Alternatives to antibiotic growth promoters in prevention of diarrhoea in weaned piglets: a review

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ABSTRACT: The weaning time is a crucial period in the management of piglets. The risk of development of post-weaning diarrhoea (PWD) in piglets is high. PWD is the cause of serious economic losses in pig herds. Since 2006, the use of antibiotic growth promoters for prevention of diarrhoeal diseases in piglets has been banned. This measure also led to the investigation of alternative suitable feed supplements that would be reasonably efficient in protecting and sustaining animal health and performance. Various natural materials such as probiotics, prebiotics, organic acids, zinc and plant extracts have been tested as effective alternatives to antibiotics. Recently, owing to their high adsorption capacity, research efforts have been conducted on the application of natural clays and clay-based feed supplements. The purpose of this review is to summarize the effect of different alternative components as growth promoters on the health and performance of weaned and growing piglets.

Keywords: infection; swine; antibiotics; intestinal microflora; performance; probiotics; prebiotics, organic acids; zinc; phytobiotics; aluminosilicate

List of abbreviations

CFU = colony-forming unit; **ETEC** = enterotoxigenic *Escherichia coli*; **MMT** = montmorillonite; **PWD** = post weaning diarrhoea; **SCFA** = short chain fatty acids; **VFA** = volatile fatty acids

Contents

1. Introduction
2. Ban of antibiotic growth promoters
3. Intestinal microflora in piglets
4. Improvement of intestinal microflora
   4.1. Probiotics
   4.2. Prebiotics
   4.3. Synbiotics
   4.4. Organic acids
   4.5. Zinc oxide
   4.6. Plant extracts
5. Clay minerals
   5.1. Adsorption capacity and effects of clay minerals
   5.2. Modified forms of clay minerals
6. Conclusions
7. References

1. Introduction

Postweaning diarrhoea (PWD) is one of the most frequent causes of heavy economic losses in pig herds. PWD can be caused by a number of causative agents. The major bacteria which cause diarrhoea are *Escherichia coli* and members of the genera *Clostridium*, *Lawsonia* and *Brachyspira.* Regarding viral agents, rotaviruses (Bertschinger, 1999; Jung et al., 2006a,b; Thomsson et al., 2008), coronavi-

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ruses (Moller et al., 1998; Song et al., 2006), transmissive gastroenteritis virus (Hampson et al., 1987; Alexa et al., 1995, 2001; Chae et al., 2000; Madec et al., 2000; Melin et al., 2004; Song et al., 2006; Thomsson et al., 2008) and others have been most frequently diagnosed.

Early-weaned piglets are exposed to several stress factors, with nutrition, etiology and indoor environment of housing being particularly implicated (Madec et al., 2000; Laine et al., 2008). Non-infectious stress factors which are involved in the development of gastroenteric disorders are:

- age of piglets when they are weaned from their dam (Svensmark et al., 1989; Skirrow et al., 1997),
- sudden change of feed from sow milk that provides piglets with immunoglobulins (Bailey et al., 1992),
- irregular feed intake (Bark et al., 1986; Spencer and Howell, 1989; Svensmark et al., 1989; Madec et al., 1998; Laine et al., 2008),
- feed structure (Amezcua et al., 2002),
- animal hygiene and housing conditions (Le Dividich, 1981; Le Dividich and Herpin, 1994; Madec et al., 1998),
- inadequate feeder space per piglet in the pen (Amezcua et al., 2002).

Numerous changes in the early-weaned piglet body can initiate PWD, such as morphological (Hampson, 1986) and functional alterations of the small intestine (Kidder and Manners, 1980; Hampson and Kidder, 1986), changes in intestinal colonization with predominance of *E. coli* (Melin et al., 1997; Katouli et al., 1999) and weakening of the immune system (Blecha et al., 1983; Wattrang et al., 1998).

2. Ban of antibiotic growth promoters

Regarding the fact that weaning greatly affects general health condition of piglets, it is necessary to stimulate the indigenous intestinal microflora and keep it well-balanced because it provides protection to animals against invasion by pathogenic microorganisms (Jensen, 1998; Katouli et al., 1999). Before 2006, health strategies widely used antibiotic growth promoters to reduce enteric infections and the occurrence of pathogens able to adhere to intestinal mucosa (Barton, 2000; Budino et al., 2005). These are added to feed for piglets from birth to weaning with the aim of improving the composition of intestinal microflora in piglets and thus ameliorate the potential consequences of PWD (Sorensen et al., 2009).

The increased use of antibiotics has given rise to a fear of the development of resistant pathogenic bacterial strains (Wegener et al., 1998; Kyriakis et al., 1999; Budino et al., 2005) and residual contamination of the food chain with antibiotics (Chen et al., 2005; Roselli et al., 2005). This has led to the adoption of safety measures and a gradual withdrawal of antibiotic promoters from pig diets. In 2006, the use of antibiotics as growth promoters was forbidden in the EU (Castro, 2005; Chen et al., 2005).

Due to the fact that the use of antibiotics as growth promoters has been banned and that the expansion of this policy to other countries can now be expected, intensive research has focused on the development of alternative strategies with the aim of maintenance of animal health and performance (He et al., 2002; Castro, 2005; Chen et al., 2005; Bomba et al., 2006; Castillo et al., 2008). Various natural materials, many of which are commercially available, have been investigated as efficient alternatives to antibiotic growth promoters. At present, probiotics, prebiotics, organic acids, enzymes, phytobiotics, clay adsorbents and others are under investigation. These materials can exert beneficial effects on the microflora composition and consequently affect animal health and growth performance.

3. Intestinal microflora in piglets

Intestinal microflora contains high numbers of various species of bacteria involved in the process of digestion. Microbial composition is determined by mutual interactions between the host and the microorganisms, and also among different microorganisms. These factors are designated as autogenic (Fuller et al., 1978; Jensen, 1998; Budino et al., 2005). On the other hand, pH in the stomach, digestive enzymes, intestinal peristalsis, nutrients and immunity of the host are termed allogenic factors (Jensen, 1998; Budino et al., 2005; Roselli et al., 2005). The predominance of beneficial species of microorganisms over pathogens is essential for stability of the immune system of the intestines and consequently of the entire body (Apgar et al., 1993; Makala et al., 2000; Mikkelsen et al., 2003).

Enteric bacteria constitute a wide range of mostly strictly anaerobic or facultatively anaerobic species with counts giving an estimate of 10^{11} to 10^{14} CFU/g of digesta (Ghnassia, 1979; Makala et al., 2000). Ducluzeau (1983) reported that numbers of microorganisms can reach values of between 10^8 and 10^9 CFU/g of faeces already 10 to 12 h after the birth of piglets and that their numbers stabilize within 24 to 48 hours after birth. However, the composition of microflora is not definitive. It develops gradually and numerous changes occur during weaning (Rojas and Conway, 1996; Mikkelsen, 2003; Roselli et al., 2005).

The highest number of microorganisms is found in the caudal part of the intestines (Table 1), where around 500 different species of microorganisms have been described and identified to date (Rojas and Conway, 1996; Budino et al., 2005). In a well-balanced microbial environment, members of the following genera prevail: *Streptococcus, Lactobacillus, Bifidobacterium, Enterococcus, Eubacterium, Fusobacterium, Peptostreptococcus, Enterobacter, Bacteroides, Porphyromona,* while the numbers of coliform bacteria *E. coli* and *Clostridium* sp. are lower (Fuller et al., 1978; Maxvell et al., 2004; Stokes et al., 2004; Budino et al., 2005; Pasca et al., 2009). Due to a relatively high pH value in the stomach, members of the genus *Lactobacillus* are more commonly found in the non-secretory part of the stomach, from which they further spread to the small intestine (McAllister et al., 1979; Rojas and Conway, 1996).

Anaerobic conditions, favourable temperature, pH and slow passage of the digesta are the preconditions for the presence of large numbers of bacteria in the large intestine and caecum, up to 10^{10} CFU/g (Kidder and Manners, 1980; Ambroz, 1981; Mikkelsen et al., 2003). In the caecum Gram-negative bacteria predominate, whilst these are outnumbered by Gram-positive bacteria in the colon (Robinson et al., 1981; Rojas and Conway, 1996; Mikkelsen et al., 2003).

Intestinal microorganisms participate in various physiological functions, by which they influence their hosts (Jensen, 1998). Protection against pathogenic and conditionally pathogenic microorganisms in the form of colonization resistance is most important (Gedek, 1992a; Jin et al., 1997; Roselli et al., 2005). Natural intestinal microflora adhere to intestinal mucosa and inhibit colonization by pathogens. This inhibition effect is caused by competition for nutrients and binding sites, bacteriocin production, lactobacilli fermentations and short chain fatty acid production (Teitelbaum and Walker, 2002; Roselli et al., 2005). Subsequently, immunological memory is created and intestinal immunity develops (Jin et al., 1997; Makala et al., 2000; Stokes et al., 2004).

Even though avirulent strains of *E. coli* are a common constituent of intestinal microflora, pathogenic strains of the same bacteria are the most frequent causes of PWD in piglets (Madej et al., 1999; Madec et al., 2000; Melin et al., 2004). The acquiring of virulence factors happens by the enrichment of some *E. coli* strains through special mobile genes and with the joint influence of bacteriophages or plasmids. Mobile genes are localized in pathogenicity islands. Pathogenic strains are endowed with at least six virulence factors: shiga toxin, adhesion factor intimin, hemolyzin, serin protease, thermostable enterotoxin and special katalase system (Alexa et al., 2001; Hayashi et al., 2001; Sousa, 2006).

4. Improvement of intestinal microflora

The health of organisms largely depends on the composition of the intestinal microflora. Its composition and function can be beneficially influenced by many factors. These factors can be significantly supported by probiotics, prebiotics, organic acids, zinc oxide and plant extracts.

Table 1. Comparison of microflora in the stomach, small and large intestines (CFU $\log_{10}$/g chyme; Fuller, 1989; Rojas and Conway, 1996; Stokes et al., 2004)

Microorganisms	Microflora in the stomach	Microflora in the small intestine		Microflora in the large intestine	
		proximal	caudal	caecum	colon
Total count	5.5–9.5	5.5–8.5	5.5–9.5	8.5–9.5	8.0–10.0
Lactobacillus	5.0–9.0	5.5–8.5	6.0–9.5	8.0–9.5	7.5–9.5
Streptococcus	4.0–7.0	4.0–6.5	5.0–7.5	7.5	6.0–8.0
Bifidobacterium	4.5–6.5	4.0–5.5	5.5–7.5	5.0–8.0	5.5–8.5

4.1. Probiotics

Probiotics are live microbial feed supplements which beneficially affect the host by improving its intestinal microbiocenosis. Probable mechanisms of action of probiotics are based on the following:
– competition between them and pathogenic microorganisms for binding sites in the intestinal mucosa (Fuller, 1989; Sissons, 1989; Bomba et al., 2002),
– nutrient availability (Guerra et al., 1997; Bomba et al., 2002)
– total inhibition of pathogen growth by production of organic acids and antibiotic-like compounds (MantereAlhonen, 1995; Guerra et al., 1997; Zimmermann et al., 2001; Bomba et al., 2002; Marinho et al., 2007).

Probiotics are mainly active in the caudal segments of the ileum, in the caecum and the ascending colon. Probiotics influence the digestive process in the body by increasing the activity of microbial probiotic enzymes and the digestibility of food (Roselli et al., 2005). They stimulate the immune system and the regeneration of intestinal mucosa. They can elicit an increase in immunoglobulin a production (Perdigon et al., 1995) and stimulate macrophages and NK cells (natural killers cells; Chiang et al., 2000; Matsuzaki and Chin, 2000). They can also regulate anti- and pro-inflammatory cytokine production (Lessard and Brisson, 1987; Shu et al., 2001; Roselli et al., 2005).

The effect of probiotics depends on the combination of selected bacterial genera (Table 2), their doses, and on the interactions of probiotics with some pharmaceuticals, feed composition, storage conditions and feed technology (Kyriakis et al., 1999; Park et al., 2001; Chen et al., 2005). Probiotic application to piglets is important especially in the post-weaning period for the prophylaxis of PWD which is usually caused by enterotoxigenic *E. coli* strains (MantereAlhonen, 1995; Roberfroid, 1998; Bomba et al., 2002; Roselli et al., 2005; Marinho et al., 2007).

Multiple studies have confirmed the stimulating effects of probiotics on the intestinal environment (Barrow et al., 1977; Roth et al., 1992b; Depta et al., 1998; Mathew et al., 1998; Jadamus et al., 2001; Scharek et al., 2005, 2007; Reiter et al., 2006; Lodemann et al., 2008). By lowering the pH value in the small intestine and producing organic acids and antibacterial substances probiotic supplements inhibit pathogenic microorganisms, improve the intestinal microflora and stimulate immune function (Kumprecht et al., 1994; MantereAlhonen, 1995; Marinho, 2007).

Table 2. Microorganisms used as probiotics (Kumprecht et al., 1994; Fooks and Gibson, 2002; Ouwehand et al., 2002; Lodemann et al., 2006)

Genus	Bacterial species
Lactobacillus	*L. acidophilus*
	L. casei
	L. rhamnosus
	L. reuteri
	L. plantarum
	L. fermentum
	L. brevis
	L. helveticus
	L. delbrückei
Lactococcus	*L. lactis*
Enterococcus	*E. faecium*
Streptococcus	*S. thermophilus*
Pediococcus	*P. pentosaceus*
Bacillus	*B. subtilis*
	B. cereus
	B. toyoi
	B. natto
	B. mesentericus
	B. licheniformis
Bifidobacterium	*B. bifidum*
	B. pseudolongum
	B. breve
	B. thermophilum
Saccharomyces	*S. cerevisiae*
Avirulent *Escherichia coli*	*E. coli*

Intestinal microflora can be modulated by the supplementation of feeds with probiotic bacterial species of the genera *Lactobacillus, Bifidobacterium* (Bomba et al., 1998), *Bacillus* (Roth et al., 1992b; Alexopoulos et al., 2001; Scharek et al., 2005, 2007), *Enterococcus* and *Streptococcus* (Scharek et al., 2005, 2007) and their combinations (Mathew et al., 1998; Bomba et al., 2002). The bacteria *Enterococcus faecium* were found to be able to prevent the K88 positive ETEC strain from adhering to the intestinal mucous membrane of piglets (Jin and Zhao, 2000; Scharek et al., 2005). Regulating intestinal microbial balance by increasing the activity of microbial digestive enzymes, it improves digestion, feed digestibility and nutrient utilization (Bomba et al., 2002). In consequence, the morbidity and mortality of farm animals decrease and performance increase (Mathew et al., 1998; Taras et al., 2005; Lodemann et al., 2006).

