The Effect of Mycobacterium avium subspecies paratuberculosis Exposure on Animal Health

Katelyn McSpadden, Kyle Caires & Ricardo Zanella

ABSTRACT

Background: Johne’s disease is an incurable wasting condition that affects ruminant and non-ruminant animals. Each year, Johne’s is responsible for losses in the billions of dollars in the United States cattle industry alone. Mycobacterium avium subspecies paratuberculosis (MAP) is the microorganism responsible for Johne’s disease. MAP can spread very fast among animals, and this patogen has been isolated across the world and in several different animal species including humans. Therefore, MAP is classified as having a major impact on both animal and human health, and therefore the economy. MAP has also been associated with Crohn’s disease in humans, which necessitates great concerns regarding public health. The objective of this literature review is to identify problems and challenges associated with this illness and highlight possible approaches to minimize the economic losses and the incidence of Johne’s, two avenues to reduce human exposure with this pathogen.

Review: Following ingestion and exposure to MAP, the bacterium will infect the host through the ileum and than it will proliferate inside of host-cells; MAP can therefore be considered an intracellular parasite. After infection, this pathogen goes to a latency period that can be from months to several years without causing the presence of clinical signs in the host. This bacteria can cause an inflammatory response in the intestine, decreasing the ability of the animal to absorb nutrients. Depending on the level of infection bacteria strain and the genetic composition of the animal, individuals can become or not infected, if infected they can shed variable levels of MAP into the environment, increasing the exposure to other animals. Thus, it is of importance to eliminate MAP infected animals from herds; aiming to reduce the environment contamination with this bacteria. Several chromosomal regions have been associated and linked with MAP infection in cattle. It is proposed that Johne’s disease has a polygenic effect with multiple genes involved in the process of susceptibility and tolerance to the disease. Selection for animals that are tolerant to Johne’s disease has also been proposed, whereby tolerance was defined as the ratio of MAP tissue infection and MAP fecal shedding. Animals that are shedding low or no levels of MAP in the environment were considered tolerant, thus are preferred in comparison with the ones that are eliminating high levels of MAP. Some positional and functional candidate genes have been identified and explored. The major problem with genetic studies with Johne’s disease is the correct classification of the phenotype. ELISA, PCR and fecal culture are methods of testing for Johne’s disease but variation exists regarding the degree of accuracy and effectiveness for each test, as discussed further within this review.

Discussion: In this study, we presented the importance of preventive control of MAP transmission amongst animals and humans, respectively. Several approaches to reduce the incidence of this illness among animals were evaluated; however the number of infected animals is still increasing annually, especially within dairy herds. Genetic selection for animals that are less susceptible might be one solution to reduce the spread and contamination of other animals with these bacteria. This necessitates the better understanding of the genes involved with host-immune-defense mechanisms for development of an accurate selection method. Questions related to the zoonotic potential of MAP, the causative agent of Johne’s disease, and Crohn’s disease in humans is still of great concern to the population, therefore efforts to control and eradicate this disease are needed.

Keywords: Johne’s disease, Mycobacterium avium subsp. paratuberculosis.
I. INTRODUCTION

Bovine paratuberculosis, also known as Johne’s disease, is a chronic progressive condition caused by a bacterial infection in the gastrointestinal tract [2]. *Mycobacterium avium* subspecies *paratuberculosis* (MAP) is the bacteria responsible for this disease in ruminants as well as many monogastric animals [56]. This bacteria was first described in the late 19th century in European cattle from the work of H. A. Johne and L. Frothingham. However, it was not until the 1900’s that the bacteria was isolated in the US [8]. In Brazil, limited work has been conducted regarding Johne’s disease, but this is an emerging field, and several reports indicate the existence of this condition in cattle, buffaloes, sheep and goats [30, 31, 60].

Johne’s disease is incurable and highly transmissible to other animals especially because MAP bacteria can be shed in the feces and milk of infected animals and easily infects others [7]. The herd prevalence in US dairy herds, in 1997, and was estimated at about 21.6%, and it was estimated at about 68.1% in 2007 [2, 50]. These prevalence levels result in annual economic losses of over $1.5 billion in the dairy industry alone [18, 48]. It is generally believed that younger animals are the most susceptible to MAP infection because their immune system are not fully developed [32].

