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REVIEW ARTICLE

Non-antibiotic possibilities in prevention and treatment of calf diarrhoea

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Abstract

Due to increasing bacterial antibiotic resistance and the consumers' tendency to choose organic products, cattle farmers are interested in alternative methods of calf diarrhoea treatment. This is a major challenge for veterinarians. Few methods of non-antibiotic treatment that bring satisfactory results have been reported in the related literature so far. In this article, the authors compare different non-antibiotic methods of diarrhoea prevention and treatment in calves. Among the alternatives discussed are herbs, probiotics, prebiotics and synbiotics, lactoferrin, and bacteriophages. It was found that the best results could be achieved through the use of pro-, pre- and synbiotics. However, the authors would like to point out that with the expansion of knowledge about the practical use of broad-scale bacteriophages, they could be the best alternative to antibiotics.

Keywords: calf diarrhoea, non-antibiotic treatment, probiotics.

Introduction

Bacterial resistance to antibiotics is a naturally occurring phenomenon in nature. It was observed during the first studies of penicillin (15, 32).

Selective environmental pressure from the presence of antimicrobial substances allows and promotes the survival of bacteria carrying resistance genes. This phenomenon occurs especially when antibiotic doses are too low or therapy is too short (1). An extremely dangerous consequence of the formation of antibiotic-resistant strains is the possibility of their transfer from animal organisms to the environment and people through direct or indirect contact with food of animal origin (9). Dairy cows and calves can be the source of many zoonotic microorganisms including bacteria from the digestive tract, such as the enterotoxigenic *Escherichia coli* strain. This and enteropathogenic strains of *E. coli* are most often the causes of diarrhoea and high mortality in newborn calves, and these animals may also be a reservoir of the enterohaemorrhagic strain which produces the dangerous Shiga toxin. Shigatoxigenic *E. coli* induces apoptosis of intestinal cells and is associated with haemolytic-uraemic syndrome and haemorrhagic colitis in humans. Careless use of antibacterial agents in animals has led to the resistance of these *E. coli* strains to β-lactam antibiotics (ampicillin and amoxicillin with clavulanic acid), aminoglycosides (gentamicin), fluoroquinolones, or combined preparations containing trimethoprim (58). In recent years, the occurrence of coagulase-negative staphylococci (CNS) in the dairy cattle population has increased. CNS are resistant to many antibiotics, including penicillin G, ampicillin, amoxicillin, gentamycin, and oxytetracycline (57). Other bacteria isolated from cattle (including calves) which are a potential zoonotic threat and show resistance to antibiotics are *Salmonella* and *Campylobacter* (24). These bacteria cause the highest number of clinically differentiated diseases in humans, especially in the digestive tract (43). Vancomycin-resistant enterococci (VRE) and methicillin-resistant staphylococci (MRSA), which may be the cause of dangerous nosocomial infections, are occasionally isolated from cows (56). Restrictions on the use of veterinary antimicrobial agents to limit the spread of resistance in bacterial populations have already been introduced in several countries (12). Belgium has set itself the goal of reducing the use of antibiotics by 50% by 2020, and in the Netherlands, with declining sales of antibiotics for veterinary use since 2007, the resistance level of commensal *E. coli* in chickens, pigs, and calves has significantly decreased since 2010 (5, 19). Given the above risks, it is necessary to develop and implement new strategies to counter bacterial calf diarrhoea which can replace and thus limit antibiotic therapy and prevent the spread of antibiotic-resistant bacteria in human and animal populations.

Diarrhoea in calves

According to data from the National Animal Health Monitoring System (60) in the USA, diarrhoea in newborn calves is the most common disorder of dairy heifers in the pre-production period, affecting almost 19% of the animal population. It is also the leading cause of death in premature calves (59). The most common pathogens for diarrhoea in young calves (less than 4 weeks old) are rotaviruses, coronaviruses, *Cryptosporidium parvum*, and *E. coli* (16). Due to the multifactorial nature of the disease, it is necessary to use antibacterial, antiprotozoal, and immunomodulatory agents in its treatment.

Alternative therapies can be carried out with many agents available on the market, *i.a.* preparations containing pre- and probiotics, synbiotics or herbal mixtures and extracts. Diet supplementation with vitamins and minerals and fluid therapy also have a supporting effect. There are also promising methods not yet fully developed, such as phage therapy. This publication contains information about the mechanisms of action of substances as herbs, probiotics, prebiotics, synbiotics, lactoferrin, and bacteriophages when administered for prevention and treatment of diarrhoea (Table 1).

