hardness, succulence, and characteristic flavour attributes decreased whereas the bitter taste increased in both LAB applications, this effect of LAB application being significant at the end of storage time (p < 0.05). In general, samples with commercial culture BLC35 addition were considered harder and less succulent than those with Lb. sakei ST153Ch addition, but these differences were not statistically significant. A conformity evaluation of the samples, using a 5-point hedonic scale that allowed perceiving potential defects that were not expressed in the attributes (data not shown) scored all samples above the conformity level (>3) during the 124 days of storage.

Discussion

The antibacterial spectrum of activity of Lb. sakei ST153Ch has been previously studied and results indicated their potential for use in a mixed starter culture for the fermentation of meat products (Todorov et al., 2013). In the present study the best strain with respect to Listeria safety was Lb. sakei ST153Ch. Another study, using the same strains but added to another cured-smoked pork product, reported samples containing BLC35 being harder and less succulent than the ones containing Lb. sakei ST153Ch although the “conformity” attribute was not influenced by the type of starter culture and MAP conditions during 120 days of storage. (Jácome et al., 2014).

Conclusion

Lactobacillus sakei ST153Ch, an autochthonous strain of Portuguese cured-smoked pork products, combined with MAP, can be regarded as an effective tool for increasing safety in ready-to-eat sliced cured-smoked products with respect to Listeria spp.

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References


06. Is competitive exclusion a valuable alternative for classical cleaning and disinfection of pig-growing units?

Luyckx, K.; Millet, S.; Van Weyenberg, S.; Herman, L.; Heyndrickx, M.; Dewulf, J. and De Reu, K.

Abstract

Colonization of the environment of pig-growing units by pathogenic microorganisms is an important factor in development of endemic diseases in pigs and, in spreading of zoonotic diseases. These pathogens are mostly controlled by the use of antibiotics and disinfection during vacancy. Because, the past years an increasing resistance against these measures is noticed, alternative methods such as competitive exclusion (CE) are promoted as promising. In this study the effect of a CE protocol on the bacterial infection in pig-growing units was compared to a classical cleaning and disinfection (C&D) protocol. Tests were performed during three successive production rounds using multiple identical pig-growing units. CE protocol consisted of cleaning (no disinfection) after loading piglets and spraying probiotic bacteria (Bacillus spp. spores) during vacancy and production. The cleaning product also contained Bacillus spores. Sampling was performed at different time-points: immediately after pig loading (manure still present); 24 hours after cleaning (CE units) or after disinfection (control units); after one week and five weeks of production (piglets present). At each time point, swab samples for analyses were taken. Enumerations of bacterial spores, Enterococcus spp., E. coli, fecal coliforms and MRSA and detections of E. coli, fecal coliforms and MRSA were performed. Next to bacterial analyses, also feed conversion and fecal consistency was monitored. This study showed that, although probiotic spores were administered well, the analyzed bacteria were not decreased after three production rounds in CE units and remained on the same level as the control units (C&D). Also, the infection pressure in CE units during vacancy was not as much reduced as after the disinfection-step in control units. Finally, no differences in feed conversion and fecal consistency were found. These results indicate that the used CE protocol is not a valuable alternative for classical C&D.

Introduction

Colonization of the environment of nursery units by pathogenic micro-organisms is an important factor in the persistence and spread of endemic diseases in pigs and zoonotic pathogens. These infections are generally controlled by the use of antibiotics and disinfectants. However, the past years an increasing level of resistance against these measures is noticed (Russell, 1998; Mateu and Martin, 2001; Soumet et al., 2012; Callens et al., 2013) It is important to be informed on its susceptibility to antimicrobial agents. In the current study, the Minimum Inhibitory Concentration (MIC). Wong et al. (2013) which has led to concern about its spread into the community. Disinfectants play an important role in reduction of contamination in both animal husbandry and food-preparation, helping control spread of organisms from foodstuffs, including raw meat. Plasmid-borne antibiotic resistance (AR described the presence of antibiotic (disinfectant) resistance genes in methicillin resistant Staphylococcus aureus sequence type 398 (MRSA ST398). Although the minimum inhibitory concentrations (MIC) and minimum bactericidal concentrations (MBC) of resistant strains remain lower than the recommended working concentrations of disinfectants, there is concern that an impairment of the used disinfectant (e.g. presence of organic material) resulting in exposure to lower active levels of these agents, could lead to selection of more resistant strains harboring these genes (Wong et al., 2013). Randall et al. (2004)“ISBN” 0305-7453 (Print suggested that the use of biocides alone or combined with antibiotic treatment may also increase selective pressure towards antibiotic resistance of Salmonella enterica. Beier et al. (2008) showed
that β-hemolytic enterotoxigenic *E. coli* strains isolated from neonatal pigs, were resistant to chlorhexidine and individual quaternary ammonium monomer compounds (QAC). Some of these isolates had also multiple antibiotic resistance. Because of the ongoing concern over excessive use of biocides and potential resistance development and cross-resistance to clinically important antibiotics, the use of bacterial biocontrol agents has been suggested many times as an alternative method to antagonize the growth of these pathogens. The aim of this study was to compare the effectiveness of a commercial competitive exclusion (CE) protocol with a classical cleaning and disinfection (C&D) protocol in nursery units.

