RESEARCH REPORT
PORK SAFETY

Title: Systematic Review of the Magnitude of Change in Prevalence and Quantity of Salmonella after Administration of Pathogen Reduction Treatments on Pork Carcasses – **NPB # 14-287**

Investigator: Annette M. O'Connor (oconnor@iastate.edu)

Institution: Iowa State University

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Industry Summary: Our objective was to summarize the current research on the effect of various washes, rinses, sprays, and steam against *Salmonella* on pork carcasses or parts of pork carcasses with the skin on. In January 2015, we searched the scientific literature and the bibliographies of 24 review articles. This search identified 4001 references. These were assessed for relevance by two reviewers. In all, 14 studies were found to be relevant and to contain extractable data. Five of these studies were conducted in commercial abattoirs. The following treatments were examined: citric acid + steam, steam + ultrasound, saline (saltwater), water + acetic acid, acidified sodium chlorite, electrolyzed oxidizing water, distilled water, water (at various temperatures), stannous chloride, hydrogen peroxide, trisodium phosphate, sodium hypochlorite, lactic acid, and acetic acid. There was little evidence of a consistent positive or negative effect of washes containing acid compared to washes containing only water. There was also no consistently positive effect of using warm or hot water or steam compared to cooler water. There was no strong evidence that any one treatment regime (concentration and temperature of acid treatment, application of hot water vs application of cooler water) was clearly superior to others for the control of *Salmonella* on skin-on pork carcasses. The most consistent finding was a positive effect of acid treatments on the prevalence of *Salmonella* on carcasses; however, this was based on the results from the individual studies. It was not possible to combine the results across the different studies to produce a summary measure of effectiveness because the treatment protocols differed too much from study to study (e.g. different doses and durations of treatment, some studies measured prevalence of *Salmonella* as an outcome, some measured concentration; some studies applied *Salmonella* to the carcasses prior to treatment, other studies were of naturally contaminated carcasses).

Keywords: Salmonella, systematic review, carcass wash, acid washes

Scientific Abstract: In this systematic review, we summarize the change in *Salmonella* prevalence and/or quantity associated with pathogen reduction treatments (washes, sprays, steam) on pork carcasses or skin-on carcass parts in comparative experimental designs (natural or artificial contamination). In January 2015, Centre for Agriculture and Biosciences International Abstracts (1910–2015), Science Citation Index and Conference Proceedings Citation Index – Science (1900–2015), Medline® and Medline® In-Process (1946–2015) (OVIDSP interface), Science.gov, and Proceedings of the International Symposium on Epidemiology and Control of *Salmonella* in Pork (1996–2012) were searched with no language or publication type restrictions. Reference lists of 24 review articles were checked. Two independent reviewers screened 4001 titles/abstracts and assessed 122 full-text articles for eligibility using pre-tested forms. Non-English-language records were not translated or extracted. Data were extracted from 14 studies and risk of bias was assessed independently by two reviewers. Five of these studies were conducted in commercial abattoirs. We considered risk of bias due to systematic error to be moderate overall; a major source of bias was the potential differential recovery of *Salmonella* from treated carcasses due to knowledge of the intervention. There was no strong evidence that any one intervention protocol (acid temperature, acid concentration, hot water, cool water) was clearly superior to others for control of *Salmonella*. The most consistently observed association was a positive effect of acid washes on measures of *Salmonella* that were categorical; however, this was based on individual results, not a summary effect measure.

ABSTRACT

In this systematic review, we summarize the change in *Salmonella* prevalence and/or quantity associated with pathogen reduction treatments (washes, sprays, steam) on pork carcasses or skin-on carcass parts in comparative experimental designs (natural or artificial contamination). In January 2015, Centre for Agriculture and Biosciences International Abstracts (1910–2015), Science Citation Index and Conference Proceedings Citation Index – Science (1900–2015), Medline® and Medline® In-Process (1946–2015) (OVIDSP interface), Science.gov, and Proceedings of the International Symposium on Epidemiology and Control of *Salmonella* in Pork (1996–2012) were searched with no language or publication type restrictions. Reference lists of 24 review articles were checked. Two independent reviewers screened 4001 titles/abstracts and assessed 122 full-text articles for eligibility using pre-tested forms. Non-English-language records were not translated or extracted. Data were extracted from 14 studies and risk of bias was assessed independently by two reviewers. Five of these studies were conducted in commercial abattoirs. We considered risk of bias due to systematic error to be moderate overall; a major source of bias was the potential differential recovery of *Salmonella* from treated carcasses due to knowledge of the intervention. There was no strong evidence that any one intervention protocol (acid temperature, acid concentration, hot water, cool water) was clearly superior to others for control of *Salmonella*. The most consistently observed association was a positive effect of acid washes on measures of *Salmonella* that were categorical; however, this was based on individual results, not a summary effect measure.

INTRODUCTION

Non-typhoidal *Salmonella* is one of the most common causes of human foodborne illness in the world (1). In the United States, an estimated 1.0 to 10.1% of human salmonellosis cases can be attributed to the consumption of contaminated pork (2). A benefit-cost analysis of pre-harvest and processing interventions against *Salmonella* in the pork production chain indicated that rinsing carcasses with water at different temperatures (with and without sanitizer) is a more cost-effective way of reducing the number of human salmonellosis cases than on-farm interventions (3). While EU Regulation (EC) No 853/2004 of the European parliament and of the Council (available from the EUR Lex website: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2004:139:0055:0205:EN:PDF. Accessed 20 September 2015.) allows water to be used for the decontamination of pork carcasses, in the United States other substances, including organic acids, are permitted for this purpose, according to the United States Department of Agriculture Food Safety and Inspection Service (USDA/FSIS) Directive 710.1 Revision 29 (http://www.fsis.usda.gov/wps/wcm/connect/bab10e09-aefa-483b-8be8-809a1f051d4c/7120.1.pdf?MOD=AJPERES. Accessed 20 September 2015.). However, we do not know the comparative effectiveness of these approaches, since at the time of our review, a systematic review of the effect of washes, rinses, and sprays on pork carcasses has not yet been conducted. Such a review could allow summarization of the magnitude of *Salmonella enterica* reduction associated with each product across several studies, with the expectation of providing information for better decision-making by commercial abattoir operators and regulatory bodies.

The objective of this review was to describe the changes in the prevalence and/or quantity of *Salmonella* on pork carcasses or parts of pork carcasses after receiving pathogen reduction treatments during processing. The PICOS question was: What is the change in *Salmonella* prevalence or quantity (O-Outcome) associated with the use of pathogen reduction treatments applied as washes, rinses, or sprays (I - interventions) to pork carcasses or parts of pork carcasses (P-Population) in study designs (S-Study design) employing randomized or non-randomized comparative experiments? An interim report of this review was presented at the 11th International Conference on the Epidemiology and Control of Biological, Chemical and Physical Hazards in Pigs and Pork (4).

MATERIALS AND METHODS

Protocol and registration. A protocol documenting the research question, eligibility criteria, intended information sources, search strategy, study selection, data collection process (including a draft of the data extraction form), assessment of risk of bias (including a draft of the risk-of-bias form), and planned synthesis of results was written in advance. This protocol is not registered. It was decided after the review began to modify the protocol objective to include studies on parts of pork carcasses so that potentially relevant studies would not be excluded and data from laboratory-based studies would become eligible.