Johne’s disease in cattle is characterized by chronic diarrhea, decreases in milk production, reproductive failure and significant weight loss [2]. MAP colonizes and proliferates within the ileum the MAP leading to chronic inflammation. As a response to this inflammation, the lining of the ileum becomes thickened leading to: (1) impaired function as nutrients are not properly absorbed and (2) chronic weight loss, despite nutritionally sufficient [51].

MAP infection has also been linked to inflammatory bowel syndrome (IBS) and Crohn’s disease in humans. Although the definitive causative agent for Crohn’s disease has not been identified yet, it is concerning that virtually all known mycobacterial pathogens are transmissible to humans and have the ability to cause disease. Interestingly, higher levels of MAP have been observed in the gastrointestinal tract of Crohn’s disease patients when compared to healthy people [22]. Of concern, if MAP can be transmissible from cattle to humans, milk or meat might be the vehicle. The possibility that the public is being exposed to MAP contaminated products on a daily basis is real and has increased the focus on the human health ramifications Johne’s disease [28]. Conflicting studies have been published related to the efficiency of the pasteurization process in killing the bacteria [44]. This review intends to clarify the problems associated with Johne’s disease and highlight possible approaches that could be used to overcome the incidence of this illness.

II. THE PATHOGEN

A bacterial infection of *Mycobacterium avium* subspecies *paratuberculosis* (MAP) in the ileum of the gastrointestinal tract of ruminants causes Johne’s disease. The immune response associated with the bacterial infection recruits and further releases macrophages to the site of infection to prevent further infection and eliminate MAP via phagocytosis. However, when macrophages fail to kill the bacteria MAP continues to multiply inside of the macrophages as hosts. MAP uses a two part mechanism that inhibits apoptosis in infected macrophages, while simultaneously up-regulating apoptosis in uninfected macrophages [19]. MAP. With MAP still present in the ileum, the immune system elicits an inflammatory response that stimulates macrophages and lymphocytes to release cytokines, an event that leads to thickening of the ileum concomitant with decreases in absorption of nutrients subsequent weight loss and diarrhea [51].

Calves are considered the most susceptible class of animals to the bacterial infection due to having an underdeveloped, or immature, immune system. Several factors including age of the animal at time of infection genetic composition of the animal and immune status will play an important role in determining whether the animal will develop Johne’s disease or not [47]. Although calves may be exposed to the MAP at
younger ages, the clinical presentation of the disease will likely only appear 2-4 years after the exposure [56]. The amount of MAP needed to cause infection is very small; it only takes about $10^3$ CFU/animal to cause MAP infection and it is known that clinically affected cows can shed up to $10^8$ CFU/g [56]. Asymptomatic, subclinical, cows are very hard to detect using traditional methods, like ELISA and fecal culture, but these animals can still shed the bacteria into the environment [50].

A study by Whitlock and Buergelt subdivided the progression of Johne’s disease into four different stages [53].

1) The first stage is a silent infection, in which the animal shows no signs of infection. Detection of disease at this stage can only be done through postmortem tissue culture.

2) The second stage is subclinical disease. These animals are often adult carriers of MAP and show no signs of disease. However, about 15-25% of these animals will test positive for the presence of MAP in feces or MAP antibodies.

3) The third stage is clinical disease. Animals in stage three will show a drop in milk production and weight loss. Histopathological lesions may be present in the ileum or in other parts of the gastrointestinal tract. The majority of stage three animals will test positive for MAP in feces and MAP antibodies.

4) The fourth and final stage is the advanced clinical stage. Progression from stage three to stage four results in the animal becoming more emaciated. Lesions continue to be present in the gastrointestinal tract and in some cases may spread to other parts of the body such as the liver or lymph nodes; followed by death.

Cattle can be exposed to the bacteria in several different ways, including: fecal oral transmission; lactational transmission; prenatal transmission; and cross-species transmissions [32]. The most common mode of transmission is fecal oral transmission. This usually takes place when calves suckle from an under contaminated with feces. Infection can also occur when the calf is born, due to environmental exposure or transmission from dam to MAP offspring through lactation [32]. It is known that up to 35% of dairy cows with clinical signs of Johne’s disease also shed the bacteria into their milk [17] and even though asymptomatic cows shed less MAP, the bacteria MAP was detected in the colostrum of 36% of asymptomatic cows with $>3000$ cfu/g [46]. In support, dams that tested positive for MAP bacteria have a 26.4% risk of producing calves that are infected [32]. MAP has also been found in semen and fetal tissue [24, 11] leading several researchers to suggest that prenatal exposure to MAP may lead to MAP infection.

