

RESEARCHREPORT



INTERNATIONAL TRADE

Title: Review of Literature and Needs Assessment for Use of Pathogen Reduction Technologies (PRTS) In Fresh Pork – NPB #12-213

Investigators: X. YANG, S. T. HOWARD, D. R. WOERNER, R. J. DELMORE, J. D. TATUM, AND K. E. BELK

Institution: COLORADO STATE UNIVERSITY
FORT COLLINS, CO 80523-1171

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For more information contact:

National Pork Board • PO Box 9114 • Des Moines, IA 50306 USA • 800-456-7675 • Fax: 515-223-2646 • pork.org

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Introduction

The United States is the world's third-largest pork producer and the largest pork exporter. In 2012, U.S. pork production was about 10.4 million metric tonnes (MT), over 2.3 million MT of which was exported (ERS-USDA, 2013). Per capita pork consumption in the U.S. has been estimated to be approximately 46.2 lb per year and has been relatively steady over the past decade (National Pork Board, 2013). Since domestic pork consumption is stagnant, the leading importers of U.S. pork (Japan, Mexico, China and Canada) are critically important to profitability of the U.S. pork industry (USMEF, 2013). Food safety and regulatory issues are a major concern in both foreign and domestic markets for U.S. pork.

Bacterial pathogens are the primary cause of most serious meat safety issues. Foodborne illness can result in economic losses to producers due to recalls from the market place and loss of reputation (Sofos, 2008). Each year, foodborne disease causes an average of 9.4 million illnesses and 1351 deaths in the U.S. (Scallan et al., 2011). Cost due to illness caused by the five major foodborne pathogens (non-typhoidal *Salmonella enterica*, *Campylobacter spp.*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Toxoplasma gondii* and norovirus) has been estimated to be \$12.6 billion per year (Buzby et al., 1996). Scharff (2010) reported that the U.S. spends \$152 billion each year on acute medical care and long-term health-related costs that result from foodborne illnesses.

Pathogens of Concern to the Pork Industry

Salmonella

On August 28, 2013, the USDA-FSIS announced that it would consider expanding testing for *Salmonella* to classes of meat products not currently subject to routine sampling; among these were pork trim and ground pork (FSIS, 2013a). *Salmonella* is one of the three most common bacterial pathogens associated with pork, the other two being *Campylobacter* and *Yersinia enterocolitica* (Fosse et al., 2009). The prevalence of *Salmonella* at various stages of swine production was summarized to be 1% to 75% in various pre-harvest production environments (Baer et al., 2013). Controlling the prevalence of *Salmonella* in the production environment is essential since reduced prevalence of *Salmonella* on carcasses has been reported for swine from farms with lower incidences of the bacteria (Swanenburg et al., 2001). *Salmonella* has been found to become present in lymphatic tissue previously free of the bacteria as quickly as 3 h post-exposure (Fedorka-Cray et al., 1995); consequently, shipping and handling procedures are influential to reducing *Salmonella* in pigs (Hurd et al., 2002; Baer et al., 2013).

Salmonella can survive over a wide range of intrinsic and extrinsic factors, including: temperatures from 5°C to 45°C (Baer et al., 2013), pH values from 4 to 9 (Doyle and Cliver 1990), and in solutions with up to 2% NaCl depending on temperature (Montville and Matthews, 2008). The pH (approx.. 5.6 to 5.8) and water activity of pork ($a_w = 0.99$) are such that these are seldom the limiting factors for growth of most bacteria (Chung and Goepfert, 1970). The conditions most bactericidal to *Salmonella* have been reported to be high salt and nitrite. Brine concentrations greater than 9% and nitrite used at low pH values have both been found to inactivate *Salmonella* (Jay et al., 2005). Ionizing radiation (5 to 7.5 kGy) has been reported to be an effective intervention to destroy *Salmonella* (Jay et al., 2005).

Prevalence of *Salmonella* on pork carcasses was 1.7% in 2011, which was lower than the baseline prevalence of 8.7% reported by USDA Food Safety and Inspection Service (FSIS) in 1996 (FSIS, 1996a; Englejohn, 2013). Another report found fewer than 4% of pork carcasses were positive for *Salmonella spp.* post-chilling, and only 0.6% of positive samples were enumerable (Schmidt et al., 2012). Englejohn (2013) reported that for those pork carcasses that were positive for *Salmonella* following chilling, counts could be extremely high (10 MPN/cm²). *Salmonella* counts that high have not been observed for any other FSIS inspected meat product (Englejohn, 2013). Only 0.07% of ready-to-eat (RTE) pork products were found to be positive for *Salmonella* (Englejohn, 2013). The limited prevalence of *Salmonella spp.* on pork carcasses and RTE pork could be perceived as good news for the industry; however, the presence of *Salmonella* in lymph nodes of pigs harvested in the U.S. has been reported to be as high as 10% to 35% for non-mesenteric lymph (Baer et al., 2013). If FSIS testing for *Salmonella* in pork is expanded to include lymph nodes, further research would be required to determine interventions and prevention of cross-contamination from lymph to muscle tissue.

The CDC estimated that over 11% of the cases of Salmonellosis associated with food were caused by pork products (Englejohn, 2013). Pork was the second leading foodborne source of *Salmonella* infections that resulted in Salmonellosis behind broiler carcasses (Englejohn, 2013). The attribution of cases of Salmonellosis to pork has increased from just over 5% (2003 to 2005) to over 11% (Englejohn, 2013). From 2008 to 2012, CDC has linked four *Salmonella* serotypes to outbreaks of salmonellosis linked to pork products. These outbreaks included two linked to serotype Infantis and one each to serotypes Adelaide and Reading (Englejohn, 2013). *Salmonella* Derby has typically been found to be multidrug resistant (MDR) and an increase has been observed in MDR strains of Infantis (Englejohn, 2013). Current isolates of serotype

Heidelberg have been found to be resistant to ampicillin, chloramphenicol, gentamicin, kanamycin, streptomycin, sulfisoxazole and tetracycline (Hurd, 2013); fortunately, this serotype has not been commonly identified in pork (Englejohn, 2013).

Salmonella Typhimurium definitive phage-type (DT) 104 has been the most recognized strain of MDR *Salmonella* as it is resistant to ampicillin, chloramphenicol, streptomycin, tetracycline, sulphonamides and gentamicin. *Salmonella* Typhimurium DT 104 became an epidemic within cattle in the UK in the early 1990's (Ministry of Agriculture, Fisheries and Food; Welsh Office Agriculture Department; Scottish Office, Agriculture and Fisheries Department, 1998). Infections in humans by DT 104 have been associated with consumption of beef, pork and chicken (Wall et al., 1994). *Salmonella* Typhimurium DT 104 has been reported to have a higher rate of hospitalization and death associated with infection (Varma et al., 2005), and has been reported to be “a foodborne threat” (Doyle et al., 2013). The emergence of MDR strains of *Salmonella* has prompted multiple investigations on antimicrobial resistance of *Salmonella* that are summarized in **Table 1**. Haley et al. (2012) and Futagawa-Saito et al. (2008) reported an increase in the antibiotic resistance of *Salmonella* isolated from pigs. The works summarized in Table 1 are from multiple countries and represent many different serotypes of *Salmonella*; however, resistance genes appear to be widely distributed in populations of *Salmonella* found in various phases of pork production.