Eligibility criteria. No restrictions were placed on language of publication, publication date, or publication status. Relevant populations were pork carcasses or parts of pork carcasses from commercial swine in commercial abattoirs; however, after screening of titles and abstracts it was decided to include laboratory-based studies. As the search and the screening would have captured laboratory-based studies, it was considered that no bias would occur due to this change. Traditional smallholder slaughter approaches were not applicable to the expected target population, who were commercial packers in the USA. Relevant interventions and comparators were pathogen reduction treatments applied as washes, rinses, and/or sprays including organic acids, aqueous ozone, electrolyzed water, potassium hydroxide, potassium sorbate, sodium hypochlorite (NaClO), trisodium phosphate (TSP), chlorine, sodium chlorite, hot or cold water treatments, steam vacuuming and steam pasteurization or any combination of these treatments, to pork carcasses. The outcomes of interest were the presence and/or quantity of *Salmonella* on the carcass as measured by methods including bacterial culture, enzyme-linked immunosorbent assay (ELISA), PCR, or other antigen-detecting methods. Relevant study designs were those employing comparative experiments on pork carcasses or carcass parts, whether naturally or artificially contaminated with *Salmonella*.

Information sources. The following information resources were searched in January 2015: Centre for Agricultural Biosciences International (CABI) Abstracts (1910–2015) (Web of Science interface), Science Citation Index (SCI) and Conference Proceedings Citation Index – Science (CPCI-S) (1900–2015) (Web of Science interface), Medline® and Medline® In-Process (1946–2015 (OVIDSP interface), Science.gov (via http://www.science.gov/scigov/), and International Conference on the Epidemiology and Control of Biological, Chemical and Physical Hazards in Pigs and Pork (Safe Pork) (1996–2012) (http://lib.dr.iastate.edu/safepork/). The last search was run on 31 January 2015. A limited updated literature search was performed in CABI Abstracts on 20 March 2015, which produced three new records. To find further relevant records that had not been captured by the information resources listed above, the reference lists of relevant review articles identified during the above search were scanned. The reference lists of all records that proceeded to the data extraction phase of the review were also checked for relevant references not captured by the database searches.

Search. A search strategy was developed to capture the concepts of pork carcass decontamination by an information specialist in consultation with food safety content experts. The concept of pork carcasses was combined with the second concept involving search terms for decontamination methods, using the AND operator. As well as the two concepts combined, there was also a general search for carcass decontamination (animal unspecified) to capture reports of the general issue. A more detailed search strategy was initially run in CABI Abstracts (Table S1). The final search strategy run in CABI Abstracts is shown in Table 1 (showing the number of records returned). Search strategies were adapted to run appropriately in other databases taking into account interface differences and database differences, such as the availability of subject indexing terms. The full search strategies for SCI and CPCI-S, Medline® and Medline® In-Process, Science.gov (via http://www.science.gov/scigov/), and Safe Pork are shown in Table S2, Table S3, Table S4, and Table S5, respectively. No restrictions were made with respect to language or document type for any of the searches.

Study selection. Records were imported into DistillerSR® (Evidence Partners, Ottawa, Ontario, Canada) and de-duplicated. Two reviewers working independently screened the records for eligibility. Level 1 screening was based on the title and abstract alone, although the reviewers were not blinded to the author(s), title, abstract, and year of publication of each record during screening. The Level 1 form was pretested on 100 records before actual screening began. The Level 1 form consisted of a single question: "Does the study appear to be primary research on pathogen reduction washes/rinses/sprays for pork carcasses or parts of a pork carcass?" The options for answering were: "Yes", "No", "Can't tell", and "No, but this is a relevant review." If even one reviewer answered "Yes" for a given record (even if the other reviewer disagreed), that record was passed to the second level of screening. For all other answer combinations, records were excluded. Level 2 screening was based on the full text of the record. Disagreements between the reviewers at Level 2 were resolved by consensus. The Level 2 form consisted of the following questions and answer options:

Q1. Is the full text available in English?
Yes (Include)
No (Exclude)
Q2. Based on the full text, is the study about primary research on pathogen reduction washes/ rinses/sprays for pork carcasses or parts of a pork carcass?
Yes (Include)
No (Exclude)
Q3. Was *Salmonella* found in any of the samples tested?
Yes (Include)
No, but the investigators did look at *E. coli*, Enterobacteriaceae, coliforms, and/or Total Plate Count (TPC) (Exclude)
No, the authors did not look at any of the above (Exclude)
Q4. Based on the full text does the study have a parallel comparative group?
Yes (Include)
No (Exclude)
Q5. Did the investigators measure the outcome *only* > 24h after application of the intervention?
Yes (Exclude)
No (Include)
Q6. What type of study is this?
Challenge (artificial contamination)
Control (natural contamination)

Question 3 was revised during screening as the wording in the protocol ("Q3. Did the investigators look at *Salmonella*?") would otherwise have resulted in the inclusion of studies in which the investigators looked for, but did not find, *Salmonella* in any of the tested samples. These studies would have had no relevant data to extract, so it was deemed best to exclude them at Level 2. The purpose of Question 4 was to eliminate studies in which only one intervention was tested by examining samples before and after intervention application. The purpose of Question 5 was to eliminate studies examining only the effects of storage on *Salmonella*.

Data collection process. All studies passing Level 2 screening underwent data extraction. Non-English language records were not translated; however, these were marked for reference so that if funds become available in the future, they can be translated and extracted. The data extraction forms for the study-level information and the intervention/outcome-level information were pilot-tested by both reviewers and revised for clarity and ease of use before the review began.

Two reviewers extracted intervention and outcome data independently from all relevant studies into DistillerSR®, with the exception of Epling (5). Study-level and risk-of-bias data for Epling (5) were extracted by two independent reviewers. However, due to the amount of outcome data in Epling (5) and the time constraints involved, only one reviewer extracted these data; a second reviewer then verified that the outcome data had been correctly extracted. All conflicts between reviewers at the data extraction stage were resolved by discussion and, as needed, by consulting a content expert (J. Dickson). No investigators were contacted to confirm or obtain missing or unpublished data.

Data items. The final drafts of the study-level and intervention-outcome forms are presented in Table S6 and Table S7, respectively.

Risk of bias in individual studies. Risk of bias was assessed in a non-blinded manner by two reviewers. The risk-of-bias assessment form is reported in Table S8. The risk-of-bias form was based on The Cochrane Collaboration's Risk-of-Bias Tool (6, Table 8.5a), and was modified during the review process as follows: It was decided to remove the questions relating to the method used to conceal allocation of the experimental units to intervention groups, because none of the extracted papers discussed their allocation methods. Additionally, when assessing performance bias, the method used to collect samples to measure the outcome was considered. If swab samples were taken and the method of swabbing was not described in detail or was such that it might be subject to individual variation, performance bias was considered to be high. This was because many of the carcass interventions would have affected the experimental units in such a way (i.e. through smell, temperature, colour change) that the person collecting the sample might have been able to determine that it had undergone an intervention (vs the control group) and might have expended more or less effort in swabbing the carcass, thus affecting the concentration or prevalence estimates for the experimental units in that group. If a more objective sampling method was used (excision of tissue of specified dimensions) or an objective swabbing technique was used, e.g. FSIS Method (7), the risk of performance bias was deemed to be low. Additionally, after consultation with our content expert (J. Dickson), outcome bias was considered to be low if the outcome was prevalence or concentration of *Salmonella* (reported as CFU, Most Probable Number, or number of organisms).

Summary measures. The summary measures of interest specified in the protocol were mean differences for continuous outcomes and summary risk ratio or summary odds ratio for categorical outcomes.

Planned methods of analysis. Depending on how comprehensively the results of the studies were reported, we proposed in the protocol to conduct a mixed-treatment comparison meta-analysis with ranking of the interventions. However, post hoc (evaluating the data available) it was decided to conduct a pairwise meta-analysis looking at specific pairwise comparisons of

interest, in particular the comparison of lactic acid to water, and acetic acid to water. Prior to conducting this analysis it was decided that calculation of a summary effect would likely not be conducted as few studies used the same concentration of an acid. Other interventions that were rare were summarized with text only.