### III. PREVALENCE AND INCIDENCE

Johne’s disease was first described in the 1800’s [2]. However, it was only in recent years that many dairy producers have become knowledgeable about the disease and made efforts to manage MAP transmission rates. A study using ELISA in 1996 identified that 21.6% of dairies tested were positive for Johne’s disease, having more than 10% of their cattle infected with MAP [2]. U.S. Department of Agriculture’s (USDA) National Animal Health Monitoring System (NAHMS) collected fecal samples from 524 dairies in the US to test for MAP in the environment, identifying 68% of the dairies evaluated as positive for MAP [2].

While this 2007 study estimated herd prevalence at 68.1% in the US, the prevalence throughout the world greatly varies. In Norway, cattle have been almost free of MAP infection [10] whereas herd prevalence in Australia is estimated at about 9-22% [27]. As a result of different testing methods and variable sensitivities, comparing herd prevalence rates between countries can be difficult. In Brazil, work conducted in a dairy Gyr herd identified 50% of animals as being positive for Johne’s disease using fecal culture in comparison to 32.3% when using the ELISA test on the same animals [30]. Interestingly Johne’s disease has been identified in a herd of dairy buffaloes [31].

Incidence rates in cattle are greater within dairy herds in comparison to beef herds [26]. Improved technology and testing methods have increased the ability to detect Johne’s disease in cows and facilitated the selection of animals to be culled. The ability to identify cattle that would have previously gone undetected is one possible explanation for an increase in incidence. Management strategies and Johne’s certification programs aim to decrease incidence of disease; in 2007, 31.7% of operations were using a Johne’s disease control or certification program, compared to only 11.2% in 2002 and 0.9% in 1996 [2].
IV. DETECTION METHODS

When testing for Johne’s disease, there are two approaches that can be taken: test for MAP or MAP antibodies. Some of the methods used to test for MAP include fecal cultures, enzyme linked immunosorbent assays (ELISA) and polymerase chain reactions (PCR). Clinical pathological signs can be found using gross and microscopic evaluation and immunologic markers of infection [32].

It is of great importance to detect the animals that are shedding the bacteria into the environment so these animals can be culled. Fecal culture tests, which have a sensitivity of 42% [54], are currently the best method to detect MAP shedding into the environment. Along those lines, fecal culture is highly regarded as the gold standard for diagnosing sub-clinically infected cattle; however, it is a time consuming and expensive process when compared to alternatives [44].

Enzyme linked immunosorbent assay (ELISA) tests are useful for detecting an immune response. ELISA testing is more effective in animals considered heavy shedders because most light shedders are sub-clinically infected animals and have not developed an overt immune response. In heavy shedders, the sensitivity of ELISA is 75%, compared to only 15% for light shedders [54]. Since ELISA is much less expensive and takes less time than the fecal tests, ELISA is a more practical method of testing. The heavy shedders are the individuals that pose the most risk to the herd and ELISA can identify these animals so they can be culled. Unfortunately, other detection methods like ELISA are unreliable in detecting sub-clinically infected cattle.

Polymerase chain reaction (PCR) testing is effective to identify the presence of MAP in cattle. The differences observed between ELISA and PCR testing is that PCR testing can detect the presence of the bacterial infection before the antibodies form. Another advantage of the PCR testing is its ability to provide quantitative results of infectivity [4]. With such results, producers can distinguish between the cows that are shedding the most and need to be removed from the herd immediately, versus the cows that are only mild or light shedders and are not posing large inherent risks. The cost of PCR testing is still relatively high, especially when compared to the cost of ELISA testing. Table I shows the differences in sensitivity and specificity among those methods of MAP detection.

<table>
<thead>
<tr>
<th>Method</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>ELISA</td>
<td>31.3%</td>
<td>97.8%</td>
<td>[6]</td>
</tr>
<tr>
<td>Fecal culture</td>
<td>42%</td>
<td>100%</td>
<td>[54]</td>
</tr>
<tr>
<td>PCR</td>
<td>70.2%</td>
<td>85.3%</td>
<td>[6]</td>
</tr>
</tbody>
</table>

V. THE DISEASE

The economic impact of Johne’s disease on the dairy industry is estimated at around $1.5 billion per year [18,48]. These costs are a result of premature culling, reduction in milk yield, loss of valuable animals for reproduction, decreased fertility, increased mortality rate and increased susceptibility to other diseases [55]. These factors contribute to a loss of approximately $100 per cow in Johne’s positive herds compared to Johne’s negative herds [35], with greater losses in cows showing clinical signs (between $401 and $959 per cow) of Johne’s disease when compared to subclinical cows (ranging from $123 and $696 per cow) [35].