Campylobacter jejuni/coli

Jay et al. (2005) summarized that *C. jejuni* may be the most frequent cause of diarrhea in humans. The Food Safety Inspection Service has established performance standards for *Campylobacter* on young chicken and turkey carcasses (FSIS, 2011a); however, no such standards exist for pork carcasses. *Campylobacter* can grow from 30° to 47° C, but can be

inactivated under freezing conditions and are more sensitive to changes in pH and oxygen availability (Gill and Harris, 1984; Doyle, 1990; Kelana and Griffiths, 2003). *Campylobacter* are microaerophilic and can grow in the presence of carbon dioxide and survive under vacuum packaged conditions (Jay et al., 2005; Reynolds and Draughon, 1987).

Data collected from March to August of 2013 by the National Antimicrobial Resistance Monitoring System (NARMS) indicated that 29% and 31% cecal samples from sows and market swine were positive for *Campylobacter*, respectively (Englejohn, 2013). The incidence of *Campylobacter spp.* in the gastrointestinal tract of pigs been reported to be between 70% and 100%, with an especially high prevalence of *Campylobacter coli* (Oosterom et al., 1985; Harvey et al., 1999; Fosse et al., 2009). *Campylobacter* are common to animals and are not typically found in the environment. *Campylobacter jejuni/coli* were the most frequently detected pathogens on raw pork carcass surfaces (31.5%) prior to 1995 (USDA, 1996). Recent data indicated a prevalence of *Campylobacter* on pork between 0% and 33%, depending on the phase of harvest (Baer et al., 2013). The incidence of *Campylobacter* on pork products at the retail level was found to be 1.3% (Duffy et al., 2001).

The serotype of *Campylobacter* may be more important than the prevalence when assessing the risk to human health. The CDC concluded that inappropriate antibiotic use in livestock was a contributing factor to the “serious threat level” of antibiotic resistant strains of *Campylobacter* (CDC, 2013). Resistance of *Campylobacter* to ciprofloxacin, a fluoroquinolone, has increased such that 25% of infections are now resistant (CDC, 2013). Antibiotic resistance of *Campylobacter* isolated from pigs on conventional farms (those that use antibiotics) in the Midwest was reported to be 50% higher for azithromycin and erythromycin, and 25% higher for tetracycline compared to isolates from antibiotic free production systems (Rollo et al., 2010).

These findings were supported by those of Tadesse et al. (2011) who reported *Campylobacter* from conventional swine production systems in the Midwest were 1.4 times more likely to be resistant to erythromycin, although the overall prevalence was still less than 5.0%. The previous work concluded that other factors in addition to antibiotic use contributed to antimicrobial resistance of *Campylobacter* (Tadesse et al., 2011). Other studies reported a high prevalence of antibiotic resistant *Campylobacter* in populations of swine raised with and without antibiotics (Gebreyes et al., 2005; Thakur and Gebreyes, 2005; Quintana-Hayashi and Thakur, 2012). Rosengren et al. (2009) reported an association between macrolide use in swine and strains of *Campylobacter* resistant to erythromycin. The results of studies that have evaluated the prevalence of antibiotic resistance in *Campylobacter* isolated from various phases of swine production are summarized in **Table 2**.

Yersinia enterocolitica

Jay et al. (2005) summarized that pigs are believed to be the most common source of *Yersinia enterocolitica*, while others have found that pigs are the only known livestock reservoir (Bhaduri et al., 2009). Pigs carry *Y. enterocolitica* in their oral cavity; particularly, on their tongue, tonsils and lymph nodes (Nesbakken et al., 2003). The prevalence of *Y. enterocolitica* in market hogs in the U.S. has been reported to be over 90% (Funk et al., 1998). The presence of *Y. enterocolitica* on pork tongues was reported to be > 98% in Finland (Fredriksson-Ahomaa et al., 1999). *Yersinia enterocolitica* can be found at any step of pork production (Davies et al., 2004; Reij and Den Aantrekker, 2004).

Few effective pre-harvest interventions to control *Y. enterocolitica* have been identified because of limited epidemiological research (Davies, 2011; Fosse et al., 2009). *Yersinia enterocolitica* has been documented to grow from -2° C to 45° C (Jay et al., 2005) and can be

destroyed in 1 to 3 min at 60° C (Hanna et al., 1977). Not all strains of *Y. enterocolitica* are toxigenic, but the toxin produced by the bacteria can remain virulent at 100° C for 20 minutes (Jay et al., 2005). *Yersinia enterocolitica* can grow over a range of pH values from 4.2 to 4.8 depending on temperature. Lactic acid has been identified as the second most effective to acetic for raising the minimum pH for growth of *Y. enterocolitica* (Brocklehurst and Lund, 1990). Salinity has been documented to raise the minimum temperature for growth of *Y. enterocolitica*, and, at a pH of 4.6, under the same high salinity, no growth occurred (Stern et al., 1980).

Staphylococcus aureus

Staphylococcus aureus and methicillin-resistant strains of the bacteria (MRSA) have received more attention recently than many of the pathogens common to swine. *Staphylococcus aureus* is found many places in nature, particularly on human skin (CDC, 2006). *Staphylococcus aureus* is species specific and during production of certain pork products, populations of *S. aureus* have been reported to be altered from initially consisting of strains of animal origin to strains specific to humans (Siems et al., 1971 as cited by Jay et al., 2005). Jay et al. (2005) summarized that staphylococci are likely to be found in all foods of animal origin, or those handled by humans. Staphylococci have differing levels of virulence based on their ability to produce enterotoxins that cause morbidity in humans. *Staphylococcus aureus* is classified as a mesophile, but growth has been reported at temperatures as low as 6.7° C (Angelotti et al., 1961); however, temperatures for toxin production to occur been found to be greater than 10° C (Smith et al., 1983). Populations of staphylococci must exceed 10⁵ cells/g for enterotoxin to be produced (ICMSF, 2005). Staphylococci are extremely salt-tolerant, can grow at low water activities (above 0.85) and prefer neutral pH values (Jay et al., 2005). The incubation period for

staphylococcal food poisoning is very short and people can get sick 30 minutes post-consumption (CDC, 2006).

