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Brachyspira pilosicoli: Colitis of growing swine

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Introduction

Recent changes in the structure and management of the US pig industry, including the adoption of more intensive production practices, have led to a decrease in the prevalence of infection by the severely pathogenic *Brachyspira* (formerly *Serpulina*) *hyodysenteriae*, the cause of swine dysentery. Conversely, the newly emerging pathogen *Brachyspira* (formerly *Serpulina*) *pilosicoli* has been recognized as the cause of porcine colonic spirochetosis (PCS). Because of decreased weight gain and the need for medication with specific antimicrobial agents, PCS is now recognized as an important cause of economic losses to swine production worldwide. This paper reviews current information about PCS and on-farm measures for control of *B. pilosicoli* infection.

Worldwide distribution with a broad host range

Infection with *B. pilosicoli* or lesions of colonic spirochetosis have been recorded in a wide range of hosts in addition to swine,[1-13] including human beings,[14-18] non-human primates,[19] dogs,[14,19,20] opossums,[19] commercial chickens, and various species of wild and zoo birds.[15,21] Although valid epidemiologic field investigations to determine the prevalence of *B. pilosicoli* infection of swine have not been done, PCS has been found in Denmark,[22] Finland,[23] Sweden,[8,9] the United Kingdom,[13] Australia,[6] New Zealand,[24] Brazil,[7] Canada,[10,25] and in all major swine producing states in the US.[1,12,25]

The prevalence within individual farms may range between 1% and 50% of grower and finisher pigs.[13] However, bacteriological isolation of intestinal spirochetes is affected by the administration of antimicrobial agents; thus, the results of field surveys might not indicate the true prevalence of PCS.[26] This indirectly is supported by findings of drastically different *B. pilosicoli* infection prevalence rates between farms where pigs are medicated compared to farms where they are not. In Brazil, *B. pilosicoli* was found in fecal specimens collected from growing swine with diarrhea on 1 of 16 farms (6.3%) that fed receiving diets containing antimicrobial agents compared with 10 of 22 farms (45.5%) that did not.[7,26] Similarly, three of four farms (75%) without feed additives in Finland had *B. pilosicoli* whereas *B. pilosicoli* was isolated from only 1 of 20 farms (5%) where pigs were fed 40-50 ppm of carbadox or 10 of 26 farms (38%) where pigs received 40-50 ppm of olaquindox in the feed.[23] In Sweden, *B. pilosicoli* was isolated from pigs on 46.5% of farms without antimicrobial agents, but was absent from pigs on four farms feeding rations containing 100 ppm of olaquindox.[8]

Potential public health significance

The prevalence of colonic spirochetosis among human adults in the US and Europe ranges between 4.5% and 32.2%.[16] In Australia, *B. pilosicoli* has been isolated from aborigines living in the remote northwest and from homosexual men in Sydney, half of them seropositive for human immunodeficiency virus.[17] In Papua New Guinea, infection with *B. pilosicoli* is endemic where 93.6% of the population is infected for a calculated average duration of about four months.[18] Human infection with *B. pilosicoli* is often asymptomatic; however, an healthy adult volunteer that ingested a pure culture of the organism became heavily colonized and developed abdominal discomfort, nausea, and headaches.[15] Because human strains of *B. pilosicoli* are genetically closely related to porcine, canine, avian and non-human primate strains, and cross-species transmission has been shown with laboratory mice, chicks, and pigs,[6,14,27,28] transmission of *B. pilosicoli* from animals to human beings is a concern.

Diarrhea and depression of weight gain

The onset of PCS is characterized by loose stools with the consistency of wet cement which usually occurs within the first weeks after mixing grower aged pigs from different sources.[1,2,8,13] Diarrhea is transient and typically resolves within seven to ten days, and although the infection may persist, diarrhea is uncommon in pigs older than 20 weeks.[6] When there is a concurrent infection with either *Lawsonia intracellularis*, Salmonellae, or *Yersinia pseudotuberculosis*, the disease is more severe.[1,13] Persistent infection causes a reduction in feed efficiency that increases the number of days required to reach market weight.[2-4] Because of the disruption in the flow of pigs,

PCS is a major problem in intensive swine productions with multi-site and all-in/all-out systems.

