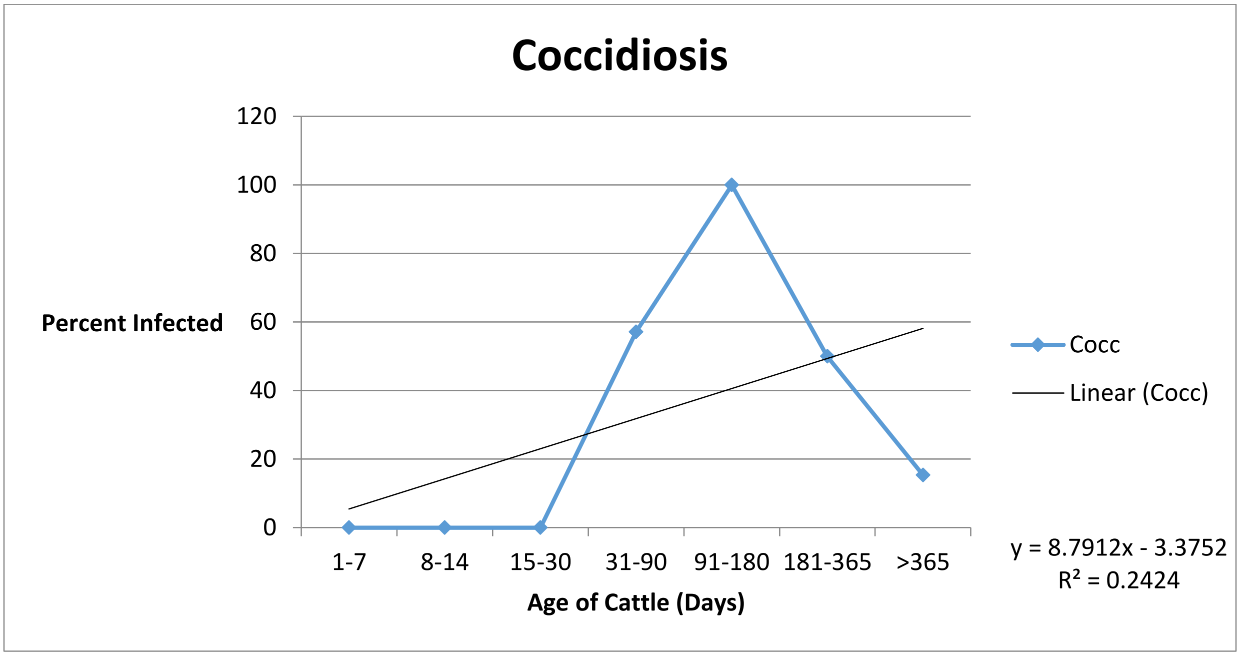

In calves ages 1 to 7 days, 2 of the 41 tested (5%) were found to have salmonellosis. In ages 8 to 14 days 3 of the 28 (11%) received a diagnosed. In the 15 to 30 day age range 2 of the 22 calves (9%) were found to be positive for salmonella. No isolates were detected in animals between the ages of 31 and 180 days. One in the 181 to 365 day old age group and 2 from the set of animals older than one-year tested positive for salmonella. The linear trend line showed that the number of cases with Salmonella increased with each age group. A 19% correlation was observed with increasing age and the number of cases with Salmonella (Figure 5.)

**Figure 6 *Clostridium perfringens* isolated obtained from cattle submitted to the ADL**




In calves between the age of 1 and 7 days, 7 of 41 (17%) were positive for *Clostridium perfringens*. In those calves between the ages of 8 to 14 days, 5 of the 28 (18%) were diagnosed. In animals aged 15 to 30 days *C. perfringens* was detected in 1 of the 22 (5%). In the 31 to 90 day age group 4 of the 14 (29%) tested positive. *C. perfringens* was isolated from one animal each in the 91 to 180 days and the 181 to 365 day age group. No animals over one year tested positive for *Clostridium perfringens*. From the linear trend line it was observed that as the age increased the number of animals with *Clostridium perfringens* declined. A 0.003% correlation was observed with decreasing age and the number of cases with *Clostridium perfringens* (Figure 6.)