Members of the genus *Bacillus* support natural intestinal microflora in the digestive tract, compete with undesirable microorganisms, and reduce the numbers of *Enterococci, Bacteroides* and coliforms (Ozawa et al., 1981; Adami and Cavazzoni, 1999; Kyriakis et al., 1999; Alexopoulos et al., 2001; Link et al., 2005; Link and Kovac, 2006) whilst *Saccharomyces cerevisiae* do not markedly alter the composition of host microflora (Mathew et al., 1998). These results are in contradiction to information provided by Mathew et al. (1993, 1996) who observed changes in the microbial environment due to *S. cerevisiae* activity. This can be explained by the ability of various strains of *S. cerevisiae* to alter cellulotic microflora (Newbold et al., 1995).

Inconsistent data have also been reported for other probiotic genera (Table 3). While some authors have observed beneficial effects of several members of the genera *Lactobacillus* and *Bacillus* (Depta et al., 1998; Bomba et al., 1999, 2002; Kyriakis et al., 1999; Reiter et al., 2006; Scharek et al., 2007), others have failed to confirm any stimulating effect on the composition of the pig intestinal microflora (Spriet et al., 1987; De Cupere et al., 1992; Newbold et al., 1995). These contradictory results can be explained by the low doses of the used species of probiotic bacteria, their stability, interactions with other medications, and the health condition, age and genetic predisposition of animals (Kyriakis, 1999; Reiter et al., 2006).

Improvement in the health condition and growth performance (Tables 4 and 5) was observed in newborn and weaned piglets after feeding with probiotics of the genera *Lactobacillus, Streptococcus* and *Bacillus* (Roth et al., 1992b; Mathew et al., 1998; Alexopoulos et al., 2001; La Ragione et al., 2001; Chen et al., 2005; Reiter et al., 2006).

E. faecium (Ushe and Nagy, 1985; Roth and Kirchgessner, 1986; Rekiel and Wiecek, 1996; Jadamus et al., 2001; Lodemann et al., 2008) and *Bifidobacterium lactis* (Alexopoulos et al., 2001; Shu et al., 2001) has been shown to be effective in reducing the occurrence and intensity of diarrhoea and increasing the performance of piglets. On the other hand, Kirchheim and Schone (1995) failed to find any positive effect of these bacteria in reducing diarrhoea incidence.

Whilst nonpathogenic *E. coli* strains lacking virulence genes have been used for the prophylaxis of *E. coli* infection (Davidson and Hirsch, 1976; Setia et al., 2009), Smith and Huggins (1983) used phages that specifically bind with K-antigens. The effects in both experiments were strictly specific and no direct protective function was observed. On the other hand, Melin and Wallgren (2002) applied non-virulent *E. coli* in combination with zinc oxide

Table 3. The effect of probiotics on the intestinal microflora of pigs

Genus/species	Beneficial effects	Nonsignificant beneficial effects
Lactobacillus	Bomba et al. (1998); Depta et al. (1998); Mathew et al. (1998); Alexopoulos et al. (2001); Scharek et al. (2005, 2007); Reiter et al. (2006)	De Cupere et al. (1992); Newbold et al. (1995)
Bifidobacterium	Bomba et al. (1998, 2002); Depta et al. (1998); Alexopoulos et al. (2001); Reiter et al. (2006)	none
Bacillus	Ozawa et al. (1981); Mathew et al. (1998); Adami and Cavazzoni (1999); Kyriakis et al. (1999), Link et al. (2005); Scharek et al. (2007)	Spriet et al. (1987); De Cupere et al. (1992)
Streptococcus	Bomba et al. (1998); Mathew et al. (1998); Alexopoulos et al. (2001)	none
Enterococcus/ E. faecium	Jin and Zhao (2000); Scharek et al. (2005, 2007)	none
Saccharomyces/ S. cerevisiae	Mathew et al. (1993, 1996)	Newbold et al. (1995); Mathew et al. (1998)

Table 4. The effect of probiotics on the occurrence of diarrhoeal diseases in pigs

Genus/species	Beneficial effects	Nonsignificant beneficial effects
Lactobacillus/L. casei	Danek et al. (1990); Mathew et al. (1998); Chen et al. (2005)	
Bifidobacterium/B. lactis	Alexopoulos et al. (2001); Shu et al. (2001)	
Bacillus/B. subtilis, B. licheniformis	Adami and Cavazzoni (1999); Kyriakis et al. (1999); La Ragione et al. (2001); Link et al. (2005)	Martelli (1992)
Streptococcus/Sp.	Mathew et al. (1998); Alexopoulos et al. (2001); Chen et al. (2005); Reiter et al. (2006)	
Enterococcus/E. faecium	Ushe and Nagy (1985); Roth and Kirchgessner (1986); Rekiel and Wiecek (1996); Jadamus et al. (2001); Lodemann et al. (2006)	Kirchheim and Schone (1995)
Escherichis/Avirulent *E. coli*	Melin and Wallgren (2002)	Davidson and Hirsch (1976); Duval-Iflah et al. (1983); Setia et al. (2009)

and a mixture of lactulose and fibre and observed a markedly reduced incidence of PWD in comparison with the control group.

The genus *Bacillus*, especially its members *B. subtilis* and *B. licheniformis* have been described to markedly reduce the severity of diarrhoea caused by *E. coli* (Adami and Cavazzoni, 1999; Kyriakis et al., 1999; La Ragione et al., 2001; Link et al., 2005) and at the same time improve growth parameters in weaned piglets (Roth et al., 1992b; Alexopoulos et al., 2001), whilst no improvement in health was observed after application of *B. toyoi* to sows and their piglets in a previous study (Martelli, 1992). However, Link and Kovac (2006) noted improvement in nutrient digestibility after feed supplementation with the same bacterium.

Improved nutrient digestibility resulting from increased enzymatic activity was also observed after feeding other probiotic bacteria such as *Lactobacillus plantarum, L. acidophillus, L. casei* and *E. faecium* (Collington et al., 1990; Scheuermann, 1993; Bhatt et al., 1995; Kumprecht and Zobac, 1998) and a significantly higher activity of carbohydrates was detected in mucosal tissues of pigs (Collington et al., 1990) in response to such treatment. However, the effects of probiotic bacteria of the genus *Lactobacillus* on growth parameters were limited to the first stage of fattening in some cases, and thus no positive effect on growth and feed conversion was detected in growing-finishing pigs (Pollman et al., 1980; Park et al., 2001).

Table 5. The effect of probiotics on the growth performance of piglets

Genus/species	Beneficial effects	Nonsignificant beneficial effects
Lactobacillus/L. plantarum, L. acidophillus, L. casei	Collington et al. (1990); Scheuermann (1993); Bhatt et al. (1995); Kumprecht and Zobac (1998)	Pollman et al. (1980); Harper et al. (1983); Lessard and Brissom (1987); Park et al. (2001)
Bifidobacterium/B. lactis	Mathew et al. (1998); Shu et al. (2001)	none
Bacillus/B. subtilis, B. licheniformis, B. toyoi	Alexopoulos et al. (2001)	none
Streptococcus/Sp.	Mathew et al. (1998); Alexopoulos et al. (2001); Chen et al. (2005); Scharek et al. (2005, 2007); Reiter et al. (2006)	none
Enterococcus/E. faecium	Roth and Kirchgessner (1986); Collington et al. (1990); Rekiel and Wiecek (1996); Jadamus et al. (2001); Lodemann et al. (2008)	none
Saccharomyces/S. cerevisiae	Mathew et al. (1998)	Burnett and Neil (1977); Dusel et al. (1994)

S. cerevisiae has been reported to have a varying influence on pig efficiency. Mathew et al. (1998) observed increased feed intake and body weight gain of piglets after feed supplementation, but Burnett and Neil (1977) and Dusel et al. (1994) did not see any positive effect.

To obtain the highest effect of probiotic preparations, supplementation of piglets shortly after birth is recommended. Martin et al. (2009) reported a beneficial effect of probiotic stimulation of the immune responses in pregnant sows before parturition. Feeding of probiotics to sows before farrowing and during lactation and to neonatal piglets caused a fall in numbers of pathogenic microorganisms present in sow and piglet faeces, and resulted in the reduced occurrence of digestive disturbances and mortality (Danek et al., 1991; Martin et al., 2009).

4.2. Prebiotics

Prebiotics are dietary short-chain carbohydrates (oligosaccharides), which cannot be digested by pigs, but are specifically utilizable by intestinal microflora. They have been referred to as the bifidus factor, because they support the growth and/or activities of probiotic microorganisms in the gastrointestinal tract (Gibson and Roberfroid, 1995; Gibson, 2004; Marinho et al., 2007; Rayes et al., 2009).

Prebiotic supplementation of the diet influences volatile fatty acid content (VFA), branched-chain proportion, lactic acid concentrations and ammonia concentrations in the gut (Pie et al., 2007). Increased concentrations of short-chain fatty acids (SCFA) stimulate natural bacterial activity and proliferation of bifidobacteria and lactic acid bacteria. The production of butyrate which is a dominant energy source for enterocytes also increases (Houdijk et al., 2002).

Table 6 lists the non-digestible oligosaccharides, which are used as prebiotics. The mannanoligosaccharides are important because they modify the microbial gut ecosystem by binding to the receptors present in the intestinal epithelium, thereby preventing the colonization of bacterial pathogens (Zimmermann et al., 2001; Shim et al., 2005). Mannanoligosaccharides isolated from the *S. cerevisiae* cell wall also have a beneficial effect on the intestinal microflora (Lyons and Bourne, 1995) and animal growth (Kumprecht et al., 1994; Kumprecht and Zobac, 1998; Shim et al., 2005). It was found that they suppress the growth of *E. coli*, *Salmonella typhimurium*, *Clostridium botulinum* and *C. sporogenes,* and conversely stimulate the growth of *B. longum*, *L. casei, L. acidophillus* and *L. delbrückei*. Lyons and Bourne (1995) reported that mannanoligosaccharides from the yeast cell wall stimulate the local immune system by increasing the activities of macrophages and T-lymphocytes.

Bifidogenic effects of galactooligosaccharides, fructooligosaccharides and soybeanoligosaccharides have been repeatedly confirmed by many *in vitro* and *in vivo* experiments, where they selectively interacted with the intestinal bacterial ecosystem (Roberfroid, 1998; Smiricky-Tjardes et al., 2003; Tuohy et al., 2005). Smiricky-Tjardes et al. (2003) observed an increase in *Bifidobacterium* and *Lactobacillus* genera numbers in the intestine, a concomitant increase in SCFA concentration and improved small intestine morphology (Rayes et al., 2009). Fructoologosaccharides and galactooligosac-

Table 6. Non-digestible oligosaccharides (NDO) used as prebiotics (Grizard and Barhomeuf, 1999; Zimmermann et al., 2001; Tuohy et al., 2005)

NDO	Mode of production
Mannanoligosaccharides	enzymatic synthesis from mannose
Galactooligosaccharides	trans-galactosylation of lactose with *Aspergillus oryzae* β-galactosidase
Fructooligosaccharides	partial enzymatic hydrolysis of inulin/transfructosylation from saccharose
Soybeanoligosaccharides	extraction from soyabean whey
Isomaltooligosaccharides	enzymatic hydrolysis of starch/transglucosylation of maltose
Xylooligosaccharides	enzymatic hydrolysis of xylan
Lactulose	alkali isomeration
Inulin	isolation from chicory root

charides stimulate acetate and lactate production, thereby reducing faecal *Bacteroides*, *Clostridia* and *Fusobacteria* shedding in pigs (Gibson et al., 1995; Zimmermann et al., 2001).

In contrast, Mikkelsen et al. (2003) and Visentini et al. (2008), while observing significantly increased numbers of *S. cerevisiae*, failed to find a stimulating effect of galactooligosacharides and fructooligosacharides on *Bifidobacterium* sp. growth in weaned piglets and on the improvement of pig efficiency.

Lactulose, which is formed by lactose isomerisation, is due to its high prebiotic activity widely used in the prevention and treatment of enteric disorders in animals and humans. Lactulose cannot be hydrolyzed by the β-galactosidase of the digestive tract and hence, it cannot be absorbed from the small intestine. It passes down to the large intestine where resident microflora consume it and produce lactic and/or acetic acid (Gibson, 2004; Bohacenko et al., 2007). Consequently, it stimulates the growth and/or activity of indigenous intestinal microflora, especially of the genera *Bifidobacterium* and *Lactobacillus,* and reduces the activity of proteolytic bacteria. Immobilization of proteolytic bacteria results in the reduction of ammonia release into the outer environment. Pig diets are usually supplemented with one per cent lactulose (Gibson, 2004; Marinho et al., 2007).

Inulin is present in a number of raw materials of vegetable origin such as onion, garlic, asparagus, banana and chicory root, which is one of the richest sources of dietary inulin. Chicory inulin-type fructans for animal nutrition typically contain more than 70 per cent of inulin, some flower sugars, organic acids, protein fragments and minerals. Dietary supplementation with inulin has a positive effect on SCFA production, sufficient height of intestinal villi, stimulation of natural microflora and improvement of efficiency parameters (Crittenden and Playne, 1996).

4.3. Synbiotics

A list of the possibilities of how to influence the intestinal microflora and thus improve the health condition and growth of pigs would not be complete if we did not mention the combination of probiotics and prebiotics – synbiotics. Preparations containing probiotics and prebiotics increase the passage of probiotic bacteria through the upper part of the intestine and help the colonization of local receptors in the intestine (Robefroid, 1998; Bomba et al., 2002; Maxwell et al., 2004).

Some studies have confirmed a significant synergistic growth-stimulating effect (Kumprecht and Zobac, 1998; Shim et al., 2005), reducing mortality and increasing the counts of species that are components of natural microfloras (Nemcova et al., 1999; Frece et al., 2009). Furthermore, it was found that synbiotic preparations increase SCFA production (Shim et al., 2007; Bird et al., 2009) and reduce faecal noxious gas emission (Lee et al., 2009).

Protective effects have been attributed to different combinations. The positive contribution of maltodextrins and *L. paracasei* (Bomba et al., 2002, 2006) or *E. faecium* (Kumprecht and Zobac, 1998) on increasing the efficiency of piglets, reducting pathogenic *E. coli* growth in the digestive tract and their adhesion to intestinal mucosa have often been observed. Improved efficiency was also observed after the feeding of animals with biological preparations containing *L. fermentum, L. brevis, L. salivarius* or *E. faecium* with lactulose or lactitol (Bomba et al., 2002; Piva et al., 2005).