Mucosal colonization, invasion, and colitis

Multiplication of *B. pilosicoli* in close proximity with the mucosal surface and inside the lumena of colonic crypts is followed by intimate attachment along enterocytes causing effacement of microvilli.[1-4,6,17,19] With time, the surface epithelium becomes attenuated and focally eroded.[1-4,11,13] Because of the presence of bacteria other than spirochetes at the site of epithelial damage, it is believed that the colonic microflora participates in development of PCS lesions.[29] Clinical signs of absorption failure or diarrhea are attributable to massive colonization and disruption of absorptive function below the reserve capacity of the large intestine.

From the damaged epithelial surface, *B. pilosicoli* spread extracellularly in the underlying lamina propria where they are phagocytosed by macrophages and also enter capillary blood vessels.[1,12,27,29] Pigs and human beings naturally-infected with *B. pilosicoli* have groups of intact spirochetes within vacuoles of submucosal colonic macrophages, and until now the significance of these observations was unknown.[1,19,29]

Brachyspira pilosicoli is a facultative intracellular pathogen

Because macrophages are one of the key defenses against enteroinvasive bacterial pathogens and intact *B. pilosicoli* are present within submucosal macrophages, we investigated the interaction of *B. pilosicoli* with macrophages.[30] In contrast to other bacteria that are taken up by conventional phagocytosis, uptake of *B. pilosicoli* by macrophages involves a novel mechanism called coiling phagocytosis. Further studies also have shown that, instead of undergoing normal degradation within macrophage phagolysosomes, *B. pilosicoli* localize and replicate inside the endoplasmic reticulum of infected cells, suggesting altered intracellular trafficking.[31] Because these changes correlate with increasing numbers of viable intracellular *B. pilosicoli* over time, it is likely that survival and replication of *B. pilosicoli* within macrophages plays a role in the development of PCS.

Diet modulates infection and colitis

The pathogenesis of PCS is incompletely understood; however, diet has been suggested as a possible risk factor for infection and clinical disease expression.[32] Field observations suggest higher prevalence of diarrheal disease when feeding pelleted compared with mash rations, and changing diets from a pelleted to a meal form decreases the prevalence of diarrhea on farms with PCS.

We proposed that *B. pilosicoli* infection and colitis are modulated by the microenvironment of the colon which, in turn, is determined by the type and the amount of substrates available for fermentation in the large intestine.[33] To test this hypothesis, we used a soybean hypersensitivity pig model to induce small intestinal malabsorption. Because of an increase in the amount of substrates and bacterial fermentation, pigs with soybean hypersensitivity developed colonic acidosis. Although infection rates were similar between pigs fed the corn- soybean meal-based diet and a control corn-based diet, diarrhea and infection were more persistent and colonic lesions were more widespread in pigs with the diet-induced colonic acidosis. From these observations, it was concluded that changes in the colonic microenvironment predispose for colonization and damage caused by *B. pilosicoli.*

Demonstration of Brachyspira pilosicoli in diagnostic specimens

Animals and humans infected with *B. pilosicoli* have lesions limited to the large intestine.[2-4,6,13,29] The contents of the cecum and spiral colon may be loose and the mucosal surface may have coalescing superficial erosions and adhered fibrinonecrotic exudate or feed particles; however, these changes are not specific to PCS and can be seen with other causes of colitis.[3,5,13,29] Spirochetes are visible on routine histologic examination and Warthin-Starry silver stained tissue sections, but specific demonstration requires immunohistochemical staining with *Brachyspira* species-specific mouse monoclonal antibody,[2,21] or fluorescent ribosomal RNA in situ hybridization.[11] Attachment of *B. pilosicoli* to enterocytes is seen in less than 50% of experimentally infected, non-medicated pigs, and this lesion occurs only within the first three weeks post-inoculation.[2,3,6] In contrast, attachment of spirochetes along the colonic epithelium is seen rarely in field cases of PCS, more often spirochetes are seen inside the lumena of colonic glands which appear dilated and filled with mucus.[13] These differences may be attributable to the transient nature of this lesion or to the patchy distribution of the spirochetal attachment which makes it difficult to identify during routine diagnostic evaluations.