**Figure 7 Coccidiosis isolated obtained from cattle submitted to the ADL**



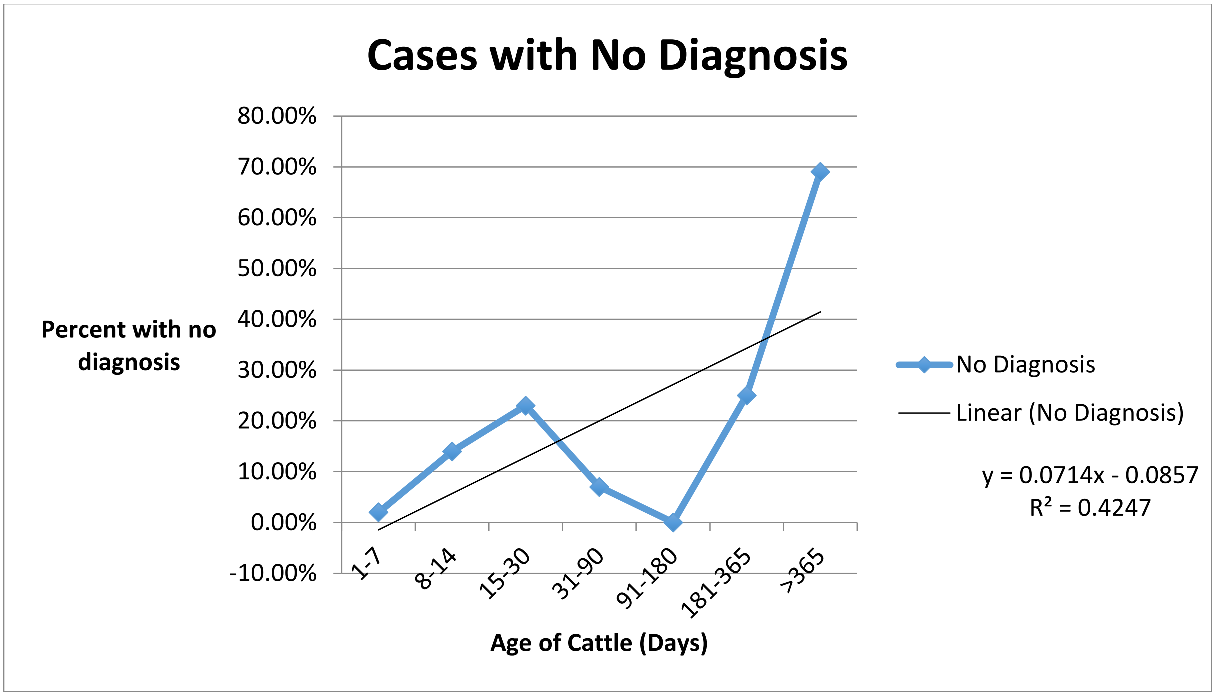

No animals under the age of 30 days were diagnosed with coccidiosis. In the 31 to 90 day age group, 8 of the 14 (57%) calves were diagnosed. In animals ages 91 to 180 days old 3 out of 3 (100%) were positive. In the 181 to 365 day old group 2 of the 4 (50%) were diagnosed. Two of the 13 (15%) samples from cattle older than one year were positive for the presence of coccidia. A linear trend line was created and determined that the number of reported cases with Coccidiosis increased with each age group. A 24% correlation was observed with increasing age and the number of cases with Coccidiosis (Figure 7.)

## Stronglyosis, Giardia, Johne's Disease and Bovine Viral Diarrhea Virus

Two calves were positive for strongyles, one between the ages of 15 and 30 days and another between 181 and 365 days. Three calves aged 8-14 days were positive for the presence of giardia. Paratuberculosis (Johne's Disease) was detected in two animals, each older than one year. Bovine Viral

Diarrhea Virus (BVD) was not detected in either of the two animals tested. However, due to the low occurrence of these pathogens within this sample set they were eliminated from further analysis.

**Figure 8 Cases with No Diagnosis from cattle submitted to the ADL**




There were a number of cattle in each age group that did not receive a diagnosis. One of the 41 calves between 1 and 7 days did not receive a diagnosis. Four out of the 28 calves between the ages of 8 and 14 days were not diagnosed. In the 15 to 30 day age group, 5 out of the 22 were not diagnosed. One of the 14 in the 31 to 90 day group did not get a diagnosis. All animals between the ages of 90 and 180 days were diagnosed. Of the animals 181 to 365 days old, 1 was not diagnosed. Nine of the 13 animals over a year old were not diagnosed. The linear trend line created depicted an increase in non-diagnosed cases with increasing age. There was a 43% correlation (Figure 8.)