Feeding a mixture of fructooligosaccharides and *L. paracasei* has been demonstrated to have a stimulating effect on the growth of natural intestinal microorganisms, to decrease the numbers of undesirable microflora including coliforms, *Clostridium* and Enterobacteriaceae and to improve the morphology of intestinal villi (Spencer et al., 1997; Nemcova et al., 1999; Bomba et al., 2002; Shim et al., 2005). Moreover, a higher effectiveness of synbiotics was shown when they were given to animals during the preweaning period (Shim et al., 2005). In the experiments performed by Bird et al. (2009), proliferation of *Bifidobacteria* in the intestinal tract occurred as a consequence of diet supplementation with *B. animalis* subsp. *lactis* and/or *B. choerinum* with fructooligosaccharides.

The incorporation of prebiotics with unsaturated fatty acids also yielded a superior synbiotic effect (Ringo et al., 1998; Bomba et al., 2002; Bontempo et al., 2004).

4.4. Organic acids

Besides antimicrobial function, organic acids and their salts have a beneficial effect on digestibility, nutrient resorption (Roth and Kirchgessner, 1998; De Freitas et al., 2006) and performance of weaned and growing piglets (Kirchgessner et al., 1995; Roth and Kirchgessner, 1998; Partanen and Mroz, 1999; De Freitas et al., 2006). Antimicrobial effects can be explained by two mechanisms: First, by a pH fall below 6 in the stomach, which inhibits the growth of pathogenic microorganisms and, second, the ability of organic acids to penetrate in their non-dissociated form through the bacterial wall and destroy some specific microorganisms (Eidelsburger et al., 1992; Roth et al., 1992a; Hansen et al., 2007). Bactericidal or bacteriostatic effects of organic acids consist above all in a direct effect of the organic acid anions on bacterial cell walls. This ability is primarily characteristic of acids with low numbers of carbon atoms in their molecules, such as formic, acetic, propionic and lactic acids (Kirchgessner et al., 1992; Roth et al., 1998; Partanen et al., 2001).

According to their effects, organic acids can be categorized into two groups. The first group comprises lactic, fumaric and citric acids and is characterized by an indirect effect in reducing bacterial populations by decreasing pH in the stomach**.** The second group comprises formic, acetic, propionic and sorbic acids, and are characterized by a direct effect of lower pH in the digestive tract on the cell wall of gram-negative bacteria thereby preventing their deoxyribonucleic acid replication (Roth et al., 1992a; Hansen et al., 1997; Castro, 2005).

Furthermore, organic acids contribute to the stabilization of intestinal microflora due to their effective biocidal qualities and their ability to improve enzymatic digestion (Kirchgessner et al., 1995; Roth and Kirchgessner, 1998; Marinho et al., 2007). The digestion of proteins by pepsin in the stomach begins when pH is low, while high pH values reduce enzyme activity. This is associated with the utilizability of feed minerals because solubility of minerals increases with a decrease in pH (Kirchgessner et al., 1995; Hansen et al., 2007). For these reasons, the use of organic acids in piglets is important because in the first days they suffer from a post-weaning syndrome associated with a typical starvation period which is followed by ingestion of excessive amounts of feed, resulting in mild acidification of the stomach chyme. Consequently, higher pH values cause weakening of the protective barrier function of the stomach against ingested bacteria and leads to intensive bacterial propagation in the small and large intestine (Castro 2005; De Freitas et al., 2006; Marinho et al., 2007).

Organic acids cause a decline in the number of pathogenic microorganisms such as *E. coli* in the stomach and small intestine (Kluge et al., 2006; Hansen et al., 2007). After application of organic acids in previous studies, a significant decrease in the number of microorganisms (*E. coli,* Bacteroidaceae, *Eubacterium, Lactobacillus* sp. and *Bifidobacterium* sp.) occurred in the duodenum, jejunum and ileum (Gedek et al., 1992b; Kirchgessner et al., 1992), whilst in the caecum and colon, the microflora was not greatly affected by the ingested organic acids (Gedek et al., 1992b). *In vitro* studies have shown that formic acid can inhibit the growth of pathogenic bacteria. Lactic acid was most effective in this regard, whereas the effect of acetic acid and propionic acid was low (Gedek et al., 1992b; Kirchgessner et al., 1992).

The effect of organic acids is initiated by their addition to the feed. Antibacterial and antifungal effects are elicited by a decrease in the feed pH (Eidelsburger et al., 1992; Castro 2005), which contributes to feed preservation. Acids in powder form are most suitable for preservation of feeds (Overland et al., 2008).

Feed acidifiers commonly used in animal nutrition, usually based on formic acid, are effective even when their proportion in a diet is low (less than 10 grams per kilogram; Eidelsburger et al., 1992; Gedek et al., 1992a; Partanen et al., 2002, 2007). Even though formic acid alone is an efficient acidifier; it is often combined with other organic acids such as sorbic acid (Kirchgessner et al., 1995), fumaric acid (Gedek et al., 1992b), citric, propionic and benzoic acids (Partanen and Mroz, 1999) so as to increase its effect. The antimicrobial effects of different organic acids vary greatly and depend on their concentration and pH. Specific effects of different organic acids towards different bacteria have been observed. It is necessary to note that an increased intake of organic acids can cause a reduction in feed intake, body weight gain and general efficiency of young pigs (Roth and Kirchgessner, 1998; Partanen et al., 2007).

Partanen et al. (2001) documented that formic acid tended to decrease bacterial nitrogen in different parts of the small intestine in pigs and improved apparent ileal digestibility of protein sources, certain essential amino-acids, lipids, calcium and phosphorus. The results showed a similar or higher effect in comparison with that of antibiotic growth promoters. The growth efficiency of market pigs was significantly increased by the addition of formic acid and this effect was further enhanced by combination of formic acid with potassium sorbate

(Partanen et al., 2002). Piva et al. (2002) supplemented piglet diet with a combination of citric, fumaric and malic acids. A beneficial effect of organic acids on the growth performance, feed intake and general health condition was observed. Biagi et al. (2006) described a favourable effect of gluconic acid on intestinal microflora, morphology of intestinal villi and general condition of piglets.

Positive combinatory effects can be achieved with lactic, formic, fumaric, benzoic and sorbic acids by suppressing *E. coli*, *Salmonella* and *Enterococcus* in the intestinal content. Several studies documented an improvement in growth efficiency have been reported in the literature (Knarreborg et al., 2002; Franco et al., 2005; Overland et al., 2008; Missotten et al., 2009).

A combination of organic acids with highly concentrated extracts, i.e., essential oils, shows synergistic activity. The bacterial cell wall damaging effect of essential oils permits the organic acids to enter. This effect is ascribed to thymol and carvacrol from plant extracts (Varley, 2002; Namkung et al., 2004; Kommera et al., 2006).

The significance of organic acids can also be assessed from the standpoint of sensory attributes. The addition of organic acids to diets can reduce the concentration of skatole present in the faeces and thereby reduce the boar taint problem of the meat (Claus et al., 1994; Babol and Squires, 1995; Bonneau et al., 2000).

4.5. Zinc oxide

Zinc is a component of several enzymes, is involved in their activation and is thus central to biochemical processes occurring in the body. It is a significant dietetic factor that regulates amino-acid and protein metabolism (Ou et al., 2007; Wang et al., 2009). Furthermore, it contributes to stabilisation of sensitive intestinal mucosa, diversity and functions of bacterial microflora, inhibits the growth of some pathogens and enhances the immune responses of the body against infections (Hahn and Baker, 1993; Srivastava et al., 1995; Case and Carlson, 2002; Hojberg et al., 2005; Han and Thacker, 2009). Some zinc compounds such as zinc sulphate, zinc lysine and zinc methionine stimulate the activity of macrophages (Van Heugten et al., 2003).

Zinc oxide has been included in piglet weaning diets for many years. High doses of zinc oxide (2500 to 3000 mg Zn/kg of feed) added to feed have been used as a preventative measure against PWD. Feeding zinc to weaned piglets has been demonstrated to reduce diarrhoea incidence and to improve growth rates (Hahn and Baker, 1993; Scott and Koski, 2000; Ou et al., 2007; Wang et al., 2009).

However, the mechanisms of zinc action on growth promotion and against diarrhoea are still not exactly known. Several hypotheses have been advanced to explain the effect of zinc oxide on weaned piglets. According to Srivastava et al. (1995) zinc is important for maintaining the integrity of the cell membrane, probably via translocation of redox-active metal ions, which can be responsible for inducing peroxidative damage at the target site. Higher cell-mediated immunity in rats fed with zinc has been observed as far back as in the study of Chandra and Au (1980). Liao et al. (2008) and Rhoads and Wu (2009) ascribed the decrease in the incidence of diarrhoea and the increase in average body weight gain in piglets to the regulation of cellular signalling pathways. Another hypothesis regarding zinc oxide activity was formulated by Wang et al. (2009) who assumed that the beneficial effect of zinc oxide may be related to its involvement in protein expression associated with glutathione metabolism and oxidative stress in the gut.

Dietary supplementation with zinc oxide has a beneficial effect on the stability of the intestinal microflora of weaned piglets (Jensen Waern et al., 1998; Huang et al., 1999; Katouli et al., 1999; Owusu-Asiedu et al., 2003) and morphology of their small intestine – increased height of villi in comparison with the depths of crypts (Katouli et al., 1999; Castillo et al., 2008; Hedemann et al., 2009). Zinc deficiency can contribute to intestinal villus ulceration and to the damage of the immune barrier function (Hennig et al., 1992; Wapnir, 2000; Roselli et al., 2005).

Furthermore, zinc can decrease the virulence of some microorganisms such as *Staphylococcus aureus* (Walsh et al., 1994), *Salmonella typhi* and *Aeromonas hydrophylia* (Fiteau and Tomkins, 1994). A comparable zinc content, which positively affected microbial environment of the intestine, also reduced the intensity of ETEC-elicited diarrhoea (Mores et al., 1998; Bosi et al., 2003; Li et al., 2006). Roselli et al. (2005) also observed a beneficial effect of glutamate zinc on the course of infection.

The beneficial effect of Zn^{2+} on the activity of some enzymes present in the pancreatic and intestinal juices, mucin production, digestion, nutrient absorption (Hedemann and Jensen, 2004; Hedemann et al., 2009) and intestinal growth of weanling piglets is exerted through increasing IGF-1 (insulin like growth factor) and IGF-IR (IGF receptor) expression in the small-intestinal mucosa (Li et al., 2006).

The stimulating effect of zinc oxide is best harnessed during the first two weeks after weaning when the assumed protective effect occurs (Katouli et al., 1999). An adverse effect of a long-term elevation in Zn^{2+} content in piglet faeces can constitute a potential environmental hazard (Carter et al., 2002; Bosi et al., 2003).

4.6. Plant extracts

Plants and their extracts had been used in traditional medicine for centuries. After the ban on antibiotic growth promoters, natural feed additives (plants and spices) started to be used as alternatives in broiler and pig diets (Borovan, 2004; Hernandez et al., 2004). Plant extracts and essential oils have been exploited in animal nutrition, particularly for their antimicrobial (Namkung et al., 2004; Brambilla and De Filippis, 2005; Costa et al., 2007), anti-inflammatory, anti-oxidative (Liu et al., 2008), and anti-parasite properties (Magi et al., 2006). It was found for some natural materials that due to their immunological and pharmacological qualities, these can have prophylactic effects against a series of enteric disorders (Laine et al., 2008).

The antibacterial activity of essential oils depends on their chemical composition and concentration (Russo et al., 1998). The existing data shows that their effect depends on the type of plant extract, correct combination and appropriate dosage (Oetting et al., 2006; Utiyama et al., 2006). A synergistic effect of plant extracts and organic acids has been also demonstrated (Namkung et al., 2004; Kommera et al., 2006).

Garlic extract is generally considered as the most effective antimicrobial agent of plant origin (Arnault et al., 2003) and *in vitro* studies have shown the antibacterial effects of thyme (Azaz et al., 2004), oregano, cloves, cinnamon (Carson et al., 2006) and juniper (Pepeljnjak et al., 2005).

Regarding the prevention of diarrhoeal diseases, it is important to focus on the extracts and essential oils that inhibit the proliferation of *E. coli* which is a common cause of intestinal diseases in piglets (Laine et al., 2008). Khan et al. (2009) reported that pathogenic strains of *E. coli* are sensitive to the following plant extracts: *Acacia nilotica, Syzygium aromaticum* and *Cinnamum zeylanicum*. However, a major disadvantage of plant extracts lies in the fact that their compositions are unstable (Bomba et al., 2006). This is due to the influence of many factors such as climate, season, harvesting method and others (Borovan, 2004). Probably for this reason there is a multiplicity of controversial results

Table 7. Plant extracts in pig nutrition

Plant	The effect observed	Reference
Oregano, cinnamon and Mexican pepper	decreased ileum total microbial mass, increased the lactobacilli : enterobacteria ratio	Manzanilla et al. (2004)
Sangrovit (alkaloids of *Macleaya cordata*)	increased body weight gain and feed conversion by growing pigs	Borovan (2004)
Cinnamon, thyme, oregano	inhibited pathogenic *E. coli* in piglet intestine	Namkung et al. (2004)
Thyme, clove, oregano, eugenol and carvacrol	improved pig performance	Oetting et al. (2006)
Clove, oregano	growth performance of pigs close to pigs fed antimicrobials	Costa et al. (2007)
Specific blend of herbal extract	increase ADG, decrease feed conversion ratio in finishing pigs	Liu et al. (2008)
Aged garlic extract, allicin	improved body weight, morphological properties of intestine villi and non-specific defense mechanisms of piglets	Tatara et al. (2008)
Camellia sinensis	decrease of clostridia and enterococci counts in the faeces of piglets	Zanchi et al. (2008)

obtained in different scientific studies examining the effect of these substances in animal nutrition. Accordingly, it is necessary to select effective plant extracts, standardize them and investigate a potential synergistic benefit deriving from their combination (Budzinski et al., 2000; Oetting et al., 2006).

5. Clay minerals

Another possibility for PWD prevention is the use of natural and modified clay minerals. Clay minerals, designated as silicates in systemic mineralogy, are stratified clay minerals formed by a net of tetrahedral and octahedral layers. They contain molecules of silicon, aluminium and oxygen. The layers can be interconnected by a system of hydrogen bonds or a group of cations, and this space is termed an interlayer. Phyllosilicates are classified into the main groups according to the type of the layers, interlayer content, charge of the layers and chemical formulas. The natural extracted clays (bentonite, zeolite, kaolin etc.) are a mixture of various clay minerals differing in chemical composition (Velebil, 2009). The best-known are montmorillonite (MMT), smectite, illite, kaolinite, biotite and clinoptilolite.