Table 1. Mechanisms of actions of discussed substances

Substances	Mechanism of action
Herbs	Disintegration of the cell membrane of the bacteria and instigation of ion migration outside the cell through the agency of essential oils
	Increase in the phagocytic activity of macrophages, and the number of stimulated B and T lymphocytes, and stimulation of the synthesis of interferon through the agency of phenolic compounds, terpenes, alkaloids and many others
	Increase in the number of lactobacilli in the digestive tract
Probiotics	Production of antibacterial substances by organisms colonising the digestive tract: organic acids – rapid reduction in pH below the optimum for the growth of pathogenic microorganisms and also inhibition of bacterial activity by undissociated acid molecules that acidify their cytoplasm hydrogen peroxide – oxidation of disulphide bridges in bacterial cell proteins bacteriocins – nisin, acidolin, acidophilin, lactacin, lactocidine, lactoline, and enterocine with bacteriostatic and bactericidal activity secreted outside the probiotic bacteria cell
	Formation of natural biofilm in the mucosa of the intestine – a barrier against potentially pathogenic factors
	Increase in immunoglobulin level, γ-interferon production and activity of lymphocytes and macrophages
Yeasts	Impact on the metabolic activity of lactic acid-producing bacteria
	Production of B vitamins, positively affecting the growth of positive bacterial flora
	Glucan and mannan (components of cell wall) activity against pathogenic bacteria growth
Prebiotics	Selective stimulation of growth or increase in the activity of the positive intestinal microflora by these food ingredients resistant to the action of digestive enzymes
Synbiotics	Selective promotion of the probiotic component in these mixtures of probiotics and prebiotics (synergy effect)
	Provision of energy and carbohydrates for the rumen microbes and increase in the amount of propionate and short-chain fatty acids
Lactoferrin	Increase in mobility of some bacteria (including *E. coli*) in the intestines, hindrance of adhesion to epithelial cells and biofilm formation
	Bacteriostatic effect – binding iron ions and limiting access to them
	Bactericidal effect – interaction of the strong positive end of the molecule on the wall of the bacterial/fungal cell, degrading it and leading to the leakage of intracellular components
Bacteriophages	Lytic cycle – activation of the lytic proteins (by the critical mass of phage progeny inside the host cell) which hydrolyse the peptidoglycan cell wall releasing novel phages

Herbs

Thyme, oregano, and sage have the strongest antimicrobial activity. The active substances in these herbs are essential oils that disintegrate the cell membrane of the bacteria and cause the migration of ions outside the cell. Other active substances are limonene and lemon aldehyde, which cause changes in long-chain fatty acids of *E. coli* cell membranes (11, 68). Available test findings also indicate the probiotic effect of herbs. The results of studies published by Castillo *et al.* (13) showed that the use of a 0.03% plant extract standardised with 5% carvacrol extracted from *Origanum* spp., 3% cinnamaldehyde extracted from *Cinnamomum* spp., and 2% capsicum oleoresin from *Capsicum annum* leads to an increase in the number of lactobacilli in the digestive tracts of animals. Herbal preparations can also be immunostimulating in many ways. Phenolic compounds, terpenes, alkaloids, saponins, essential oils, polysaccharides, glycoproteins, tannins, mucus, and many other substances contained in herbs increase the phagocytic activity of macrophages and the number of stimulated B and T lymphocytes, and stimulate the synthesis of interferon. These substances are included in *Echinacea*, garlic, aloe, mountain arnica, oregano, and nettle (18, 21, 23).

Research carried out by Nowak *et al.* (41) showed that thyme containing phytobiotics and watery oregano extract (used at 200 g/t feed) reduced dry matter concentration of ileal digesta in pigs, which could possibly indicate a faster digestion of feed or proliferation of microorganisms. The group that received phytobiotic extract had a significantly lower number of yeasts and moulds in caecal digesta, but no effect was observed on the *E. coli* or *Clostridium* count. The study also showed that administering herbal extracts separately (without addition of pro- or prebiotics) improved animal performance significantly. Another study, carried out by Klebaniuk *et al.* (31), showed that addition of dried herbs (3% of dry matter per day per animal) for dry cows had a positive effect on the subsequent composition of colostrum. It contributed to a higher level of immunoglobulins in comparison with the control group not receiving the supplement. A significant reduction in diarrhoea and a higher concentration of IgG antibodies in the serum of calves born to cows receiving herbal supplements were observed. Research on the effectiveness of herbal mixtures in the treatment of calves' diarrhoea was also carried out in India, where mixtures containing *Catechu acacia*, Bengal quince, Indian barberry, greater plantain, or common pomegranate were administered at a 15 g dose twice a day until recovery. These herbs have anti-diarrhoeal, styptic, analgaesic, anti-inflammatory, and antiprotozoidal properties and absorb excess water from the intestines. In an experiment comparing the efficacy of herbal anti-diarrhoeal products with antibiotics, calves were divided into three groups: two of them received herbal mixtures and the third chemotherapy (ciprofloxacin and tinidazole). The administration of six doses of mixtures or chemotherapeutics cured all animals from the three groups, which proves that herbal mixtures are as effective as ciprofloxacin and tinidazole (48).

Another plant used in the treatment of diarrhoea on some organic farms is garlic in the form of extract added to feed. An evaluation of the effects of feeding garlic (*Allium sativum*) powder or garlic with probiotics on diarrhoeal incidence and immunoglobulin response in pre-weaned Holstein calves was performed recently. One of the treated groups was supplemented with 5 g/d of garlic powder, a second was supplemented with 4 g/d of probiotics (total viable count: 1.3×10^7 CFU/g), and the third received both garlic and probiotics. Calves fed garlic and garlic-probiotic mixture had higher serum IgG levels than the control group and calves fed probiotics. Moreover, calves fed the garlic-probiotic mixture had lower faecal scores, fewer days of diarrhoea, and higher final body weight compared to the control group (30).

An evaluation of the effect of oral administration of chestnut tannins (*Castanea sativa mill.*) on the duration of diarrhoea in newborn calves was carried out in 2018. Two litres of warm water was mixed with 10 g of chestnut tannins as extract powder (750 g/kg of dry matter equivalent of tannic acid). It was found that the occurrence of diarrhoea was significantly higher in the control group, which was not given the powdered chestnut tannin extract in water (8). However, it should be borne in mind that obtaining curative effects similar to antibiotics requires the use of high concentrations of extracts and essences. For this reason, phytotherapy is safer to use in the prevention of infections when doses are low than in their treatment when they must be higher.

