Chapter 6

How to Control *Campylobacter* in Poultry Farms?: An Overview of the Main Strategies

Michel Federighi

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/64846

Abstract

It is now recognized that *Campylobacter* is one of the main bacterial hazard involved in foodborne diseases around the world leading to an increasing number of gastrointestinal campylobacteriosis in humans. Also, it is known that this disease has a very high-social cost. According to researchers of Emerging Pathogens Institute (EPI) (University of Florida, the United States), the combination poultry/*Campylobacter* is the greatest cause of human campylobacteriosis. It is well known all around the world that intestinal carriage of *Campylobacter* is very large and frequent; it can be reached 100% of animal infected. Reducing this biological hazard can be exercised at different stage levels in the food chain. Intervention at the farm level by reducing colonization of the birds should be taken into account in the overall control strategy. This chapter gives an up-to-date overview of suggested on-farm control measures to reduce the prevalence and colonization of *Campylobacter* in poultry.

Keywords: *Campylobacter*, poultry, breeding, control strategies

1. Introduction

These days, the majority of human zoonotic microbial infections have a food origin (**Table 1**). Contamination of the food matrix can occur at all stages of the food production chain. In the search for causes of contamination, any stage of the production chain must not be neglected. This fact requires a global approach of problems and a good knowledge of the characteristics of the microorganisms involved. For the latter, the precise knowledge of their privileged reservoir and their potential ability to colonize other reservoirs will identify or clarify some contamination scenarios. Thus, it is known that the psychrotrophic nature of *Listeria monocytogenes* and their

affinity for inert surfaces and biofilms found in food industries are promoting the contamination of the food matrix during the industrial stages of product processing [1].

Disease	Number of confirmed (a) human cases	Hospitalized cases	Reported deaths	Case-fatality (%)
Campylobacteriosis	236,851	18,303	25	0.01
Salmonellosis	88,715	9830	65	0.15
Yersiniosis	6625	442	5	0.13
VTEC infections	5955	930	7	0.20
Listeriosis	2161	812	210	15.0
Echinococcosis	801	122	1	0.51
Q-fever	777	NA	1	0.26
Brucellosis	347	142	0	0.00
Tularemia	480	92	0	0.00
Trichinellosis	319	150	2	0.84
West Nile fever (a)	77	48	7	13.7
Rabies	3	NA	2	100.0

(a) Exception made for West Nile fever where the total number of cases was included; NA: not applicable.

Table 1. Reported hospitalization, deaths, and case-fatality rates due to zoonoses in confirmed human cases in the EU, 2014.

Contamination of food appears as a necessary step to trigger disease in humans. In some cases, and for certain microorganisms, this phase must necessarily be followed by another phase involving a multiplication of microorganisms in food, concomitantly, or not, with a toxin synthesis. This second phase will allow microorganisms to reach sufficient numbers (minimum infectious dose) to cause disease in consumers. Thus, some microbial hazards should multiply in food (such as *Salmonella* or *L. monocytogenes*) and others not (*Campylobacter*, VTEC, for example).

Researchers from the Emerging Pathogens Institute (EPI) of the University of Florida in the United States have recently focused on infectious diseases of food origin. They estimated that 31 foodborne pathogens are responsible for 9.4 million cases of human infections each year in the United States, leading to 55,961 hospitalizations and 1351 deaths (http://www.epi.ufl.edu/?q=RankingTheRisks). Among all of these cases, 59% cases are associated with viruses, 39% cases with bacteria, and 2% cases by parasites. Among viruses, norovirus is involved in 58% of cases and for bacteria, *Campylobacter*, *Salmonella*, and *Clostridium perfringens* occupy the first three places of the ranking. The first two of this classification are confirmed in Europe where campylobacteriosis exceeds salmonellosis since 2008.

http://dx.doi.org/10.5772/64846

In fact, *Campylobacter* is considered the most abundant zoonotic agent in the European Union. Indeed, 190,566 cases of *Campylobacter* infections were reported in 2008, increasing annually to reach 236,851 in 2014 (**Table 1**). Salmonellosis are still in second place in this epidemiological study, with 88,175 cases in 2014 (against 131,468 cases reported in 2008), the *Listeria* bacteria are responsible for 2161 cases of infection in 2014 (against 1381 in 2008) with a high mortality rate (15%), especially among vulnerable people [2]. To illustrate the importance of *Campylobacter* infections in France, it is interesting to recall that the report of the "Institut de Veille Sanitaire" (InVS) in 2004 estimated the number of confirmed cases of campylobacteriosis in France to 21,652 cases, of which 17,322 cases were from food origin [3]. According to this report, 3516 cases had required hospitalization and 18 cases would have conducted to death. *Campylobacter* infections not only indirectly cause a high processing cost but also a high number of days of work stopping. For example, the treatment of campylobacteriosis in the UK is 465 € while it is 77 € in the Netherlands [4]. In Europe, the annual cost of campylobacteriosis treatment is of the order of 2.4 billion euros [5].

In Africa, the situation is most worrying. It is known that the first *Campylobacter* infection occurs early in life, by food or nonfood way. Indeed, children under 5 years are the most exposed at the campylobacteriosis [6]. So, Goualie et al. [7] reported an estimated incidence of campylobacteriosis between 40,000 and 60,000 for 100,000 children in developing countries of this continent. These numbers are increasing in most African countries. So, the means of control and prevention are more than necessary even though it has been shown that repeated infections in some children gave them a protection in front of the next infection [8].

Transmission by direct contact with reservoirs like pets, human being, or contaminated bathing water, although rare, should not be neglected. It can cause disease, especially for high-risk professions, namely: farmers, veterinarians, and slaughterhouse workers [9]. Notwithstanding, in most cases, transmission to humans is done indirectly by ingestion of water or food contaminated by certain species of *Campylobacter* called thermophilic (named *C. jejuni* ssp. *jejuni*, *C. coli*, *C. lari*, *C. upsaliensis*), naturally present in many farm animals (poultry, cattle, swine, etc.) [10]. In fact, researchers from the Emerging Pathogens Institute (EPI), mentioned above, were then interested in food vehicles of microbial hazards. Of the 14 most frequent pathogens involved in infectious disease of food origin and the 12 most consumed foods in the United States (or the 168 food-pathogen combinations studied), *Campylobacter*-chicken combination is the one that is causing the greatest number of cases followed by *Toxoplasma*-pork combination, *Listeria*-ready to eat meats combination, *Salmonella*-poultry, and *Listeria*-dairy products combinations [11]. In the United States, the campylobacteriosis is mainly sporadic cases of which it turns out that the contaminated chicken consumption is the cause of most cases. It seems that in Europe we can also highlight the *Campylobacter*-chicken combination since a study conducted in Belgium during the dioxin crisis, has shown that the number of campylobacteriosis decreased by 40% during the withdrawal period of the sale of poultry [12]. **Table 2** highlights the importance of chicken as *Campylobacter* vector compared with pork and beef meat (**Table 2**).