5.1. Adsorption capacity and effects of clay minerals

A common feature of clay minerals is a high sorption capacity determined by their stratified structure. Within the layers of these clays, substitution of other metal ions for silicon or aluminium can occur resulting in a net negative charge on the surface of the clay platelet. This charge imbalance is offset by hydrated cations, the predominant ones being Na^+ and Ca^{2+}. These interlaminar cations can be exchanged with other metal cations. In aqueous solutions, water is intercalated into the interlaminar space of clay, leading to an expansion of the minerals (Ma and Guo, 2008).

Due to their sorption capacity and absence of primary toxicity, claystone minerals are regarded as a simple and effective tool for chemical prevention of a series of toxic materials, not only in the environment, but also in the living bodies. Clays added to a diet can bind and immobilize toxic materials in the gastro-intestinal tract of animals and largely reduce their biological availability and toxicity for the body (Phillips, 1999; Lemke et al., 2001; Phillips et al., 2002; Trckova et al., 2004). Multiple studies have confirmed the decontaminating properties of clay minerals. They can bind:

- aflatoxins (Phillips et al., 1987, 1988; Lindemann et al., 1993; Schell et al., 1993a,b; Phillips et al., 2002; Abdel-Wahhab et al., 2002; Huebner and Phillips, 2003; Rizzi et al., 2003; Shi et al., 2005, 2007; Maciel et al., 2007; Magnoli et al., 2008; Nguyen et al., 2008),
- plant metabolites (Dominy et al., 2004),
- heavy metals (Hassen et al., 2003; Katsumata et al., 2003; Lin et al., 2004; Xu et al., 2004; Yu et al., 2005, 2008; Abbes et al., 2007),
- toxins (Knezevich and Tadic, 1994).

Kaolin-based medication is used for the therapy of diarrhoea and digestive disorders in human medicine (Heimann, 1984; Madkour et al., 1993; Kasi et al., 1995; Knezevich, 1998; Gebesh et al., 1999; Narkeviciute et al., 2002; Gonzalez et al., 2004). The ability of clay minerals to allow adsorption of pathogens and enterotoxins to the surface layer by means of hydrogen bonds or if needed, into the interlayer sheets and to reduce their numbers in the intestinal tract has been demonstrated *in vivo* (Trckova et al. 2009) and *in vitro* (Novakova, 1968; Brouillard and Rateau, 1989; Ramu et al., 1997; Hu et al., 2002; Hu and Xia, 2006). The question of how much clay minerals reduce the occurrence and severity of PWD in piglets due to their adsorption qualities is still under discussion.

The extent of adsorption is determined by the chemistry of the clay minerals, exchangeable ions, the surface properties (Williams et al., 2008) and the fine structure of clay particles (Hassen et al., 2003). An important role is also played by pH, dosage and exposure time (Brouillard and Rateau, 1989). Hence, the resulting effect of clay mineral treatment is preconditioned by these factors. There is an abundance of published data which indicate that the dietary use of clay minerals reduces the incidence and decreases the severity and the duration of diarrhoea in pigs (Castro and Elias, 1978; Said et al., 1980; Vrzgula et al., 1982; Bartko et al., 1983; Brouillard and Rateau, 1989; Ramu et al., 1997; Narkeviciute et al., 2002; Dominy et al., 2004; Gonzalez et al., 2004; Martinez et al., 2004; Papaioannou et al., 2004; Castro, 2005).

The improvement of clinical manifestations of diarrhoeal diseases can also be explained by the elimination of various factors that are associated with the occurrence of these diseases in piglets, primarily at the period of weaning. These factors include intestinal hypersensitivity to feed antigens or weaning-induced changes leading to the manifestation of malabsorption syndrome caused by a reduction in digestive enzyme activity, which can both result in a predisposition to infectious enteritis (Wilson et al., 1989; Papaioannou, et al., 2004, 2005). The use of clay minerals retards the rate of digestive passage through the intestines and their ability to absorb water results in more compact and better shaped faeces.

The capacity of clay minerals to immobilize antinutritional components in feeds (Albengres et al., 1985; Ma and Guo, 2008), reduce the number of pathogenic microorganisms and depress the activity of bacterial enzymes in the small intestinal digesta (Gonzalez et al., 2004; Xia et al. 2004, 2005; Trckova et al., 2009), prevents irritation and damage and improves the morphological characteristics of mucosa (Albengres et al., 1985; Xia et al. 2004, 2005; Trckova et al., 2009).

The supplementation of a diet with one to three per cent of clay-based adsorbents is generally recommended. Nevertheless, diets supplemented with a higher proportion of such adsorbents are also voluntarily ingested by animals (Castro and Elias, 1978; Vrzgula et al., 1982; Bartko et al., 1983; Castro and Iglesias, 1989), and do not exert any adverse effect on their growth and performance (Castro and Elias, 1978; Castro and Iglesias, 1989). Many studies have documented significant improvements in body weight gain and feed conversion after supplementation of a diet with clay minerals (Pond et al., 1981; Vrzgula et al., 1982; Bartko et al., 1983; Pond and Yen, 1987; Pond et al., 1988; Castro and Iglesias, 1989; Cabezas et al., 1991; Papaioannou et al., 2004, 2005; Chen et al., 2005; Kolacz et al., 2005; Alexopoulos et al., 2007; Prvulovic et al., 2007; Trckova et al., 2009).

5.2. Modified forms of clay minerals

Modification of clay minerals by the addition of Cu^{2+} can improve their antibacterial activity (Ye et al., 2003; Zhou et al., 2004; Xia et al. 2004; 2005; Hu and Xia, 2006), which is then a result of two mechanisms: one is electrostatic attraction which promotes the adherence of *E. coli* to the surface of clay minerals and the other is the slow release of Cu^{2+}, which can kill bacteria (Xia et al., 2004). For example, Hu and Xia (2006) reported that whilst calcium MMT, sodium MMT and acid activated MMT showed some ability to reduce bacterial plate counts by 13 to 37 per cent, their modified cupric containing forms reduced this number by 96 to 99 per cent (Hu and Xia, 2006).

Xia et al. (2004) reported that Cu^{2+}-enriched MMT significantly decreased the frequency of diarrhoea by up to 72 per cent and significantly increased the villus height and the villus height to crypt depth ratio in the jejunal mucosa by 19 and 37 per cent, respectively. Specific physical and chemical modifications – nanocomposites (Lin et al., 2004; Xu et al., 2004; Shi et al., 2005, 2007; Yu et al., 2008), and complexes with organic materials (Xu et al., 1997; Herrera et al., 2000, 2004; Koh and Dixon, 2001), are characterized by much higher adsorptive abilities than other regular clay minerals. The effect of clay minerals can also be reinforced by the addition of other components such as charcoal or antioxidants (Bonna et al., 1991).

6. CONCLUSIONS

The body of literature on the characteristics of different alternative components intended as antibiotic growth promoters strongly argues that these can be used as effective means of preventing diarrhoea in weaned piglets. The modification of intestinal microflora composition via probiotic and prebiotic preparations and providing the body with species of microorganisms which have a beneficial effect on the intestinal tract stimulates the immune system and decreases the risk of diarrhoea. Altering the acidity of the intestinal content, stabilization of the intestinal mucosa and supporting the digestive process contribute to the maintenance of good intestinal health. Due to their adsorption and detoxifying capabilities, non-conventional feed supplements in the form of clay minerals appear to be effective agents for protection against infection. Provided the recommended dosage is observed, the above mentioned raw materials can be used as suitable alternatives to antibiotics for the prevention of diarrhoeal diseases and the enhancement of piglet growth in the crucial period of weaning.

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REFERENCES

Abbes S, Ben Salah-Abbes J, Nahdi K, Ben Younes R, Hetta MM, El-Kady AA, Abdel-Wahhab MA, Oueslati R (2007): Inactivation of cadmium induced immunotoxicological alterations in rats by Tunisian montmorillonite clay. International Immunopharmacology 7, 750–760.

Abdel-Wahhab MA, Nada SA, Khalil FA (2002): Physiological and toxicological responses in rats fed aflatoxin-contaminated diet with or without sorbent materials. Animal Feed Science and Technology 97, 209–219.

Adami A, Cavazzoni V (1999): Occurence of selected bacterial groups in the faeces of piglets fed with *Bacillus coagulans* as probiotic. Journal of Basic Microbiology 39, 3–9.

Albengres E, Urien S, Tillement JP, Oury P, Decourt S, Flouvat B, Drieu K (1985): Interactions between smectite, a mucus stabilizer, and acidic and basic drugs – *in vitro* and *in vivo* studies. European Journal of Clinical Pharmacology 28, 601–605.

Alexa P, Salajka E, Salajkova Z, Machova A (1995): Combined parenteral and oral immunization against diarrhea in weaned piglets caused by enterotoxigenic strains of *Escherichia coli*. Veterinarni Medicina 40, 365–370.

Alexa P, Stouracova K, Hamrik J, Rychlik I (2001): Gene typing of the colonisation factors K88 (F4) in enterotoxigenic *Escherichia coli* strains isolated from diarrhoeic piglets. Veterinarni Medicina 46, 46–49. http://old.vri.cz/docs/vetmed/46-2-46.pdf

Alexopoulos C, Karagiannidis SK, Kritas SK, Boscos C, Georgoulakis IE, Kyriakis SC (2001): Field evaluation of a bioregulator containing live *Bacillus cereus* spores on health status and performance of sows and their litters. Journal of Veterinary Medical Science 48, 137–145.

Alexopoulos C, Papaioannou DS, Fortomaris P, Kyriakis CS, Tserveni-Goussi A, Yannakopoulos A, Kyriakis SC (2007): Experimental study on the effect of in feed administration of a clinoptilolite-rich tuff on certain biochemical and hematological parameters of growing and fattening pigs. Livestock Science 111, 230–241.

Ambroz Z (1981): General Microbiology. University of Agriculture in Brno. 100 pp.

Amezcua R, Friendship R, Dewey C, Gyles C (2002): A case-control study investigating risk factors associated with postweaning *Escherichia coli* diarrhea in southern Ontario. Journal of Swine Health and Production 10, 245–249.

Apgar GA, Kornegay ET, Lindemann MD, Wood CM (1993): The effect of feeding various levels of *Bifidobacterium globosum* a on the performance, gastrointestinal measurements, and immunity of weanling pigs and on the performance and carcass measurements of growing-finishing pigs. Journal of Animal Science 71, 2173–2179.

Arnault I, Christides JP, Mandon N, Haffner T, Kahane R, Auger J (2003): High performance ion pair chromatography method for simultaneous analysis of alliin, deoxyalliin, allicin and dipeptide precursors in garlic products using multiple mass spectrometry and UV detection. Journal of Chromatography 991, 69–75.

Azaz AD, Irtem HA, Kurkcuoglu M, Baser KHC (2004): Composition and the *in vitro* antimicrobial activities of the essential oils of some Thymus species. Journal of Biosciences 59, 75–80.

Babol J, Squires EJ (1995): Quality of meat from entire male pigs. Food Research International 28, 201–212.

Bailey M, Clarke CJ, Wilson AD, Williams NA, Stokes CR (1992): Depressed potential for interleukin-2-production following early weaning of piglets. Veterinary Immunology and Immunopathology 34, 197–207.

Bark LJ, Crenshaw TD, Leibbrandt VD (1986): The effect of mal intervals and weaning on feed intake of early weaned pigs. Journal of Animal Science 62, 1233–1239.

Barrow PA, Fuller R, Newport MJ (1977): Changes in the microflora and physiology of the anterior intestinal tract of pigs weaned at 2 days with special reference to the pathogenesis of diarrhea. Infection and Immunity 18, 586–595.

Bartko P, Chabada J, Vrzgula L, Solar I, Blazovsky J (1983): The fortification of pig feed ration with zeolite in the cage rearing system (in Czech). Veterinarni Medicina 28, 429–435.

Barton MD (2000): Antibiotic use in animal feed and its impact on human health. Nutrition Research Reviews 13, 279–299.

Bertschinger K, Exner GU, Hodler J (1999): Diagnostic imaging of bone and soft tissue tumors. Tumordiagnostik and Therapie 20, 71–78.

Bhatt RS, Katoch BS, Dogra KK, Gupta R, Sharma KS, Sharma CR (1995): Effect of dietary supplementation of different strains of *Saccharomyces cerevisiae* on the biological performance of broilers. Indian Journal of Animal Nutrition 12, 61–66.

Biagi G, Piva A, Moschini M, Vezzali E, Roth FX (2006): Effect of gluconic acid on piglet growth performance, intestinal microflora, and intestinal wall morphology. Journal of Animal Science 84, 370–378.

Bird AR, Vuaran M, Crittenden R, Hayakawa T, Playne MJ, Brown IL, Topping DL (2009): Comparative effects of a high-amylose starch and a fructooligosacharide on fecal bifidobacteria numbers and short-chain fatty acids in pigs fed *Bifidobacterium animalis*. Digestive Diseases and Sciences 54, 947–954.

Blecha F, Pollmann DS, Nichols DA (1983): Weaning pigs at an early age decreases cellular immunity. Journal of Animal Science 56, 396–400.

Bohacenko I, Pinkrova J, Peroutkova J, Pechackova M (2007): Fermentation of lactose and lactulose mixture by *Lactobacillus acidophilus* strain (in Czech). Chemicke Listy 101, 911–915.

Bomba A, Gancarcikova S, Nemcova R, Herich R, Kastel R, Depta A, Demeterova M, Ledecky V, Zitnan R (1998): The effect of lactic acid bacteria on intestinal metabolism and metabolic profile of gnotobiotic pigs. Deutsche Tierarztliche Wochenschrift 105, 384–389.

Bomba A, Nemcova R, Gancarcikova S, Herich R, Kastel R (1999): Potentiation of the effectiveness of *Lactobacillus casei* an the prevention of *E. coli* induced diarrhea in conventional and gnotobiotic pigs. Mechanisms in the Pathogenesis of Enteric Diseases 473, 185–190.

Bomba A, Nemcova R, Gancarcikova S, Herich R, Guba P, Mudronova D (2002): Improvement of the probiotic effect of microorganisms by their combination with maltodextrins, fructo-oligosacharides and polyunsaturated acids. British Journal of Nutrition 88, 95–99.

Bomba A, Jonecova Z, Koscova J, Nemcova R, Gancarikova S, Mudronova D, Scirankova L, Buleca V, Lazar G, Posivak J, Kastel R, Marekova M (2006): The improvement of probiotics efficasy by synergistically acting components of natural origin: a review. Biologia 61, 729–734.

Bonna RJ, Aulerich RJ, Bursian SJ, Poppenga RH, Braselton WE, Watson GL (1991): Efficasy of hydrated sodium calcium aluminosilicate and activated charcoal in reducing the toxicity of dietary aflatoxin to mink. Archives of Environmental Contamination and Toxicology 20, 444–447.