Risk of bias across studies. We assessed studies to have an overall high risk of bias if they had at least one risk-of-bias domain with a high risk of bias. Presentation of risk-of-bias data was made using Review Manager (RevMan) software (version 5.3; Cochrane Collaboration, The Nordic Cochrane Centre, Copenhagen, http://tech.cochrane.org/revman/download). We originally planned to conduct an analysis for small-study effects, acknowledging that it might not be possible to detect small-study effects as most of the eligible studies were expected to be small. The criteria for defining a study as small were based on the number of experimental units, not on the number of pseudo-replicates, where pseudo-replicates were subsamples taken from each experimental unit.

Additional analyses. We proposed, a priori, that if the sample size was sufficient, we would conduct a meta-regression to determine what factors were associated with the magnitude of the effect size based on the demographic factors collected. After identifying relevant studies and extracting the data, the small number of relevant studies and the heterogeneity of interventions among those studies, this analysis was not considered likely to be informative and was therefore not conducted. We did not propose, a priori, to do any other additional analyses.

RESULTS

Study selection. The search of the electronic databases returned 5299 records. After de-duplication, 3993 records remained. Table 2 reports the number of records found before and after de-duplication for each database searched. After reading the reference lists of 24 relevant review articles as well as the bibliographies of all articles that went through data extraction, an additional 8 potentially relevant records were added to the total to be screened, resulting in a total number of 4001 records to be screened at Level 1. The total number of records screened, assessed for eligibility, and included in the review, with reasons for exclusion at each stage, is shown in Fig. 1. Table S9 lists the citation information for each record excluded at the full-text stage (Level 2 screening), along with the reason for its exclusion. Fourteen studies involving 17 experiments were eligible for inclusion in the review.

Study characteristics. The characteristics of the included studies are reported in Table 3. Of the included studies, only Machado et al. (2013) reported the year in which their data were collected (in 2008). Of the included studies taking place at commercial abattoirs, the slaughter capacity was reported by Hamilton et al. (9) (>500,000/y and 5.5 carcasses/min during the study), Trivedi et al. (10) (50-60 hogs/day twice each week at Plant A, and 24-40 hogs/week once each week at Plant C), and Epling et al. (11) (700-800 hogs/h). Although Eggenberger-Solorzano et al. (12) and van Netten et al. (13) conducted experiments at commercial abattoirs, they did not report the slaughter capacity of these establishments. The methods used to inoculate the experimental units in the challenge studies are presented in Table 4. The methods used in each included study to measure the outcome of prevalence and/or quantity of *Salmonella* in the experimental units are presented in Table 5.

Risk of bias within studies. A graphical depiction of the risk-of-bias summary across and within studies is shown in Fig. 2. None of the studies provided a detailed description of the approach to allocating carcasses/skin/jowl to the interventions, and therefore for all studies the risk of bias due to confounding of the intervention effect by other factors was unclear. The risk of bias due to failure to blind the outcome assessor was considered low for all studies.

Results of individual studies. The results of included studies for all outcomes considered are presented in Table 6. Data from a thesis (5) are not shown. Epling (5) reported the reduction of *Salmonella* in experimentally inoculated skin samples after treatment with 1%, 2%, 5%, and 10% lactic acid, a combination of 2% lactic acid and 2% acetic acid, 2% acetic acid alone, and water. Each of these interventions was applied at three different temperatures (25°C, 55°C, and 10°C) via immersion, no-charge sprayer (applied 40 cm away from the sample at a pressure of 137.9 kPa), or by an electrostatic sprayer (run at 1000 V) and tested for effect of the intervention at 10 min, 2.5 h, and 24 h (according to the Results section, 25 h according to the Methods section) after application. This resulted in 189 different intervention permutations, which would have been unwieldy to present in a table. Further, regrettably, Epling (5) reported neither the sample sizes for each intervention nor the precision estimates of the reported concentrations. Epling (5) did report results of a separate study of the effect of 2% lactic acid on naturally contaminated pork carcasses, and these results appear to have been published in a peer-reviewed journal (11) whose results are reported in Table 6.

Trivedi et al. (10) examined the effects of a commercial household steam cleaner (Steam Fast SF 275, Top Innovation, Inc., Riverside, Missouri, USA, steam capacity 1500 W, water tank maximum capacity 1.44 L, steam chamber and hosepipe with nozzle) applied to the ham, belly, and jowl of carcasses in 100 cm^2 areas for 60 s with the nozzle held 6 to 7 cm away after the final carcass wash but before any organic acid solutions were sprayed on the carcass. The investigators performed their pork carcass experiments in two slaughter plants (Plant A and Plant C). However, the results of this study are difficult to translate to application due to the approach to sampling and analysis. First, the authors presented the *Salmonella* data for the two plants combined so that it was not possible to separate the data by plant. Statistical analysis of the *Salmonella* results was not undertaken. The authors presented results as "Number of samples positive" but did not specify the number of carcasses that were positive. Three samples were taken per carcass, but the authors did not clarify if more than one of the four positive samples in the control group came from the same carcass or from different carcasses. The review team felt the impact of the intervention was very different, if for example three samples on one carcass were positive compared to one sample on three carcasses. This reporting approach rendered interpretation of the results difficult and inclusion of the results into the conclusions of this review impossible.

Morild et al. (14) examined the effect of steam ultrasound applied to a 10 x 10 cm piece of inoculated jowl skin. As this was the only study to examine this intervention it is not summarized in a forest plot. Steam ultrasound was applied in a test cabinet (85 x 79 x 57 cm, Force Technology, Brøndby, Denmark) using steam (130°C) at 354.6 to 506.6 kPa applied through nine nozzles. Only the upper surface of the sample could be treated with this apparatus; the lower surface was therefore left untreated. Ultrasound (30 to 40 kHz) was generated through nozzles that were 10 to 12 cm from the surface of the sample, and kinetic energy was delivered by steam

pressure. The authors did not report the results of their control groups. The authors did report that the reduction in *S. enterica* serovar Typhimurium was significantly different after 0.5 s vs 1.0 s of steam-ultrasound and after 1.0 s vs 2.0 s of steam-ultrasound. The mean reduction in *S.* Typhimurium between the two inoculation levels (10^4 vs 10^7) did not differ significantly for the 0.5 s (p=0.073), 1.0 s (p=0.095), 1.5 s (p=0.084), and 2.0 s (p=0.066) treatments.

Morris et al. (15) studied the effect of trisodium phosphate (TSP) (AvGARD^{TM}, Rhone-Poulenc Inc., Cranbury, New Jersey, USA) on samples of pork skin collected < 45 min post-exsanguination and inoculated with rifampicin-resistant *S.* Typhimurium prior to treatment. As this intervention was only assessed in one study, it was not summarized in a forest plot. The authors did not report results for the three samples that received no treatment. For the samples dipped for any length of time (5 s, 10 s, or 15 s), the mean post-treatment concentration of *Salmonella* was lower for all samples dipped in actual TSP solutions compared to samples dipped for the same length of time in the corresponding 0% TSP control solution ($p<0.05$). Within each concentration group (4%, 8%, or 12% TSP), samples dipped for different durations (5 s or 10 s or 15 s) did not have significantly different mean post-treatment concentrations of *Salmonella* ($p>0.05$).

van Netten et al. (16) studied the effects of 2% lactic acid (pH 2.3; temperature 21^{o}C $\pm$ 2^{o}C) vortexed (200 rev/min for 2 s) with inoculated skin cell suspensions (25 cm^2 pork belly skin stomached in Seward Stomacher 400 and placed in 45 ml sterile peptone water (0.5% NaCl, 1% peptone, pH 6.9.)). Immediately after treatment the lactic acid activity was quenched using a 4 mL solution of 0.05 mol/L K_3PO_4, 3% (wt/vol) tryptic soy broth, and 0.3% (wt/vol) yeast extract to bring the pH of the skin suspension up to 7.4. The authors did not report data for the control group (sprayed with water for 90 s) for the pork belly experiments, which focused on the effect of washing on muscle and were not relevant to this review.