The Centers for Disease Control and Prevention recently reported a 31% decline in the number of MRSA infections. The greatest reduction in MRSA was reported in those cases attributed to previous hospitalization (CDC, 2013). Hospitals are still the source of the most serious MRSA infections (CDC, 2013); however, proximity to swine production has been suggested to be linked to MRSA infections in humans (Casey et al., 2013). Methicillin is a beta-lactam antibiotic that has a similar mode of action to penicillin. Resistance to beta-lactams among gram-negative bacteria is widespread (CDC, 2013), but MRSA represents a gram positive bacteria that has developed beta-lactam resistance. The prevalence of violative sulfa-drug residues in swine (approx. 1%; FSIS, 2011b) indicated use of beta-lactam antibiotics by the industry. Baer et al. (2013) summarized that the MRSA strain common to pigs (ST398) was not a serious threat to human health since the serotype lacks genes to produce enterotoxin. Studies conducted outside the U.S. have found other strains of MRSA in swine (Wagenaar et al., 2009; Battisti et al., 2010; Gómez-Sanz et al., 2010; Lo et al., 2012). Gómez-Sanz et al. (2010) concluded that lineages of MRSA other than ST398 in food animals required further exploration to determine the risk of MRSA in food.

Clostridium perfringens

Clostridium perfringens is a spore-forming, rod-shaped bacterium that produces a toxin during sporulation that causes illness in humans (Juneja et al., 2010). Unlike *C. botulinum* that is found in soil and water, *C. perfringens* is more widely distributed in the environment and can be found in the intestinal tract of humans and animals (Jay et al., 2005). *Clostridium perfringens* is more likely to be introduced during the harvest process, whereas *C. botulinum* needs to be

present in the plant environment to contaminate pork cuts. Growth of *C. perfringens* has not been observed below 20° C (Jay et al., 2005; Labbe and Juneja, 2006) and sporulation occurs between 37° and 40°C (Rey et al., 1975). Strains of *C. perfringens* have been documented to grow between pH values of 5.5 and 8.0, but not in media with NaCl concentrations greater than 5% (Jay et al., 2005). The resistance of *C. perfringens* spores to heat is strain dependent (Jay et al., 2005). *Clostridium perfringens* has low survival rates during freezing (< 5%); however, spores can persist under freezing conditions (Strong and Canada, 1964). The toxin that causes illness as a result of *C. perfringens* is released when cells lyse to release spores. The toxin produced by *C. perfringens* is heat liable, but requires exposure at 60° C for 10 minutes to be inactivated (Stark and Duncan, 1971).

Clostridium perfringens receives less attention than other pathogens since the true incidence of intoxication is unknown due to milder symptoms (Jay et al., 2005). Inappropriate cooling of food can allow sporulation by *C. perfringens*, which makes the pathogen of particular concern in meat products that are re-heated, or in ready RTE products (Juneja et al., 2010). Since many cuts of pork are processed into RTE products, introduction of *C. perfringens* during the harvest of swine should be a concern. The work of Saide-Albornoz et al. (1995) reported no prevalence of *C. perfringens* on pork carcasses, but did find a low prevalence (< 1%) on boneless loins. These findings could indicate that *C. perfringens* was introduced from the plant environment during fabrication. The prevalence of *C. perfringens* on pork carcasses was previously reported to be much higher (Smart et al., 1979), but little research either in the U.S. or abroad exists to contradict or confirm these findings.

Listeria monocytogenes

Listeria monocytogenes is the only pathogenic *Listeria* species (Baer et al., 2013). Only three serotypes have been identified as causing listeriosis in humans: 1/2a, 1/2b and 4b (Rocourt and Bille, 1997). The most common serotypes found in raw pork were 1/2a, 1/2b and 1/2c (Thevenot et al. 2005). Giovannacci et al. (1999) and Lundén et al. (2002) reported that *L. monocytogenes* can survive in swine processing environments for over a year and can grow in a meat grinder for up to three years. *Listeria spp.* are mesophilic and growth has been documented to occur between 0.5° and 45° C making the pathogen of particular concern during refrigerated storage. The range in pH values at which *Listeria* will grow has been reported to be 4.1 to 9.6; however, most growth has been documented to occur at relatively acidic pH values of 4.5 to 5.0 (Jay et al., 2005). Acetic and lactic acid have been found to be antimicrobials for *Listeria*, with acetic acid being slightly more effective (Sorrells et al., 1989). Increasing levels of NaCl at lower pH values has been reported to increase the amount of time required for visible growth of *L. monocytogenes* to occur (Cole et al., 1990).

Baer et al. (2013) summarized that the prevalence of *L. monocytogenes* was extremely low in swine; however, given the ability of the bacteria to persist in processing environments, introduction of the pathogen to commercial plants poses a risk to cross-contaminate fresh pork. *Listeria monocytogenes* has extreme consequences in RTE pork products because the pathogen can grow at refrigerated temperatures (Sim et al., 2002). The Food Safety Inspection Service has a zero-tolerance policy for *L. monocytogenes* in RTE meat due to high mortality rates (approx. 10%; FSIS, 2003; Morbidity and Mortality Weekly Report, 2013). Recent data indicated a 0.52% prevalence of *L. monocytogenes* in RTE products from various species; however, only one positive test result was obtained from pork (Mamber, 2010). Duffy et al. (2001) reported

that 22.9% of ground pork and fresh sausages were contaminated by *L. monocytogenes*. *Listeria monocytogenes* has been found in 2% of samples obtained from the plant environment, with over half of the positive samples found in/on drains, floors and equipment wheels (Mamber, 2010). Although only 121 cases of listeriosis were reported in 2012 (CDC, 2013), 96% required hospitalization and the estimated cost per case was reported to be as high as \$1.2 million when the cost of a recall and loss of business for purveyors was considered (Scharff, 2011).

Toxoplasma gondii

The protozoan *Toxoplasma gondii* is a parasite that can infect livestock as well as humans (Dubey et al., 2003). Transmission of *T. gondii* oocysts to livestock occurs via the ingestion of contaminated substances, such as soil or water (Baer et al., 2013). Consumption of raw or undercooked meat that is contaminated with *T. gondii* may cause illness in humans and other species (Davies, 2011). Studies have found up to 50% of citizens in the U.S. have been exposed to *T. gondii* and have circulating antibodies to the parasite (Roghmann et al., 1999; Jones et al., 2001). The prevalence of *T. gondii* in pigs has been reported to have declined from 24% to 2.7% during the past two decades (APHIS, 2011; Hill et al., 2010). *Toxoplasma gondii* cysts can be destroyed by heating above 60° C or through exposure to radiation 0.3 kGy or higher (Fayer and Dubey, 1985). Eight percent of hospitalizations and 24% of deaths associated with foodborne illness were caused by *Toxoplasma* (CDC, 2011). *Toxoplasma gondii* is of particular concern to pregnant women due to the possibility of abortion and stillbirth (Cliver, 1990).