VI. GENETICS OF JOHNE’S DISEASE

Despite efforts to reduce the incidence of Johne’s disease by elimination of infected animals, the prevalence has not been greatly affected by current methods. Resistance or susceptibility and tolerance to MAP infection have been shown to have a hereditary component in cattle [14,29,57]. Thus, marker assisted selection (MAS) selection presents an important alternative strategy to produce individuals that are not susceptible to MAP infection. Several approaches were used to identify genetic regions involved with susceptibility and tolerance to Johne’s disease [14,42,58,59]. The identification of genetic variants associated with susceptibility to Johne’s disease could also lead to a more accurate genetic selection against susceptible animals [23,58,59].
Candidate genes identified to be involved in the process of MAP infection in animals are shown (Table 2). Interleukins, more specifically IL5, are involved in the humoral immune response to Mycobacterium infections [49]. An animal infected with MAP will stimulate the production of antibodies to fight the bacteria, and as a result of this response, animals will have increased levels of IL5 [9]. Interleukin 5 (IL5) is a cytokine involved in regulating the production and function of B cells [49]. Cattle infected with MAP show an increase in IL5 levels compared to uninfected animals [9]. These lymphocytes are associated with the humoral immune response (HIR) pathway that leads to antibody production, and as the number of MAP antigens in the body increases, a similar up-regulation HIR signaling pathways will initiate production of more antibodies.