Meat	2006	2005	2004	2003	2002
Poultry	34.6%	30.5%	37.8%	35%	30.2%
Pork	0.7%	0.3%	1.6%	1.2%	1.4%
Beef	0.7%	0.9%	0.6%	0.3%	0.3%

Table 2. Comparison of different sources of *Campylobacter* contamination of poultry, pork and beef meat in Europe (years 2002–2006).

Some gestures made during the preparation of foods in the kitchen is often the cause of contamination transfers, including the use, for cutting the roasted poultry, of the board on which was cut or eviscerated raw poultry. Furthermore, studies have shown that the transfer of *Campylobacter* from chicken skin to kitchen work surfaces was possible at significant rate (from 0.05 to 36%), as well as to the hands of users (2.9–3.8%) [13]. It has, moreover, been shown that *Campylobacter* was able to survive for several hours on surfaces and stainless steel utensils, and sponges used for cleaning surfaces could also be sources of contamination [14].

Campylobacter jejuni is responsible for over 85% of campylobacteriosis. When it occurs, campylobacteriosis occurs typically after incubation for 24–72 h, by intestinal manifestations during a week. The most frequently described manifestation is acute gastroenteritis characterized by inflammation, severe abdominal pains in the periumbilical region, mucous diarrhea that can be bloody, accompanied sometimes by fever. It should be noted that the clinical manifestation is often less severe in developing countries where campylobacteriosis is manifested only by a significant watery diarrhea, this could be related to immune protection settling in individuals with frequent contact with *Campylobacter* [8].

This disease can be serious for certain populations or during postinfection complications, like Guillain-Barré syndrome or Miller-Fisher syndrome [15]. It seems that some serogroups of *C. jejuni*, as the serogroup O19 Penner, are particularly involved in this type of complications. Having a minimum growth temperature of 30°C, being intolerant to ambient oxygen and also being sensitive to technological stress (such as cold, heat, acidification, and drying), *Campylobacter jejuni* was always considered as a delicate and fragile organism [16]. Despite these nutritional requirements and the sensitivity to environmental stresses which prevent to grow and multiply outside the host or in food, *C. jejuni* is still able to survive and persist throughout the food production chain to cause campylobacteriosis constituting, in fact, a real paradox [17].

Although *Campylobacter* is found in the intestinal tracts of most red meat animals (cattle, pigs, and small ruminants) and pets (cats and dogs), the avian reservoir remains predominant due to the high carrier rate in animals and to the bacterial load per gram of feces, up to 10^7 CFU/g [13, 18]. Due to the large intestinal asymptomatic carriage in production animals, manure, slurry, soil, and water can also be reservoirs of *Campylobacter*. In fact, a study conducted in Italy showed that about 30% of water samples from rivers were contaminated with *Campylobacter jejuni* [19].

The colonization of the intestine of broilers by *Campylobacter* during rearing is responsible for contamination of carcasses after processing [20–22]. Worldwide, the average prevalence of

http://dx.doi.org/10.5772/64846

Campylobacter on poultry carcasses is about 60–80% [2, 23]. The carcass contamination occurs during the slaughtering process, even if certain operations are contaminating more than others. Thus, it is recognized that the contamination occurs more favorably during defeathering and evisceration, with feces leaking from the cloaca and the rupture of caeca, causing massive contamination by *Campylobacter* [24–26]. In addition, various transfers of contamination (or cross contamination) can intervene. Among these, there is a carcass to carcass contamination by contact and contamination transfer via vectors such as equipment and personnel, mostly [24].

All of these works clearly show that intestinal carriage of *Campylobater* by poultry is a key element of the transmission of this hazard to humans. Although beyond the breeding, control measures of this danger exist (good hygienic practices transformations, physical, and chemical treatments sanitizers), their effectiveness will be strengthened if the number of *Campylobacter* present at this stage is as low as possible. Therefore, the reduction or eradication of intestinal carriage in chickens is a strategic element of major importance for risk control *Campylobacter* in this sector especially as the contamination of poultry carcasses is proportional to the amount of *Campylobacter* present in caeca before slaughter [22]. In fact, a recent study of EFSA based on a quantitative microbiological assessment of risks (QMRA) evaluated the performance of implementation of interventions in primary production on reducing the risk of campylobacteriosis cases for the chicken consumers [27]. Following this study, the potential reduction of campylobacteriosis cases can be predicted after the application of breeding measures and is presented in **Table 3**.

Interventions in primary production		**Reduction of campylobacteriosis cases**
Improved hygiene/biosecurity		16%
Systematic use of screen fly in broiler houses (Denmark)		60%
Discontinued thinning		1.8–25%
Reduction of slaughter age	42 days	0–5%
	35 days	0.6–18%
	28 days	21–43%
Reduction colonization in cecal contents	1 log	48–83%
	2 logs	76–98%
	3 logs	90–100%
	6 logs	100%

Table 3. Effect of interventions in primary production on the reduction of human campylobacteriosis cases.

The study found that the most effective measures are those aimed at reducing the number of *Campylobacter* in caeca of chickens. However, other simpler measures also have a real impact, we now do an overview of all these measures.

2. Control *Campylobacter* in chicken farms

The few quantitative risk assessment studies available on the *Campylobacter*-chicken combination, conducted in different countries, often focus on the effectiveness of physical and chemical means of eliminating this zoonotic agent during the slaughter and the transformation process [28]. These methods have often proven their efficacy but are often oversized and sometimes poorly accepted by the consumer [10]. Preventive approaches such good hygiene practices and biosecurity now find some interest and may be a strategy to prevent the colonization of animals by *Campylobacter* and participate in the control of this zoonotic agent in the production of poultry meat. This interest is reinforced by the development of indirect measures, complementary of best practices, to reduce the intestinal number of *Campylobacter* in poultry. For some authors, these interventions would significantly reduce campylobacteriosis [29]. Thus, a Belgian study showed that the incidence of human campylobacteriosis in the country could be reduced by 32, 53, and 77% if the prevalence of broiler batches colonized by *Campylobacter* was reduced by 25, 50, or 75%, respectively [30].

2.1. Good hygienic practices and biosecurity

Thus, in addition to reducing the risk *Campylobacter* obtained at later stages of the human food chain, this reduction can also be achieved by the establishment of biosecurity measures at the breeding stage. These interventions are designed to protect a population of animals from the introduction of infectious agents transmissible like *Campylobacter*. In poultry, the biosecurity program includes all measures that must or may be taken to prevent the entry of this agent and changing the health status of the chicken population. These measures, collectively known as biosecurity, cover the hygienic practices during the rearing period. These include washing hands before entering in a poultry house, the use of different boots to enter each house, the cleaning, and disinfection of shoes before entering the room, a high level of hygienic water quality beverage.