Bonneau M, Walstra P, Claudi-Magnussen C, Kempster AJ, Tornberg E, Fischer K, Diestre A, Siret F, Chvillon P, Claus R, Dijksterhuis G, Punter P, Matthews KR, Agerhem H, Beague MP, Oliver MA, Gispert M, Weiler U, Von Seth G, Leask H, Furnols MFI, Homer DB, Cook GL (2000): An international study on the importance of androstenone and skatole for boar taint: IV. Simulation studies on consumer dissatisfaction with entire male pork and effect of sorting carcasses on the slaughter line, main conclusions and recommendations. Meat Science 54, 285–295.

Bontempo V, Sciannimanico D, Pastorelli G, Rossi R, Rosi F, Corino C (2004): Dietary conjugated linoleic acid positively affects immunological variables in lactating sows and piglets. Journal of Nutrition 134, 817–824.

Borovan L (2004): Plant alkaloids enhance performance of animals and improve the utilizability of amino acids (in Czech). Krmivarstvi 6, 36–37.

Bosi P, Merialdi G, Sarli G, Casini L, Gremokolini C, Preziosi R, Brunetti B, Trevisi P (2003): Effects of doses of ZnO or Zn-glutamate on growth performance, gut characterictics, health and immunity of early weaned pigs orally challenged with *E. coli* K88. In: Proceedings of the A.S.P.A., 15th Congress, Parma, Italy. Journal of Animal Science 2, 361–363.

Brouillard MY, Rateau JG (1989): Adsorption of bacterial enterotoxins by smectite and kaolin clays – *in vitro* study in cell-culture and in the newborn mouse (in French). Gastroenterologie Clinique et Biologique 13, 18–24.

Brambilla G, De Filippis S (2005): Trends in animal feed composition and possible consequences on residue tests. Analytica Chimica Acta 529, 7–13.

Budino FEL, Thomaz MC, Kronka N, Nakaghi LSO, Tucci FM, Fraga AL, Scandolera AJ, Huaynate RAR (2005): Effect of probiotic and prebiotic inclusion in weaned piglet diets on structure and ultra-structure of small intestine. Brazilian Archives of Biology and Technology 6, 921–929.

Budzinski JW, Foster BC, Vandenhoek S, Arnason JT (2000): An *in vitro* evaluation of human cytochrome P450 3A4 inhibition by selected commercial herbal extracts and tinctures. Phytomedicine 7, 273–282.

Burnett GS, Neil EL (1977): A note on the effect of probioticum feed additive on the live-weight gain, feed conversion and carcass quality of bacon pigs. Animal Production 25, 95–98.

Cabezas MJ, Salvador D, Sinisterra JV (1991): Stabilization-activation of pancreatic-enzymes adsorbed on to a sepiolite clay. Journal of Chemical Technology and Biotechnology 52, 265–274.

Carter S, Westerman P, Van Kempen T, Cromwell G, Hill G, Shurson G, Richert B, Casey K (2002): Estimation of manure nutrient excretion from swine based upon diet composition and feed intake. Journal of Animal Science 80, 137–138.

Carson CF, Hammer KA, Riley TV (2006): *Melaleuca alternifolia* (tea tree) oil: a review of antimicrobial and other medicinal properties. Clinical Microbiology Reviews 19, 50–62.

Case CL, Carlson MS (2002): Effect of feeding organic and inorganic sources of additional zinc on growth performance and zinc balance in nursery pigs. Journal of Animal Science 80, 1917–1924.

Castillo M, Martin-Orue SM, Taylor-Pickard JA, Perez JF, Gasa J (2008): Use of mannan-oligosacharides and zinc chelate as growth promoters and diarrhea preventative in weaning pigs: Effects on microbiota and gut function. Journal of Animal Science 86, 94–101.

Castro M (2005): Use of additives on the feeding of monogastric animals. Cuban Journal of Agricultural Science 39, 439–445.

Castro M, Elias A (1978): Effect of inclusion of zeolite in final molasses-based diets on performance of growing and fattening pigs. Cuban Journal of Agricultural Science 12, 69–75.

Castro M, Iglesias M (1989): Effect of zeolite on traditional diets for fattening pigs. Cuban Journal of Agricultural Science 23, 289–291.

Chae C, Kim O, Choi C, Min K, Cho WS, Kim J, Tai JH (2000): Prevalence of porcine epidemic diarrhoea virus and transmissible gastroenteritis virus infection in Korean pigs. Veterinary Record 147, 606–608.

Chandra RK, Au B (1980): Single nutrient deficiency and cell-mediated immune response. American Journal of Clinical Nutrition 33, 736–738.

Chen YJ, Kwon OS, Min BJ, Son KS, Cho JH, Hong JW, Kim IH (2005): The effects of dietary Biotite V supplementation as an alternative substance to antibiotics in growing pigs. Asian-Australasian Journal of Animal Sciences 18, 1642–1645.

Chiang BL, Sheih YH, Wang LH, Liao CK, Gill HS (2000): Enhancing immunity by dietary consumption of a probiotic lactic acid bacterium (*Bifidobacterium lactis* HN019): optimization and definition of cellular immune responses. European Journal of Clinical Nutrition 54, 849–855.

Claus R, Weiler U, Herzog A (1994): Physiological aspects of adrostenone and skatole formation in the boar – a review with experimental data. Meat Science 38, 289–305.

Collington GK, Parker DS, Armstrong DG (1990): The influence of inclusion of either an antibiotic or a probiotic in the diet on the development of digestive enzyme activity in the pig. British Journal of Nutrition 64, 59–70.

Costa LB, Panhoza Tse ML, Miyada VS (2007): Herbal extracts as alternatives to antimicrobial growth for weanling pigs. Brazilian Journal of Animal Science 36, 589–595.

Crittenden RG, Playne MJ (1996): Production, properties, and applications of food-grade oligosaccharides. Trends in Food Science and Technology 7, 353–361.

Danek P, Novak J, Semeradova H, Diblikova E, Dlabolova O, Adama L (1990): Investigation of the effect of *Lactobacillus casei* CCM 4160 on reared piglets (in Czech). In: Proceedings of Conference on Probiotics in Animal Nutrition, Brno, Czech Republic, 143–152.

Danek P, Novak J, Semeradova H, Diblikova E (1991): The effect of probiotic *Lactobacillus casei* CCM4160 application to sows on piglet performance (in Czech). Zivocisna Vyroba 36, 411–415.

Davidson JN, Hirsh DC (1976): Bacterial competetion as a means of preventing neonatal diarrhea in pigs. Infection and Immunity 13, 1773–1774.

De Cupere F, Deprez P, Demeulenaere D, Muylle E (1992): Evaluation of the effect of 3 probiotics on experimental *Escherichia coli* enterotoxaemia in weaned piglets. Journal of Veterinary Medicine 39, 277–284.

De Freitas LS, Lopes DC, De Freitas AF, Carneiro JDC, Corassa A, Pena SDM, Costa LF (2006): Effects of feeding organic acids for piglets from 21 to 49 days. Brazilian Journal of Animal Science 35, 1711–1719.

Depta A, Rychlik R, Nieradka R, Rotkiewicz T, Kujawa K, Bomba A, Grabowska-Swiecicka G (1998): The influence of alimentary tract colonization with *Lactobacillus* sp. strains on chosen metabolic profile indices in piglets. Polish Journal of Veterinary Science 1–2, 3–7.

Dominy NJ, Davoust E, Minekus M (2004): Adaptive function of soil consumption an *in vitro* study modelling the human stomach and small intestine. The Journal of Experimental Biology 207, 319–324.

Ducluzeau R (1983): Implantation and development of the gut flora in the newborn animal. Annales de Recherches Veterinaires 14, 354–359.

Dusel G, Behringer J, Rohrmoser G, Durst L (1994): Effects of *Saccharomyces cerevisiae* on fattening pigs. 1. Results of the fattening investigation. Zuchtungskunde 66, 383–394.

Duval-Iflah Y, Chappuis JP, Ducluzeau R, Raibaud P (1983): Intraspecific interaction between *Escherichia coli* strains in human newborns and in gnotobiotic mice and piglets. Progress in Food and Nutrition Science 7, 107–116.

Eidelsburger U, Kirchgessner M, Roth FX (1992): Influence of fumaric acid, hydrochloric acid, sodium formate, tylosin and tyocerin on daily weight gain, feed intake, feed conversion rate and digestibility. 11. investigations about the nutritive efficasy of organic acids in the rearing of piglets. Journal of Animal Physiology and Animal Nutrition 68, 82–92.

Fiteau SM, Tomkins AM (1994): Micronutrients and tropical infections. Transaction of Royal Society of Tropical Medicine and Hygiene 88, 1–3.

Fooks LJ, Gibson GR (2002): Probiotics as modulators of the gut flora. British Journal of Nutrition 88 (Suppl. 1), 39–49.

Franco LD, Fondevila M, Lobera MB, Castrillo C (2005): Effect of combinations of organic acids in weaned pig diets on microbial species of digestive tract contents and their response on digestibility. Journal of Animal Physiology and Animal Nutrition 89, 88–93.

Frece J, Kos B, Svetec IK, Zgaga Z, Beganovic J, Lebos A, Suskovic J (2009): Synbiotic effect of *Lactobacillus helveticus* M92 and prebiotics on the intestinal microflora and immune system of mice. Journal of Dairy Research 76, 98–104.

Fuller R, Barrow PA, Brooker BE (1978): Bacteria associated with the gastric epithelium of neonatal pigs. Applied and Environmental Microbiology 35, 582–591.

Fuller R (1989): Probiotics in man and animals. Journal of Applied Bacteriology 66, 365–368.

Gebesh VV, Ianchenko VI, Sukhov IUA (1999): Kaopectate in the combined treatment of patients with intestinal infection (in Russian). Likarska Sprava 3, 140–142.

Gedek B, Roth FX, Kirchgessner M, Wiehler S, Bott A, Eidelsburger U (1992a): Influence of fumaric acid, hydrochloric acid, sodium formate, tylosin and toyocerin on the microflora in different segments of the gastrointestinal tract. 14. Investigations about the nutritive efficacy of organic acids in the rearing of piglets. Journal of Animal Physiology and Animal Nutriton 68, 209–217.

Gedek B, Kirchgessner M, Eidelsburger U, Wiehler S, Bott A, Roth FX (1992b): Influence of formic acid on the microflora in different segments of the gastrointestinal tract. 5. Investigations about the nutritive efficacy of organic acids in the rearing of piglets. Journal of Animal Physiology and Animal Nutrition 67, 206–214.

Ghnassia JC (1979): Quantitative study of gastrointestinal bacterial-flora. Medecine et Maladies Infectieuses 9, 507–510.

Gibson GR (2004): From probiotics to prebiotics and a healthy digestive system. Journal of Food Science 69, 141–143.

Gibson GR, Roberfroid M (1995): Dietary modulation of the human colonic microbiota-introducing the concept of prebiotics. Journal of Nutrition 125, 1401–1412.

Gibson GR, Beatty ER, Wang X, Cummings JH (1995): Selective stimulation of bifidobacteria in the human colon by oligofructose and inulin. Gastroenterology 108, 975–982.

Gonzalez R, De Medina FS, Martinez-Augustin O, Nieto A, Galvez J, Risco S, Zarzuelo A (2004): Anti-inflammatory effect of diosmectite in hapten-induced colitis in the rat. British Journal of Pharmacology 141, 951–960.

Grizard D, Barthomeuf C (1999): Non-digestible oligosaccharides used as prebiotic agents: mode of production and beneficial effects on animal and human health. Reproduction Nutrition Development 39, 5–6.

Guerra NP, Bernardez PF, Mendez J, Cachaldora P, Castro LP (1997): Production of four potentially probiotic lactic acid bacteria and their evaluation as feed additives for weaned piglets. Animal Feed Science and Technology 134, 89–107.

Hahn JD, Baker DH (1993): Growth and plasma zinc responses of young pigs fed pharmacologic levels of zinc. Journal of Animal Science 71, 3020–3024.

Hampson DJ (1986): Alterations in piglet small intestinal structure at weaning. Research in Veterinary Science 40, 32–40.

Hampson DJ, Kidder DE (1986): Influence of creep feeding and weaning on brush border enzyme activities in the piglet small intestine. Research in Veterinary Science 40, 24–31.

Hampson DJ, Fu YF, Robertson ID (1987): Investigation of the source of haemolytic *Escherichia coli* infecting weaned pigs. Epidemiology and Infection 99, 149–153.

Han YK, Thacker PA (2009): Performance, nutrient digestibility and nutrient balance in weaned pigs fed diets supplemented with antibiotics or zinc oxide. Journal of Animal and Veterinary Advances 8, 868–875.

Hansen LL, Larsen AE, Jensen BB, Hansen-Moller J (1997): Short time effect of zinc bacitracin and heavy fouling with faeces plus urine on boar taint. Animal Science 64, 351–363.

Hansen CF, Riis AL, Bresson S, Hojbjerg O, Jensen BB (2007): Feeding organic acids enhances the barrier function against pathogenic bacteria of the piglet stomach. Livestock Science 108, 206–209.

Harper AF, Kornegay ET, Bryant KL, Thomas HR (1983): Efficasy of virginiamycin and a commercially available *Lactobacillus* probiotic in swine diets. Animal Feed Science and Technology 8, 69–86.

Hassen A, Jamoussi F, Saidi N, Mabrouki Z, Fakhfakh E (2003): Microbial and cooper adsorption by smectitic clay – an experimental study. Environmental Technology 24, 1117–1127.

Hayashi T, Makino K, Ohnishi M, Kurokawa K, Ishii K, Yokoyama K, Han CG, Ohtsubo E, Nakayama K, Murata T, Tanaka M, Tobe T, Iida T, Takami H, Honda T, Sasakawa C, Ogasawara N, Yasunaga T, Kuhara S, Shiba T, Hattori M, Shinagawa H (2001): Complete genome sequence of enterohemorrhagic *Escherichia coli* O157:H7 and genomic comparison with a laboratory strain K-12. DNA Research 8, 11–22.

Hedemann MS, Jensen BB (2004): Variations in enzyme activity in stomach and pancreatic tissue and digesta in piglets around weaning. Archives of Animal Nutrition 58, 47–59.

Hedemann MS, Theil PK, Knudsen KEB (2009): The thickness of the intestinal mucous layer in the colon of rats fed various sources of non-digestible carbohydrates is positively correlated with the pool of SCFA but negatively correlated with the proportion of butyric acid in digesta. British Journal of Nutrition 102, 117–125.

Heimann G (1984): Pharmacotherapy of acute infant enteritis (in German). Monatsschrift fur Kinderheilkunde 132, 303–305.

Hennig B, Wang Y, Ramasamy S, McClain CJ (1992): Zinc deficiency alters barrier function of cultured porcine endothelial cells. Journal of Nutrition 122, 1242–1247.