Christiansen et al. (17) studied the effect of 1% or 2.5% (vol/vol) lactic acid or hot sterile water on inoculated pork jowl skin samples (10 x 10 cm). The samples were placed vertically, and the hot water or lactic acid were poured over them with a watering device to simulate in-line cabinet hot water carcass decontamination. For this experiment, 10 interventions were assessed. The comparisons of greatest interest to this review are presented in Table 6.

Epling et al. (11) used an electrostatic dispersion sprayer (air pressure 137.9 kPa; electrode potential 1000 V) to apply approximately 150 ml of 2% (v/v) L-lactic acid solution evenly over half of the carcass (n=75 carcasses) from a distance of 40 cm. The lactic acid spray decreased the prevalence of *Salmonella* immediately after treatment ($p<0.05$) and 24 h after treatment ($p<0.01$) when applied to the ham or shoulder.

Fabrizio and Cutter (18) examined the effect of spraying (using a food-grade hand-held garden sprayer (Hudson, Hastings, Minnesota, USA; Model 67220)) distilled water, sodium hypochlorite, electrolyzed oxidizing water, or aged electrolyzed oxidizing water on inoculated pork bellies hung vertically on a stainless steel rack in a biological safety hood. The acidic electrolyzed oxidizing water (1150 mV oxidation reduction potential; 50 ppm free chlorine) was produced by passing a 12% NaCl solution across a charged bipolar membrane (EO water generator, ROX Water Electrolyzer, Hoshizaki America, Inc., Peachtree City, Georgia, USA).

Aged acidic electrolyzed oxidizing water was created by storing acidic electrolyzed oxidizing water for 24 h at 4^{o}C in an airtight bottle. All treatments (including the distilled water) were significantly different from the control (no-treatment) group ($p<0.05$).

In their laboratory experiment, Eggenberger-Solorzano et al. (12) examined pieces of skin (1 cm x 1 cm x 0.5 cm), obtained from scalded hog carcasses, placed in 50 ml centrifuge tubes with water at different temperatures and vortexed for varying lengths of time. The authors found no significant difference ($p>0.05$) between any of the water treatments in the level of *Salmonella* after treatment. In their commercial experiment, the authors studied the effects of acetic acid and/or hot water on carcasses. The hot water was applied using a low-pressure spray wash system at 172.4 kPa. Acetic acid (1.8% vol/vol) was applied at < 172.4 kPa using the commercial acid rinse cabinet already installed at the abattoir so that the entire carcass was covered in approximately 3 s. When both treatments were used, hot water was applied first, followed, 7 s later, by acetic acid.

Biemuller et al. (19) in their pilot plant studies sprayed acetic acid (pH 1.5 or 2.0) or stannous chloride or hydrogen peroxide onto inoculated pork carcasses from a distance of 8 cm from the carcass. To achieve pH 1.5 and 2.0 acetic acid, the authors added hydrochloric acid to 0.1 N acetic acid solutions. As hydrochloric acid is not approved for use in pork processing (FSIS/USDA, Directive 710.1 Revision 29 http://www.fsis.usda.gov/wps/wcm/connect/bab10e09-aefa-483b-8be8-809a1f051d4c/7120.1.pdf?MOD=AJPERES. Accessed 20 September 2015), it was decided not to include this study in a forest plot summary. The authors also investigated the effect of steam applied for 10 s (according to their Results section) or 30 s (according to their Methods section) 2.5 cm from the carcasses (apparatus used to apply the steam not reported).

van Netten et al. (13) examined the effect of water or lactic acid spray (400 ml/min) at 11^{o}C or 55^{o}C applied after the final carcass wash at a commercial abattoir. The lactic acid solutions were at a concentration of 2% (pH 2.3) or 5% (pH 1.9) and were made from a 50% L(+)-lactic acid stock solution (Chemie Combination, Amsterdam). Spraying was carried out using a spray nozzle at a distance of 20 cm from the carcass with an electrostatic spray apparatus (Wezer, Assendelft, The Netherlands). After treatment, three samples were taken per carcass and if even one of these tested positive for *Salmonella*, the carcass was considered to be positive.

Morild et al. (20) examined the effects of hot water or 1% lactic acid rinses on *Salmonella*. They used swabbing to detect superficially attached bacteria and stomaching of tissue samples to detect firmly attached bacteria. The number of bacteria attached to surfaces did not differ significantly ($p=0.06$) between 5-s vs 15-s treatments, so these data were pooled. The total number of superficially and firmly attached bacteria was higher after decontamination with both hot water and lactic acid compared to non-decontaminated (i.e. control) skin ($p<0.0001$).

Synthesis of results. As mentioned above it was originally hoped to conduct a mixed-treatment comparison meta-analysis; however, the sparse dataset precluded this option. It was possible, however, to construct three forest plots that help to identify patterns within the dataset. Fig. 3 presents a forest plot of the data from studies that assessed lactic acid-based interventions and reported measures of *Salmonella* that were concentration-based. We used the standardized mean

difference as the effect measure because the studies used different metrics; Christiansen et al. (17) used the mean reduction in *Salmonella* between two time points, while Fabrizio and Cutter (18) and Morild et al. (20) used the mean concentration post-treatment as the metric for comparison. Fig. 3 has no summary effect because it graphs all possible pairwise comparisons and therefore reuses control arm data; as such, sample size estimates would be inflated and variance underestimated if a summary effect were calculated. For example, if a three-arm trial existed, with two acid-based interventions and one control arm, these data would be presented as two pairwise comparisons with the control arm providing data twice, but no summary effect would be presented. In Fig. 3 there is little evidence of a consistent positive or negative effect of acid washes compared to water washes, as most estimates center around the null value of zero.

In Fig. 4 we present data from studies that compared the prevalence of *Salmonella* after treatment with one form of water or steam to another treatment, which was usually a standard carcass treatment or cooler treatment. In Fig. 5 we present a forest plot for the *Salmonella* prevalence estimates for studies that used various acid treatments with some type of water treatment (either standard or warm/hot water). Again, it is important to note that a summary effect was not reported because the control arms were repeated. Fig. 5 provides a more consistent picture of a positive effect of acid-based treatments on *Salmonella* prevalence.

Risk of bias across studies. As a meta-analysis was not conducted, we did not evaluate the effect of small-study effects on the outcome.

Additional analyses. No additional analyses were conducted.

DISCUSSION

Summary of evidence. When discussing the conclusions we used the GRADE evidence framework (21) (strength of association, consistency, directness) as a basis for considering the conclusions; however, we did not conduct a formal GRADE panel meeting. With respect to the strength of association, there does not seem to be strong evidence that one intervention protocol (acid temperature, acid concentration, hot water, cool) is clearly superior to others for the control of *Salmonella* on pork carcasses. Overall Fig. 4 shows a generally more favorable effect of warmer water over cooler or standard water washes for reducing the prevalence of *Salmonella*; however, only the Hamilton et al. (9) study showed a significant difference between the treatments. The preliminary conclusion we reached from these data is that there is no strong evidence for the efficacy of one particular intervention. With respect to consistency, the most consistently observed association is a positive effect of acid washes on measures of *Salmonella* that were categorical; however, this is based on individual results and not a summary result, which was not calculated for reasons already discussed. The directness of the findings to pork production is mixed; some studies were conducted in abattoir settings, which are clearly relevant to the target population. Laboratory-based challenge studies predominate in this review for obvious reasons i.e., it is an unacceptable public health risk to introduce *Salmonella* into an abattoir. The validity of findings from challenge studies in our opinion should be viewed as such; the results were probably optimal because the laboratory studies occurred in controlled settings, and we would expect the same interventions to have smaller and more variable effects when applied in commercial settings.