Trichinella spiralis

Trichinella spiralis is a parasite that can infect pigs. People are infected by consumption of raw or uncooked pork (Gamble et al., 1999a). Historically, prevalence of trichinae in pigs was 2.5%, which has declined to virtually non-detectable levels over the past 60 years in the U.S.

(Gamble et al., 1999b; Pyburn et al., 2005). Twenty percent of the approximately 20 cases of trichinellosis each year result from consumption of pork (Kennedy et al, 2009). The low prevalence of *Trichinella* in the domestic U.S. swine herd indicates the pathogen poses a low risk to human health; however, trading partners such as the EU and Russia enforce policies for testing and/or vaccination.

Escherichia coli O157:H7

Pork can easily be contaminated with *E. coli* O157:H7 and other strains of shiga-toxin producing *E. coli* (STEC); however, the pork industry is less concerned with these pathogens than the beef industry. Baer et al. (2013) summarized the work of Milnes et al. (2009) who reported that the incidence of *E. coli* O157-H7 in swine (0.6%) was low enough that *Campylobacter* and *Salmonella* were substantially greater concerns. The CDC linked one STEC outbreak cluster to a strain of *E. coli* O111 in pork products during the period of 2007 to 2012 (Englejohn, 2013). Nonetheless, barring a major change in prevalence, the threat to public health associated with STEC in pork is low.

Pathogen Interventions

On July 25, 1996, USDA-FSIS issued the Final Rule for Pathogen Reduction; Hazard Analysis and Critical Control Points (HACCP) Systems that required all establishments under federal inspection to: 1) implement sanitation standard operating procedures (sSOP); 2) implement testing for fecal contamination as indicated by generic *E. coli*; 3) implement pathogen reduction standards for *Salmonella*; and, 4) implement a HACCP plan (FSIS, 1996b). Sofos (2009) summarized that effective pathogen control should minimize contamination on live animals, minimize the transfer of contamination to carcasses and inactivate or control the growth of microorganisms that persist on the carcass. Sofos and Smith (1998) reviewed animal washing, chemical de-hairing, knife trimming, steam vacuuming, carcass washing and non-acid chemical as interventions for microbial contamination. It is generally accepted that the synergistic effect of multiple interventions used in succession, or the multiple hurdle concept, is the best approach to reduce microbial contamination. The reduction in bacterial count following any one intervention is dependent on the initial microbial load (Dorsa et al., 1996b, 1997; Graves-Delmore et al., 1997) which is partially determined prior to animals entering packing facilities.

Pre-Harvest Interventions

Baer et al. (2013) concluded that farm management and biosecurity were both effective pre-harvest interventions for controlling pathogens in pork. Narvaez-Bravo et al. (2013) found that increased prevalence of *Salmonella* and *E. coli* O157-H7 at pre-harvest phases of beef production increased the relative-risk that carcasses in the cooler would have some level of contamination. The principles of HACCP dictate prevention of problems before they occur. This could easily be applied to pre-harvest management of swine production given the integration of the industry. Pre-harvest interventions investigated in swine have included

vaccination to control *Salmonella*, *T. gondii*, and *Trichinella*; use of bacteria phages to control *Salmonella*; feed additives to reduce shedding of *Campylobacter*; sanitation to minimize cross-contamination between pigs; and, reduced holding time at the packing facility (Baer et al., 2013).

Vaccination

Denagamage et al. (2007) published a review of literature on the ability of vaccination to reduce the shedding of *Salmonella* by swine. The previous work concluded vaccination may be effective at reducing the shedding of *Salmonella* in market weight hogs; however, it also cited that the methodology of studies that have evaluated the effectiveness of vaccination against *Salmonella* in swine may have reporting biases due to design flaws (Denagamage et al., 2007). Denagamage et al. (2007) concluded that most vaccination studies failed to identify bacterial count data, making determination of the total reduction in microbial load impossible. The validity of any pathogen intervention must be evaluated not only based on statistical differences, but also on the basis of microbiological practicality, or the ability to reduce bacterial counts by at least one log CFU/g (NACMCF, 2009). Most studies agree that vaccination reduces shedding of *Salmonella* (Baer et al., 2013); however, to the best of our knowledge, none have published the corresponding reduction in bacterial counts on carcasses.

Vaccination of swine against *T. gondii* was reviewed by Baer et al. (2013) who concluded that biosecurity measures that minimized the exposure of swine and feed sources to felines were more effective than vaccination to control *T. gondii*. Issues surround vaccination for *T. gondii* since the parasite can readily adapt to immunity acquired by the host and the potential exists for the vaccine to cause toxoplasmosis in swine (Baer et al., 2013). The previous work summarized that if solutions to those problems could be found, vaccination might be effective to control *T. gondii*. Control of *T. gondii* has been achieved through post-harvest handling, such as freezing

(< 10° F) and through cooking to a temperature of greater than 152.6° F (Kijlstra and Jongert, 2008; Baer et al., 2013).

Baer et al. (2013) reviewed the control measures for *Trichinella* in swine, including vaccination. More important than this was the report by Gamble (2011) that summarized testing of over 20,000 pigs from the 1990's, from which 0% were positive for *Trichinella*. The low or non-existent prevalence of *Trichinella* in the domestic U.S. swine herd is superseded by trade barriers that influence the interventions that are used against the parasite. These trade barriers dictate the post-harvest interventions and testing that are required to be used prior to export (9 CFR 318.10), most notably for pork sent to either the EU or Russia. Research that might assist in changing political policy on *Trichinella* would be beneficial to the swine industry, but it is still likely that non-scientific trade barriers would persist.

Feed Additives

Carbadox is a quinoxaline used to control bacterial enteritis and promote growth. The label approved use of carbadox is as a growth promotant and to control swine dysentery. Carbadox will not be discussed as a microbial intervention with respect to food safety as it is more often used at the nursery level. Violative residues of carbadox have been reported in sows and market swine (FSIS, 2011b). The EU, Canada and Australia all forbid use of Carbadox in food producing animals as several metabolites of the drug have been found to be carcinogenic (Ungemach, 2012). Carbadox persists in kidney, liver and fat tissue longer than muscle making residues of particular concern to emerging markets like China that consume larger quantities of variety meats. The swine industry will continue to be reliant on export markets and must analyze the effect that a ban on growth promotants like carbadox might have.

Phage Therapy

Carvalho et al. (2012) summarized that phages offer advantages to antibiotics because they are highly specific to the target bacteria and do not negatively affect other microflora beneficial to the host. Bacteriophages do not replicate outside of hosts where the target bacteria are not present eliminating concerns over residues in food producing animals (Goodridge and Abedon, 2003). Phages for both *Campylobacter* and *Salmonella spp.* have been identified; however, due to issues associated with growing *Campylobacter* in the lab, identification of phages for that pathogen has been more limited (Carvalho et al., 2012). All studies that have evaluated phage therapy for control of *Campylobacter* have been conducted in poultry.