### Table 2. Candidate genes associated with susceptibility to Johne's disease.

<table>
<thead>
<tr>
<th>Gene</th>
<th>Gene ID</th>
<th>Gene name</th>
<th>Chromosome</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDN2</td>
<td>319094</td>
<td>Endothelin 2</td>
<td>3</td>
<td>[42, 59]</td>
</tr>
<tr>
<td>SLC11A1</td>
<td>282470</td>
<td>solute carrier family 11 member 1</td>
<td>2</td>
<td>[29]</td>
</tr>
<tr>
<td>TLR1</td>
<td>574090</td>
<td>toll like receptor 1</td>
<td>6</td>
<td>[29]</td>
</tr>
<tr>
<td>TLR2</td>
<td>281534</td>
<td>toll like receptor 2</td>
<td>17</td>
<td>[29]</td>
</tr>
<tr>
<td>TLR4</td>
<td>281536</td>
<td>toll like receptor 4</td>
<td>8</td>
<td>[29]</td>
</tr>
<tr>
<td>VDR/NR1I2</td>
<td>533656</td>
<td>Vitamin D receptor</td>
<td>5</td>
<td>[59]</td>
</tr>
<tr>
<td>CXADR</td>
<td>281733</td>
<td>coxsackie virus and adenovirus receptor</td>
<td>1</td>
<td>[59]</td>
</tr>
<tr>
<td>TGF-β</td>
<td>282089</td>
<td>transforming growth factor beta 1</td>
<td>18</td>
<td>[45]</td>
</tr>
<tr>
<td>IL5</td>
<td>280825</td>
<td>interleukin 5</td>
<td>7</td>
<td>[9]</td>
</tr>
<tr>
<td>IL10</td>
<td>281246</td>
<td>interleukin 10</td>
<td>16</td>
<td>[43]</td>
</tr>
<tr>
<td>IFN-γ</td>
<td>281237</td>
<td>interferon gamma</td>
<td>5</td>
<td>[9]</td>
</tr>
<tr>
<td>NOD2/CARD15</td>
<td>444867</td>
<td>nucleotide binding oligomerization domain 2</td>
<td>18</td>
<td>[36]</td>
</tr>
<tr>
<td>IL1α</td>
<td>281250</td>
<td>interleukin 1 alpha</td>
<td>11</td>
<td>[40]</td>
</tr>
<tr>
<td>IL4</td>
<td>280824</td>
<td>interleukin 4</td>
<td>7</td>
<td>[40]</td>
</tr>
<tr>
<td>IL17</td>
<td>282863</td>
<td>interleukin 17A</td>
<td>23</td>
<td>[40]</td>
</tr>
<tr>
<td>GATA3</td>
<td>505169</td>
<td>GATA binding protein 3</td>
<td>13</td>
<td>[40]</td>
</tr>
<tr>
<td>FoxP3</td>
<td>506053</td>
<td>forkhead box P3</td>
<td>X</td>
<td>[40]</td>
</tr>
<tr>
<td>TRAF1</td>
<td>540944</td>
<td>TNF receptor associated factor 1</td>
<td>8</td>
<td>[1]</td>
</tr>
<tr>
<td>IL6</td>
<td>280826</td>
<td>interleukin 6</td>
<td>4</td>
<td>[9]</td>
</tr>
<tr>
<td>TIMP1</td>
<td>282092</td>
<td>TIMP metalloproteinase inhibitor 1</td>
<td>X</td>
<td>[9]</td>
</tr>
<tr>
<td>TIMP2</td>
<td>282093</td>
<td>TIMP metalloproteinase inhibitor 2</td>
<td>19</td>
<td>[9]</td>
</tr>
<tr>
<td>TNFR1/TRADD</td>
<td>504707</td>
<td>TNFRSF1A associated via death domain</td>
<td>18</td>
<td>[9]</td>
</tr>
<tr>
<td>HIVEP3</td>
<td>509866</td>
<td>human immunodeficiency virus typr 1 enhancer binding protein 3</td>
<td>3</td>
<td>[58]</td>
</tr>
<tr>
<td>IL16</td>
<td>506314</td>
<td>Interleukin 16</td>
<td>21</td>
<td>[42]</td>
</tr>
<tr>
<td>IL12A</td>
<td>281856</td>
<td>Interleukin 12A</td>
<td>1</td>
<td>[16]</td>
</tr>
<tr>
<td>IL12B</td>
<td>281857</td>
<td>Interleukin 12B</td>
<td>7</td>
<td>[16]</td>
</tr>
<tr>
<td>IL18</td>
<td>281249</td>
<td>Interleukin 18, interferon-gamma-inducing factor</td>
<td>15</td>
<td>[16]</td>
</tr>
<tr>
<td>TNF</td>
<td>280943</td>
<td>tumor necrosis factor</td>
<td>23</td>
<td>[16]</td>
</tr>
</tbody>
</table>
Some interleukins such as IL10 interfere with the function and expression of other genes that inhibit or facilitate the infection of cells with MAP. Two of these genes that may be affected by high IL10 levels are IFN-γ and TNF; both genes associated with macrophage up-regulation [12, 13]. If a mutation in the IFN-γ or TNF genes is observed, then macrophages cannot effectively fight the MAP and the animal will be susceptible to infection [33].

Interferon gamma (IFN-γ) is another biomarker that can be important in detecting Johne’s infected cattle. IFN-γ is a type II interferon that can be secreted by B cells as well as CD4+ T helper lymphocytes and CD8+ cytotoxic lymphocytes [3]. IL10 and TGF are both known to negatively regulate IFN-γ levels [41]. IL10 levels in cattle infected with MAP have been found to be elevated [43]. Increased levels of IL10 have a negative effect on production of IFN-γ, which indicates that these MAP infected cattle have low levels of IFN-γ. In the presence of IFN-γ, it is known that macrophages synthesize and secrete pro-inflammatory cytokines [13]. The low level of IFN-γ helps explain why the macrophages are unsuccessful in killing the MAP. Without up regulation, the macrophages do not function effectively enough to destroy the MAP.

Both IL5 and IL10 have genes that have been associated with intestinal immune network for IgA production (Supplementary Figure 1S).

Tumor necrosis factor (TNF) is produced by macrophages or activated B cells, T cells or NK cells [34]. Within the macrophages, TNF works with IFN-γ to help the macrophage eliminate the bacteria through phagocytosis. TNF also aids in inflammatory cell recruitment and in turn regulates the inflammatory response [12]. Some bacteria, such as MAP, are not effectively killed by macrophages there and instead use the macrophage as a host to proliferate. To prevent this occurrence, TNF induces apoptosis of the macrophage [12]. Much like IFN-γ, IL10 is believed to inhibit TNF [43]. Cattle infected with Johne’s disease will show low levels of TNF [52]. Apoptosis will not occur effectively and MAP will continue to proliferate within the macrophages. Supplementary Figure 2S shows the pathway analysis within genes IL5, IL10, TNF and IFN involved with cytokine-cytokine receptor interaction and Supplementary Figure 3S shows the interaction of those same genes in a T cell receptor signaling pathway.