Hernandez F, Madrid J, Garcia V, Orengo J, Megias MD (2004): Influence of two plant extracts on broilers performance, digestibility, and digestive organ size. Poultry Science 83, 169–174.

Herrera P, Burghardt RC, Phillips TD (2000): Adsorption of *Salmonella enteritidis* by cetylpyridinium-exchanged montmorillonite clay. Veterinary Microbiology 74, 259–272.

Herrera P, Burghardt R, Huebner H, Phillips TD (2004): The efficacy of sand-immobilized organoclays as filtration bed materials for bacteria. Food Microbiology 21, 1–10.

Hojberg O, Canibe N, Poulsen HD, Hedemann MS, Jensen BB (2005): Influence of dietary zinc oxide and copper sulfate on the gastrointestinal ecosystem in newly weaned piglets. Applied and Environmental Microbiology 71, 2267–2277.

Houdijk JGM, Hartemink R, Verstegen MWA, Bosch MW (2002): Effects of dietary non-digestible oligosacharides on microbial characteristics of ileal chyme and faeces in weaner pigs. Archives of Animal Nutrition 56, 297–307.

Hu CH, Xia MS (2006): Adsorption and antibacterial effect of copper-exchanged montmorillonite on *Escherichia coli* K-88. Applied Clay Science 31, 180–184.

Hu XR, Lu GL, Chen LS, Gu JM, Zhang Y (2002): Study on the mechanism of the interaction between montmorillonite and bacterium (in Chinese). Yao Xue Xue Bao 37, 718–720.

Huang SX, McFall M, Cegielski AC, Kirkwood RN (1999): Effect of zinc supplementation on *Escherichia coli* septicemia in weaned pigs. Swine Health and Production 7, 109–111.

Huebner HJ, Phillips TD (2003): Clay-based affinity probes for selective cleanup and determination of aflatoxin B1 using nanostructured montmorillonite on quartz. Journal of AOAC International 86, 534–539.

Jadamus A, Vahjen W, Simon O (2001): Growth behaviour of a spore forming probiotic strain in the gastrointestinal tract of broiler chicken and piglets. Archives of Animal Breeding 54, 1–17.

Jensen BB (1998): The impact of feed additives on the microbial ecology of the gut in young pigs. Journal of Animal and Feed Sciences 7, 45–65.

Jensen-Waern M, Melin L, Lindberg R, Johannisson A, Petersson L, Wallgren P (1998): Dietary zinc oxide in weaned pigs – effects on performance, tissue concetrations, morphology, neutrophil functions and faecal microflora. Research in Veterinary Science 64, 225–231.

Jin LZ, Zhao X (2000): Intestinal receptors for adhesive fimbriae of enetrotoxigenic *Escherichia coli* (ETEC) K88 in swine – a review. Applied Microbiology and Biotechnology 54, 311–318.

Jin LZ, Ho YW, Abdullah N, Jalaludin S (1997): Probiotics in poultry: modes of action. World's Poultry Science Journal 53, 351–368.

Jung K, Kim J, Ha Y, Choi C, Cae C (2006a): The effects of transplacental porcine circovirus type 2 infection on porcine epidemic diarrhoea virus-induced enteritis in preweaning piglets. Veterinary Journal 171, 445–450.

Jung K, Ahn K, Chae C (2006b): Decreased activity of brush border membrane-bound digestive enzymes in small intestines from pigs experimentally infected with porcine epidemic diarrhoea virus. Research in Veterinary Science 81, 310–314.

Kasi M, Kausar P, Naz R, Miller LC (1995): Treatment of diarrhoea in infants by medical doctors in Balochistan, Pakistan. Journal of Diarrhoeal Diseases Research 13, 238–241.

Katouli M, Melin L, Jensen-Waern M, Wallgren P, Mollby R (1999): The effect of zinc oxide supplementation on the stability of the intestinal flora with special reference to composition of coliforms in weaned pigs. Journal of Applied Microbiology 87, 564–573.

Katsumata H, Kaneco S, Inomata K, Itoh K, Funasaka K, Masuyama K, Suzuki T, Ohta K (2003): Removal of heavy metals in rising wastewater from plating factory by adsorption with economical viable materials. Journal of Environmental Management 69, 197–191.

Khan R, Islem B, Akram M, Shakil S, Ahmad A, Ali SM, Siddiqui M, Khan AU (2009): Antimicrobial activity of five herbal extracts against multi drug resistant (MDR) strains of bacteria and fungus of clinical origin. Molecules 14, 586–597.

Kidder DE, Manners MJ (1980): The level of distribution of carbohydrases in the small intestine mucosa of pigs from 3 weeks of age to maturity. British Journal of Nutrition 43, 141–153.

Kirchgessner M, Gedek B, Wiehler S, Bott A, Eidelsburger U, Roth FX (1992): Influence of formic-acid, calcium-formate and sodiumhydrogencarbonate on the microflora in different segments of the gastrointestinal tract. 10. Investigations about the nutritive efficacy of organic acids in the rearing of piglets. Journal of Animal Physiology and Animal Nutrition 68, 73–81.

Kirchgessner M, Roth FX, Paulicks BR (1995): Nutritive efficacy of sorbic acid in the rearing of piglets. Journal of Animal Physiology and Animal Nutrition 74, 235–242.

Kirchheim U, Schone F (1995): Performance of piglets fed diets containing a probiotic (*Enetrococcus faecium*). In: Conference Information, 5th Symposium on Vitamins and Additives in Human and Animal Nutrition, 28–29 September, 1995, Jena Thuringen, Germany, 522–525.

Kluge H, Broz J, Eder K (2006): Effect of benzoic acid on growth performance, nutrient digestibility, nitrogen digestibility, nitrogen balance, gastointestinal microflora and parameters of microbial metabolism in piglets. Journal of Animal Physiology and Animal Nutrition 90, 316–324.

Knarreborg A, Miquel N, Granli T, Jensen BB (2002): Establishment and application of an *in vitro* methodology to study the effects of organic acids on coliform and lactic acid bacteria in the proximal part of the gastrointestinal tract of piglets. Animal Feed Science and Technology 99, 131–140.

Knezevich M (1998): Geophagy as a therapeutic mediator of endoparasitism in a free-ranging group of rhesus macaques (*Macaca mulatta*). American Journal of Primatology 44, 71–82.

Knezevich DL, Tadic V (1994): Decontamination with clay or alcoholate of pigs percutaneously poisoned with VX and soman (in Croatian). Vojnosanitetski Pregled 51, 488–491.

Koh S, Dixon JB (2001): Preparation and application of organominerals as sorbents of phenol, benzene, and toluene. Applied Science 18, 111–122.

Kolacz R, Korniewicz A, Dobranski Z, Korniewicz D, Trziszka T (2005): Effects of dietary halloisyte on the performance and meat quality of pigs. Medycyna Weterynaryjna 61, 1266–1269.

Kommera SK, Mateo RD, Neher FJ, Kim SW (2006): Phytobiotics and organic acids as potential alternatives to the use of antibiotics in nursery pig diets. Asian-Australasian Journal of Animal Sciences 19, 1784–1789.

Kumprecht I, Zobac P (1998): Study of the effect of a combined preparation containing *Enterococcus faecium* M-74 and mannan-oligosacharides in diets for weanling piglets. Czech of Journal Animal Science 43, 477–481.

Kumprecht I, Zobac P, Gasnarek Z, Robosova E (1994): The effect of continuous application of probiotics based on *Saccharomyces cerevisiae* var. *ellipsoideus* and *Streptococcus faecium* C-68 (SF68) on the performance of broiler chickens (in Czech). Zivocisna Vyroba 39, 491–503.

Kyriakis SC, Tsiloyiannis VK, Vlemmas J, Sarris K, Tsinas AC, Alexopoulos C, Jansegers L (1999): The effect of probiotic LSP 122 on the control of post-weaning diarrhoea syndrome of piglets. Research in Veterinary Science 67, 223–228.

Laine TM, Lyytikainen T, Yliaho M, Anttila M (2008): Risk factors for post-weaning diarrhoea on piglet producing farms in Finland. Acta Veterinaria Scandinavica 50, 21.

La Ragione RM, Casua G, Cutting SM, Woodward MJ (2001): *Bacillus subtilis* spores competetively exclude *E. coli* O78:K80 in poultry. Veterinary Microbiology 79, 133–142.

Le Dividich J (1981): Effects of environmental temperature on the growth rates of early-weaned piglets. Livestock Production Science 8, 75–86.

Le Dividich J, Herpin P (1994): Effects of climatic conditions on performance, metabolism and health status of weaned piglets: a review. Livestock Production Science 38, 79–90.

Lee SJ, Shin NH, Ok JU, Jung HS, Chu GM, Kim JD, Kim IH, Lee SS (2009): Effects of dietary synbiotics from anaerobic microflora on growth performance, noxious gas emission and fecal pathogenic bacteria population in weaning pigs. Asian Australasian Journal of Animal Sciences 22, 1202–1208.

Lemke SL, Mayura K, Reeves WR, Wang NY, Fickey C, Phillips TD (2001): Investigation of organophilic montmorillonite clay inclusion in zearalenone-contamined diets using the mouse uterine weight bioassay. Journal of Toxicology and Environmental Health, part A 62, 243–258.

Lessard M, Brisson GJ (1987): Effect of a *Lactobacillus* fermentation product on growth, immune response and fecal enzyme activity in weaned pigs. Canadian Journal of Animal Science 67, 509.

Li XL, Yin JD, Li DF, Chen XJ, Zang JJ, Zhou X (2006): Dietary supplementation with zinc oxide increases IGF-I and IGF-I receptor gene expression in the small intestine of weanling piglets. Journal of Nutrition 136, 1786–1791.

Liao XH, Majithia A, Huang Xl, Kimmel AR (2008): Growth control via TOR kinase signaling, an intracellular sensor of amino acids and energy availability, with crosstalk potential to proline metabolism. Amino Acids 35, 761–770.

Lin XL, Xu ZR, Zou XT, Wang F, Yan XH, Jiang JF (2004): Effects of montmorillonite nanocomposite on mercury residues in growing/finishing pigs. Asian-Australasian Journal of Animal Sciences 17, 1434–1437.

Lindemann MD, Blodgett DJ, Kornegay ET, Schurig GG (1993): Potential ameliorators of aflatoxicosis in weanling/growing swine. Journal of Animal Science 71, 171–178.

Link R, Kovac G (2006): The effect of probiotic BioPlus 2B on feed efficiency and metabolic parameters in swine. Biologia 61, 783–787.

Link R, Kovac G, Pistl J (2005): A note on probiotics as an alternative for antibiotics in pigs. Journal of Animal and Feed Sciences 3, 513–519.

Liu GM, Wei Y, Wang ZS, Wu D, Zhou AG, Liu GL (2008): Effects of herbal extract supplementation on growth performance and insulin-like growth factor (IGF)-I system in finishing pigs. Journal of Animal and Feed Sciences 17, 538–547.

Lodemann U, Hubener K, Jansen N, Martens H (2006): Effects of *Enterococcus faecium* NCIMB 10415 as probiotic supplement on intestinal transport and barrier function of piglets. Archives of Animal Nutrition 1, 35–48.

Lodemann U, Lorenz BM, Weyrauch KD, Martens H (2008): Effects of *Bacillus cereus* var. *toyoi* as probiotic feed supplement on intestinal transport and barrier function in piglets. Archives of Animal Nutrition 62, 87–106.

Lyons TP, Bourne S (1995): Principles of the effect of probiotics on the basis of yeasts and mannans. In: Proceedings of Conference on Probiotics in animal nutrition, Pohorelice, Czech Republic, 13–22.

Ma YL, Guo T (2008): Intestinal morphology, brush border and digesta enzyme activities of broilers fed on a diet containing Cu2+ – loaded montmorillonite. British Poultry Science 49, 65–73.

Maciel RM, Lopes STDA, Santurio JM, Martins DB, Rosa AP, Emanuelli MP (2007): Hepatic and renal functions in broilers fed on diets with aflatoxins and natural clinoptilolite. Pesquisa Agropecuaria Brasileira 42, 1221–1225.

Madec F, Bridoux N, Bounaix S, Jestin A (1998): Measurement of digestive disorders in the piglet at weaning and related risk factors. Preventive Veterinary Medicine 35, 53–72.

Madec F, Bridoux N, Bounaix S, Cariolet R, Duval-Iflah Y, Hampson DJ, Jestin A (2000): Experimental models of porcine post-weaning colibacillosis and their relationship to postweaning diarrhoea and digestive disorders as encountered in the field. Veterinary Microbiology 72, 295–310.

Madej M, Lundh T, Lindberg JE (1999): Activities of enzymes involved in glutamine metabolism in connection with energy production in the gastrointestinal tract epithelium of newborn, suckling and weaned piglets. Biology of the Neonate 75, 250–258.

Madkour AA, Madina EMH, Elazzouni OEZ, Amer MA, Elwalili TMK, Abbass T (1993): Smectite in acute diarrhea in children – a double-blind placebo-controlled clinical-trial. Journal of Pediatric Gastroenterology and Nutrition 17, 176–181.

Magi E, Jarvis T, Miller I (2006): Effects of different plant products against pig mange mites. Acta Veterinaria Brno 75, 283–287.

Magnoli AP, Tallone L, Rosa CAR, Dalcero AM, Chlacchiera SM, Sanchez RMT (2008): Commercial bentonites as detoxifier of broiler feed contaminated with aflatoxin. Applied Clay Science 40, 63–71.

Makala LHC, Kamada T, Nishikawa Y, Nagasawa H, Igarashi I, Fujisaki K, Suzuki N, Mikami T, Haverson K, Bailey M, Stokes CR, Bland PW (2000): Ontogeny of pig discrete Peyer's patches: distribution and morphometric analysis. Pathobiology 68, 275–282.

MantereAlhonen S (1995): Propionibacteria used as probiotics – a review. Lait 75, 447–452.

Manzanilla EG, Perez JF, Martin M, Kamel C, Baucells F, Gasa J (2004): Effect of plant extracts and formic acid on the intestinal equilibrium of early-weaned pigs. Journal of Animal Science 82, 3210–3218.

Marinho MC, Lordelo MM, Cunha LF, Freire JPB (2007): Microbial activity in the gut of piglets: I. Effect of prebiotic and probiotic supplementation. Livestock Science 108, 236–239.

Martelli P (1992): Nutritional and sanitary effects of *Bacillus subtilis* use as feed additive in pigs. Rivista-di-Suinicoltura 33, 35–39.