## Alternate Age Division

Although there was no statistical difference in the causative agents across the age groups used in this study, if the data are divided into animals that are less than 90 days and those older than 90 days, the results do become significant for some agents. In this case, *E. coli*, cryptosporidium and *C. perfringens* were found to be more common in cattle less than 90 days of age at a significant level when evaluated with a T-test (Table 3 and Figure 9.) This result is expected and supported by literature stating that *E. coli*, cryptostoridium and *C. perfringes* are most commonly pathogens of calves. This finding would have supported the hypothesis had the age groups originally been divided in this way.

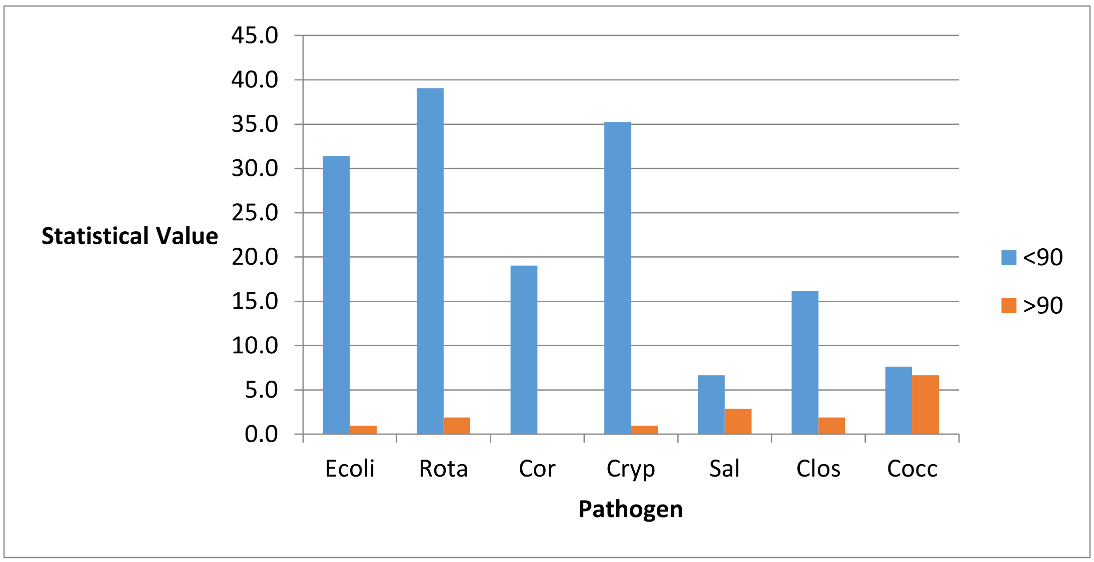
**Table 3 Association of Age and Pathogen: >90 Days and <90 days**

| Age (days) | *E. coli* | Rotavirus | Coronavirus | Cryptosporidium | Salmonella | *C. Perfringens* | Coccidia |
|---|---|---|---|---|---|---|---|
| <90 | 31.4 | 39.0 | 19.0 | 35.2 | 6.7 | 16.2 | 7.6 |
| >90 | 1.0 | 1.9 | 0.0 | 1.0 | 2.9 | 1.9 | 6.7 |
| Student's t-test (p-value <0.05)) | 3.363* (0.021) | 2.227 (0.056) | No Data | 3.329* (0.022) | 0.878 (0.209) | 2.769* (0.034) | 0.164 (0.439) |
| Significance | yes | no | unavailable | yes | no | yes | no |

When reexamined in these new age groups, rotavirus, salmonella and coccidia were still found to have no significant differences between groups. Due to the lack of any animals positive for coronavirus older than 90 days, there was no significance information available

Though rotavirus was found to not have a significant difference in commonality between age groups (p value of 0.056) a significant difference may have been detected if the sample size was larger. Though the incidence of salmonella was not significant, this was not entirely unexpected. Salmonella is a primary pathogen of all ages, meaning that there likely would never be a significant commonality between any age group. Results of coccidia were found to be not significant. However, coccidia were overall one of the least commonly found pathogens. So, it likely would have required an even larger sample size to glean statistically significant results.