Wagenaar et al. (2005) reported that *Campylobacter* counts were approximately 1 log CFU/g lower in broilers treated with phages. Carvalho et al. (2012) summarized that phage therapy controlled *Campylobacter* in poultry and reported that the initial reduction that occurred in bacterial counts was not maintained, but an overall lower population was still observed in treated birds. Further evaluation of the potential for phage therapy to be used to control *Campylobacter* in swine should be conducted.

Use of phages to control *Salmonella* in swine has been explored. Wall et al. (2010) reported a 2 to 3 log CFU/g reduction in *Salmonella* colonization of pigs treated with *S. Typhimurium*. This agreed with the findings of *in vitro* work that used phages introduced to *Salmonella spp.* resistant to pig gastric juices (O'Flynn et al., 2006). Baer et al. (2013) summarized the issue with these studies was that the small sample sizes were small. Baer et al. (2013) concluded that phages may only be effective if administered immediately prior to harvest. Administration of phages immediately before harvest might be feasible for the swine industry considering group housing offers potential for phages to be administered via water or feed. It is

more likely that the highly specific nature of phages would limit their effectiveness as a microbial intervention.

Post-Harvest Interventions

In April 2012 USDA-FSIS issued the FSIS Compliance Guideline HACCP Systems Validation document that reported many establishments have not adequately validated their HACCP system. The Food Safety Inspection Service requires that all HACCP systems must have documentation supporting that an intervention is able to function as intended (FSIS, 2012). Validation studies establish the range in technical parameters over which an intervention is effective against a pathogen. These studies provide the documentation required by FSIS for an intervention to be included in a HACCP system. If documentation does not exist to support an intervention, then it should not be included as part of a HACCP system.

Interventions likely to be used as critical control points in swine processing HACCP systems include thermal treatment, water washes, application of organic acids and pre-fabrication application of chemical interventions. A survey of major packers found the following interventions were commonly applied during swine processing in the U.S.: pre-evisceration washes and application of peroxyacetic acid; post-evisceration steam vacuuming; final wash application of acetic, lactic, hypobromous or peroxyacetic acid, as well as aqueous ozone and/or hot water; re-conditioning application of lactic acid; cooler application of citric acid; pre-fabrication application of peroxyacetic acid and aqueous ozone; and, application of acetic or peroxyacetic acid to primals and trim. No two HACCP systems are the same; however, literature should exist that has evaluated the previously mentioned interventions if they are to be used as critical control points in swine processing.

Heat/Thermal Treatment

High temperature is one of most effective interventions to kill bacteria. The U.S. Food Safety Inspection Service (FSIS) allows the use of thermal interventions as a critical control point in HACCP plans. Scalding pork carcasses at 56° C for 1 to 1.25 minutes has been reported to reduce *Campylobacter*, *Salmonella* and *Escherichia coli* counts from 10⁶/ml to less than 10/ml (Sorquist and Danielsson-Tham, 1990). Similar results were found for *Yersinia spp.* when pork carcasses were scalded at 60°C for 2.5 minutes (Sorquist and Danielsson-Tham, 1990). Pearce et al. (2003) reported that 1% of pork carcasses were positive for *Salmonella* following scalding. Total viable bacterial counts have been reported to be < 2.0 CFU/cm² following scalding; however, 12% of carcasses have been found to be positive for *Enterobacteriaceae* following scalding (Spescha et al., 2006). Generic *E. coli* was typically not detected after scalding in a Canadian swine processing facility (Namvar and Warriner, 2006).

Hot water washes, steam vacuuming and steam pasteurization have been validated for the use as critical control points on meat carcasses (Dorsa et al., 1996a; Nutsch et al., 1997; Phebus et al., 1997; Retzlaff et al., 2004; Sommers et al., 2002). Hot water (85° C) yielded a 2 log₁₀ CFU reduction in *E. coli* counts when applied for 15 second to un-eviscerated pig carcasses (Bolton, 2004). Eggenberger-Solorzano et al. (2002) reported that the reduction in *Enterobacteriaceae* on pork muscle tissues was increased when the temperature of water used for washes was increased from 65° to 80°C. Loretz et al. (2011) published a review of studies that found that hot water washes applied to pork carcasses at many different parameters (e.g, temperature, time) were a microbiologically practical intervention for most pathogens of concern.

Sofos and Smith (1998) pointed out that spray systems (washes) that use high pressure may cause penetration of bacteria into tissue. Steam is an alternative intervention to hot water that is commonly applied to hog carcasses in the U.S. (Bolton, 2004). The advantage of steam is the capability to penetrate areas of the carcass that may not be reached by water washes (James et al., 2007; Hugas and Tsigarida, 2008). Steam pasteurization has been evaluated as a pathogen intervention applied to beef carcasses (Phebus et al., 1997; Sofos and Smith, 1998; Huffmann, 2002; Retzlaff et al., 2004); however, there have been fewer studies that have evaluated the use of steam applied to pork carcasses. Trivedi et al. (2008) found that three household types of steam/vacuum cleaning systems used on pork carcasses were capable of reducing the population of *L. monocytogenes* from 7.6 to 3.2 log₁₀ CFU/cm². A vacuum-steam-vacuum surface pasteurizer was used to inactivate *L. innocua* on the surface of hot dogs and ham skins (Kozempel et al., 2000; Sommers et al., 2002). Populations of psychrophilic and mesophilic bacteria on pork carcasses were reduced following application of steam and lactic acid singly or in combination (Pipek et al., 2006). The effective parameters for steam vacuuming as a microbial intervention used on pork carcasses have not been extensively reported. Bolton et al. (2001) stated that steam vacuuming is typically used as a GMP, but if a GMP is used as an upstream control to reduce the likelihood of a hazard occurring, the technology would still need to be validated. Bolton et al. (2001) summarized the critical limits for hot water washes, steam vacuuming and organic acids as pathogen interventions applied to beef carcasses, but no such review exists for pork.

Organic Acids

Studies have validated the use of organic acids on beef carcasses and beef products. Fewer validation studies have been completed to justify the use of organic acids or chemical

compounds in pork plants. Nevertheless, FSIS has approved several organic acids and their salts as antimicrobial interventions to be used during pork production (FSIS, 2013b). Lactic acid at a concentration of 2 to 5% and a temperature of 55°C has been approved for application to pork primals and trim. *Salmonella* Typhimurium can be reduced to an undetectable level on the surface of pork after treatment with a 2% lactic acid solution for 30 seconds (van Netten et al., 1995). Acetic and peroxyacetic acid have been approved as antimicrobials for red meat carcasses (21 CFR 173.370). A blend of lactic acid (45% to 60%), citric acid (20 to 35%) and potassium hydroxide (> 1%) can be sprayed on pork carcasses at maximum level of 2.5% by weight. A combination of hydrochloric and citric acid at a concentration of 1 to 2%, a pH value of 1.5, applied at a pressure of 2.07 to 27.5 BAR, for 2 to 5 seconds has been approved for pork carcasses (FSIS, 2003). In cured meat, Sofos (1989) and Shelef (1994) found that a mixture of lactates, acetates and diacetates in cured meat formulations was able to reduce *L. monocytogenes*. Citric acid up to 10% can be applied to bologna in an inedible casing before slicing. Chlorine dioxide (a food contact substrate) can be applied as a spray or dip to control bacteria on RTE products, provided residues are less than 3 ppm prior to packaging. Barmpalia et al. (2005) reported that sodium lactate, sodium diacetate, and glucono-delta-lactone (singly or in combination) included in the formulation of pork bologna inhibited the growth of *L. monocytogenes*.