Martin R, Delgado S, Maldonado A, Jimenez E, Olivares M, Fernandez L, Sobrino OJ, Rodriguez JM (2009): Isolation of lactobacilli from sow milk and evaluation of their probiotic potential. Journal of Dairy Research 76, 418–425.

Martinez M, Castro M, Hidalgo K, Ayala U, Perez R, Hernandez L, Baez L (2004): Effective utilization of natural zeolite for diarrhea control. Cuban Journal of Agricultural Science 38, 387–390.

Mathew AG, Sutton AL, Scheidt AB, Patterson JA, Kelly DT, Meyerholtz KA (1993): Effect of galactan on selected microbial populations and pH and volatile fatty acids in the ileum of the weanling pig. Journal of Animal Science 71, 1503–1509.

Mathew AG, Franklin MA, Upchurch WG, Chattin SE (1996): Influence of weaning age on ileal microflora and fermentation acids in young pigs. Nutrition Research 16, 817–827.

Mathew AG, Chattin SE, Robbins CM, Golden DA (1998): Effects of a direct-fed yeast culture on enteric microbial populations, fermentations acids, and performance of weanling pigs. Journal of Animal Science 76, 2138–2145.

Matsuzaki T, Chin J (2000): Modulating immune responses with probiotic bacteria. Immunology and Cell Biology 78, 67–73.

Maxwell FJ, Duncan SH, Hold G, Stewart CS (2004): Isolation, growth and prebiotics and probiotic potential of novel *Bifidobacteria* from pigs. Anaerobe 10, 33–39.

McAllister JS, Kurtz HJ, Short EC (1979): Changes in the intestinal flora of young pigs with postweaning diarrhea of edema disease. Journal of Animal Science 49, 868–879.

Melin L, Wallgren P (2002): Aspects on feed related prophylactic measures aiming to prevent post weaning diarrhoea in pigs. Acta Veterinaria Scandinavica 43, 231–245.

Melin L, Jensen-Waern M, Johannisson A, Ederoth M, Katouli M, Wallgren P (1997): Development of selected faecal microfloras and of phagocytic and killing capasity of neutrophils in young pigs. Veterinary Microbiology 54, 287–300.

Melin L, Mattsson S, Katouli M, Wallgren P (2004): Development of post-weaning diarrhoea in piglets. Relation to presence of *Escherichia coli* strains and rotavirus. Journal of Veterinary Medicine, Series B-Infectious Diseases and Veterinary Public Health 51, 12–22.

Mikkelsen LL, Bendixen CH, Jakobsen M, Jensen BB (2003): Enumeration of bifidobacteria in gastrointestinal samples from piglets. Applied and Environmental Microbiology 69, 654–658.

Missotten JAM, Goris J, Michiels J, Van Coillie E, Herman L, De Smet S, Dierick NA, Heyndrickx M (2009): Screening of isolated lactic acid bacteria as potential beneficial strains for fermented liquid pig feed production. Animal Feed Science and Technology 150, 122–138.

Moller K, Jensen TK, Jorsal SE, Leser TD, Cartensen B (1998): Detection of *Lawsonia intracellularis*, *Serpulina hyodysenteriae*, weakly beta-haemolytic intestinal spirochaetesllа enterica, *Salmonella enterica*, and haemolytic *Escherichia coli* from swine herds with and without diarrhoea among growing pigs. Veterinary Microbiology 62, 59–72.

Mores N, Cristani J, Piffer IA, Barioni W, Lima GMM (1998): Effects of zinc oxide on postweaning diarrhea control in pigs experimentally infected with *E. coli*. Arquivo Brasileiro De Medicina Veterinaria, E Zootecnia 50, 513–523.

Namkung H, Li M, Gong J. Yu H. Cottrill M, De Lange CFM (2004): Impact of feeding blends of organic acids and herbal extracts on growth performance, gut microbiota and digestive function in newly weaned pigs. Canadian Journal of Animal Science 84, 697–704.

Narkeviciute I, Rudzeviciene O, Leviniene G, Mociskiene K, Eidukevicius R (2002): Management of Lithuanian children's acute diarrhoea with Gastrolit solution and dioctahedral smectite. European Journal of Gastroenterology & Hepatology 14, 419–424.

Nemcova R, Bomba A, Gancarcikova S, Heich R, Guba P (1999): Study of the effect of *Lactobacillus paracasei* and fructooligosacharides on the faecal microflora in weanling piglets. Berliner und Munchener Tierarztliche Wochenschrift 112, 225–228.

Newbold CJ, Wallace RJ, Chen XB, McIntosh FM (1995): Different strains of *Saccharomyces cerevisiae* differ in their effects on ruminal bacterial numbers *in vitro* and in sheep. Journal of Animal Science 73, 1811–1818.

Nguyen QT, Ogle B, Pettersson H (2008): Efficacy of bentonite clay in ameliorating aflatoxicosis in piglets fed aflatoxin contaminated diets. Tropical Animal Health and Production 40, 649–656.

Novakova J (1968): Effect of bentonite and kaolinite on growth curve of *E. coli.* Folia Microbiologica 13, 543.

Oetting LL, Utiyama CE, Giani PA, Ruiz UD, Miyada VS (2006): Effects of herbal extracts and antimicrobials on apparent digestibility, performance, organs morphometry and intestinal histology of weanling pigs. Brazilian Journal of Animal Science 35, 1389–1397.

Ou DY, Li DF, Ca YH, Li XL, Yin JD, Qiao SY, Wu GY (2007): Dietary supplementation with zinc oxide decreases expression of the stem cell factor in the small intestine of weanling pigs. Journal of Nutritional Biochemistry 18, 820–826.

Ouwehand AC, Salminen S, Isolauri E (2002). Probiotics: an overview of beneficial effects. Antonie van Leeuwenhoek 82, 279–289.

Overland M, Kjos NP, Borg M, Skjerve E, Sorum H (2008): Organic acids in diets for entire male pigs: effect on skatole level, microbiota in digesta, and growth performance. Livestock Science 115, 169–178.

Owusu-Asiedu A, Nyachoti CM, Marquardt RR (2003): Response of early weaned pigs to an enterotoxigenic *Echerichia coli* (K88) challenge when fed diets containing spray-dried porcine plasma or pea protein isolate plus egg yolk antibody, zinc oxide, fumaric acid, or antibiotic. Journal of Animal Science 81, 1790–1798.

Ozawa K, Yokota H, Kimura M, Mitsuoka T (1981): Effects of administration of *Bacillus subtilis* strain BN on intestinal flora of weanling piglets. Japanese Journal of Veterinary Science 43, 771–775.

Papaioannou DS, Kyriakis CS, Alexopoulos C, Tzika ED, Polizopoulou ZS, Kyriakis SC (2004): A field study on the effect of the dietary use of a clinoptilolite-rich tuff, alone or in combination with certain antimicrobials, on the health status and performance of weaned, growing and finishing pigs. Research in Veterinary Science 76, 19–29.

Papaioannou D, Katsoulos PD, Panousis N, Karatzias H (2005): The role of natural and synthetic zeolites as feed additives on the prevention and/or the treatment of certain farm animal diseases: a review. Microporous and Mesoporous Materials 84, 161–170.

Park DY, Namkung H, Paik IK (2001): Effects of supplementary enzymes or probiotics on the performance and ammonia gas production in weanling pigs. Korean Journal of Animal Science 43, 485–496.

Partanen KH, Mroz Z (1999): Organic acids for performance enhacement in pig diets. Nutrition Research Reviews 12, 117–145.

Partanen K, Jalava T, Valaja J, Perttila S, Siljander-Rasi H, Lindeberg H (2001): Effect of dietary carbadox or formic acid and fibre level on ileal and faecal nutrient digestibility and microbial metabolite concentrations in ileal digesta of the pig. Animal Feed Science and Technology 93, 137–155.

Partanen K, Siljander-Rasi H, Alaviuhkola T, Suomi K, Fossi M (2002): Performance of growing-finishing pigs fed medium or high fibre diets supplemented with avilamycin, formic acid or formic acid-sorbate blend. Livestock Production Science 73, 139–152.

Partanen K, Siljander-Rasi H, Pentikainen J, Pelkonen S, Fossi M (2007): Effects of weaning age and formic acid-based feed additives on pigs from weaning to slaughter. Archives of Animal Nutrition 61, 336–356.

Pacsa I, Marghitas LA, Groza I, Pusta D, Morar R, Oroian T, Cimpean A, Bogdan L, Bogdan I, Bogdan S, Oroian R (2009): The importance of probiotics administration to sucking pigs. Journal of Food Agriculture and Environment 7, 485–491.

Pepeljnjak S, Kosalec I, Kalodera Z, Blazevic N (2005): Antimicrobial activity of juniper berry essential oil (*Juniperus communis* L., Cupressaceae). Acta Pharmaceutica 55, 417–422.

Perdigon G, Alvarez S, Rachid M, Aguero G, Gobbato N (1995): Immune system stimulation by probiotics. Journal of Dairy Science 78, 1597–1606.

Phillips TD (1999): Dietary clay in the chemoprevention of aflatoxin-induced disease. Toxicological Sciences 52, 118–126.

Phillips TD, Kubena LF, Harvey RB, Taylor DR, Heidelbaugh ND (1987): Mycotoxin hazards in agriculture: new approach to control – abstract. Journal of the American Veterinary Medical Association 190, 1671.

Phillips TD, Kubena LF, Harvey RB, Taylor DR, Heidelbaugh ND (1988): Hydrated sodium calcium aluminosilicate: A high affinity sorbent for aflatoxin. Poultry Science 67, 243–247.

Phillips TD, Lemke SL, Grant PG (2002): Characterization of clay-based enterosorbents for the prevention of aflatoxicosis. Mycotoxins and Food Safety 504, 157–171.

Pie S, Awati A, Vida S, Falluel I, Williams BA, Oswald IP (2007): Effects of added fermentable carbohydrates in the diet on intestinal proinflammatory cytokine-specific mRNA content in weaning piglets. Journal of Animal Science 85, 637–683.

Piva A, Prandini A, Fiorentini L, Morlacchini M, Galvano F, Luchansky JB (2002): Tributyrin and lactitol synergistically enhanced the trophic status of the intestinal mucosa and reduced histamine levels in the gut of nursery pigs. Journal of Animal Science 80, 670–680.

Piva A, Casadei G, Gatta PP, Luchansky JB, Biagi G (2005): Effect of lactitol, lactic acid bacteria, or their combinations (synbiotic) on intestinal proteolysis *in vitro*, and on feed efficiency in weaned pigs. Canadian Journal of Animal Science 85, 345–353.

Pollman DS, Danielson DM, Peo ER (1980): Effect of microbial feed additives on performance of starter and growing-finnishing pigs. Journal of Animal Science 51, 577–581.

Pond WG, Yen JT, Lindvall RN, Maurer RR (1981): Effect of pig breed group and of soil or clinoptilolite (a-zeolite) in the pen environment on preweaning weight-gain and survival. Nutrition Reports International 24, 443–449.

Pond WG, Yen JT (1987): Effect of supplemental carbadox, and antibiotic combination, or clinoptilolite on weight-gain and organ weigts of growing swine fed maize or rye as the grain sources. Nutrition Reports International 35, 801–809.

Pond WG, Yen JT, Varel VH (1988): Response of growing swine to dietary copper and clinoptilolite supplementation. Nutrition Reports International 37, 795–803.

Prvulovic D, Jovanovi-Galovic A, Stanitic B, Popovic M, Grubor-Lajsic G (2007): Effects of a clinoptilolite supplement in pig diets on performance and serum parameters. Czech Journal of Animal Science 52, 159–164.

Ramu J, Clark K, Woode GN, Sarr AB, Phillips TD (1997): Adsorption of cholera and heat-labile *Escherichia coli* enterotoxins by various adsorbents: An *in vitro* study. Journal of Food Protection 60, 358–362.

Rayes N, Seehofer D, Neuhaus P (2009): Prebiotics, probiotics, synbiotics in surgery-are they only trendy, truly effective or even dangerous? Langenbecks Archives of Surgery 394, 547–555.

Reiter K, Eggebrecht S, Drewes B, Riess M, Weyrauch KD (2006): Effect of *Enterococcus faecium* and *Bacil-*

lus cereus var. *toyoi* on the morphology of the intestinal mucous membrane in piglets. Biologia Bratislava 61, 1–7.

Rekiel A, Wiecek J (1996): Effect of preparations Biogen and Microferm-fer on the rearing results of piglets. Veterinary Medicine 52, 187–189.

Rhoads JM, Wu G (2009): Glutamine, arginine, and leucine signaling in the intestine. Amino Acids 37, 111–122.

Ringo E, Bendiksen HR, Gausen SJ, Sundsfjord A, Olsen RE (1998): The effect of dietary fatty acids on lactic acid bacteria associated with the epithelial mucosa and from faecalia of *Arctic charr*, *Salvelinus alpinus*. Journal of Applied Microbiology 85, 855–864.

Rizzi L, Simioli M, Roncada P, Zaghini A (2003): Aflatoxin B1 and clinoptilolite in feed for laying hens: Effects on egg quality, mycotoxin residues in livers, and hepatic mixed-function oxygenase activities. Journal of Food Protection 66, 860–865.

Roberfroid MB (1998): Prebiotics and synbiotics: concepts and nutritional properties. British Journal of Nutrition 80 (Suppl. 2), 197–202.

Robinson IM, Allison MJ, Bucklin JA (1981): Characterization of the cecal bacteria of normal pigs. Applied and Environmental Microbiology 41, 950–955.

Rojas M, Conway PL (1996): Colonization by lactobacilli of piglet small intestinal mucus. Journal of Applied bacteriology 81, 474–480.

Roselli M, Finamore A, Britti MS, Bosi P, Oswald I, Mengheri E (2005): Alternatives to in-feed antibiotics in pigs: Evaluation of probiotics, zinc or organic acids as protective agents for the intestinal mucosa. A comparison of *in vitro* and in vivo results. Animal Research 54, 203–218.

Roth FX, Kirchgessner M (1986): Nutritive effects of Streptococcus-faecium (strain M-74) in broiler chicks. Archiv fur Geflugelkunde 50, 225–228.

Roth FX, Kirchgessner M (1998): Organic acids as feed additives for young pigs: Nutritional and gastrointestinal effects. Journal of Animal and Feed Sciences 7, 25–33.

Roth FX, Eidelsburger U, Kirchgessner M (1992a): Influence of fumaric-acid, hydrochlorid-acid, sodium formate, tylosin and toyocerin on dry-matter content, pH value, concentration of carbonic-acids and ammonia in different segments of the gastrointestinal tract. 12. Investigations about the nutritive efficacy of organic-acids in the rearing of piglets. Journal of Animal Physiology and Animal Nutrition 68, 93–103.