Probiotics

Probiotics are products containing living cells (*e.g.* lyophilised cells or active bacteria), improving the health of humans and animals and exerting beneficial effects in the oral cavity, alimentary tract, respiratory tract, or urogenital system (4, 42). The effect of probiotics is based on several biological and biochemical mechanisms. The first of these is the production of antibacterial substances by organisms colonising the digestive tract. These include organic acids, hydrogen peroxide, and bacteriocins. The antibacterial effect of organic acids is the result of a rapid reduction in pH lower than the optimum for the growth of pathogenic microorganisms and the inhibition of bacterial activity by undissociated acid molecules that acidify their cytoplasm. This activity is characteristic of lactic acid. Acetic acid, in turn, can cause denaturation of intracellular proteins lowering the pH inside the cell. Hydrogen peroxide is particularly effective against microbes that have low levels of enzymes, such as peroxidase or catalase. It causes the oxidation of disulphide bridges in bacterial cell proteins. However, the most interesting substances produced by probiotic bacteria are bacteriocins that have bacteriostatic and bactericidal activity. These are protein elements produced by the probiotic bacteria and secreted outside the cell. The best-known bacteriocins are nisin, acidolin, acidophilin, lactacin, lactocidine, lactoline, and enterocine, and they have antibacterial activity against, for example, *E. coli*, *S.*

Typhimurium, *C. perfringens*, or *S. aureus* (45, 49, 52). According to the studies carried out by Perdigón *et al.* (43), the administration of fermented milk with probiotic bacteria to mice at a dose of 100 µg/day for 8–11 consecutive days stimulates the efficiency of the immune system. Probiotic bacteria form a natural biofilm in the mucosa of the intestine and constitute a barrier against potentially pathogenic factors, boosting the immune system. Immunostimulation is also manifested in increased immunoglobulin and γ-interferon production and increased lymphocytes and macrophages activity (20, 43).

The most common probiotic bacteria used in calves are those of the *Lactobacillus* and *Bacillus* genera and *Enterococcus faecium*. It was shown that early colonisation of ruminants' intestines by *Lactobacillus* can reduce pathogenic flora adhesion to the mucous membrane, and administration of 1.85×10^7 CFU/L of *Lactobacillus* species in young calves has been shown to improve weight gain and immunocompetence (2).

A large number of studies have been devoted to the probiotic properties of yeast, in particular *Saccharomyces cerevisiae*. The presence of yeast affects the metabolic activity of lactic acid–producing bacteria. Yeasts also produce B vitamins, encouraging the growth of positive bacterial flora. Furthermore, the cellular components of the cell wall of these organisms (glucan and mannan) protect the digestive tract from the growth of pathogenic *E. coli* and *Klebsiella pneumoniae* (37). It was also shown that feeding yeast culture at 2% of grain dry matter improved health, minimised frequency of treatment need, and reduced risk of morbidity and mortality in dairy calves (39).

Prebiotics

These are undigested feed ingredients that are resistant to the action of digestive enzymes. They beneficially affect the host organism by selectively stimulating growth or increasing the activity of the positive intestinal microflora (28). Examples of prebiotics used in ruminants are mannan oligo-saccharides (MOS), fructo-oligo-saccharides (FOS) and galactosyl-lactose (GL). MOS are the complex mannose sugars blocking the colonisation of pathogens in the digestive tract, the term FOS is used for short-chain fructans containing 2–4 fructosyl units linked by glycosidic linkages (33) and GL is a trisaccharide produced during the digestion of whey with β-galactosidase. The beneficial effect of GL addition at 1% of dry matter in the milk replacer on the growth and health of dairy calves has been demonstrated (46). MOS, FOS, and GL supplementation may improve calf growth; however, changes in the microbial fermentation activity of these sugars have not yet been studied in detail.

Synbiotics

The preparations which are a mixture of probiotics and prebiotics are called synbiotics, the term applying to products of which the prebiotic part selectively promotes the probiotic component. Due to the synergy effect, they are given together in order to restore the intestinal microbial balance (55). Examples of synbiotics used in ruminants are products that combine oligosaccharides (*e.g.* citrus pectin) with bacterial cultures. Research on the effectiveness of probiotic/synbiotic preparations in calves with diarrhoea has also been carried out in Poland, when calves with symptoms of diarrhoea were given a preparation containing *Bacillus licheniformis* and *Bacillus subtilis* (in the ratio 1:1 and amount 6.4×10^{10} CFU), *Enterococcus faecium* (3×10^{10} CFU), betaine, vitamins, and macroelements at a dose of 20 g of the mixture per calf for 8–10 days. During this period, the condition, appetite, thirst, and diarrhoea were observed and, except for condition, were categorised as present, absent, or mild/poor in the experimental group. After 10 days, all of the parameters assessed had improved. Diarrhoea no longer affected 90% of the calves examined at all, and affected the other 10% only mildly (26).

The reason for the positive effect of prebiotics and synbiotics on the calf organism may be the provision of energy and carbohydrates for the rumen microbes necessary in the fermentation process, thus increasing the amount of propionate and short-chain fatty acids. The administration of oligosaccharides to weaned calves appears to be beneficial because the formation of the desired microbial flora in the rumen and lower parts of the gastrointestinal tract can prevent diarrhoea (61).