**Figure 9 Comparisons of pathogens between cattle <90 and >90 days of age submitted to the ADL**



# Chapter 4

## Discussion

While some trends indicating differences in pathogens between the age groups were apparent, the data was found overall to be not significant (table 3.) This finding can primarily be attributed to the retrospective nature of the study.  Many of these aspects would ideally be addressed if this protocol were to be repeated in a prospective manner.

The small and uneven sample population is largely responsible for the lack of statistical significance of the data. Since there were considerably more young animals, the value of the findings in the younger age groups are greater in that the volume of samples provides a better statistical view of potential causes of diarrhea. However, there were so many fewer animals in the older age groups that it is not possible to make a valid comparison. If performing this study in a proactive manner, more data would be requested to increase overall sample size and ensure that each age group was equally represented. This would enable identification of statistically significant outcomes.

However, it could be argued that the lack of adult data provides information in and of itself. All cases examined were only from animals that had samples submitted to ADL due to diarrhea. The lack of adult data may indicate that diarrhea is simply less common in adults, since there were so many less samples submitted in the same 21-month time period. This could be a fair conclusion, considering that the majority of pathogens studied were indeed associated with age infecting predominantly calves.

Additionally, there were factors within each tested disease that played a role in the findings that resulted in data that was not significant.

*Escherichia coli*

As discussed in the introduction, *E. coli* is generally part of normal gut flora. However, it is evident in Figure 2 that it was not found in all animals. It is known that it is generally only a pathogen in calves. For this reason, most samples from older animals brought into ADL were not evaluated for the presence of *E. coli* resulting in no positive diagnoses being found. If the study were to be repeated, testing of all specimens for all pathogens would result in more sound representation of the incidence of the agents across age groups.

Also, it is important to note that nearly all cattle older than 8 days of age diagnosed with *E. coli* had concurrent infections with other known pathogens. The significance of this fact is two fold. First, knowing that *E. coli* is a part of natural gastrointestinal flora, it is realized that it is always present. When examining a culture to determine if *E. coli* could be a pathogen in that animal, the degree to which it grows is evaluated. Consequently, errors in laboratory techniques or simply differences in interpreting the heaviness of the growth on the plates can greatly affect the test results. So, while the animals may be reported to be positive for *E. coli* it may be that diarrhea is caused by one of the other pathogens and the *E. coli* cultured is an incidental finding. Second, if an animal is infected by another pathogen then their immune system is compromised and an animal with compromised immunity will be more susceptible to pathogens like *E. coli*.

Not all varieties of *E.coli* have virulence properties that impart the isolate with pathogenic abilities. As mentioned in materials and methods, virulence testing is available but was not used in all samples incorporated into this study. So, it is not possible to know if these isolates of *E.coli* were necessarily pathogenic.

*Rotavirus*

The significance of the rotavirus testing results were also likely affected by the retrospective nature of this study. As mentioned in the introduction, rotavirus primarily infects the small intestine. So, if the tissue samples were submitted rather than feces and the tissues did not include small intestine, rotavirus could be missed in a positive animal.

It is important to note that the vaccination history of the animals was unknown and the presence of antibodies resulting from a rotavirus vaccination may yield a positive test result although the animal is not actually infected with the pathogen.

*Coronavirus*

Unlike rotavirus, coronavirus resides in both the small and large intestine. This means that both tissue samples and fecal samples would be an accurate way to test for its presence. However, like rotavirus, the vaccination history was unknown and some positive results could potentially be attributed to previous exposure through vaccination with a modified live vaccine.

*Cryptosporidiosis*

Literature states that cryptosporidiosis is most often contracted by calves between the ages of 7 and 21 days. However, it is also discussed that the pathogenic nature of cryptosporidium makes relapsing common. Since the health histories of these animals are unknown, this could potentially play a role in the reason for a few older calves testing positive. In a repeat of the study, complete health histories of the animals would be obtained.

Also, as calves get older they develop increasing resistance to *Cryptosporidium* infection. So, while an animal may test positive for cryptosporidium, their diarrhea may be caused by another agent or issue.