Roth FX, Kirchgessner M, Eidelsburger U, Gedek B (1992b): Nutritive efficacy of *Bacillus cereus* as a probiotic in the fattening of calves. 1. Influence on fattening performance, carcass criteia and microbial activity in the small intestine. Agribiological Research-Zeitschrift fur Agrarbiologie Agrikulturchemie okologie 45, 294–302.

Roth FX, Windisch W, Kirchgessner M (1998): Effect of potassium diformate (Formi (TM) LHS) on nitrogen metabolism and nutrient digestibility in piglets at graded dietary lysine supply. Agribiological Research 51, 167–175.

Russo M, Galletti GC, Bocchini P, Carnacini A (1998): Essential oil chemical composition of wild populations of Italian oregano spice (*Origanum vulgare* ssp. *hirtum letswaart*): A preliminary evaluation of their use in chemotaxonomy by cluster analysis. 1. Inflorescences. Journal of Agricultural and Food Chemistry 46, 3741–3746.

Said SA, Shibl AM, Abdullah ME (1980): Influence of various agents on adsorption capacity of kaolin for pseudomonas-aeruginosa toxin. Journal of Pharmaceutical Sciences 69, 1238–1239.

Scott ME, Koski KG (2000): Zinc deficiency impairs immune responses against parasitic nematode infections at intestinal and systemic sites. Journal of Nutrition 130, 1412–1420.

Scharek L, Guth J, Reiter K, Weyrauch KD, Tara D, Schwerk P, Schierack P, Schmidt MF, Wieler LH, Tedin K (2005): Influence of a probiotic *Enterococcus faecium* strain on development of the immune system of sows and piglets. Veterinary Immunology and Immunopathology 105, 151–161.

Scharek L, Altherr BJ, Tolke C, Schidt MFG (2007): Influence of the probiotic *Bacillus cereus* var. *toyoi* on the intestinal immunity of piglets. Veterinary Immunology and Immunopathology 120, 136–147.

Schell TC, Lindemann MD, Kornegay ET, Blodgett DJ, Doerr JA (1993a): Effectiveness of different types of clay for reducing the detrimental effects of aflatoxin-contaminated diets on performance and serum profiles of weanling pigs. Journal of Animal Science 71, 1226–1231.

Schell TC, Lindemann MD, Kornegay ET, Blodgett DJ (1993b): Effects of feeding aflatoxin-contaminated diets with and without clay to weanling and growing pigs on performance, liver function, and mineral metabolism. Journal of Animal Science 71, 1209–1218.

Scheuermann SE (1993): Effect of the probiotic Paciflor (C.I.P. 5832) on energy and protein metabolism in growing pigs. Animal Feed Science and Technology 41, 181–189.

Setia A, Bhandari SK, House JD, Nyachoti CM, Krause DO (2009): Development and *in vitro* evaluation of an *Escherichia coli* probiotic able to inhibit the growth of pathogenic *Escherichia coli* K88. Journal of Animal Science 87, 2005–2012.

Shi YH, Xu ZR, Feng JL, Xia MS, Hu CH (2005): Effects of modified montmorillonite nanocomposite on growing/finishing pigs during aflatoxicosis. Asian-Australasian Journal of Animal Sciences 18, 1305–1309.

Shi YH, Xu ZR, Wang CZ, Sun Y (2007): Efficacy of two different types of montmorillonite to reduce the toxicity of aflatoxin in pigs. New Zealand Journal of Agricultural Research 50, 473–478.

Shim SB, Verstegen WA, Kim IH, Kwon OS, Verdonk JMAJ (2005): Effects of feeding antibiotic-free creep feed supplemented with oligofructose, probiotics or synbiotics to suckling piglets increases the preweaning weight gain and composition of intestinal microbiota. Archives of Animal Nutrition 59, 419–427.

Shim SB, Verdonk JMAJ, Pellikaan WF, Verstegen MWA (2007): Differences in microbial activities of faeces from weaned and unweaned pigs in relation to *in vitro* fermentation of different sources of inulin-type oligofructose and pig feed ingredients. Asian-Australasian Journal of Animal Sciences 20, 1444–1452.

Shu Q, Qu F, Gill HS (2001): Probiotic treatment using *Bifidobacterium lactis* HN019 reduces weaning diarrhea associated with rotavirus and *Escherichia coli* infection in a piglet model. Journal of Pediatric Gastroenterology Nutrition 33, 171–177.

Sissons JW (1989): Potential of probiotic organisms to prevent diarrhea and promote digestion in farm animals: a review. Journal of Food and Agriculture Science 49, 1–13.

Skirrow SZ, Buddle JR, Mercy AR, Madec F, Nicholls RR (1997): Epidemiological studies of pig diseases: 2. Post-weaning diarrhoea and performance in Western Australian pigs. Australian Veterinary Journal 75, 282–288.

Smiricky-Tjardes MR, Grieshop CM, Flickinger EA, Bauer LL, Fahey GC (2003): Dietary galactooligosaccharides affect ileal and total-tract nutrient digestibility, ileal and fecal bacterial concentrations, and ileal fermentative characterisctics of growing pigs. Journal of Animal Science 81, 2535–2545.

Smith HW, Huggins MB (1983): Effectiveness of phages in treating experimental *Escherichia coli* diarrhoea in calves, piglets and lambs. Journal of General Microbiology 129, 2659–2675.

Song DS, Kang BH, Oh JS, Ha GW, Yang JS, Moon HJ, Jang YS, Park BK (2006): Multiplex reverse transcription-PCR for rapid differential detection of porcine epidemic diarrhoea virus, transmissible gastroenteritis virus, and porcine group A rotavirus. Journal of Veterinary Diagnostic Investigation 18, 278–281.

Sorensen MT, Vestergaard EM, Jensen SK, Lauridsen C, Hojsgaard S (2009): Performance and diaahoea in piglets following weaning at seven weeks of age: Challenge with *E. coli* O 149 and effect of dietary factors. Livestock Science 123, 314–321.

Sousa CP (2006): The versatile strategies of *Escherichia coli* pathotypes: a mini review. Journal of Venomous Animals and Toxins Including Tropical Diseases 12, 363–373.

Spencer BT, Howell PG (1989): Some husbandry factors influencing weaning stresses in piglets. Journal of the South African Veterinary Association 60, 62–64.

Spencer KJ, Touchette KJ, Liu H, Allee GL, Newcomb MD, Kerley MS, Pace LW (1997): Effect of spray-dried plasma and fructooligosacharide on nursery performance and small intestinal morphology of weaned pigs. Journal of Animal Science 75 (Suppl. 1), 199.

Spriet SM, Decuypere JA, Henderickx HK (1987): Effect of *Bacillus toyoi* (Toyocerin) on the gastrointestinal microflora, concentration of some bacterial metabolites, digestibility of the nutrients and the small intestinal mean retention time in pigs. Medelingen van de Faculteit Landbouwwetenschappen, Rijksuniversiteit Gent 52, 71–75.

Srivastava RC, Ferookh A, Ahmad N, Mistra M, Hasan SK, Husain M. (1995): Reduction of Cis-platinum induced nephrotoxicity by zinc histidine complex: The possible implication of nitric oxide. Biochemistry and Molecular Biology International 36, 855–862.

Stokes CR, Bailey M, Haverson K, Harris C, Jones P, Inman C, Pie S, Oswald IP, Williams BA, Akkermans ADL, Sowa E, Rothkoetter HJ, Miller BG (2004): Postnatal development of intestinal immune system in piglets: implications for the process of weaning. Animal Research 53, 325–334.

Svensmark B, Nielsen K, Willeberg P, Jorsal SE (1989): Epidemiological studies of piglet diarrhoea in intensively managed Danish sow herds. 2. Post-weaning diarrhoea. Acta Veterinaria Scandinavica 30, 55–62.

Taras D, Vahjen W, Macha M, Simon O (2005): Response of performance characteristics and faecal consistency to long-lasting dietary supplementation with the probiotic strain *Bacillus cereus* var. *toyoi* to sows and piglets. Archives of Animal Nutrition 59, 405–417.

Tatara MR, Sliwa E, Dudek K, Gawron A, Piersiak T, Dobrowolski P, Mosiwicz J, Siwicki AK, Studzinski T (2008): Aged garlic extract and allicin improve performance and gastrointestinal tract development of piglets reared in artificial sow. Annals of Agricultural and Environmental Medicine 15, 63–69.

Teitelbaum JE, Walker WA (2002): Nutritional impact of pre- and probiotics as protective gastrointestinal organisms. Annual Review of Nutrition 22, 107–138.

Thomsson A, Rantzer D, Botermans J, Svedsen J (2008): The effect of feeding system at weaning on performance, health and feeding behaviour of pigs of different

sizes. Acta Agriculturae Scandinavica, Section A-Animal Science 58, 78–83.
Trckova M, Matlova L, Dvorska L, Pavlik I (2004): Kaolin, bentonite, and zeolites as feed supplements for animals: health advantages and risks. Veterinarni Medicina 49, 389–399. http://www.vri.cz/docs/vetmed/49-10-389.pdf
Trckova M, Vondruskova H, Zraly Z, Alexa P, Hamrik J, Kummer V, Maskova J, Mrlik V, Krizova K, Slana I, Leva L, Pavlik I (2009): The effect of kaolin feeding on efficiency, health status and course of diarrhoeal infections caused by enterotoxigenic *Escherichia coli* strains in weaned piglets. Veterinarni Medicina 54, 47–63. http://www.vri.cz/docs/vetmed/54-2-47.pdf
Tuohy KM, Rouzaud GCM, Bruck WM, Gibson GR (2005): Modulation of the human gut microflora towards improved health using prebiotics-assessment of efficacy. Current Pharmaceutical Design 11, 75–90.
Ushe TC, Nagy B (1985): Inhibition of small intestinal colonization of enterotoxigenic *Escherichia coli* by *Streptococcus faecium* M74 in pigs. Zentralblatt fur Bakteriologie Mikrobiologie und Hygiene, Serie B-Umwelthygiene Krankenhaushygiene Arbeitshygiene Praventive Medizin 181, 374–382.
Utiyama CE, Oetting LL, Giani PA, Ruiz UDS, Miyada VS (2006): Effects of antimicrobials, prebiotics, probiotics and herbal extracts on intestinal microbiology, diarrhea incidence and performance of weanling pigs. Brazilian Journal of Animal Science 35, 2359–2367.
Van Heugten E, Spears JW, Kegley EB, Ward JD, Qureshi MA (2003): Effects of organic forms of zinc on growth performance, tissue zinc distribution, and immune response of weanling pigs. Journal of Animal Science 81, 2063–2067.
Varley M (2002): Real future for herbal nutraceuticals. Pig Progress 18, 34–35.
Velebil D (2009): Clay minerals (in Czech). The National Museum, Prague. http://www.velebil.net/mineraly/jilove-mineraly
Visentini PRS, Berto DA, Wechsler FS, Wechsler FS, Cardoso TA, Castro VD (2008): Feeding piglets in the nursey with rations containing fructooligosacharides. Ciencia Rural 38, 1402–1406.
Vrzgula L, Bartko P, Blazovsky J, Kozac J (1982): The effect of the administration of clinoptilolite on the health condition, blood picture and weight gains of pigs (in Czech). Veterinarni Medicina 27, 267–274.
Walsh CT, Sandstead HH, Prasad AS, Newberne PM, Fraker PJ (1994): Zinc: health effects and research priorities for the 1990s. Environment and Health Perspective 102, 5–46.
Wang JJ, Chen LX, Li P, Li XL, Zhou HJ, Wang FL, Li DF, Yin YL, Wu GY (2009): Gene expression is altered in piglet small intestine by weaning and dietary glutamine supplementation. Journal of Nutrition 138, 1025–1032.
Wapnir RA (2000): Zinc deficiency, malnutrition and gastrointestinal tract. Journal Nutrition 130, 1388–1392.
Wattrang E, Wallgren P, Lindberg A, Fossum C (1998): Signs of infections and reduced immune functions at weaning of conventionally reared and specific pathogen free pigs. Journal of Veterinary Medicine Series B-infectious Diseases and Veterinary Public Health 45, 7–17.
Wegener HC, Aarestrup FM, Jensen LB, Hammerum AM, Bager F (1998): The association between the use of antimicrobial growth promoting and therapeutic antimicrobials. Journal of Animal and Feed Sciences 7, 7–14.
Williams LB, Haydel SE, Giese RF, Eberl DD (2008): Chemical and mineralogical characteristics of French green clays used for healing. Clays and Clay Minerals 56, 437–452.
Wilson AD, Stokes CR, Bourne FJ (1989): Effect of age on absorption and immune-responses to weaning or introduction of novel dietary antigens in pigs. Research in Veterinary Science 46, 180–186.
Xia MS, Hu CH, Xu ZR, Ye Y, Zhou YH, Xiong L (2004): Effects of copper-bearing montmorillonite (Cu-MMT) on *Escherichia coli* and diarrhea on weanling pigs. Asian-Australasian Journal of Animal Sciences 17, 1712–1716.
Xia MS, Hu CH, Xu ZR (2005): Effects of copper bearing montmorillonite on the growth performance, intestinal microflora and morphology of weanling pigs. Animal Feed Science and Technology 118, 307–317.
Xu S, Sheng G, Boyd SA (1997): Use of organoclays in pollution abatement. Advances in Agronomy 59, 25–62.
Xu ZR, Han XY, Wang YZ (2004): Effects on growth and cadmium residues from feeding cadmium-added diets with and without montmorillonite nanocomposite to growing pigs. Veterinary and Human Toxicology 46, 238–241.
Ye Y, Zhou YH, Xia MS, Hu CH, Ling HF (2003): A new type of inorganic antibacterial material: Cu-bearing montmorillonite and discussion on its mechanism. Journal of Inorganic Materials 18, 569–574.
Yu D.Y., Xu Z.R., Yang X.G. (2005): Effects of lead and particulate montmorillonite on growth performance, hormone and organ weight in pigs. Asian-Australasian Journal of Animal Sciences, 18, 1775–1779.
Yu DY, Li XL, Li WF (2008): Effect of montmorillonite superfine composite on growth performance and tis-

sue lead level in pigs. Biological Trace Element Research 125, 229–235.
Zanchi R, Canzi E, Molteni L, Scozzoli M (2008): Effect of *Camellia sinensis* L. whole plant extract on piglet intestinal ecosystem. Annals of Microbiology 58, 147–152.
Zhou YH, Xia MS, Ye Y, Hu CH (2004): Antimicrobial ability of $Cu2^{+}$-montmorillonite. Applied Clay Science 27, 215–218.
Zimmermann B, Bauer E, Mosenthin R (2001): Pro- and prebiotics in pig nutrition – potential modulators of gut health? Journal of Animal and Feed Sciences 10, 47–56.

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