Lactoferrin

Lactoferrin is naturally found in body fluids and secretions, including milk. It is a bioactive protein with a number of virtues, including immunoregulatory, anti-inflammatory, bacteriostatic, antibacterial, antiviral, and antifungal properties (22). The use of this protein in the treatment of calf diarrhoea is associated with its effect on pathogens in the gastrointestinal tract and the impact on the immune system. In the intestines, lactoferrin increases the mobility of some bacteria (including *E. coli*), hindering adhesion to epithelial cells and biofilm formation. In addition, it has a bacteriostatic effect, which involves binding iron ions, thus limiting other bacteria's access to them, and as a result inhibits the growth and expansion of microorganisms. The antibacterial and antifungal activity is based on the interaction of the strong positive end of the molecule on the wall of the bacterial/fungal cell, degrading it and leading to the leakage of intracellular components and consequently to the cell's death (22). However, the antiviral activity has been confirmed only in relation to HIV and HBV

(22). There is a need for further research to determine the utility of this protein in the treatment of calf diarrhoea. It is also important that not only lactoferrin is involved in these processes, but also peptides resulting from the digestion of its molecules, including lactoferricin (22). Experimentally, it was found that in calves with diarrhoea which were administered lactoferrin (in the form of a lyophilisate dissolved in distilled water in the proportion of 3 g of protein per 30 mL of water) the risk of death was halved on day 120 after the diagnosis compared to the control group which received only water. However, there were no significant differences in the duration of the disease or intensity of symptoms (25). Attention should also be paid to the usefulness of lactoferrin in the prevention of calf diarrhoea. In a study by Pempek *et al.* (43), it was found that the prophylactic administration of lactoferrin (1 g of powder added to the feed mixture administered in the evening) reduced the incidence of diarrhoea in calves by 2% compared to the control group. Further research into the use of lactoferrin in both the prevention and treatment of diarrhoea may recommend it as a very important alternative to antibiotic therapy.

Bacteriophages

Bacteriophages are a separate group of viruses that infect bacteria. Two types of bacteriophage impact on bacterial cells are distinguished: depending on the cycle, it may be bacterial death (the lytic cycle) or continued vitality with latent viral DNA being carried (the lysogenic cycle). Phages typically bind to their specific receptor on the bacterial cell surface, inject their genetic material into the host cell, integrate it into the bacterial genome, and then reproduce vertically. Attainment of a critical mass of phage progeny inside the host cell activates the lytic proteins, which hydrolyse the peptidoglycan cell wall, releasing novel phages to reinitiate the lytic cycle (63). Lysogenic phages integrate their genetic material into the bacterial chromosome in the form of endogenous prophages. Phage DNA can also remain separate in plasmids and be stably transmitted across bacterial generations; however, this unintegrated DNA is less common (36). Prophage genes can be beneficial to the bacterial host, because they can encode virulence factors (*e.g.* Shiga and botulinum toxins) and metabolic or antibiotic resistance genes (17, 40, 44, 47). Environmental stressors on the bacteria host may lead to transformation of the lysogenic cycle into the lytic cycle (63). Conventional phage therapy is based on the lytic phages and mechanism of the lytic cycle that obligately kill their bacterial host.

Bacteriophage therapy is an interesting alternative or complement to antibiotic therapy. Due to a different mechanism of action on the bacterial cell, it can bring satisfactory results even in the treatment of those infections that have been caused by bacteria resistant to most known antibiotics (36). Recent studies using animal models have demonstrated phage activity against vancomycin-resistant *E. faecium* (7), extended-spectrum β-lactamase–producing *E. coli* (64), imipenem-resistant *Pseudomonas aeruginosa* (65), *S. aureus* (54), and *Acinetobacter baumannii* (67). The effectiveness of phages against multidrug-resistant *E. coli* O25:H4-ST131 at a dose of 2×10^9 PFU per 1 mL of injection solution was confirmed in several studies on calves, as was the action of IP phage pVp-1 against *Vibrio parahaemolyticus* in 2.0×10^8 PFU of phages per mouse in a single injection or oral administration (29). There are also reports that phages are able to restore sensitivity to antibiotics in resistant bacteria (14). An important aspect is also the high selectivity of these microorganisms in relation to bacteria and the lack of harmful effects on both the animal's body and the positive bacterial flora living in its digestive system (35). Thus, phage therapy causes less unwanted modification of the gut microbiome and is still effective against gut-carried pathogens (36). The impact on the immune system seems to be promising because bacteriophage-induced stimulation of this system has been observed by scientists (10, 36). However, a disadvantage of this type of therapy is the possibility of resistance to phages developing and the production of specific antiphage antibodies being induced (62).

Taking into consideration the disadvantages associated with phage therapy, it appears interesting to use engineered recombinant phage lytic proteins. There are two major proteins employed by the majority of phage species during the lysis of a bacterial host: a transmembrane-penetrating protein called holin and a peptidoglycan cell wall hydrolase called lysin. These two proteins act together and trigger the lysis of bacterial host cells. At the end of the lytic cycle, the holin creates openings in the cell membrane on the cytoplasmic side, and this allows lysin to hydrolyse the cell wall (50). Phage lysins may be highly specific or exhibit a broad spectrum of activity between strains or even between species. Lysins alone are capable of bacterial cell lysis. Studies in mice show that lysins exhibit activity against multidrug-resistant *A. baumannii* (38), *Streptococcus pneumoniae* (69), and MRSA (53). It was also demonstrated that combining lysins and antibiotics may be more effective in eliminating infection than using antibiotics alone (66). Moreover, phage lytic proteins may be easier to mass produce and administer, and neither are susceptible to nor cause the formation of neutralising antibodies, in contrast to preparations containing live phages (27).