Other measures such as cleaning and effective disinfection of poultry house between two batches of animals, as reducing the number of visits, as strict control of entry into the breeding of rodents, wild birds, and flying insects. Thus, studies in Denmark have shown that the use of mosquito nets preventing the entry of flying insects in the broiler house, potential vectors of *Campylobacter*, significantly reduced the contamination of poultry by *Campylobacter* during the seasonal peak (**Figure 1**) [31].

The application of all these measures greatly reduces the risk of *Campylobacter* infections. So, Gibbens et al. estimated that this application would lower the prevalence of *Campylobacter* in batches of chickens from 80 to less than 40% [32]. The respect of a good personal and clothing hygiene for staff and good measures of biosecurity, including the control of rodents and insects in two Dutch farms, reduced prevalence of *Campylobacter* in batches of chickens from two different farms of 34% in the first farm, and of 20% in the second [33].

http://dx.doi.org/10.5772/64846

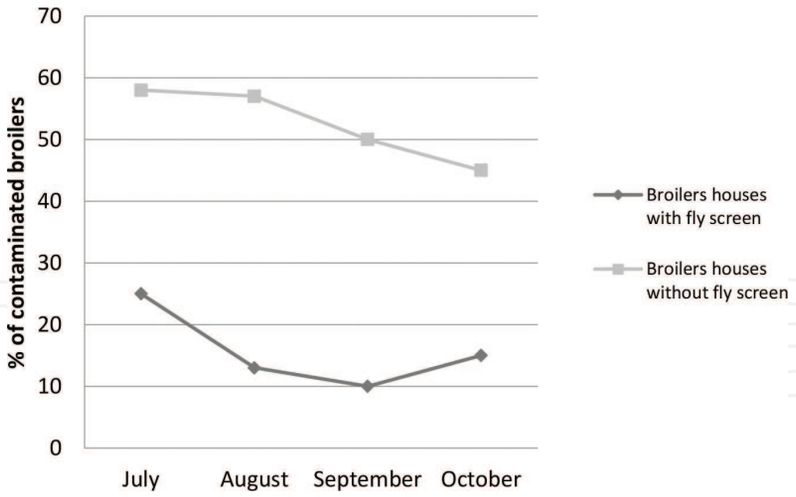

Figure 1. Effect of the use of fly screen in broilers houses on the percentage of contaminated broilers.

2.2. Treatment of drinking water

Another important factor is the quality of drinking water. Several studies have shown that poor quality water (untreated water from wells) may increase the transmission of *Campylobacter* in animals [34, 35]. The microbiological quality of drinking water should be monitored by the analysis and can be improved on the farm by techniques such as filtration, chlorination, ozonation, UV rays. For some authors, the impact of these interventions on *Campylobacter* infection is uncertain, but they point out that the absence of interventions may be worse [32, 36]. Studies by Byrd et al. showed that adding 0.44% (vol/vol) of lactic acid in drinking water prior to slaughter, has reduced the level of contamination of carcasses with *Campylobacter* [37]. Hilmarsson et al. showed that the addition of glycerol monocaprate (monocaprin) the last 3 days before slaughter, has reduced the number of *C. jejuni* in feces samples of chickens naturally or artificially infected [38].

2.3. Use of antimicrobial from vegetal origin

In addition to their application in drinking water, organic acids can also be used as additives in foods to reduce the prevalence of *Campylobacter* in poultry. Thus, 0.7% caprylic acid reduced colonization when used preventively on old chickens of 10 days and achieves a significant reduction of *C. jejuni* in broiler feces up to three to four decimal reductions [39]. In contrast, Van Deun et al. [40] observed that butyrate did not reduce colonization of caeca by *Campylobacter* in broiler chickens, but the addition of fatty acids short chain at a concentration of 1% reduces the risk of colonization of farms [41]. However, Hermans et al. [42] did not find any effect of these medium chain fatty acids (caproic, caprylic, and capric) on the number of *Campylobacter* in the caeca of broilers 28 days fed therewith 3 days before slaughter. Moreover, they observed that injection of a highly concentrated solution of sodium caprate directly into the caeca did not prevent colonization and did not reduce the caeca contents by *Campylobacter*. Thereafter, these authors showed that this ineffectiveness was explained by the presence of the intestinal mucus whom protecting *C. jejuni* in the caeca vis-a-vis of the bactericidal effect

of organic acids observed *in vitro*. Conversely, another research group found a significant reduction (several logs) of *Campylobacter* in caeca of chickens when caprylic acid was given 3 days before slaughter [43]. In addition, another study showed that adding monocaprin to chicken feed the last 3 days before slaughter, resulted in a significant reduction of *C. jejuni* on feces samples of animals artificially or naturally infected, compared to controls [38]. These results, apparently contradictory, demonstrate the need to continue the investigations necessary to establish with greater certainty the effectiveness or lack of effectiveness of this strategy in the control of *Campylobacter* in chicken farms

2.4. Vaccination

The principle of vaccination of chickens against *Campylobacter* is to administer a product capable of inducing immunity directed specifically against this pathogen and confer immune memory enabling rapid activation (much shorter of lag period) of defenses in case of contamination. Vaccination could complement the use of biosecurity measures and other intervention strategies to reduce the level of contamination of poultry by *Campylobacter*. The development, production, and application of available vaccines would be beneficial to all parties involved and help enhance food safety and improve public health. Several studies on vaccination to reduce the sensitivity of broilers with *Campylobacter* colonization were performed. Khoury and Meinersmann [44] vaccinated chickens using a hybrid protein consisting of a portion of the FlaA *Campylobacter jejuni* flagellin (flagellar subunit) and the B subunit of heat-labile toxin (LT-B) of *Escherichia coli*. This results in a significant reduction of colonization of chickens by *Campylobacter*, and the production of specific antibodies against FlaA. Comparing with unvaccinated chicks, Rice et al. [45] have demonstrated a reduction of *Campylobacter* in chicks vaccinated orally with a combination of dead *Campylobacter jejuni* cells coupled to the heat labile toxin of *E. coli*.