*Clostridium perfringens*

As referenced earlier, *C. perfringens* is most often found in calves under the age of 2 weeks old. The few calves older than 15 days that were diagnosed were less anticipated, yet due to the infrequencies of diagnoses in these more advanced age groups, understanding of the disease might grant some explanation. *C. perfringens* is often part of the natural flora of the gastrointestinal system, much like *E. coli.* So, the presence of the bacteria on the culture does not always indicate its presence as a pathogen. This is especially meaningful in this sample set since the majority of animals older than 15 days that had *C. perfringens* also had accompanying diagnoses. Making it a possibility that the *C. perfringens* was not a pathogenic cause of the diarrhea and simply an incidental finding.

*Undetermined Causes*

One common diarrhea-causing pathogen that was not found in this sample set was Bovine Viral Diarrhea Virus. However, only two animals were tested due to the fact that, at the ADL, this test requires a tissue or serum sample and many of these cases only had a fecal sample available. Given this, BVD may have been present in some cases of diarrhea but would not have been detected. As shown in Figure 9, a few cases in each age group did not lead to any definitive diagnosis, bringing up the important aspect of causes that could not be addressed by this study. There are factors other than pathogenic agents that can lead to diarrhea including stress, feeding changes and nutritional imbalances. In neonates separation from the dam, change in milk replace or poor environmental conditions could serve as stressors that may lead to development of diarrhea. In adults and neonates alike management changes, nutritional changes, poor ventilation and inadequate shelter could cause stress resulting in gastrointestinal upset that manifests as diarrhea. It is also critical to note that any one of these undeterminable causes could also have played a part in the diarrhea of animals that did have a pathogenic diagnosis.

It is evident that there were more undiagnosed cases in the older age groups. This could potentially be explained by increased stress from management that the animals experience as they reach production age. Especially in the case of dairy cattle, first calving and the accompanying diet and management changes could cause intermittent diarrhea. Yet, it was unknown if the animals were dairy or beef, in a repeat of the study this information would be advantageous.

# Chapter 5

## Conclusion

Due to the retrospective nature of the study, the significance of the results was negatively impacted by lack of complete histories, lack of control over testing variables, overall lack of adequate sample size and uneven numerical representation within each age group. Although the results were not significant for the age classes chosen, overall trends indicate that if the study were repeated with attention to increasing sample size and controlled testing design there is potential to find some significance differences between causes of diarrhea in cattle and their age. Appropriate improvements would need to be made, and the study would likely find success if produced in a prospective manner. This information could continue to prove helpful to herd managers and owners alike when attempting to best manage the health of their animals, ensuring that both excellent welfare and profitability are maintained.

# Appendix

## Percent Isolation of Pathogens Associated with Diarrhea in Cattle

| Age (days) | Escherichia Coli | Rotavirus | Coronavirus | Cryptosporidiosis | Salmonellosis | Clostridium Perfringens | Coccidiosis | Strongylosis | Giardia | Johne's Disease | Ruminitis | Bovine Viral Diarrhea Virus | No Diagnosis |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1-7 | 68.2 (28/41) | 46.3 (19/41) | 12.1 (5/41) | 36.6 (15/41) | 4.9 (2/41) | 17.1 (7/41) | 0 (0/41) | 0 (0/41) | 0 (0/41) | 0 (0/41) | 0 (0/41) | 0 (0/41) | 2.4 (1/41) |
| 8-14 | 50 (14/28) | 57.1 (16/28) | 25 (7/28) | 42.9 (12/28) | 10.7 (3/28) | 17.9 (5.28) | 0 (0/28) | 0 (0/28) | 10.7 (3/28) | 0 (0/28) | 0 (0/28) | 0 (0/28) | 14.3 (0/28) |
| 15-30 | 50 (11/22) | 18.2 (4/22) | 18.2 (4/22) | 31.8 (7/22) | 9.1 (2/22) | 4.6 (1/22) | 0 (0/22) | 4.6 (1/22) | 0 (0/22) | 0 (0/22) | 0 (0/22) | 0 (0/22) | 22.7 (5/22) |
| 31-90 | 57.1 (8/14) | 14.3 (2/14) | 28.6 (4/14) | 21.4 (3/14) | 0 (0/14) | 28.6 (4.14) | 57.4 (8/14) | 0 (0/14) | 0 (0/14) | 0 (0/14) | 0 (0/14) | 0 (0/14) | 7.1 (1/14) |
| 91-180 | 0 (0/3) | 0 (0/3) | 0 (0/3) | 0 (0/3) | 0 (0/3) | 33.3 (1/3) | 100 (3/3) | 0 (0/3) | 0 (0/3) | 0 (0/3) | 0 (0/3) | 0 (0/3) | 0 (0/3) |
| 181-365 | 25 (1/4) | 50 (2/4) | 0 (0/4) | 25 (1/4) | 25 (1/4) | 25 (1/4) | 50 (2/4) | 25 (1/4) | 0 (0/4) | 0 (0/4) | 0 (0/4) | 0 (0/4) | 25 (1/4) |
| >365 | 0 (0/13) | 0 (0/13) | 0 (0/13) | 0 (0/13) | 15.4 (2/13) | 0 (0/13) | 15.4 (2/13) | 0 (0/13) | 0 (0/13) | 15.4 (2/13) | 0 (0/13) | 0 (0/13) | 69.2 (9/13) |