Phage therapy was used to treat diarrhoea in calves, pigs, and lambs, but these studies were published without detailed information on the phages used (3). It is experimentally possible to isolate a broad spectrum phage demonstrating bactericidal activity *in* *vitro* against 47.3% of *E. coli* isolates causing calf diarrhoea (34).

In 2011, Bicalho *et al*. (6) attempted to treat calves with phages isolated from manure from two large local farms. A cocktail of four isolated bacteriophages was administered orally to six of the ten calves in the study, which were administered the drug twice daily after a meal for 11 days. The assessment was based on morphological tests of the blood and measurement of the amount of bacteria in the stool. A reduction in the number of bacteria in the faeces was noted, but it was too low to be diagnostically significant. The authors pointed out that the explanation for this result may be the excessively small number of animals used in the experiment, the method of obtaining bacteriophages or the route of administration (6). Nevertheless, Rozema *et al.* (51) in 2009 compared two methods of phage administration: *per os* and *per rectum* and concluded that the longer retention time experienced by the orally administered phages within the digestive tract increased the opportunity for the phages to interact with *E. coli* cells and decreased their population size. In *in vitro* studies, Bicalho *et al.* (6) noted that phage efficacy at pH 1 and 2 was nonexistent, whereas at pH between 3 and 11 their viability was difficult to determine. Hence the hypothesis that the reason for the low effectiveness of the therapy was not the exposure of phages to the low pH of the stomach because they were administered after a meal when the pH level was 3. Bicalho *et al.* (6) assumed that the reason for their unencouraging results was probably the method of obtaining phages, in which dairy cow uterine *E. coli* strains were used to multiply and purify the culture, while better results could have been obtained with those isolated from the gastrointestinal tract. Further studies concerning phages should be carried out on a larger group of animals. It is appropriate to mention that no side effects were observed in any of the calves.

Although the origins of phagotherapy research date back to 1919, this topic still raises many unknowns, such as effects on the host organism, interaction between phages and bacteria, and duration of the therapy (36). In veterinary medicine, it is still not available on a large scale.

Conclusion

Due to the increasing antibiotic resistance among bacteria, non-antibiotic methods of treatment of diarrhoea are crucial and relevant, which is reflected in the growing interest in this topic. Over the past few years, the number of studies and scientific publications in this area has increased, although there are still no clearly schematised and effective treatment programmes.

The effects of herbs have been experimentally confirmed, but their role seems to be greater in the prevention than in the treatment of diarrhoea. A big difficulty in their use is appropriate dose selection. Other elements to take into account are their origin, cultivation (traditional/ecological), and standardisation in terms of active substance content, stringent specifications for which may attach high costs to this method. In the case of lactoferrin, the effects achieved in the prevention and treatment of diarrhoea should prompt physicians to undertake further research and put the findings into use on a large scale. Bacteriophages are a kindred subject; due to a high level of interest in this subject in human medicine, we can expect that their importance in veterinary medicine will also grow. Unfortunately, just as phage therapy is not widely available in humans, it is also unavailable on a large scale in animals. The best results from the methods presented in this work come from the use of probiotics, prebiotics, and synbiotics. This reflects the fact that they are the best-known products and the most widely available in the form of commercial preparations. The active substances referred to in this article act on many levels and can therefore be used alone or in combination with traditional therapy.

The reduction in the use of antibiotics in animals is written into policy and serves the real needs of animals. The methods discussed in this paper are already applied on organic farms around the world. From the trends prevailing also among consumers, we can conclude that there is impetus to enhance the alternative methods of diarrhoea treatment in calves and attend to the further research into their effectiveness which they require.

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References

1. Albrich W.C., Monnet D.L., Harbarth S.: Antibiotic selection pressure and resistance in *Streptococcus pneumoniae* and *Streptococcus pyogenes*. Emerg Infect Dis 2004, 10, 514–517.
2. Al-Saiady M.Y.: Effect of probiotic bacteria on immunoglobulin G concentration and other blood components of newborn calves. J Anim Vet Adv 2010, 9, 604–609.
3. Anand T., Vaid R.K., Bera B.C., Barua S., Riyesh T., Virmani N., Yadav N., Malik P.: Isolation and characterization of a bacteriophage with broad host range, displaying potential in preventing bovine diarrhoea. Virus Genes 2015, 51, 315–321.
4. Barbosa T.M.: The impact of antibiotic use on resistance development and persistence. Drug Resist Updat 2000, 3, 303–311.
5. BelVet-SAC. Belgian Veterinary Surveillance of Antibacterial Consumption National Consumption Reports 2016 and 2017.