**Materials and methods**

**Management in control and CE units**

This study was carried out in six identical nursery units at the experimental pig farm of ILVO during three successive production rounds. Three units were assigned to the control group (classical C&D protocol) and three to the treatment group (CE protocol). After six weeks, piglets were transported to fattening units and pens were cleaned (and disinfected) according to the test protocols. Classical C&D protocol was carried out in the control units after pig loading. CE protocol in treatment units consisted of the following steps: pens were hosed with cold water under low pressure, foamed with 1.5% PIP AHC (Probiotics In Process Animal House Cleaner, Chrisal, Lommel, Belgium) and rinsed with warm water. PIP AHC consists of cleaning compounds, *Bacillus* spp. spores and enzymes. In CE units, no disinfection was carried out. Besides, during vacancy and production, CE units were sprayed 2 – 3 times a week with pure PIP AHS (Animal Housing Stabilizer, Chrisal) in order to bring and retain biocontrol agents in the stable environment. The used AHC and AHS PIP products contained *Bacillus* spores of five different species in a concentration of 8.5 and 7.5 log CFU/ ml, respectively. Both protocols were carried out according to the manufacturers guidelines.

**Sampling scheme**

Sampling was performed at different time points (i.e. sampling moments):

- Immediately after pig loading, but before cleaning (manure still present) (BC)
- 24 hours after cleaning (CE units) (AC) or 24 hours after disinfection (control units) (AD)
- After one week of production (piglets present) (W1)
- After five weeks of production (piglets present) (W5)

Five locations per pen and three pens per compartment were sampled at each time point with premoistened sponge swab samples. A surface of 625 cm² (i.e. A4 format) was swabbed whenever possible. Since the surface of the drinking nipples was smaller than 625 cm², two drinking nipples per pen were sampled.

**Sample processing**

Swab samples were analyzed for *Enterococcus* spp., *E. coli*, fecal coliforms and MRSA enumerations. Besides, spore enumerations were carried out, in order to test if *Bacillus* spp. spores from the PIP products were well distributed and sufficiently present in pens. After overnight incubation of the swab samples, detection of MRSA and *E. coli*, fecal coliforms and *Salmonella* was carried out.

**Other Analyses**

Next to bacterial analyses, feed conversion ratio of every pen was calculated. In addition, faecal consistency was evaluated according to Pedersen and Toft (2011): a score from 1 (no diarrhea) to 4 (serious diarrhea) was assigned per pen. Finally, the number of treatment days per 100 risk days or TD100 was calculated per pen for each protocol. This was done by calculating the ratio of treatments days (the number of days that piglets were treated with antibiotics) and the number of days at risk (the time that pigs could be exposed to antibiotics). This ratio was then multiplied by 100 to determine or TD100.

**Results**

**Spores**

At every sampling moment and at each production round, higher spore enumerations were found for CE units compared to control units (P< 0.01). Besides, spore-enumerations increased after every round in CE units (P< 0.01).

**Enterococcus spp.**

After disinfection of the control units, lower *Enterococcus* spp. enumerations were observed compared to cleaned CE units (P< 0.01). The mean difference was 2.28 log CFU/ sampling surface. Microbial cleaning in CE units caused a reduction of 0.70 log CFU/ sampling surface, while in control units a reduction of 3.24 log CFU/ sampling surface was noticed after disinfection. During production (piglets present) and before cleaning, no differences in *Enterococcus* spp. enumerations were found between units.

**E. coli, fecal coliforms and MRSA.**

More *E. coli* countable samples were found for CE units after cleaning compared to control units after disinfection (P< 0.01). Proportion of countable samples was reduced by 9% after cleaning of CE units, while a reduction of 41% was obtained after disinfection of control units. During production and before cleaning, no differences were found in amount of countable *E. coli* samples between both types of units. Same observations were made for fecal coliforms and MRSA enumerations and detections results.

**Performance results.**

No significant differences in mean feed conversion and scores of fecal consistency were found between piglets raised in CE and control units.

**Antibiotic treatment**

No significant differences in mean TD100 were found between protocols.

**Discussion**

Several hypotheses have been proposed to explain the mechanisms of CE cultures. One is that CE bacteria should compete with other bacteria for nutrients and energy, which results in preventing growth and proliferation of pathogenic bacteria in the environment (Cummings and Macfarlane, 1997). Another hypothesis is that these bacteria influence the quorum sensing communication and therefore inhibit expression of virulence and colonization genes of pathogens (Vila et al., 2010). Besides CE bacteria, also enzymes are administered during cleaning, that would help to eliminate biofilms. In this study, no reduction of the analyzed bacteria after three production rounds in CE units was seen.