This review was focussed on studies of *Salmonella* on pork skin because of the issue of directness. The use of other organisms (*E coli*, etc.), other animal species (beef or poultry), or tissue type (muscle) were considered too indirect to be useful for answering the review question. Note that we use the term directness because it matches the GRADE evidence frame work, however "applicability" would be suitable synonym.

Limitations. The conduct of this review is consistent with current standards for systematic reviews. Steps were taken to ensure that an *a priori* protocol was developed and made available (Protocol S1). An extensive search was conducted along with reference checking; duplicate record screening, eligibility assessment and data extraction were undertaken, the data were reported comprehensively, and conservative and thoughtful analysis (within the limitations of the data available) are provided to the end-user of the review. Our ability to explicitly address the review question about the magnitude of reduction we would expect based on pathogen reduction treatments was limited by the approach to reporting the underlying data and the absence of repeated protocols assessed by independent groups. The most common issue was the failure of authors of the included studies to clarify the unit of concern and the units for measures of variation. This limited our ability to use all of the potentially available data in the meta-analysis. We did not follow-up with the authors of these papers regarding missing data because our experience with previous reviews indicated that this could have potentially taken months to hear back from these authors and would have extended the time required to complete the review.

It would have been preferable to have been able to assess a specific dose and duration of either water or acid intervention. As is evidenced from the information about the study characteristics, interventions, methods of detection, and outcomes, few studies were directly comparable (Table 3, Table 4, Table 5, and Table 6).

After the cut-off date for consideration for the review, two additional review documents (22, 23) were brought to our attention. We evaluated the bibliographic lists of these reviews and identified three potentially relevant studies (Table S10), which were not evaluated as part of this review, but which can be considered for an update of this review in the future.

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TABLES

TABLE 1 Search strategy run in CABI Abstracts on 21 January 2015 for a systematic review of pork carcass decontamination against *Salmonella*

Search no.	No. records identified	Search string
#1	25,552	TS=((pork or swine or pig or pigs or hog or hogs or boar or boars or sow or sows) near/7 (carcass* OR slaughter* or abattoir* or bellies))
#2	7614	TS=(pathogen near/4 reduc*) OR TS=prt
#3	57,209	TS=(wash or washes or washing or washed or rinse or rinses or rinsing or rinsed)
#4	192,274	TS=(spray or sprays or spraying or sprayed)
#5	157	TS=(Organic NEAR/5 (decontaminat* or saniti*))
#6	46,191	TS=(PEROXYACETIC OR LACTIC)
#7	56,989	TS=(ACETIC OR hypobromous or citric or "mineral acid$")
#8	6609	TS=((HYDROCHLORIC OR NITRIC OR PHOSPHORIC OR ACID) NEAR/5 (spray* or decontaminat* or saniti* or wash*))
#9	134	TS=NONACID
#10	26,217	TS=((hot or cold or electrolyzed or electrolysed or warm) NEAR/3 water)
#11	70,878	ts="water treatment$"
#12	24,497	TS=steam
#13	70	TS="AQUEOUS OZONE"
#14	2,975	TS=("POTASSIUM HYDROXIDE" OR "POTASSIUM SORBATE")
#15	10,343	ts=("sodium hypochlorite" OR NaClO or "sodium acetate" or "sodium citrate" or "sodium chlorite" or "sodium lactate")
#16	144,840	ts=(TSP or phosphate$)
#17	139,833	TS=(CHLORINE OR ALCIDE OR ULTRAVIOLET OR UV OR IRRADIAT* OR "DRY HEAT" OR ULTRASOUND)
#18	15,540	TS=((Prevent* or reduc*) near/4 contaminat*) or TS=decontaminat*

#19	34,856	TS=(Chilling or "freezing air" or "high air velocity" or blasting)
#20	756,777	#19 OR #18 OR #17 OR #16 OR #15 OR #14 OR #13 OR #12 OR #11 OR #10 OR #9 OR #8 OR #7 OR #6 OR #5 OR #4 OR #3 OR #2
#21	1516	#20 AND #1
#22	473	TI=((DECONTAMINAT* OR CONTAMINAT*) AND CARCASS*)
#23	1943	#22 OR #21

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TABLE 2 The number of studies found for each electronic database before and after de-duplication in a systematic review of pathogen reduction treatments against *Salmonella* in pork carcasses

Database	Date of search	No. records identified	No. records after deduplication
CABI Abstracts	21 January 2015	1946[a]	1931
SCI and CPCI-S	25 January 2015	1099	363
Medline® and Medline® In-Process	25 January 2015	1440	901
Science.gov	30 January 2015	163	160
Safe Pork	31 January 2015	651	643
Total		5299	3993

[a] Includes three references found during an updated search conducted on 20 March 2015.

94 **TABLE 3** Characteristics of included studies in a systematic review of pathogen reduction treatments against *Salmonella* on pork
95 carcasses

96

Source	Country	Setting[a]	Interventions investigated	Exptl Unit	Did investigators inoculate the exptl units with *Salmonella*?
Machado et al. (8)	Brazil	Lab	Organic acid[b], saline, steam, steam + citric acid	Pork leg (mean wt 3.5 kg)	Yes
Hamilton et al. (9)	Australia	CA	Hot water, acidified sodium chlorite	Carcass	No
Trivedi et al. (10)	USA	CA	Steam	Carcass half	No
Fabrizio and Cutter (18)	USA	Lab	Electrolyzed oxidizing water, distilled water, sodium hypochlorite, lactic acid	Section of pork belly	Yes
Eggenberger-Solorzano et al. (12) Commercial	USA	CA	Hot water, acetic acid, hot water + acetic acid	Pork jowl	Yes
Eggenberger-Solorzano et al. (12) Laboratory	USA	Lab	Hot water	Skin	Yes

[a] CA = Commercial abattoir; Lab = Laboratory; URS = University/Research Slaughter Plant

[b] Citric acid was the main constituent (other constituents not reported)

Epling et al. (11)	USA	CA	Lactic acid	Carcass half	No
Biemuller et al. (19) Second pilot study	NR[c]	URS	Acetic acid + water	Carcass	Yes[d]
Biemuller et al. (19) First pilot study	NR	URS	Acetic acid, stannous chloride, hydrogen peroxide, steam	Carcass	Yes
Morris et al. (15)	USA	Lab	Trisodium phosphate	Skin	Yes
van Netten et al. (13)	The Netherlands	CA	Lactic acid, hot water	Carcass	Yes
Morild et al. (14)	NR	Lab	Steam ultrasound	Pork jowl	Yes
Morild et al. (20)	Denmark	Lab	Lactic acid, hot water	Pork jowl	Yes
Christiansen et al. (17)	Denmark	NR	Lactic acid, hot water	Pork jowl	Yes
van Netten et al. (16)	The Netherlands	Lab	Lactic acid	Pork skin suspension	Yes
Epling (5)	USA	NR		Skin	Yes

[c] NR = Not Reported

[d] Some samples were naturally infected.