More recent studies involving a larger number of animals were used to test the use of recombinant vaccines. Thus, 840 SPF chicks were used to evaluate the effectiveness of the vaccine derived from *Salmonella enterica* Typhimurium ΔaroA attenuated and expressing the immunogenic protein CJAA *C. jejuni*. Chicks who received the vaccine orally at the age of 1 day, then 2 weeks later, showed a reduction in fecal contamination by *C. jejuni* of 1.4log CFU/g, compared with unvaccinated chicks [46]. Layton et al. [47] used recombinant vaccines attenuated from Salmonella expressing three peptide epitopes of protein of *Campylobacter* (Omp18/CJAD protein, CJAA, and Cj0420 (ACE393)). These three vaccines were administered orally to chicks on the day 1, then 21 days later. The vaccinated chicks were inoculated with *C. jejuni*. Eleven days after the inoculation, an increase in IgG and IgA antibodies specifically against *C. jejuni* was observed and also a reduction in the number of *C. jejuni* in the ileum. Vaccination was most effective when the vectored vaccine expressing the epitope of the Omp18/CJAD protein was administered to chicks, with a considerable reduction of *C. jejuni* in the chicken intestine (4.8 decimal reductions of *C. jejuni* in the ileum) compared with unvaccinated controls and those vaccinated only with the vector (*Salmonella* 13A) or negative control (**Figure 2**).

These studies are promising and probably mean that a possible vaccination strategy for *Campylobacter* reduction is possible. They still face a lack of information on the immune system

http://dx.doi.org/10.5772/64846

of the chick that such hinders the development of an attenuated vaccine expressing the linear peptide epitope *Campylobacter* (Omp18/CJA). In addition, advances in functional genomics *Campylobacter* suggest that other proteins of this agent could be excellent candidates for testing for future vaccines.

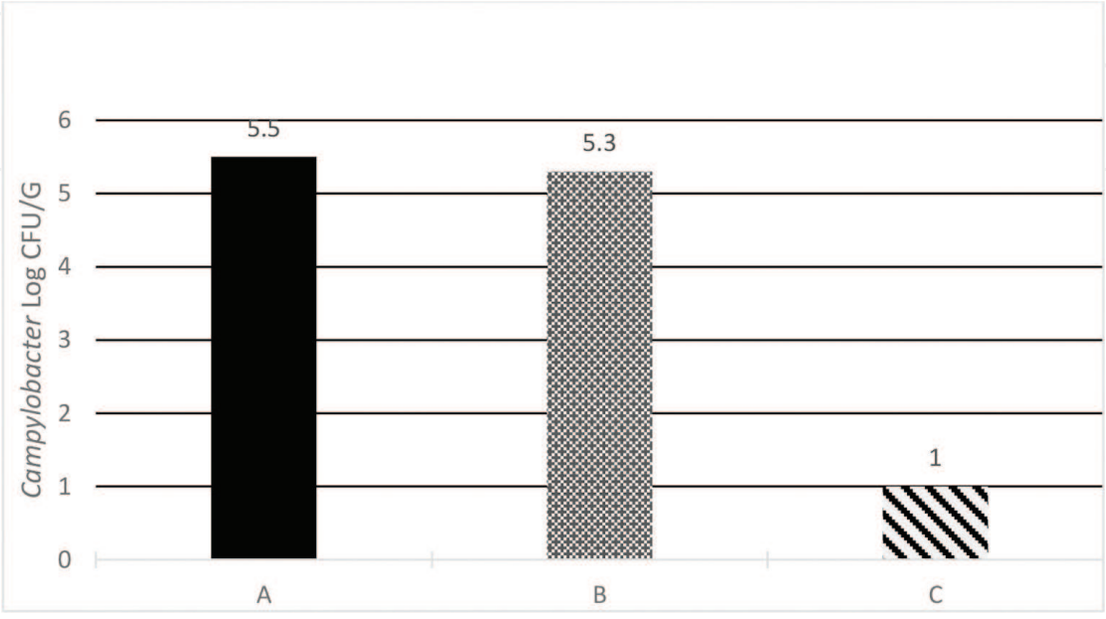

Figure 2. Enumeration of *Campylobacter* (log CFU/cecal content) on 1 day orally vaccinated chicken. Group A: salted water vaccinated; group B: *Salmonella* 13A vaccinated, and group C: *Campylobacter* CJ0113 vaccinated.

2.5. Use of phages

The lytic activity of bacteriophages can be used as a strategy to reduce the colonization of chickens with *Campylobacter*. Phages usually have a very narrow spectrum of activity, and they do not interact with other bacterial species in the intestinal flora. Phages bind and penetrate into bacterial cells by protein receptors and multiply within the cytoplasm until the death of the bacteria. At this time, the lysis of the bacteria permits the release of new bacteriophages.

Loc Carrillo et al. [48] and Wagenaar et al. [49] have shown three decimal reductions of *Campylobacter* in caeca of chickens that received the bacteriophage, compared with negative control. However, this reduction is not stable, they observed a reduction of only 1log/g 5 days later. Similarly, El-Shibiny et al. [50] observed an immediate reduction of $2\log_{10}$(CFU/g) of *Campylobacter* in caeca 2 days after ingestion of phages. Then, the number of *Campylobacter* in caeca returns to the original level a few days later, reversing the improvement achieved. These results show that this strategy is more a short-term therapeutic strategy than a preventive long term one. It could be very interesting if, for example, the treatment is taken only 2–3 days before slaughter: resistance to bacteriophages then not have time to reverse the reduction of *Campylobacter* obtained by the treatment. In this matter, other studies have also shown that administration of phages in feed is more effective than oral gavage [37]. In these conditions of administration, use of phages few days before slaughter appears to be an excellent strategy

for the reduction of *Campylobacter* in poultry, but the diversity of *Campylobacter* protein receptors requires a large diversity of phages, which also increases the complexity of this strategy.

2.6. Use of prebiotics and probiotics

Probiotics are defined as "live microorganisms which when administered in adequate amounts confer a benefit to human health." Prebiotics are generally oligosaccharides (fructo-oligosaccharide (FOS), galacto-oligosaccharides (GOS)) or polysaccharides such as inulin. These escape digestion in the small intestine and have a beneficial effect on the health of their host by stimulating the growth and/or activity of bacteria of the genera *Lactobacillus* and *Bifidobacterium*, naturally present in the colon or administered as probiotics.

The use of prebiotics and probiotics is a strategy that has been studied by several research teams in order to reduce the colonization of chickens by *Campylobacter jejuni*.

In 1997, Morishita et al. [51] used on 1-day-old chicks, a probiotic mixture containing *Lactobacillus acidophilus* and *Enterococcus faecium*. The chicks were randomly divided into two groups, one group was treated with the probiotic cocktail during the first 3 days of culture while the second batch received distilled water instead of the probiotic cocktail. Six hours after the first oral administration of probiotics, the number of *C. jejuni* in feces of the chicks was determined and was performed until the slaughter. The results showed a 70% reduction in the concentration of *C. jejuni* in chickens on day 3 and a 27% reduction for broilers at slaughter, compared to the control group. For Schoeni and Wong, administration of a mixture of different bacteria (*Citrobacter diversus*, *Klebsiella pneumonia*, and *E. coli*) appears to be effective in preventing or reducing the colonization of chickens with *Campylobacter* [52]. This protection has been strengthened by mannose which was given as a prebiotic. In 2000, Chang and Chen [53] tested on *C. jejuni*, in an *in vitro* model of the digestive tract of chicken, the effect of a mixed culture of *Lactobacillus acidophilus*, *L. fermentum*, *L. crispatus*, and *L. brevis* in a feed wherein mannose has been added, showing a inhibitory effect. Similarly, Baurhoo et al. [54] observed a significant reduction of *C. jejuni* in naturally contaminated caeca of broiler, who received a diet containing mannanoligosaccharide as a prebiotic.