Transcription growth factor beta receptor (TGF-β) is a cytokine produced by macrophages [21]. TGF-β plays a critical role in the synthesis and degradation of the extracellular matrix [39]. In cows showing clinical signs of MAP infection, increased levels of TGF-β were derived [45]. Increased levels of TGF-β have a negative effect and decrease the production of IFN-γ [25]. In turn, high levels of TGF-β likely decrease the bactericidal activity of the macrophages [20].

Endothelins are known to be highly potent vasoconstrictive peptides [37]. Endothelin-2 gene (EDN2) encodes a member of the protein family of secretory vasoconstrictive peptides and functions as a ligand for the endothelin receptors, initiating intracellular signaling events, which can modulate MAP tissue infection. SNPs located near the region of EDN2 were previously identified as being associated with MAP tissue infection [42, 58, 59]. EDN2 has the ability to recruit macrophages to the site of infection and it also play a role in macrophage activation [15]. Macrophages play important roles in the detection and elimination of the pathogenic microorganisms and initiation of an adaptive immune response [5]. Settles and colleagues [42] have identified a genetic region on BTA3 associated with susceptibility to MAP infection in Holstein cattle. This region was located near the EDN2 gene suggesting a possible involvement of this gene with Johne’s disease. A follow up study in Holstein cows confirmed and refined this region using a fine mapping approach in Holstein cows including additional information to the importance of EDN2 and MAP infection [59].

Guanine nucleotide binding protein subunit alpha 12 (GNA12) plays a role in the G protein coupled receptor (GPCR) signaling pathway and following activation, intracellular increases in cAMP and CA2+ levels signal immune cells to cluster and aggregate together [38], Zanella and colleagues identified a genetic region on BTA15 near the gene GNA12 associated with tolerance to Johne’s disease in Holstein cattle; this region was further refined and confirmed in dairy cows [58, 59], supporting the notion that GNA12 is an important positional and functional candidate gene possibly involved with mycobacterial infection in cattle. Supplementary Figure 4S shows the pathway analysis of GNA12 and its involvement with an inflammatory response and apoptosis in Bos taurus.
VII. CONCLUSIONS

Johne’s disease has proven to be a significant animal and human health problem, and thus, mitigating the spread of the disease is very important. Previous approaches attempting to reduce the incidence of bovine paratuberculosis have yielded unsatisfactory results, as observed in the increased incidence of Johne’s disease over the last decade. Vaccination has not yet been proven to prevent infection, but rather has the ability to delay the time when the animal begins to present clinical signs of infection. Therefore, new methodologies to control the spread of MAP are needed. First, we advocate testing for MAP infected animals followed by immediate removal of animals that test positive will help reduce environmental contamination, and possibly transmission between animals. In this strategy, animals need to be tested frequently for this method to be effective, since some tests (e.g.: ELISA) are less reliable for sub-clinically infected animals. Second, use of genetic testing and selection for animals that are not susceptible to or tolerant of this infection is a very optimistic possibility to control Johne’s disease on the horizon. Third, biosecurity measures on farms should be increased especially to avoid cross contamination from wildlife while also quarantining animals from other farms. Collectively, by using new technologies with the available current methodologies, we can reduce the incidence of Johne’s disease in the cattle industry and improve the welfare of both animal and human populations.

Supplementary data. Supplementary data associated with this article can be found on the Acta Scientiae Veterinariae’s website.

Figure 1S. Influence of IL4, IL5, IL6 and IL10 involved with intestinal immune network for production of IgA in Bos taurus.

Figure 2S. Shows the interaction within cytokine-cytokine receptors involved with genes IL2, IL5, IL10, TNF and IFN all genes associated with intestinal inflammatory response.

Figure 3S. Shows the interaction within T cell receptor signaling pathway involved with genes IL5, IL10, TNF and IFN all genes previously associated with intestinal inflammatory response.

Figure 4S. Shows the signaling pathway between MAPK and GNA12 in Bos taurus associated with inflammatory response and apoptosis.

Declaration of interest. The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

REFERENCES