97		Lactic acid, acetic acid, lactic + acetic acid, hot water, cold water

TABLE 4 Inoculation methods used in challenge studies in a systematic review of pathogen reduction treatments against *Salmonella* on pork carcasses

Source	Type of *Salmonella enterica* serovar inoculated	Inoculum dose	Inoculation method
Machado et al. (8)	Typhimurium phage-type DT177	10^3 CFU/mL	Pork legs were immersed in the inoculum solution. The intervention was applied 15 min after inoculation.
Fabrizio and Cutter (18)	Typhimurium ATCC 13311	10^7 CFU/mL (inoc. concn); 10^6 CFU/cm^2 (final concn)	UV-treated pork bellies were inoculated with porcine fecal suspension using a sterile spray bottle. Bacteria were allowed to attach for 15 min at room temp prior to application of the intervention.
Eggenberger-Solorzano et al. (12) Commercial	NR[a]	NR	One part fresh hog fecal material mixed with two parts distilled water was brushed once onto the jowls with a 5 cm wide foam brush. Time between inoculation and intervention application not reported.
Eggenberger-Solorzano et al. (12) Laboratory	Typhimurium ATCC 13311	10^6 CFU/g	Pork skin slices were inoculated by immersion in manure/distilled water mixture for 1 min. Interventions were then applied.
Biemuller et al. (19) Second pilot study	Enteritidis	10^6 organisms/mL	Skin of freshly slaughtered pig carcasses was inoculated using a cotton swab. Time interval between inoculation and intervention application was not reported.

[a] NR = Not Reported; CFU = colony forming units

Biemuller et al. (19) First pilot study	Enteritidis	10^6 organisms/mL	Skin of freshly slaughtered pig carcasses was inoculated using a cotton swab. Time between inoculation and intervention application was not reported.
Morris et al. (15)	Rifampicin-resistant Typhimurium ATCC 13311	10^4 organisms/cm^2	Each 10 cm^2 sample of pork skin was dipped for 10 s in the inoculum then allowed to stand for 20 min at 25°C (according to the Methods section) or 10 min (according to Table 1 of the Results section) before the intervention was applied.
van Netten et al. (13)	Acid-adapted[b] Typhimurium strain S1	1.7 ± 0.2 $\log_{10}$ CFU/cm^2	Immediately after evisceration the inoculum was poured from the top of the carcass downward. Interventions were applied about 20 min after inoculation.
Morild et al. (14)	Typhimurium 4/74 MS 21697, Typhimurium DT104 MS 14329, Derby MS 21664, and Infantis MS 21663	10^4 or 10^7 CFU/cm^2	Inoculum was spread onto the surface of the pork jowls with a Drigalski spatula. The intervention was applied after 30 min at room temp.
Morild et al. (20)	Typhimurium 4/74	2×10^9 CFU/mL (inoculum);	Within two min following the application of the intervention, the inoculum was applied to the pork jowl surface using a Drigalski spatula. Inoculated samples were left at room temp for 30 min prior to sampling for *Salmonella*.

[b] *Salmonella* selected had survived 120 s in 2% lactic acid (pH 2.3) at 21°C. Organisms were cultured in tryptic soy broth (TSB, pH 5.8) using 10% lactic acid at 30°C for 1 d then on fresh TSB-LA at 17°C for 2 d.

		7.54 ± 0.04 log_{10} CFU/cm^2 (final surface concn)	
Christiansen et al. (17)	Typhimurium 4/74	10^7 CFU/cm^2	Inoculum was spread onto the surface of 10 x 10 cm pieces of pork jowl using a sterile spatula. Application of the intervention occurred after 30 min at room temp.
van Netten et al. (16)	Typhimurium strain S1	10^8 CFU/mL	A piece ($25cm^2$) of pork skin was excised from the pork belly and stomached in 45mL 0.5% NaCl , 1% peptone and Seward Stomacher 400 (manufacturer not reported) (pH 6.9). This pork skin suspension was then inoculated by centrifuging (at 6000 *g*, make of centrifuge and model of rotor not reported) bacterial cultures and resuspending them in equal volume with pork skin. The intervention was applied 20 min later.
Epling (5)	Typhimurium	10^7 organisms/mL	Pork skin sections were inoculated (method not reported) and allowed to dry for 10 min prior to application of the intervention.

TABLE 5 Methods used to measure the prevalence and/or quantity of *Salmonella* in a systematic review of pathogen reduction treatments against *Salmonella* on pork carcasses

Source	Pre-enrichment	Enrichment	Bacterial culture	Confirmation method	Method to determine concn of *Salmonella*	Other methods used
Machado et al. (8)	Sterile BPW[c] for 20 h at 37°C	RV[d] broth for 24 h at 42°C	XLD[e] for 24 h at 37°C	None	MPN[f]/100 cm² of sampled surface	None
Hamilton et al. (9)	BPW for 18 ± 2 h at 37 ± 1°C	RV soy broth at 41.5 ± 1°C and Muller–Kauffmann tetrathionate/ novoboicin broth at 45 + 1°C and 37 ± 1°Crespectively, for 24 ± 3 h	XLD and BGA[g] for 24 ± 3 h at 37 ± 1°C	Subcultured on CLED[h] at 37°C ± 1°C for 24 h ± 3 h. Typical colonies confirmed by LA[i] using Serobact™ *Salmonella* (manufacturer NR). LA-negative colonies underwent	ND[j]	None

[c] BPW = Buffered peptone water

[d] RV = Rappaport-Vassiliadis

[e] XLD = Xylose lysine desoxycholate agar

[f] MPN = Most probable number

[g] BGA = Brilliant green agar

[h] CLED = Cystine–Lactose–Electrolyte Deficient agar

[i] LA = Latex agglutination

[j] ND = Not done

				biochemistry. (MICROBACT™12E, Oxoid Pty Ltd).		
Trivedi et al. (10)	Lactose broth for 22-24 h at 37°C	Modified RV broth (42°C) and tetrathionate broth (35°C) for 24 h	XLT4[k] for 24 h at 37°C	TSI[l] and LI[m] slants then *Salmonella* polyvalent O (poly A-1, Vi) and polyvalent H sera (Difco)	ND	None
Fabrizio and Cutter (18)	Lactose broth at 35°C for 24 h	Selenite cysteine and tetrathionate broth at 35°C for 24 h	XLD for 48 h at 35°C	Oxoid *Salmonella* latex test	Spiral plating in duplicate on XLD then enumerated manually or with Q-count image analyser (Advanced Instruments)	None
Eggenberger-Solorzano et al. (12) Commercial	BPW for 24 h, temp NR	Tetrathionate broth, time and temp NR	XLD, time and temp NR	TSI and LI slants	ND	Presumptive *Salmonella* determined using the Organon Teknika *Salmonella* ELISA

[k] XLT4 = Xylose lysine tergitol 4

[l] TSI = Triple sugar iron

[m] LI = Lysine iron

Eggenberger-Solorzano et al. (12) Laboratory[n]	NR	NR	Violet red bile glucose agar for 24 h at 37°C	ND	Spiral plater (Spiral Biotech)	None
Epling et al. (11)	ND	Brilliant green tetrathionate broth for 24 h at 42°C	BGA for 24 h at 37°C	Colonies positive on TSI slants tested serologically and biochemically (Analytab Products)	ND	None
Biemuller et al. (19) (both pilot plant studies)	ND	Brilliant green tetrathionate broth for 24 h at 37°C	BGA and bismuth sulphite agar for 24 h at 37°C	TSI slants (37°C. 24 h) then forwarded to the Southeastern *Salmonella* Serotyping Laboratory, Atlanta, for identification	ND	None
Morris et al. (15)	ND	ND	Rif-TSA[o] (24 h at 37°C)	ND	NR	None
van Netten et al. (13)		B-TSBY + equal vol of double strength Muller-Kauffamnn broth for		Subcultured on XLD-N and identified	Counts determined after catalase-mediated solid	None

[n] For this lab study, the authors tested for Enterobacteriaceae, but they knew that these were only *Salmonella* because they had previously killed off all the Enterobacteriacia already present via irradiation.