The issue with the use of organic acids and other chemical interventions by the pork industry is not effectiveness; rather it is the highly specific parameters (e.g., concentration, temperature, pressure, etc.) that these compounds have been approved to be used at. The extreme (high and low) parameters at which chemical interventions are effective for application to beef carcasses have been defined (Pittman et al., 2013). The result of this work was

determination of an acceptable range over which organic acids and other interventions could be used as part of a HACCP system. Conduction of such a study by the pork industry would allow multiple producers who use the same intervention, at different application parameters, the ability to cite one study that would provide supporting documentation for the intervention.

Chilling

Chilling improves pork quality and slows the growth of bacteria (Huff-Lonergan, 2000). Low temperatures slow enzymatic activity and extend the lag phase of microorganisms (Troller, 1976; Vanderzant et al., 1985). In the U.S., rapid (-5° C for 2 h) or ultra-rapid (-30° C for 30 min) chilling are utilized by the pork industry prior to tempering carcasses for approximately 24 h at 0° C (Cutter, 2003). Cooper (1968) and James et al. (1983) found that the anti-microbial properties of the two chilling methods were not different. Later work reported that blast or ultra-rapid chilling was able to reduce populations of *C. coli* to non-detectable levels, whereas this was not observed with conventional chilling (Chang et al., 2003). Nesbakken et al. (2008) published similar findings and determined that the prevalence of *Campylobacter spp.* on pork carcasses was reduced from over 50% to less than 2% following blast chilling. The antimicrobial effect of blast chilling on *Campylobacter* has been reported to be due to a desiccation effect (Loretz et al., 2011). Chang et al. (2003) found that blast chilling may also reduce *L. monocytogenes* and *S. Typhimurium* counts by 0.4 to 1.1 log CFUcm². Some large scale packers chill carcasses and then apply citric acid during the tempering phase.

Citric acid has been reported to decrease aerobic plate counts and total coliforms in pork loins treated with a 1.5% solution and stored for 14 d (Fu et al., 1994). Pittman et al. (2011) found that citrus essential oils (CEO) reduced populations of generic *E. coli* that served as surrogates for *E. coli* O157-H7 and *Salmonella spp.* on beef brisket flats. The previous work

reported CEO were an effective intervention against *E. coli* O157:H7 and *Salmonella spp.* during refrigerated storage. No works exist that have summarized the effectiveness of citric acid as an intervention applied to pork carcasses, particularly when applied at refrigerated temperatures. Application of organic acids during chilling requires further exploration to be validated as an effective microbial intervention to be used during pork production.

Freezing can kill microorganisms by forming ice crystals that can penetrate cell membranes and by causing permanent chemical changes to the lipid bilayer. Rust and Zimmermann (1972) found that the use of liquid nitrogen or liquid carbon dioxide at the temperature of -29°C destroyed *Trichinella*, which was confirmed by Smith (1975). Although it is rare to freeze pork carcasses and pork prior to domestic consumption, pork for export is commonly frozen which likely addresses any concern for contamination with *Trichinella*. Freezing pork has also been found to be an effective mechanism to kill *T. gondii* (Kotula et al., 1991). Freezing is likely the most practical intervention to satisfy concerns of trading partners as they relate to *Trichinella* or *T. gondii*.

Hot-Boned Pork

Harvest of sows comprises a major portion of pork production in the U.S. Sow plants fabricate pre-rigor carcasses (hot-bone) for inclusion in sausage formulations. Pork plants that hot-bone carcasses typically do not scald or singe carcasses; instead they are skinned immediately following slaughter. The microbial challenges associated with hot-boned pork are different due the intrinsic properties of the product (higher pH). The ability of bacteria to attach to pork skin was found to be different than the ability to attach to the fat surfaces of beef or lamb carcasses (Butler et al., 1979). Several works have reported differences in the ability of either *E. coli* O157:H7, *L. monocytogenes*, *S. Typhimurium*, *Y. enterocolitica* or *S. aureus* to attach to

either fat or muscle tissue, typically reporting greater attachment to muscle (Dickson and Koohmaraie, 1989; Rivas et al., 2006; Morild et al., 2011). Kinsella et al. (2007) summarized that two of the major stresses that bacteria attached to meat surfaces encountered were low temperatures and osmotic pressure that resulted from surface drying. The previous work reported that attachment of *S. Typhimurium* DT 104 to meat prevented injury to the bacteria as a result of changes in osmotic pressure due to desiccation or cold stress. Desiccation has been found to be influential to survival of *Campylobacter* on swine carcasses (Loretz et al., 2011). Pork carcasses that are hot-boned are not exposed to chilling procedures that may yield beneficial effects of desiccation. Duffy et al. (2001) reported on the prevalence of several pathogens in hot-boned pork plants; however, little work been completed on the topic since that time. To our knowledge, no study has been conducted on the differential attachment of bacteria to hot-boned pork carcasses and the resulting effectiveness of interventions used in those settings.

Irradiation

Ionizing radiation has both direct and indirect effects on pathogens. Radiation can destroy DNA and RNA and produce radicals in the cell that can cause ionization of molecules. Irradiation only affects bacteria and parasites, but does not provide any effect on viruses or toxins (Hugas and Tsigarida, 2008). Brake et al. (1985) found that 20 krad or radiation was able to slow the growth of *T. spiralis*, whereas 30 krad was capable of inactivating *T. spiralis* in ground pork. The use of gamma rays to control *Trichinella* in fresh or previously frozen pork was permitted by the amendment of the Federal Meat Inspection Act in 1986. In 1999, FSIS amended its regulation to allow the use of radiation on refrigerated and frozen uncooked meat and on meat by-products to eliminate or reduce foodborne pathogens or to extend shelf life

(FSIS, 1999). According to the Code of Federal Regulations Title 21, Volume 3, to control *Trichinella* in pork carcasses or fresh, non-heat-processed cuts of pork carcasses, the U.S. Food and Drug Administration (FDA) permits the use of irradiation at a level of 0.3 to 1.0 kGy (U.S. FDA, 2013). Pork can be labeled as “certified” if companies prove that viable trichinae have been inactivated by irradiation. In 2009, 57 countries had approved the use of irradiation (IAEA, 1999; Kume et al., 2009). The biggest concerns when pork is irradiated are quality issues such as lipid and pigment oxidation; consequently, in the U.S., only around 80,000 tons of pork are irradiated each year (Kume et al., 2009).