Faculty of Veterinary Medicine, University of Ghent, Ghent, 2016 & 2017.
6. Bicalho M.L.S., Machado V.S., Nydam D.V., Santos T.M.A., Bicalho R.C.: Evaluation of oral administration of bacteriophages to neonatal calves: phage survival and impact on fecal *Escherichia coli*. Livest Sci 2012, 144, 294–299.
7. Biswas B., Adhya S., Washart P., Paul B., Trostel A.N., Powell B., Carlton R., Merril C.R.: Bacteriophage therapy rescues mice bacteremic from a clinical isolate of vancomycin-resistant *Enterococcus faecium*. Infect Immun 2002, 70, 204–210.
8. Bonelli F., Turini L., Sarri G., Serra A., Buccioni A., Mele M.: Oral administration of chestnut tannins to reduce the duration of neonatal calf diarrhea. BMC Vet Res 2018, 14, 227.
9. Buczek K., Marć M.: Bacterial antibiotic resistance – reasons and effects. Ann Univer Mariae Curie-Sklodowska, Lublin 2009, 64, 3.
10. Budynek P., Dąbrowska K., Skaradziński G., Górski A.: Bacteriophages and cancer. Arch Microbiol 2010, 192, 315–320.
11. Burt S.: Essential oils: their antibacterial properties and potential applications in food – a review. Int J Food Microbiol 2004, 94, 223–255.
12. Callens B., Cargnel M., Sarrazin S., Dewulf J., Hoet B., Vermeersch K., Wattiau P., Welby S.: Associations between a decreased veterinary antimicrobial use and resistance in commensal *Escherichia coli* from Belgian livestock species (2011–2015). Preventive Veterinary Medicine 2018, 157, 50–58.
13. Castillo M., Martin-Orue S.M., Roca M., Manzanilla E.G., Badiola I., Perez J.F., Gasa J.: The response of gastrointestinal microbiota to avilamycin, butyrate, and plant extracts in early weaned pigs. J Anim Sci 2006, 84, 2725–2734.
14. Chan B.K., Sistrom M., Wertz J.E., Kortright K.E., Narayan D., Turner P.E.: Phage selection restores antibiotic sensitivity in MDR *Pseudomonas aeruginosa*. Sci Rep 2016, 6, 26717.
15. Chandra H., Bishnoi P., Yadav A., Patni B., Mishra A.P., Nautiyal A.R.: Antimicrobial resistance and the alternative resources with special emphasis on plant-based antimicrobials. Review. Plants 2017, 6, 16.
16. Cho Y., Yoon K.J.: 2014. An overview of calf diarrhea – infectious etiology, diagnosis, and intervention. J Vet Sci 2014, 15, 1–17.
17. Colomer-Lluch M., Jofre J., Muniesa M.: Antibiotic resistance genes in the bacteriophage DNA fraction of environmental samples. PLoS One 2011, 6, e17549.
18. Craig W.J.: Health-promoting properties of common herbs. Am J Clin Nutr 1999, 70 (Suppl.), 491–499.
19. Dorado-García A., Mevius D.J., Jacobs J.J.H., Van Geijlswijk I.M., Mouton J.W., Wagenaar J.A., Heederik D.J.: Quantitative assessment of antimicrobial resistance in livestock during the course of a nationwide antimicrobial use reduction in the Netherlands. J Antimicrob Chemother 2016, 71, 3607–3619.
20. Fooks L.J., Fuller R., Gibson G.R.: Prebiotic, probiotics, and human gut microbiology. Int Dairy J 1999, 9, 53–61.
21. Frankič T., Voljč M., Salobir J., Rezar V.: Use of herbs and spices and their extracts in animal nutrition. Acta Agricult Slov 2009, 94, 2, 95–102.
22. Gajda-Morszewski P., Śpiewak K.: Laktoferyna – białko multipotencjalne (Lactoferrin – a multipotential protein – in Polish), Zeszyty Naukowe Towarzystwa Doktorantów Univer Jagiel, Nauki Ścisłe 2015, 10, 177–188.
23. Grela E.R., Sembratowicz L., Czech A.: Immunostimulatory effects of herbs. Med Weter 1998, 54, 152–158.
24. Guardabassi L., Jensen L.B.: Guide to antimicrobial use in animals. Edited by H. Kruse, Blackwell Publishing, Oxford, 2008, pp. 1–223.
25. Habing G., Harris K., Schuenemann G.M., Piñeiro J.M., Lakritz J., Alcaraz Clavijo X.A.: Lactoferrin reduces mortality in preweaned calves with diarrhea. J Dairy Sci 2017, 100, 3940–3948.
26. Hamala A., Kotwica S., Kumala A, Rypuła K.: Probiotyki – alternatywny sposób na leczenie biegunek u cieląt. (Probiotics – an alternative treatment modality for calf diarrhoea – in Polish). Weter Teren 2012, 6, 65–68.