[o] Tryptic soy agar

	B-TSBY[p] (6 h at 25°C)	24 h for XLD samples and 48 h for BGA samples, both at 42°C	XLD-N[q] and BGA for 24 h at 37°C	biochemically and serologically	medium repair and after XLD-N overlay was applied	
Morild et al. (14)	ND	ND	XLD for 20-22 h at 37°C	ND	Red colonies (± black centers) counted on XLD	None
Morild et al. (20)	ND	ND	XLD at 37°C, time NR	NR	NR	None
Christiansen et al. (17)	ND	ND	XLD for 24 h at 37°C	ND	NR	None
van Netten et al. (16)	NR	NR	TSBYA-C[r] agar for 24 h at 37°C	ND	Two replicate plates (range > 7 to < 100 colonies) were counted.	None
Epling (5)	ND	TSA (for injured cells), 4 h at 37°C	BGA 24 h (for uninjured cells) or TSA		Serology (polyvalent and group sera) and biochemistry (API strips)	None

[p] BTSBY = Buffered trypticase soy broth with yeast extract

[q]XLD-N = Xylose lysine desoxycholate agar with novobiocin

[r] The authors did not report what the "C" stood for.

		overlaid with 10-12 mL BGA for 20 h (for injured cells) both at 37^{o}C	

111

112 **TABLE 6** Outcomes of individual studies included in a systematic review of pathogen reduction treatments against *Salmonella* on
113 pork carcasses or parts of pork carcasses

Source	Intervention information	Sample type	Prevalence after treatment	N (concn)	Concn	P-value
Trivedi et al. (10)	Final wash[a] (control)	Swab	4[b]/216[c]	NA[d]	ND[e]	ND
	Final wash then steam (80-85ºC, 60 s)		0/216	NA	ND	
Morild et al. (14)	Steam U/S[f] for 0.5s 10^4 CFU/cm^2 inoc[g]	Not reported	ND	6[h]	1.1 ± 0.21[i]	See text
	Steam U/S for 0.5s 10^7 CFU/cm^2 inoc			6	0.6 ± 0.22	
	Steam U/S for 1.0s 10^4 CFU/cm^2 inoc			6	2.2 ± 0.37	
	Steam U/S for 1.0s 10^7 CFU/cm^2 inoc			6	2.1 ± 0.34	

[a] As this study took place at a US commercial abattoir, the final wash probably consisted of water (J. Dickson, pers. comm., 13 April 2015). Water temp, pressure and pH not reported.

[b] This refers to the number of positive samples. The number of positive carcass halves was not reported.

[c] 72 carcass halves, 3 locations per carcass half (ham, belly, and jowl) = 216 samples

[d] Not applicable

[e] Not done

[f] Ultrasound

[g] Inoculum

[h] Number of replicates. Actual number of samples not reported.

[i] Reduction in *S.* Typhimurium ± SEM log CFU/cm^2

	Steam U/S for 1.5s 10^4 CFU/cm^2 inoc			6	2.8 $\pm$ 0.30	
	Steam U/S for 1.5s 10^7 CFU/cm^2 inoc			6	2.3 $\pm$ 0.30	
	Steam U/S for 2.0s 10^4 CFU/cm^2 inoc			6	2.8 $\pm$ 0.24	
	Steam U/S for 2.0s 10^7 CFU/cm^2 inoc			6	3.2 $\pm$ 0.40	
Morris et al. (15)	5s 0% TSP[j] (pH 6.6) immersion	10 cm^2 X 2 mm skin samples	ND	3	4.6[k]	See text
	10s 0% TSP (pH 6.6) immersion		ND	3	4.6	
	15s 0% TSP (pH 6.6) immersion		ND	3	4.4	
	5s 4% TSP (pH 12.5) immersion		ND	3	3.4	
	10s 4% TSP (pH 12.5) immersion		ND	3	3.5	
	15s 4% TSP (pH 12.5) immersion		ND	3	2.9	
	5s 8% TSP (pH 13.1) immersion		ND	3	2.5	
	10s 8% TSP (pH 13.1) immersion		ND	3	2.6	
	15s 8% TSP (pH 13.1) immersion		ND	3	3.1	
	5s 12% TSP (pH 13.2) immersion		ND	3	2.9	
	10s 12% TSP (pH 13.2) immersion		ND	3	2.7	
	15s 12% TSP (pH 13.2) immersion		ND	3	3.0	

[j] TSP = Trisodium phosphate (AvGARD™)

[k] Mean *Salmonella* counts (log_{10}/cm^2). Measure of precision not reported.

van Netten et al. (16)	2% LA[l] and skin vortexed[m] 30 s	Skin suspension	ND	NR[n]	0.6[o] ± 0.2	See text
	2% LA and skin vortexed 90 s		ND	NR	1.9 ± 0.2	
Christiansen et al. (17)	2.5% LA 80°C 15 s	10 X 10 cm skin sample	ND	3[p]	5.8[q] ± 0.01	ND
	2.5% LA 80°C 5 s		ND	3	3.3 ± 0.6	
	2.5% LA 55°C 5 s		ND	3	1.6 ± 0.01	
	2.5% LA 55°C 15 s		ND	3	2.4 ± 0.1	
	1% LA 80°C 5 s		ND	3	2.5 ± 0.3	
	1% LA 80°C 15 s		ND	3	4.7 ± 0.5	
	80°C sterile water 5 s		ND	3	2.9 ± 0.1	
	80°C sterile water 15 s		ND	3	3.3 ± 0.7	
	1% LA 55°C 5 s		ND	3	1.5 ± 0.5	
	1% LA 55°C 15 s		ND	3	1.7 ± 0.2	

[l] LA = lactic acid

[m] Speed: 200 rev/min

[n] NR = not reported

[o] Immediate lethality (mean reduction during the lactic acid treatment) ± SD (log_{10} CFU/ml). A quenching solution was used to neutralize lactic acid at the end of treatment.

[p] Number of replicates = 3. Number of samples not specified.

[q] Mean reduction in *S.* Typhimurium ± SEM

Epling et al. (5)	ww[r] then 5 min 2% LA spray, shoulder	Carcass swab	2/75	ND	ND	See text
	ww then 5 min 2% LA spray, ham		3/75	ND	ND	
	ww, 5 min no spray (control), shoulder		8/75	ND	ND	
	ww, 5 min no spray (control), ham		9/75	ND	ND	
	ww, 24 h no spray (control), ham		9/75	ND	ND	
	ww, 24 h no spray (control), shoulder		12/75	ND	ND	
	ww, 24 h 2% LA spray, ham		1/75	ND	ND	
	ww, 24 h 2% LA spray, shoulder		0/75	ND	ND	
Machado et al. (8)	Physiological saline, 5 s dip	Skin swab	7/8[t]	8	0.7[u]	> 0.05[v]
	Organic acid[s] (1000 ppm) 5 s dip		4/8	8	1.2	
	Steam spray (4 BAR, 140^{o}C, 15 s)		6/8	8	0.8	
	Organic acid (immerse) + steam spray		10/10	10	0.6	

[r] ww = since all carcasses (controls and treated carcasses) underwent conventional slaughter in a US commercial abattoir, all carcasses would have received a water wash (temp, pressure and pH of the water not reported) prior to any treatment (J. Dickson, pers. comm., 13 April 2015)

[s] Citric acid was the main constituent (other constituents not reported).