Other Interventions and Practices

Aqueous ozone has been approved as a contact food-sanitizing agent by the FDA and FSIS (FSIS, 2002). Several major pork packers in the U.S. have been reported to use aqueous ozone as an intervention either as a final rail or pre-fabrication wash. Unfortunately, few reports from the U.S. exist on the effectiveness of aqueous ozone used as an intervention on pork carcasses. Ozone is a powerful oxidant and not only should it be evaluated as an effective antimicrobial, but the effect of ozone on sensory attributes and shelf life must also be thoroughly explored. The cost-benefit of ozone may be such that its antimicrobial properties are not worth the decline in sensory attributes and shelf-life (e.g. color deterioration, oxidation).

The effect of high-pressure processing on pathogens of concern to the pork industry was summarized by Baer et al. (2013). The previous authors concluded that this intervention was effective against *T. gondii*, *Trichinella* and *L. monocytogenes*. Jofré et al. (2009) reported that high-pressure processing at 600 MPa was an effective intervention against *L. monocytogenes*, *Salmonella*, *Y. enterocolitica* and *C. jejuni* that had been inoculated at 3.5 log CFU/g onto sliced cooked and dry cured ham. *Staphylococcus aureus* has been shown to be resistant to high-

pressure processing applied to pork products (Jofré et al., 2009). High-pressure processing could be used in packaged subprimals and further processed pork products if the technical parameters for inclusion in a HACCP system are documented by the pork industry. High-pressure processing has been reported to have detrimental effects on color and sensory attributes. These challenges may need to be explored prior to widespread acceptance of high-pressure processing as a microbial intervention applied to pork.

Nisin is generally recognized as safe (GRAS) and has been approved in both the EU and U.S. to be used as an antimicrobial against gram-negative pathogens (Cutter and Siragusa, 1995). To increase the efficacy of nisin as an antimicrobial on meat products, it has been reported to be better to use the compound with other antimicrobials such as lactic acid (Barboza et al., 2002). Samelis et al. (2005) confirmed the anti-listerial effect of nisin used with sodium diacetate or potassium benzoate as dipping solution applied to pork bologna. Nisin is likely most applicable to processed products; however, research could be conducted on the usefulness of the compound as an antimicrobial applied to carcasses.

Lauramide arginine ethyl estermonohydrochloride (LAE) is an antimicrobial approved by the U.S. FDA and USDA to be applied to RTE and raw pork sausages (EFSA, 2007). Lauramide arginine ethyl estermonohydrochloride is GRAS and is manufactured by LAMIRSA under the common name lauric arginate. Food Safety Inspective Service allows the application of LAE within packages or on meat surfaces at concentration up to 44 ppm by weight of the finished product (FSIS, 2013b). The antimicrobial effect of LAE on commercially-prepared hams was evaluated with sprayed lethality In Container (SLICTM) that allowed the impregnation of a liquid form of LAE into shrink-wrap vacuum bags before introducing the meat product (Luchansky et

al., 2005). The previous work reported a reduction in *L. monocytogenes* of 6.5 log₁₀ CFU/ham for samples treated with 8 ml of a 5% solution of LAE.

Electrolyzed oxidizing (EO) water is considered a new microbial intervention. Electrolyzing water that has sodium chloride in it results in the formation of sodium hypochlorite, a common antimicrobial. Electrolyzed oxidizing water has been found to yield a 7 log₁₀ CFU/ml reduction of *Escherichia coli* O157:H7, *Salmonella enteritidis*, and *Listeria monocytogenes* after a five- minute exposure at 4° or 23°C (Venkitanarayanan et al., 1999). Fabrizio and Cutter (2004) found that spraying EO water on fresh pork bellies surfaces inoculated with feces for 15 seconds had the ability to reduce *Campylobacter coli* by 1.81 log₁₀ CFU/cm² at 0 d after application. Further investigation into the possibility of EO as an intervention for use by the pork industry is warranted.

Gap Analysis

Expansion of testing for *Salmonella* in pork products is almost certain. One of the critical elements of HACCP Final Rule is that “Plants must meet Pathogen Reduction Performance Standards, set by FSIS-USDA, for *Salmonella*.” Control of *Salmonella* must be of primary concern to the pork industry. Prevalence of *Salmonella* in other tissues of pork carcasses such as the lymphatic system must be fully determined. The structure of the swine industry could allow for a more integrated approach to food safety than any other red meat industry. Continued research on vaccination is likely the best approach to address *Salmonella* concerns in live animals. Development of phage therapy could offer an alternative to vaccination that might be effective if applied at the correct time before harvest and if phages are developed for multiple serotypes of *Salmonella* and possibly *Campylobacter*. Pre-harvest control of *T. gondii* is still likely to be best achieved through biosecurity and through research into how to improve a vaccine for the parasite. *Trichinella* persists as a regulatory issue with trading partners. It is unlikely that regulations will be changed concerning this pathogen. Continuing the monitoring processes that are conducted for the pathogen is the best approach for the U.S. swine industry to document the exceptionally low risk that the *Trichinella* poses to human health.

Few works exist that have evaluated the control of *Yersinia enterocolitica* in the U.S. swine population. Data exist that show a high prevalence of the pathogen in pigs. Further research is required by the U.S. swine industry to address potential concerns with *Y. enterocolitica*. Given the presence of the pathogen in tongues, *Y. enterocolitica* poses a threat to become a food safety issue in export markets for variety meats. The U.S. swine industry will continue to be dependent on exports to maintain economic viability and should explore fully the concerns of all trading partners as they relate to food safety.

The emergence of antimicrobial resistant strains of *Salmonella* and *Campylobacter* cannot be ignored by the swine industry. No data currently exist on the use of antibiotics or growth promotants by the industry. Some work conducted in the U.S. has reported an increase in the antibiotic resistance of bacteria common to swine (Haley et al., 2012). Moreover, the swine industry has an issue with violative antibiotic residues by comparison to every other class of livestock (FSIS, 2011b). If the swine industry hopes to be proactive instead of reactive, investigation into the contribution of swine production practices in the U.S. to antimicrobial resistance and drug residues must be conducted to provide a science based conclusion.