Again, this is very promising works, that requires further study in order to decide definitively on their use. They also have the merit of bringing forward an interesting and ongoing concept named "microbial solution for microbial problems."

2.7. Genetic selection of chicken

Selective breeding of resistant lines of chickens to *Campylobacter* colonization is a particularly modern intervention strategy to reduce *Campylobacter* problem in the poultry industry.

In 2005, Boyd et al. [55] have shown that the selection of chicken lines genetically resistant to *Campylobacter* germ, significantly reduces this risk in poultry. In their study, Boyd et al. [55] have inoculated 1-day-old chicks of different inbred lines with 10^7–10^8 CFU of *C. jejuni* or *C. jejuni* 81–176 ^{14}N and measured bacterial colonization levels of chickens over a period of 2–3

weeks. They have always been a difference of a factor 10–100 from four inbred lines in the number of *C. jejuni* present in chicken caeca between the four inbred chicken lines. The biggest difference was for the N line, which presented relatively high levels of *Campylobacter*, and the line 61, which had a relatively small number of bacteria. Among the four lines studied, the major histocompatibility complex does not appear to be a major factor in determining the resistance. The difference in the number of bacteria in fecal samples was observed after 24 h after inoculation and was still present at the end of the experiment. This work revealed that the difference in the number of bacteria was inherited in a consistent manner with the resistance (low number of bacteria), controlled by a single autosomal dominant locus. These data suggest that it may be possible to identify the responsible genes. Indeed, the recent knowledge of the whole sequence of the chicken genome has identified the genes involved in susceptibility to *Campylobacter* colonization [56]. These observations led to the suggestion that selective breeding could be used to select chickens resistant to *Campylobacter* colonization.

2.8. Use of bacteriocins

The use of antimicrobial peptides could be an interesting biological intervention strategy to reduce colonization of poultry by *Campylobacter* [57–61]. These studies highlight the ability of bacteriocins produced by lactic acid bacteria, such as *Lactobacillus salivarius* NRRL B-30514, *Enterococcus faecium* E50-52 E760, *Lactobacillus salivarius*, and *L. salivarius* SMXD51 1077 (NRRL B-50053)) to inhibit the growth of *Campylobacter jejuni*. In 2005, Stern et al. [62] studied the effect of the bacteriocin "SRCAM 602" produced by *P. polymixa* NRRL B-30509 on cecal colonization by *Campylobacter* of chicks artificially inoculated with 10^8 CFU of *C. jejuni* from day 1. These animals, colonized by *Campylobacter*, received from day +7 to day +10 a feed containing the bacteriocin purified (250 mg/kg). For the chicks that received during 3 days, this diet the number of *Campylobacter* in caeca was very low and undetectable (<$2\log_{10}$ CFU/g), while the control animals showed high cecal colonization by *Campylobacter* (10^6–10^8 log CFU/g) [62]. In 2008, Line et al. [57] have found similar results following administration of the bacteriocin "Enterococcine E-760" in broiler naturally infected with *Campylobacter*. Moreover, Svetoch et al. [63] administered 10.8 mg/chicken (oral gavage) of bacteriocin "E 50-52" produced by *E. faecium* NRRL B-30746 3 days before slaughter. The results showed a significant reduction of *Campylobacter* in the gut, greater than 10^5 CFU/g of feces.

Stern et al. [64] have studied the effect of the bacteriocin OR 7, produced by *L. salivarius* NRRL B-30514, in encapsulated form in a concentration of 250 mg/kg on eight groups of contaminated chicks by *C. jejuni*. In three of eight groups of chickens, there was no *Campylobacter* colonization and, in the other five groups, the level of contamination remained very low (10–100 CFU/g) compared with controls. By cons, *L. salivarius* NRRL B-30514 and *Paenibacillus polymyxa* NRRL-B-30509 showed no effect on colonization of artificially infected chicks by *C. jejuni* [64]. Finally, Svetoch et al. [63] showed that treatment with the bacteriocin L-1077, produced by *L. salivarius* strain NRRL B-50053, chickens inoculated with *C. jejuni* and *Salmonella* Enteritidis provides more than four decimal reductions in the number of bacteria per gram of cecal contents, compared with controls. Moreover, the presence of these bacteria in the liver and spleen of the animals is very greatly reduced.

3. Conclusion

Campylobacter is today a leading cause of foodborne diseases, all around the world. It is also a paradox for microbiologists, who see a contradiction between the apparent physiological fragility, its small genome and its obvious ability to survive outside its main habitat (digestive tract of birds) and to reach its main target (i.e., the consumer). Moreover, this impression is reinforced by the fact that the organism does not grow in foods and that his number would tend to decrease during processing operations, rather than increase. In fact, intestinal carriage of *Campylobacter* by animals becomes a key element of the contamination of the consumer and a series of strategies have been developed to reduce intestinal carriage in the past 15 years. Today, despite all the efforts and progress made, there is still no miracle solution but a set of interventions strategies, each with their advantages and disadvantages. The use of bacteriocins and bacteriophages is very promising because their implementation is simple: they can be easily administered with water or feed. However, their potential use requires further research work on their long-term effectiveness. In addition, the successful application of these methods as well as probiotics, prebiotics and even vaccination may be affected by genomic instability of *C. jejuni* [44] which may affect the effectiveness of these strategies in the long term. Finally, the reduction of *Campylobacter* contamination pressure of animals at the stage of livestock should be based on a strong base of Good Hygienic Practices and biosecurity, reinforced by targeted interventions selected on the criteria of efficiency, practicality, and cost in front of the type of poultry production.

Author details

Michel Federighi

Address all correspondence to: michel.federighi@oniris-nantes.fr

Oniris, Food Quality and Hygiene, Nantes, France

References

[1] Ribeiro da Silva M, Federighi M. Mint: seafood products as a source of listeriosis: myth or reality? Advanced Research in Food Science. 2005; 4, 73–78.

[2] EFSA. Mint: analysis of the baseline survey on the prevalence of *Campylobacter* in broiler batches and of *Campylobacter* and *Salmonella* on broiler carcasses in the EU, 2008, part A: *Campylobacter* and *Salmonella* prevalence estimates. EFSA Journal, 2010; 8 (3), 1503. doi:10.2903/j.efsa.2010.1503.