27. Hodyra-Stefaniak K., Miernikiewicz P., Drapała J., Drab M., Jończyk-Matysiak E., Lecion D., Kaźmierczak Z., Beta W., Majewska J., Harhala M., Bubak B., Kłopot A., Górski A., Dąbrowska K.: Mammalian host-*versus*-phage immune response determines phage fate *in vivo*. Sci Rep 2015, 5, 14802.
28. Jiang H.Q., Gong L.M., Ma Y.X., He Y.H., Li D.F., Zhai H.X.: Effect of stachyose supplementation on growth performance, nutrient digestibility and caecal fermentation characteristics in broilers. Brit Poult Sci 2006, 47, 516–522.
29. Jun J.W., Shin T.H., Kim J.H., Shin S.P., Han J.E., Heo G.J., De Zoysa M., Shin G.W., Chai J.Y., Park S.C.: Bacteriophage therapy of a *Vibrio parahaemolyticus* infection caused by a multiple-antibiotic-resistant O3:K6 pandemic clinical strain. J Infect Dis 2014, 210, 72–78.
30. Kekana T.: Effects of supplemental garlic (*Allium sativum*) powder and probiotics on diarrhea and immunoglobulin response in pre-weaned dairy calves. J Dairy Sci 2014, 97, E-Suppl 1, 565.
31. Klebaniuk R., Grela E.R., Kowalczuk-Vasilev E., Olcha M., Góźdź J.: The effectiveness of the use of organic herbal blends for cattle. Wiad Zootech 2014, 52, 56–63.
32. Kozińska A., Sitkiewicz I.: "Nowe" i "stare" antybiotyki – mechanizmy działania i strategie poszukiwania leków przeciwbakteryjnych. (New and old antibiotics – their mechanisms and the approaches to discovering antibacterial drugs – in Polish). Kosmos – problemy nauk biologicznych 2017, 314, 109–124.
33. Kubik C., Piasecka K., Anyszka A., Bielecki S.: Polyfructans and fructooligosaccharides (FOS) – occurrence, production, and application. Biotechnologia. 2006, 73, 102–116.
34. Letkiewicz S., Międzybrodzki R., Górski A.: Terapia fagowa antybiotykoopornych zakażeń bakteryjnych - powrót do źródeł terapii przeciwbakteryjnych. Part 1. (Phage therapy treating antimicrobially resistant infections – a return to the origins of antibacterial therapy. Part 1 – in Polish). Przegląd Urol 2017, 102, 40–45.
35. Letkiewicz S., Międzybrodzki R., Górski A.: Historia rozwoju terapii fagowej w Polsce. Part 2. (The history of phage therapy in Poland. Part 2 – in Polish). Przegląd Urol 2017, 103, 69–71.
36. Lin D.M., Koskella B., Lin H.C.: Phage therapy: an alternative to antibiotics in the age of multi-drug resistance. World J Gastrointest Pharmacol Ther 2017, 8, 162–173.
37. Lipiński K.: Mechanizm działania probiotyków paszowych. (The mechanism of feed probiotic action – in Polish). Trzoda Chlewna 1998, 1, 65–67.
38. Lood R., Winer B.Y., Pelzek A.J., Diez-Martinez R., Thandar M., Euler C.W., Schuch R., Fischetti V.A.: Novel phage lysin capable of killing the multidrug-resistant gram-negative bacterium *Acinetobacter baumannii* in a mouse bacteremia model. Antimicrob Agents Chemother 2015, 59, 1983–1991.
39. Magalhães V.J., Susca F., Lima F.S., Branco A.F., Yoon I., Santos J.E.: Effect of feeding yeast culture on performance, health, and immunocompetence of dairy calves. J Dairy Sci. 2008, 91, 1497–509.
40. Modi S.R., Lee H.H., Spina C.S., Collins J.J.: Antibiotic treatment expands the resistance reservoir and ecological network of the phage metagenome. Nature 2013, 499, 219–222.
41. Nowak P., Kasprowicz-Potocka M., Zaworska A., Nowak W., Stefańska B., Sip A., Grajek W., Juzwa W., Taciak M., Barszcz M., Tuśnio A., Grajek K., Foksowicz-Flaczyk J., Frankiewicz A.: The effect of eubiotic feed additives on the performance of growing pigs and the activity of intestinal microflora. Arch Anim Nutr 2017, 71, 455–469.
42. Ouwehand A.C., Isolauri E., Kirjavainen P.V., Salminen S.J.: Probiotics: mechanisms and established effects. Int Diary J 1999, 9, 43–52.
43. Pempek J.A., Holder E., Proudfoot K.L., Masterton M., Habing G.: Short communication: investigation of antibiotic