Demonstration of spirochetes by direct examination of fecal smears with fluorescent-labeled antibodies is possible.[34,35] However, fluorescent antibody tests are not highly sensitive and currently these antibodies are available only in certain parts of the world. By contrast, isolation of *B. pilosicoli* from mucosal scrapings of the large intestine, feces, or rectal swabs by anaerobic culture on selective agar media containing blood is widely used.[5,7,14] Because many non-pathogenic commensal spirochetes produce a weak ß-hemolysis like *B. pilosicoli*, additional biochemical tests are necessary before final identification.[7] Provisional identification of *B. pilosicoli* can be done

by demonstration of hippurate hydrolysis.[7,9,12,14] However, definitive identification of *B. pilosicoli* is done by amplification of 16S- or 23S-ribosomal DNA-specific sequences by polymerase chain reaction (PCR).[7,9,12,22]

On-farm control by medication and management changes

Control measures for PCS are similar to those used for swine dysentery. Antimicrobial drugs such as tiamulin, carbadox, and lincomycin administered in the feed or water are effective.[2,5,25] Field isolates obtained from pigs in Canada (n=5) and the United States (n=14) have been susceptible to carbadox and tiamulin, whereas the susceptibility to lincomycin and gentamicin appears more variable. However, a more conclusive determination of the clinical efficacy of these antimicrobial agents would require controlled evaluation with pigs on infected farms, as shown previously with zinc bacitracin.[24]

As indicated earlier, factors predisposing to increased substrate load in the large intestine, including viral- and bacterial-induced small intestinal damage, should be considered when implementing control measures to reduce infection and severity of PCS. Although a change in diet may lead to disappearance of diarrhea, infection might persist and become subclinical.

On the basis of epidemiological studies and animal infection models, transmission of *B. pilosicoli* is likely to be fecal-oral.[1-4,6,13] Pigs become infected by ingestion of spirochetes present in the environment contaminated by clinically and subclinically affected pigs that are shedding the spirochetes in their feces. Therefore, reducing environmental contamination by sanitation and strategic medication can limit transmission among susceptible pigs. Also, mice and birds are potential sources of *B. pilosicoli* infection for pigs.[15,21,28] Thus, control programs for rodents and prevention against the entry of birds should be in place.

In all-in/all-out pig flow systems, thorough cleaning and disinfection between groups in combination with antimicrobial drugs at therapeutic levels in the receiving diet are effective.[2] When control of environmental contamination is less than optimal, a waiting period of 7 to 10 days to allow natural exposure before initiation of pulse medication with therapeutic levels of an effective antimicrobial agent either in the water for 24 hours once per week for a few weeks, or in several alternating batches of feed may be cost-effective.

Natural recovery from experimental infection of conventional weaned pigs can occur within six weeks post-inoculation.[2] Pigs recovered from PCS also have serum IgG antibodies to several *B. pilosicoli* antigens.[36] In contrast, laboratory mice remain persistently infected for up to 30 days in spite of a serum IgG antibody response.[28] Vaccination might provide an alternative strategy for control of PCS; however, an experimental whole-cell bacterin was not protective when administered parenterally.[37]

Future developments

Although PCS was first identified over two decades ago by Taylor and coworkers in the United Kingdom,[7] it is only in the last five years that it has become widely recognized as a contributing factor to reduce performance of swine raised under intensive management practices. As less emphasis is placed on the use of antimicrobial agents in low concentrations to enhance performance, alternative strategies for control of PCS will emerge. Future research on PCS will focus on identifying basic mechanisms of disease and designing improved methods of prevention, including vaccination and management changes.

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