[3] INVS. Morbidity and mortality due to foodborne infections in France. Report of the French Institute of Public Health. 2004; pp. 185.

[4] Roberts JA, Cumberland P, Sockett PN, Wheeler J, Rodrigues LC, Sethi D, Roderick PJ. Mint: the study of infectious intestinal disease in England: socio-economic impact. Epidemiology and Infection. 2003; 130, 1–11.

[5] EFSA. Mint: scientific opinion on *Campylobacter* in broiler meat production: control options and performance objectives and/or targets at different stages of the food chain. EFSA Journal. 2011; 9 (4), 2105. doi:10.2903/j.efsa.2011.2105.

[6] Oberhelman R, Taylor D. *Campylobacter* infections in developing countries. In: Nachamkin I, Blaser M, editors. *Campylobacter*, 3rd ed. Washington: ASM Press; 2000. p. 139–153.

[7] Goualie GB, Karou G, S Bakayoko, KJ Coulibaly, Coulibaly KE, Niamke SL, Mr. Dosso Mint: Prevalence of Campylobacter in chickens sold in the markets of Abidjan: Pilot study in the municipality of Adjamé 2005 African Journal of Health and Animal Production. 2010; 8 (S), 31–34.

[8] Lastovica A, Skirrow M. Clinical significance of *Campylobacter* and related species other than *Campylobacter jejun*i and *C. coli*. In: Nachamkin I, Blaser M, editors. *Campylobacter*, 3rd ed. Washington: ASM Press; 2000. p. 89–120.

[9] Garénaux A Ritz-Bricaud M, Mr. Federighi Mint: Campylobacter and food safety: risk analysis, risk assessment and risk management. Bulletin of the Veterinary Academy of France, 2005; 158, 377-383.

[10] Federighi M, C Magras, Pilet MF. Campylobacter. In: Federighi M, editor. Food bacteriology, Compendium of food hygiene, Paris, Economica, 2005, p. 145-172.

[11] Scallan E, Hoekstra R, Angulo F, Tauxe R, Widdowson MA, Roy S, Jones J, Griffin P. Mint: foodborne illness acquired in the United States—major pathogens. Emerging and Infectious Disease. 2011; 17 (1), 7–15.

[12] Vellinga A, Loock VID. Mint: the dioxin crisis as experiment to determine poultry-related *Campylobacter enteritis*. Emerging and Infectious Disease. 2002; 8, 19–22.

[13] Salvat G, Chemaly M, Denis M, Robinault C, Huneau A, Le Bouquin S, Fravalo P. Mint: evolution des risques sanitaires: *Campylobacter* et Salmonelles. Sciences des Aliments. 2008, 28 (4–5), 285–292.

[14] Kusumaningrum HD, Riboldi G, Hazeleger WC, Beumer RR. Mint: survival of foodborne pathogens on stainless steel surfaces and cross-contamination to foods. International Journal of Food Microbiology. 2003; 85, 227–236. doi:10.1016/S0168-1605(02)00540-8; doi:10.1016/S0168-1605%2802%2900540-8#doilink.

[15] Zilbauer M, Dorrell N, Elmi A, Lindley KJ, Schüller S, Jones HE, Klein NJ, Núňez G, Wren BW, Bajaj-Elliott M. Mint: a major role for intestinal epithelial nucleotide oligomerization domain 1 (NOD1) in eliciting host bactericidal immune responses to

Campylobacter jejuni. Cellular Microbiology. 2007; 9, 2404–2416. DOI: 10.1111/j.1462-5822.2007.00969.x.

[16] Martinez-Rodriguez S, Mackey BM. Mint: factors affecting the pressure resistance of some *Campylobacter* species. Letters in Applied Microbiology. 2005; 41, 321–326. DOI: 10.1111/j.1472-765X.2005.01768.x.

[17] Messaoudi S, Federighi M. Campylobacter infections: epidemiology, diagnosis, control and prevention. In: Bertucci BA, editor. Campylobacter Infection: Epidemiology, Clinical Management and Prevention. New York: Novascience Publishers; 2015. p. 29–54.

[18] Newell D. Mint: the ecology of *Campylobacter jejuni* in avian and human hosts and in the environment. International Journal of Infectious Disease. 2002; 6, (Suppl. 3), S16–S21. doi:10.1016/S1201-9712(02)90179-7; doi:10.1016/S1201-9712%2802%2990179-7#doilink.

[19] Yan SS, Pendrak ML, Foley SL, Powers JH. Mint: *Campylobacter* infection and Guillain-Barre syndrome: public health concerns from a microbial food safety perspective. Clinical Applied Immunology Review. 2005; 5, 285–305. doi:10.1016/j.cair.2005.08.001; doi:10.1016/j.cair.2005.08.001#doilink.

[20] Herman L, Heyndrickx M, Grijspeerdt K, Vandekerchove D, Rollier I, De Zutter L. Mint: routes for *Campylobacter* contamination of poultry meat: epidemiological study from hatchery to slaughterhouse. Epidemiology and Infection. 2003; 131, 1169–1180. doi.org/10.1017/S0950268803001183; doi:10.1017/S0950268803001183#_blank.

[21] Rasschaert G, Houf K, Van Hende J, De Zutter L. Mint: *Campylobacter* contamination during poultry Slaughter in Belgium. Journal of Food Protection. 2006; 69, 27–33.

[22] Reich F, Atanassova V, Haunhorst E, Klein GN. Mint: the effects of Campylobacter numbers in caeca on the contamination of broiler carcasses with *Campylobacter*. International Journal of Food Microbiology. 2008; 127, 116–120. doi:10.1016/j.ijfoodmicro.2008.06.018; doi:10.1016/j.ijfoodmicro.2008.06.018#doilink.

[23] Suzuki H, Yamamoto S. Mint: *Campylobacter* contamination in retail poultry meats and by-products in Japan: a literature survey. Food Control. 2009; 20, 531–537. doi:10.1016/j.foodcont.2008.08.016; doi:10.1016/j.foodcont.2008.08.016#doilink.

[24] Allen VM, Weaver H, Ridley AM, Harris JA, Sharma M, Emery J, Sparks N, Lewis M, Edge S. Mint: sources and spread of thermophilic *Campylobacter* spp. during partial depopulation of broiler chicken flocks. Journal of Food Protection. 2008; 71, 264–270.

[25] Berrang ME, Buhr RJ, Cason JA, Dickens JA. Mint: broiler carcass contamination with *Campylobacter* from feces during defeathering. Journal of Food Protection. 2001; 64, 2063–2066.

[26] Boysen L, Rosenquist H. Mint: reduction of thermotolerant *Campylobacter* species on broiler carcasses following physical decontamination at slaughter. Journal of Food Protection. 2009; 72, 497–502.