alternatives to improve health and growth of veal calves. J Dairy Sci 2018, 101, 4473–4478.
44. Penadés J.R., Chen J., Quiles-Puchalt N., Carpena N., Novick R.P.: Bacteriophage-mediated spread of bacterial virulence genes. Curr Opin Microbiol 2015, 23, 171–178.
45. Perdigón G., Alvarez S., Medici M.: Systemic and local augmentation of the immune response in mice by feeding with milk fermented with *Lactobacillus acidophilus* and/or *Lactobacillus casei*. In: *Dynamic Nutrition Research Volume 1: Foods, Nutrition & Immunity Effects of Dairy and Fermented Milk Products. 2nd Bio-Inova/EIBET Workshop, Paris, December 1991*, edited by M. Paubert-Braquet, C.H. Dupont, R. Paoletti, S. Karger, Basel, 1992, pp. 66–76.
46. Quigley J.D., Drewry J.J., Murray L.M., Ivey S.J.: Body weight gain, feed efficiency, and fecal scores of dairy calves in response to galactosyl-lactose or antibiotics in milk replacers. J Dairy Sci 1997, 80, 1751–1754.
47. Quirós P., Colomer-Lluch M., Martínez-Castillo A., Miró E., Argente M., Jofre J., Navarro F., Munies M.: Antibiotic resistance genes in the bacteriophage DNA fraction of human fecal samples. Antimicrob Agents Chemother 2014, 58, 606–609.
48. Ranaut N., Sharma F., Ravikant K., Ganguly B.: Comparative efficacy of herbal anti diarrheal products for treatment of diarrhea in calves. Pharm Biol Eval 2018, 5, 10–13.
49. Rial D.R.: The role of probiotic cultures in the control of gastrointestinal health. J Nutr 2000, 130, 396–420.
50. Roach D.R., Donovan D.M.: Antimicrobial bacteriophage-derived proteins and therapeutic applications. Bacteriophage 2015, 5, e1062590.
51. Rozema E.A., Stephens T.P., Bach S.J., Okine E.K., Johnson R.P., Stanford K., McAllister T.A.: Oral and rectal administration of bacteriophages for control of *Escherichia coli* O157:H7 in feedlot cattle. J Food Prot 2009, 72, 241–250.
52. Salminen S.J., Ouwehand A.C., Isolauri E.: Clinical applications of probiotic bacteria. Int Diary J 1998, 8, 563–572.
53. Schmelcher M., Shen Y., Nelson D.C., Eugster M.R., Eichenseher F., Hanke D.C., Loessner M.J., Dong S., Pritchard D.G., Lee J.C., Becker S.C., Foster-Frey J., Donovan D.M.: Evolutionarily distinct bacteriophage endolysins featuring conserved peptidoglycan cleavage sites protect mice from MRSA infection. J Antimicrob Chemother 2015, 70, 1453–1465.
54. Soothill J.S.: Treatment of experimental infections of mice with bacteriophages. J Med Microbiol 1992, 37, 258–261.
55. Trafalska E., Grzybowska K.: Probiotyki – alternatywa dla antybiotyków? (Probiotics – an alternative to antibiotics? – in Polish). Wiad lek 2004, 57, 9–10.
56. Truszczyński M., Pejsak Z.: Antybiotykooporność bakterii zoonotycznych występujących u zwierząt i w żywności. (Antibiotic resistance in zoonotic bacteria present in animals and foodstuffs – in Polish). Życie Wet 2008, 85, 891–894.
57. Turutogly H., Ozturk D.: Antibiotic resistance of *Staphylococcus aureus* and coagulase-negative staphylococci isolated from bovine mastitis. Bull Vet Inst Pulawy 2006, 50, 41–46.
58. Umpierrez A., Ines B., Oliver M., Acquistapace S., Etcheverria A., Lia Padola N., Vignoli R., Zunino P.: Zoonotic potential and antibiotic resistance of *Escherichia coli* in neonatal calves in Uruguay. Microbes Environ 2017, 32, 275–282.
59. United States Department of Agriculture. Dairy Heifer Raiser, 2011. USDA–APHIS–VS, CEAH, National Animal Health Monitoring System, Fort Collins, CO 2012.
60. United States Department of Agriculture. Heifer Calf Health and Management Practices on U.S. Dairy Operations, 2007. USDA–APHIS–VS, CEAH, National Animal Health Monitoring System, Fort Collins, CO 2010.
61. Uyeno Y., Shigemori S., Shimosato T.: Effect of probiotics/prebiotics on cattle health and productivity. Microbes Environ 2015, 30, 126–132.
62. Verma V., Harjai K., Chhibber S.: Characterization of a T7-like lytic bacteriophage of *Klebsiella pneumoniae* B5055: a potential therapeutic agent. Curr Microbiol 2009, 59, 274–281.
63. Wahida A., Ritter K., Horz H.P.: The Janus-face of bacteriophages across human body habitats. PLoS Pathog 2016, 12, 6.
64. Wang J., Hu B., Xu M., Yan Q., Liu S., Zhu X., Sun Z., Reed E., Ding L., Gong J., Li Q.Q., Hu J.: Use of bacteriophage in the treatment of experimental animal bacteremia from imipenem-resistant *Pseudomonas aeruginosa*. Int J Mol Med 2006, 17, 309–317.
65. Wang J., Hu B., Xu M., Yan Q., Liu S., Zhu X., Sun Z., Tao D., Ding L., Reed E., Gong J., Li Q.Q., Hu J.: Therapeutic effectiveness of bacteriophages in the rescue of mice with extended spectrum beta-lactamase-producing *Escherichia coli* bacteremia. Int J Mol Med 2006, 17, 347–355.
66. Wang Q., Euler C.W., Delaune A., Fischetti V.A.: Using a novel lysin to help control *Clostridium difficile* infections. Antimicrob Agents Chemother 2015, 59, 7447–7457.
67. Watanabe R., Matsumoto T., Sano G., Ishii Y., Tateda K., Sumiyama Y., Uchiyama J., Sakurai S., Matsuzaki S., Imai S., Yamaguchi K.: Efficacy of bacteriophage therapy against gut-derived sepsis caused by *Pseudomonas aeruginosa* in mice. Antimicrob Agents Chemother 2007, 51, 446–452.
68. Windisch W., Schedle K., Plitzner C., Kroismayr A.: Use of phytogenic products as feed additives for swine and poultry. J Anim Sci 2008, 86, 140–148.
69. Witzenrath M., Schmeck B., Doehn J.M., Tschernig T., Zahlten J., Loeffler J.M., Zemlin M., Müller H., Gutbier B., Schütte H., Hippenstiel S., Fischetti V.A., Suttorp N., Rosseau S.: Systemic use of the endolysin Cpl-1 rescues mice with fatal pneumococcal pneumonia. Crit Care Med 2009, 37, 642–649.