**BIBLIOGRAPHY**

Araque, J. 2006. Diarrhea in Calves Induced by *Cryptosporidium parvum.* Summer 2006 Newsletter. ADDL Purdue University. West Lafayette, IN. Web.

APHIS. 2007. Bovine Viral Diarrhea Virus Info Sheet. Veterinary Services Centers for Epidemiology and Animal Health. USDA. Fort Collins, CO.

Bartels, C., M. Holzhauer, R. Jorritsma, W. Swart, and T. Lam. 2010. Prevalence, prediction and risk factors of enteropathogens in normal and non-normal faeces of young Dutch Dairy calves. Preventive Veterinary Medicine. 93(2-3):162-169.

Caserta, M. 2016. Rotavirus Infection. Merck Manual. Merck & Co. Inc. Kenilworth, NJ. Web.

Cavanagh, D. 2005. Coronaviruses in poultry and other birds. Avian Pathology. 34(6):439-48.

Constable, P. 2015a. Coccidiosis of Cattle. The Merck Veterinary Manual. Kenilworth, N.J. Web.

Constable, P. 2015b. Overview of Coccidiosis. The Merck Veterinary Manual. Kenlworth, N.J. Web.

CDC. Center for Disease Control and Prevention. E.coli (*Escherichia coli)* General Information. Atlanta, GA. Web.

Daly, R. and L. Rotert. 2007. *Clostridium perfringens* Infections in Baby Calves. Extension Extra: South Dakota Cooperative Extension Service. South Dakota State University College of Agriculture and Biological Sciences and USDA.

Divers, T. J. 2013. Non-Nutritional Causes of Diarrhea in Adult Dairy Cattle. Department of Clinical Sciences. Cornell University.

Ed, J. and L. Longe. 2013. Escherichia Coli. The Gale Encyclopedia of Environmental Health. Detroit, MI. (1)283-8.

Fox, M. 2014. Gastrointestinal Parasites of Cattle. The Merck Veterinary Manual. Kenilworth, N.J. Web.

Fray, M., Clarke, M., Thomas, L., McCauley, J., and B. Charlston. 1998. Prolonged nasal shedding and viraemia of cytopathogenic bovine virus diarrhea virus in experimental late-onset mucosal disease. Veterinary Research. 143(22):608-11.

Giannella, R. 1996. Medical Microbiology 4th Edition. The University of Texas Medical Branch at Galveston. Galveston, TX.

Goyal, S. and J. Ridpath. 2006. Bovine Viral Diarrhea Virus: Diagnosis, Management and Control. John Wiley and Sons. Hoboken, NJ.

Gruenberg, W. 2014. Intestinal Diseases in Cattle. The Merck Veterinary Manual. Kenilworth, N.J. Web.

Helman, R. G. 2000. Diagnosis and Diseases of the Digestive Tract. The Veterinary Clinics of North America| Food Animal Practice. W. B. Saunders Company. Philadelphia, PA. 16:1.

Hodges, K. and R. Gill. 2010. Infectious diarrhea: Cellular and molecular mechanisms. Gut Microbes. 1(1):4-21.