**E. coli** indicator for *Salmonella* and MRSA analyses showed that the infection pressure was not reduced to the same extent as implementing a disinfection step. In addition, during production no differences were noticed. Also no improvement in hygiene was seen: during production no differences were found in *Enterococcus* spp. (hygiene indicator) and fecal coliforms (fecal indicator) contamination between the two types of units. Improvement of feed conversion efficiency by probiotic type bacteria could be obtained by a shift in intestinal flora, stimulating growth of nonpathogenic facultative anaerobic and Gram positive bacteria forming lactic.
that β-hemolytic enterotoxigenic E. coli strains isolated from neonatal pigs, were resistant to chlorhexidine and individual quaternary ammonium compounds (QAC). Some of these isolates had also multiple antibiotic resistance. Because of the ongoing concern over excessive use of biocides and potential resistance development and cross-resistance to clinically important antibiotics, the use of bacterial biocontrol agents has been suggested many times as an alternative method to antagonize the growth of these pathogens. The aim of this study was to compare the effectiveness of a commercial competitive exclusion (CE) protocol with a classical cleaning and disinfection (C&D) protocol in nursery units.

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acid and hydrogen peroxide, inhibiting growth of pathogens, and enhancement of digestion and utilization of nutrients (Lutful Kabir, 2009). However, no differences were found between piglets raised in CE and control units in our study. In addition, no differences in fecal consistency was noticed. In addition, no differences in TD100 were found. A possible explanation for not obtaining the claimed effect of reducing the microbiological infection pressure and improved feed conversion might be the too low administered dose of CE bacteria resulting in the need to revise the product compositions and application protocols. On the other hand, there is also the possibility that CE is not a valuable alternative for classical cleaning and disinfection.

Conclusion

Very few studies about the impact of microbial cleaning and administration during production on the environment in animal houses is available. Our results showed that competitive exclusion by probiotic type bacteria could not meet the claims provided by the manufacturer. Moreover, this study showed that a good cleaning and disinfection protocol during vacancy is very important for reducing infection pressure in nursery units. However, more research should be carried out for a valuable alternative, because disinfectant resistance might be an upcoming problem.

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References


Introduction

Ireland has a high prevalence of Salmonella contamination on pork carcasses (20%) (EFSA, 2008) and this is likely related to a high level of Salmonella carriage in some pig herds within the country (Burns et al., 2013). This highlighted a need to find measures to control Salmonella shedding in pigs at primary production, especially in finishing pigs, as they are a significant source of Salmonella in the abattoir. Dietary supplementation with organic acids has shown promise in controlling Salmonella in pigs (Friendship et al., 2006). After a critical review of the literature, sodium butyrate was selected for further evaluation. Sodium butyrate acts via down-regulation of expression of hKIA (an invasion gene) in Salmonella, thereby suppressing its ability to invade porcine intestinal epithelial cells, which in-turn decreases faecal shedding of the bacterium in pigs (Buyen et al., 2008). This study investigated the ability of dietary supplementation with sodium butyrate during the last month of growth pre-slaughter to reduce faecal shedding and intestinal carriage of Salmonella in finishing pigs.

Material and Methods

(a) Farms

Farm A had a historically high Salmonella seroprevalence (according to the Irish National Pig Salmonella Control Programme) i.e. > 60%; however, prior to commencement of the trial, the seroprevalence declined to 0%. Thus, artificial contamination of pens using a Salmonella strain recovered from sows on the farm during the same time period was performed in advance of the feeding study. This strain was a monophasic variant of S. Typhimurium (4,5,12:i:-), and its antimicrobial resistance pattern was determined to be A5SuT. Farm B, on which pigs had been shown to carry S. Typhimurium, also had a historically high Salmonella seroprevalence (i.e. > 50%).

(b) Animal Housing and Diets

Farm A had 14 pens with a total of 168 pigs (72 males and 96 females, with 12 same gender pigs/pen), 220

07. Evaluating the effectiveness of a sodium butyrate feed additive for the control of Salmonella carriage in finishing pigs.


Abstract

This study aimed to investigate the effectiveness of commercially available sodium butyrate to control the shedding of Salmonella on two Irish pig farms with a history of high Salmonella seroprevalence. On both farms, pens (12–17 pigs/pen) were randomly assigned to a control (finisher feed without additives) or an acid treatment (the same feed supplemented with 0.03% sodium butyrate) for 24–26 days prior to slaughter. On Farm A, Salmonella shedding was reduced in the acid group compared to the control group at the end of the treatment period (30% vs. 57% probability of detecting Salmonella in faeces, respectively; p<0.05). However no effect of treatment was observed on Farm B, which could perhaps be explained by a concomitant infection by Lawsonia intracellularis. No significant differences in Salmonella recovery rates were observed from caecal digesta or ileocecal/omentum lymph nodes collected at slaughter in either of the trials. Furthermore, feed intake, weight gain and feed conversion efficiency did not differ significantly between control and treatment groups on either farm.