[27] Romero-Barrios P, Hempen M, Messens W, Stella P, Hugas M. Mint: quantitative microbiological risk assessment (QMRA) of food-borne zoonoses at the European level. Food Control, 2013; 29(1), 341–349. doi:10.1016/j.foodcont.2012.05.043; doi:10.1016/j.foodcont.2012.05.043#doilink.

[28] Rosenquist H, Sommer HM, Nielsen NL, Christensen BB. Mint: the effect of slaughter operations on the contamination of chicken carcasses with thermotolerant *Campylobacter*. International Journal of Food Microbiology. 2006; 108, 226–232. doi:10.1016/j.ijfoodmicro.2005.12.007; doi:10.1016/j.ijfoodmicro.2005.12.007#doilink.

[29] Lin J. Mint: novel approaches for *Campylobacter* control in poultry. Foodborne Pathogens and Diseases. 2009; 6, 755–765. doi:10.1089/fpd.2008.0247.

[30] Messens W, Hartnett E, Gellynck X, Viaene J, Halet D, Herman L, Grijspeerdt K. Quantitative risk assessment of human campylobacteriosis through the consumption of chicken meat in Belgium. In: Proceedings of the XVIII European Symposium on the Quality of Poultry Meat and XII European Symposium on the Quality of Eggs and Egg Products, 2–5 September 2007; Prague. p. 167–168.

[31] Hald B, Sommer HM, Skovgard H. Mint: use of fly screens to reduce *Campylobacter* spp., introduction in broiler houses. Emerging and Infectious Diseases. 2007; 13, 1951–1953.

[32] Gibbens JC, Pascoe SJS, Evans SJ, Davies RH, Sayers AR. Mint: a trial of biosecurity as a means to control *Campylobacter* infection of broiler chickens. Preventive Veterinary Medicine. 2001; 48, 85–99. doi:10.1016/S0167-5877(00)00189-6; doi:10.1016/S0167-5877%2800%2900189-6#doilink.

[33] Van De Giessen AW, Tilburg JJ, Ritmeester WS, Van Der Plas J. Mint: reduction of *Campylobacte*r infections in broiler flocks by application of hygiene measures. Epidemiology Infection. 1998; 121, 57–66.

[34] Lyngstad TM, Jonsson ME, Hofshagen M, Heier BT. Mint: risk factors associated with the presence of *Campylobacter* species in Norwegian broiler flocks. Poultry Science. 2008, 87, 1987–1994. doi: 10.3382/ps.2008-00132.

[35] Sparks NHC. Mint: the role of the water supply system in the infection and control of *Campylobacter* in chicken. World Poultry Science Journal. 2009; 65, 459–474. doi.org/10.1017/S0043933909000324; doi:10.1017/S0043933909000324#_blank.

[36] Mohyla P, Bilgili SF, Oyarzabal OA, Warf CC, Kemp GK. Mint: application of acidified sodium chlorite in the drinking water to control *Salmonella serotype Typhimurium* and *Campylobacter jejuni* in commercial broilers. Journal of Applied Poultry Research. 2007; 16, 45–51. doi: 10.1093/japr/16.1.45.

[37] Byrd JA, Hargis BM, Caldwell DJ, Bailey RH, Herron KL, Mac Reynolds JL, Brewer RL, Anderson RC, Bischoff KM, Callaway TR, Kubena LF. Mint: effect of lactic acid

administration in the drinking water during preslaughter feed withdrawal on *Salmonella* and *Campylobacter* contamination of broilers. Poultry Science. 2001; 80, 278–283. doi: 10.1093/ps/80.3.278.

[38] Hilmarsson H, Thormar H, Thrainsson JH, Gunnarsson E, Dadadottir S. Mint: effect of glycerol monocaprate (monocaprin) on broiler chickens: an attempt at reducing intestinal *Campylobacter* infection. Poultry Science. 2006; 85, 588–592. doi: 10.1093/ps/85.4.588.

[39] De Los Santos FS, Donoghue AM, Venkitanarayanan K, Metcalf JH, Reyes-Herrera I, Dirain ML, Aguiar VF, Blore PJ, Donoghue DJ. Mint: the natural feed additive caprylic acid decreases *Campylobacter jejuni* colonization in market-aged broiler chickens. Poultry Science. 2009; 88, 61–64. doi: 10.3382/ps.2008-00228.

[40] Van Deun K, Haesebrouck F, Van Immerseel F, Ducatelle R, Pasmans F. Mint: short-chain fatty acids and l-lactate as feed additives to control *Campylobacter jejuni* infections in broilers. Avian Pathology. 2008; 37, 379–383. doi: 10.1080/03079450802216603.

[41] Gerwe TV, Bouma A, Klinkenberg D, Wagenaar JA, Jacobs-Reitsma WF, Stegeman A. Mint: medium chain fatty acid feed supplementation reduces the probability of *Campylobacter jejuni* colonization in broilers. Veterinary Microbiology. 2010; 143, 314–318. doi:10.1016/j.vetmic.2009.11.029; doi:10.1016/j.vetmic.2009.11.029#doilink.

[42] Hermans D, Martel A, Van Deun K, Verlinden M, Van Immerseel F, Garmyn A, Messens W, Heyndrickx M, Haesebrouck F, Pasmans F. Mint: intestinal mucus protects *Campylobacter jejuni* in the ceca of colonized broiler chickens against the bactericidal effects of medium-chain fatty acids. Poultry Science. 2010; 89, 1144–1155. doi:10.3382/ps.2010-00717.

[43] De Los Santos FS, Hume M, Venkitanarayanan K, Donoghue AM, Hanning I, Slavik MF, Aguiar VF, Metcalf JH, Reyes-Herrera I, Blore PJ, Donoghue DJ. Mint: caprylic acid reduces enteric *Campylobacter* colonization in market-aged broiler chickens but does not appear to alter cecal microbial populations. Journal of Food Protection. 2010; 73, 251–257.

[44] Khoury C, Meinersmann RJ. Mint: a genetic hybrid of the *Campylobacter jejuni* flaA gene with LT-B of *Escherichia coli* and assessment of the efficacy of the hybrid protein as an oral chicken vaccine. Avian Diseases. 1995; 39 (4), 812–820. doi: 10.2307/1592418.

[45] Rice BE, Rollins DM, Mallinson ET, Carr L, Joseph SW. Mint: *Campylobacter jejuni* in broiler chickens: colonization and humoral immunity following oral vaccination and experimental infection. Vaccine, 1997; 15, 1922–1932. doi:10.1016/S0264-410X(97)00126-6; doi:10.1016/S0264-410X %2897%2900126-6#doilink.