Hogue, B., King, B., and D. Brian. 1984. Antigenic Relationships Among Proteins of Bovine Coronavirus, Human Respiratory Coronavirus OC43, and Mouse Hepatitis Coronavirus A59. Journal of Virology. 51(2):381-8.

Jeong, J., G. Kim, S. Yoon, S. Park, Y. Kim, C. Sung, O. Jang, S. Shin, H. Koh, B. Lee, C. Lee, M. Kang, H. Kim, N. Park and K. Cho. 2005. Detection and Isolation of Winter

Dysentery Bovine Coronavirus Circulated in Korea during 2002-2004. Journal of Veterinary Medical Science. 67(2): 187-189.

Kehoe, S. and J. Heinrichs. 2005. Electrolytes for Dairy Calves. College of Agriculture Department of Dairy and Animal Science. Penn State Extension. Web.

Kirkwood, C. 2010. Genetic and antigenic diversity of human rotaviruses: potential impact on vaccination programs. The Journal of Infectious Diseases. 202:S43-8.

Lanyon, S., Hill, F., Reichel, M., and J. Brownlie. 2014. Bovine viral diarrhea: Pathogenesis and diagnosis. The Veterinary Journal. 199(2):201-9.

McGurk S. and S. Peek. 2003. Salmonellosis in Cattle: A Review. American Association of Bovine Practitioners. UW-Madison School of Veterinary Medicine.

Meganck, V., G. Hoflack, S. Piepers, and G. Opsomer. 2015. Evaluation of a protocol to reduce the incidence of neonatal calf diarrhea on dairy herds. Preventative Veterinary Medicine.118(1):64-70.

Melancon, J. 1999. *Nematodis* Infections in Cattle. Merial Veterinary Bulletin. Veterinary Professional Services, Merial Limited. Iselin, NJ.

O'Handley, R., A. Buret, T. McAllister, and M. Olson. 2001. Giardiasis in dairy calves: effects of fenbendazole treatment on intestinal structure and function. International Journal for Parasitology. 31(1):73-79.

Petit, L., M, Gilbert, and M. Popoff. 1999. *Clostridium perfringens*: toxinotype and genotype. Trends in Microbiology. 7(3):104-110.

Sheng, H., S. Shringi, K. Baker, S. Minnich, C. Hovde, and T. Besser. 2015. Standardized E. Coli O157:H7 exposure studies in cattke: Evidence that bovine factors do not drive increased summertime colonization. Applied and Environmental Microbiology. AEM.02839-15

Stampfli, H. 2014. *Clostridium difficle* and *C perfringens* Infection. The Merck Veterinary Manual. Kenilworth, N.J. Web.

Sweeney, R. W. 1996. Paratuberculosis (Johne's Disease). The Veterinary Clinics of North America| Food Animal Practice. W. B. Saunders Company. Philadelphia, PA. 12:2.

Vega, C., M. Bok, L. Saif, F. Fernandez, and V. Parreno. 2015. Egg yolk IgY antibodies: A therapeutic intervention against group A rotavirus in calves. Research in Veterinary Science. 103:1-10.

Zajac, A., K. Peterson and H. Burdett. 2014. How to do the Modified McMaster Fecal Egg Counting Procedure. USDA Sustainable Agriculture and Education Program. University of Rhode Island and Virginia-Maryland Regional College of Veterinary Medicine.

Zhang, X., Q. Tan, G. Zhao, J. Ma, W. Zheng, X. Ni, Q. Zhou, and X. Zhu. 2016. Prevalence, Risk Factors and Multilocus Genotyping of *Giardia intestinalis* in Dairy Cattle, Northwest China. Journal of Eukaryotic Microbiology. doi: 10.1111/jeu.12293.

# ACADEMIC VITA

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**Education:**

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*Veterinary Assistant*

- Collected canine and feline blood, fecal and urine samples, conducted preoperative screening, completed dental cleanings monitored animals under anesthesia during surgery.
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**Hagyard Equine Medical Institute, Lexington, KY** January 2015-May 2015
*Veterinary Medical Assistant Intern*

- Provided primary care to horses of all ages with various diagnoses
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**Belpre Animal Clinic, Belpre, OH** May 2014- August 2014
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- Assisted in animal for standard procedures such as blood draws, nail trimming anal gland cleaning, enemas and IV catheter placement
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