No studies have been conducted to evaluate the range in application parameters for chemical microbial interventions that may be effective to control pathogens on pork. It may not be necessary to apply chemical compounds to pork carcasses if thermal controls work effectively. The differences in production environments between plants that harvest market pigs vs. sows are such that interventions must be evaluated in both settings. Outside of routine FSIS testing, baseline data for the frequency of carcass contamination in sow plants has not been collected since 2001 (Duffy et al., 2001). The differential ability of bacteria to attach to skin vs. muscle/fat, and the ability of those bacteria to survive following attachment, dictates that interventions must be validated in both market hog and sow plants.

Listeria monocytogenes is a concern to the manufacture of RTE pork products. Based on the prevalence of *L. monocytogenes* in live swine, it is more likely that the pathogen is introduced from the plant environment to meat products rather than as a result of initial contamination from live animals. The ability of *L. monocytogenes* to persist in plant environments is of critical importance to producers of RTE products. Many studies exist that have evaluated control mechanisms for *L. monocytogenes*; however, eliminating the pathogen

from processing environments still appears to be a challenge. Further research may be necessary to document sanitation procedures that could be more effective at controlling the presence of *L. monocytogenes* in the plant environment.

Novel interventions such as oxidizing water and high-pressure processing offer potential to control pathogens on pork. Determination of the effect of these interventions on pork quality will be required prior to widespread acceptance by the industry. High-pressure processing has the capability to sterilize meat products; however, this occurs as a result of protein denaturation. Protein denaturation negatively impacts color and functional properties of meat products. Exploration of the appropriate application parameters of high-pressure processing to pork may be required to insure consumer acceptability of pressure-treated pork products. It is more likely that high-pressure processing should be applied as one of several interventions within a system, rather than as a lethality step.

Conclusions

Salmonella and *Campylobacter* should be the safety issues of greatest concern to the swine industry, particularly in production of fresh pork. Identification by CDC (2013) of antibiotic use in livestock as a contributing factor to the serious threat of resistant strains of *Campylobacter* should be concerning to the swine industry. The antibiotic residue issue in pork is now at a level that surpasses the dairy industry (FSIS, 2011b). The incidence of pathogens on pork carcasses has been reduced, but the safety issues associated with the pathogens found on pork are dramatically more complex. Science based solutions to validate interventions and production practices must be found to improve the safety of U.S. pork.

Table 1. Summary of frequency (%) of antimicrobial resistance *Salmonella* isolated from various phases of swine production. Antibiotics (except gentamicin) are those that *Salmonella* Typhimurium DT104 has resistance to.

Study	Sample	N	Antibiotic ¹					
			Ampicillin	Chloramphenicol	Streptomycin	Tetracycline	Sulfamethoxazole	Gentamicin
Gebreyes et al., 2004	Fecal	1,314	46.6	21.5	-	85.2	-	2.1
Gebreyes and Thakur., 2005	Farm	28	79.0	79.0	75.0	86.0	79.0	0.0
Zhao et al., 2005	NARMS	166	37.3	28.9	44.0	45.8	43.4	1.8
Gebreyes et al., 2006	Fecal	85	35.3	25.9	85.9	93.0	64.7	2.4
Gebreyes et al., 2006	Carcass	115	23.5	21.7	54.8	97.4	50.4	0.0
Thakur et al., 2007	Finishing	226	13.8	13.2	28.3	89.3	21.2	-
Thakur et al., 2007	Pre Evisceration	5	0.0	0.0	0.0	100.0	0.0	-
Thakur et al., 2007	Post Evisceration	12	0.0	0.0	10.0	66.0	10.0	-
Thakur et al., 2007	Post-Chill	15	0.0	0.0	20.0	0.0	33.3	-
Thakur et al., 2007	All Isolates	400	9.7	7.5	31.5	78.5	24.7	-
Futagawa-Saito et al., 2008	Fecal	193	23.8	23.8	35.7	39.7	39.7	0.0
Perron et al., 2008	Lymph	367	29.4	25.1	-	68.4	-	-
Rosengren, 2008	Fecal – Finishing	241	9.1	7.1	8.7	27.4	16.6	0.4
Rosengren, 2008	Fecal – Sows	144	16.7	5.6	10.4	22.9	27.8	0.0
Huang et al., 2009	Tissue / Fecal	197	55.8	-	-	83.8	-	6.6
Lim et al., 2009	Fecal / Lymph	37	75.7	40.5	81.1	81.1	81.1	16.2
Yan et al., 2010	Pork at Retail	12	16.7	16.7	0.0	33.3	83.3	0.0
Malik et al., 2011	Tissue / Fecal	406	74.0	-	-	-	-	13.0
Haley et al., 2012	Fecal	584	38.7	33.2	53.3	78.6	-	2.1
Keelara et al., 2013	Fecal / Carcass	386	13.9	8.0	27.7	80.3	55.9	-
Keelara et al., 2013	Environment	536	14.1	4.1	14.5	78.3	43.4	-

¹ β -Lactam: ampicillin; Aminoglycosides: gentamicin, streptomycin; Sulfonamides: sulfamethoxazole; Quinolones: tetracycline, chloramphenicol.

Table 2. Summary of antimicrobial resistant *Campylobacter* isolated from various phases of swine production.

Study	Sample	N	<i>Campylobacter</i> Positive (%)	Erythromycin Resistant (%)	Azithromycin Resistant (%)	Tetracycline Resistant (%)	Ciprofloxacin Resistant (%)	Gentamicin Resistant (%)
Gebreyes et al., 2005 ^a	Fecal	150	56.3	55.3	--	58.8	0.5	--
Thakur and Gebreyes, 2005	Fecal	370	55.8	77.0	--	83.4	2.8	0.8
Rollo et al., 2010	Fecal	1,422	35.8	68.3	70.0	74.5	--	--
Frye et al., 2011 ^b	Fecal	196	61.0	17.3	17.3	17.9	1.5	0.5
Quintana-Hayashi and Thakur, 2011	Fecal	464	72.7	25.2	25.3	88.3	17.1	0.0
Tadesse et al., 2011	Fecal	838	56.3	63.2	--	65.9	3.2	3.8
Shin and Lee, 2010 ^c	G.I.	572	19.9	70.1	--	--	--	--
Rosengren et al., 2009 ^d	Herd	20	98.6	50.0	--	0.0	5.0	0.0
Thakur and Gebreyes, 2005	Carcass	195	2.6	81.4	--	80.8	0.0	0.5
Quintana-Hayashi and Thakur, 2011	Carcass	158	1.6	20.9	20.9	82.9	15.2	0.0
Tadesse et al., 2011	Carcass	1,173	3.2	64.7	--	47.1	0.0	5.9

^aSample population was obtained from a group of swine raised without antibiotics.

^bEstimate of *Campylobacter* positive samples reported from total sample population (N = 7,960). Subset used to determine resistance was evaluated from samples (N = 196) positive for multiple bacteria, including *Campylobacter*.

^cG.I. – sample obtained from the gastrointestinal tract.

^dData reported at the herd level, erythromycin resistance was obtained from an estimate of macrolide resistance.

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