[46] Buckley AM, Wang J, Hudson DL, Grant AJ, Jones MA, Maskell DJ, Stevens MP. Mint: evaluation of live-attenuated *Salmonella* vaccines expressing *Campylobacte*r antigens for control of *C. jejuni* in poultry. Vaccine. 2010; 28, 1094–1105.

[47] Layton SL, Morgan MJ, Cole K, Kwon YM, Donoghue DJ, Hargis BM, Pumford NR. Mint: evaluation of *Salmonella*-vectored *Campylobacter* peptide epitopes for reduction of *Campylobacter jejuni* in broiler chickens. Clinical Vaccine Immunology. 2011; 18, 449–454. doi: 10.1128/CVI.00379-10.

[48] Loc Carrillo C, Atterbury RJ, El-Shibiny A, Connerton PL, Dillon E, Scott A, Connerton IF. Mint: bacteriophage therapy to reduce *Campylobacter jejuni* colonization of broiler chickens. Applied and Environmental Microbiology. 2005; 71, 6554–6563. doi: 10.1128/AEM.71.11.6554-6563.

[49] Wagenaar JA, Bergen MAPV, Mueller MA, Wassenaar TM, Carlton RM. Mint: phage therapy reduces *Campylobacter jejuni* colonization in broilers. Veterinary Microbiology. 2005; 109, 275–283. doi:10.1016/j.vetmic.2005.06.002 doi:10.1016/j.vetmic.2005.06.002# doilink.

[50] El-Shibiny A, Scott A, Timms A, Metawea Y, Connerton P, Connerton I. Mint: application of a group II *Campylobacter* bacteriophage to reduce strains of *Campylobacter jejuni* and *Campylobacter coli* colonizing broiler chickens. Journal of Food Protection. 2009; 72, 733–740.

[51] Morishita TY, Aye PP, Harr BS, Cobb CW, Clifford JR. Mint: Evaluation of an avian-specific probiotic to reduce the colonization and shedding of *Campylobacter jejuni* in broilers. Avian Diseases. 1997; 41, 850–855. doi: 10.2307/1592338.

[52] Schoeni JL, Wong AC. Mint: Inhibition of *Campylobacter jejuni* colonization in chicks by defined competitive exclusion bacteria. Applied and Environmental Microbiology. 1994, 60, 1191–1197.

[53] Chang MH, Chen TC. Mint: reduction of *Campylobacter jejuni* in a simulated chicken digestive tract by *Lactobacilli* cultures. Journal of Food Protection. 2000; 63, 1594–1597.

[54] Baurhoo B, Ferket PR, Zhao X. Mint: effects of diets containing different concentrations of mannanoligosaccharide or antibiotics on growth performance, intestinal development, cecal and litter microbial populations, and carcass parameters of broilers. Poultry Science. 2009; 88, 2262–2272. doi: 10.3382/ps.2008-00562.

[55] Boyd Y, Herbert EG, Marston KL, Jones MA, Barrow PA. Mint: host genes affect intestinal colonisation of newly hatched chickens by *Campylobacter jejuni*. Immunogenetics. 2005; 57, 248–253. doi:10.1007/s00251-005-0790-6.

[56] Kaiser P, Howell MM, Fife M, Sadeyen JR, Salmon N, Rothwell L, Young J, Poh TY, Stevens M, Smith J, Burt D, Swaggerty C, Kogut M. Mint: towards the selection of chickens resistant to *Salmonella* and *Campylobacter* infections. Bulletin des Membres de l'Académie Royale de Médecine de Belgique. 2009; 164, 17–25; discussion 25-16.

[57] Line JE, Svetoch EA, Eruslanov BV, Perelygin VV, Mitsevich EV, Mitsevich IP, Levchuk VP, Svetoch OE, Seal BS, Siragusa GR, Stern NJ. Mint: isolation and purification of enterocin E-760 with broad antimicrobial activity against gram-positive and gram-

negative bacteria. Antimicrobial Agents Chemotherapy. 2008; 52, 1094–1100. doi: 10.1128/AAC.01569-06.

[58] Messaoudi S, Kergourlay G, Rossero A, Ferchichi M, Prevost H, Drider D, Manai M, Dousset X. Mint: identification of *Lactobacilli* residing in chicken ceca with antagonism against *Campylobacter*. International Microbiology. 2011; 14, 103–110.

[59] Stern NJ, Svetoch EA, Eruslanov BV, Perelygin VV, Mitsevich EV, Mitsevich IP, Pokhilenko VD, Levchuk VP, Svetoch OE, Seal BS. Mint: isolation of a *Lactobacillus salivarius* strain and purification of its bacteriocin, which is inhibitory to *Campylobacter jejuni* in the chicken gastrointestinal system. Antimicrobial Agents Chemotherapy. 2006; 50, 3111–3116. doi: 10.1128/AAC.00259-06.

[60] Svetoch EA, Eruslanov BV, Perelygin VV, Mitsevich EV, Mitsevich IP, Borzenkov VN, Levchuk VP, Svetoch OE, Kovalev YN, Stepanshin YG, Siragusa GR, Seal BS, Stern NJ. Mint: diverse antimicrobial killing by *Enterococcus faecium* E 50-52 bacteriocin. Journal of Agricultural Food Chemistry. 2008; 56, 1942–1948. doi: 10.1021/jf073284g.

[61] Svetoch EA, Stern NJ. Mint: bacteriocins to control *Campylobacter* spp. in poultry — a review. Poultry Science. 2010; 89, 1763–1768. doi:10.3382/ps.2010-00659.

[62] Stern NJ, Svetoch EA, Eruslanov BV, Kovalev YN, Volodina LI, Perelygin VV, Mitsevich EV, Mitsevich IP, Levchuk VP. Mint: *Paenibacillus polymyxa* purified bacteriocin to control *Campylobacter jejuni* in chickens. Journal of Food Protection. 2005; 68, 1450–1453.

[63] Svetoch EA, Eruslanov BV, Levchuk VP, Perelygin VV, Mitsevich EV, Mitsevich IP, Stepanshin J, Dyatlov I, Seal BS, Stern NJ. Mint: isolation of *Lactobacillus salivarius* 1077 (NRRL B-50053) and characterization of its bacteriocin, including the antimicrobial activity spectrum. Applied and Environmental Microbiology. 2011; 77, 2749–2754. doi: 10.1128/AEM.02481-10.

[64] Stern NJ, Eruslanov BV, Pokhilenko VD, Kovalev YN, Volodina LL, Perelygin VV, Mitsevich EV, Mitsevich IP, Borzenkov VN, Levchuk VP, Svetoch OE, Stepanshin YG, Svetoch EA. Mint: bacteriocins reduce *Campylobacter jejuni* colonization while bacteria producing bacteriocins are ineffective. Microbial Ecology and Health Diseases. 2008; 20, 74–79. doi:10.1080/08910600802030196.