[t] Number of samples showing reduction in the count of the Most Probable Number of *Salmonella* following treatment (samples with counts below 3MPN were withdrawn)

[u] Mean MPN of *Salmonella* sp. after treatment (It was not possible to extract the SEM from this paper as the data were presented in a figure and the error bars of the different treatments overlapped too much to distinguish.)

[v] For the prevalence data, none of the treatments differed significantly from the control (physiological saline) group.

Hamilton et al. (9)	SHS (control)[w] SHS+hot water 15s, 81.9°C 2.67L/s rinse SHS SANOVA™[x] (15s, 0.27L/s) spray	1-2 cm belly strip excision including skin, muscle, fat and peritoneum	24/150 4/150 7/100	ND	ND	<0.001[y]
Fabrizio and Cutter (18)	Untreated Distiller pH 7.81, 15 s spray Sodium hypochlorite[z] pH 7.82, 15 s spray 2% LA pH 2.33, 15 s spray EO[aa] water, pH 2.79, 15 s spray EO water (aged)[bb] pH 2.84, 15 s spray	25 cm^2 X 0.5 cm thick excised belly skin sample	ND ND ND ND ND ND	4 4 4 4 4 4	6.27 ± 0.09[cc] 4.91 ± 0.36 4.89 ± 0.25 4.48 ± 0.32 4.60 ± 0.74 4.72 ± 0.77	See Text

[w] SHS= standard hygienic slaughter. Although not specified in the paper, we assumed this entailed a water wash at the end of slaughter.

[x] Acidified sodium chlorite (pH 2.4-2.6), ECOLAB Inc.

[y] Both treatments were significantly different from the control group. Hot water and SANOVA™ were not significantly different from each other (p = 0.12).

[z] 19.9 ppm free chlorine

[aa] Electrolyzed oxidizing water (68.25 ppm free chlorine)

[bb] 66 ppm free chlorine

[cc] Mean $\log_{10}$ CFU/cm^2 *S.* Typhimurium immediately following treatment ± mean square error (variance). Initial pathogen level for all treatments was approx 6 $\log_{10}$ CFU/cm^2.

Eggenberger-Solarzano et al. (12) Lab Study	25°C water, 0 s vortex[dd]	Excised skin sample	ND	NR	2.2[ee]	See text
	25°C water, 5 s vortex		ND	NR	0.9	
	25°C water, 10 s vortex		ND	NR	1.0	
	25°C water, 15 s vortex		ND	NR	0.9	
	55°C water, 0 s vortex		ND	NR	2.5	
	55°C water, 5 s vortex		ND	NR	1.0	
	55°C water, 10 s vortex		ND	NR	0.9	
	55°C water, 15 s vortex		ND	NR	1.0	
	65°C water, 0 s vortex		ND	NR	2.4	
	65°C water, 5 s vortex		ND	NR	1.0	
	65°C water, 10 s vortex		ND	NR	1.0	
	65°C water, 15 s vortex		ND	NR	1.0	
	80°C water, 0 s vortex		ND	NR	2.2	
	80°C water, 5 s vortex		ND	NR	1.0	
	80°C water, 10 s vortex		ND	NR	1.0	
	80°C water, 15 s vortex		ND	NR	1.0	
Eggenberger-Solorzano et al. (12) Commercial study	No treatment (control)[ff]	Jowl swab	4/60	ND	ND	ND
	1.8% AA[gg] spray, 3 s		0/30	ND	ND	
	74°C water spray, 5 s		2/30	ND	ND	
	74°C water spray 5 s then AA spray 3 s		1/30	ND	ND	
Biemuller et al. (19) First Pilot	AA pH 2.0 spray for 10-12 s	Carcass swab	11/72	NA	NA	NR
	AA pH 1.5, spray for 10-12 s		1/6	ND	ND	

[dd] Samples were placed in water in a centrifuge tube and vortexed for 0 s.

[ee] Log_{10} CFU/cm^2 *Salmonella* present after treatment (measure of precision not reported)

[ff] The jowl was scalded prior to inoculation with the challenge organism.

[gg] AA = acetic acid

Plant Study	5% stannous chloride spray for 10-12 s		1/6	ND	ND	
	5% H_2O_2 spray for 10-12 s		2/6	ND	ND	
	Steam (10 s or 30 s?)[hh]		0/6	ND	ND	
Biemuller et al. (19) Second Pilot Plant Study	Inoc[ii] 1 h[jj]	Carcass swab	6/6	ND	ND	NR
	Inoc 24 h[kk]		4/6	ND	ND	
	Inoc ww[ll] 1h		6/6	ND	ND	
	Inoc ww 24 h		4/6	ND	ND	
	Inoc 30 s AA spray, 1h		1/6	ND	ND	
	Inoc 30 s AA spray 24 h		0/6	ND	ND	
	Inoc 30 s AA spray, ww, 1 h		6/6	ND	ND	
	Inoc 30 s AA spray, ww, 24 h		2/6	ND	ND	
	Inoc 60 s AA spray, 1 h		2/6	ND	ND	
	Inoc 60 s AA spray 24 h		2/6	ND	ND	
	Inoc 60 s AA spray, ww, 1h		2/6	ND	ND	
	Inoc 60 s AA spray, ww, 24 h		2/6	ND	ND	
	Nat[mm] ww, 1 h		1/6	ND	ND	
	Nat ww, 24 h		1/6	ND	ND	

[hh] The authors' paper specifies 10 s in the Results section and 30 s in the Methods section.

[ii] Inoculated with *Salmonella*

[jj] Outcome measured after 1 h

[kk] Outcome measured after 24 h

[ll] ww = The carcass was washed with water (duration, temp and pressure not reported). If carcass was sprayed with acetic acid, the wash occurred afterwards.

[mm] Naturally infected

	Nat 30 s AA spray, ww, 1 h		0/6	ND	ND	
	Nat 30 s AA spray, ww, 24 h		0/6	ND	ND	
	Nat 60 s AA spray, ww, 1 h		0/6	ND	ND	
	Nat 60 s AA spray, ww, 24 h		0/6	ND	ND	
van Netten et al. (13)	Inoc[nn] 11°C water spray 60 s	5 cm^2 cheek, back, and belly tissue excision	9/9	9	NR	ND
	Inoc 2% 11°C LA spray 60 s		5/9	9	NR	
	Inoc 5% 11°C LA spray 60 s		3/9	9	NR	
	Inoc 55°C water spray 120 s		15/15	15	NR	
	Inoc 55°C 2% LA spray 30 s		6/15	15	NR	
	Inoc 55°C 2% LA spray 60 s		0/15	15	NR	
	Inoc 55°C 2% LA spray 90 s		0/15	15	NR	
	Inoc 55°C 2% LA spray 120 s		0/15	15	NR	
	Inoc 55°C 5% LA spray 30 s		3/15	15	NR	
	Inoc 55°C 5% LA spray 60 s		0/15	15	NR	
	Inoc 55°C 5% LA spray 90 s		0/15	15	NR	
	Inoc 55°C 5% LA spray 120 s		0/15	15	NR	
	Fec[oo] 11°C water spray 30 s		NA	5[pp]	0.1 $\pm$ 0[qq]	
	Fec 11°C water spray 60 s		NA	5	0.2 $\pm$ 0	
	Fec 55°C water spray 30 s		NA	5	0.1 $\pm$ 0	
	Fec 55°C water spray 120 s		NA	5	0.3 $\pm$ 0.1	

[nn] Inoculated prior to treatment for a contamination level of 1.7 $\pm$ 0.2 (SD) $\log_{10}$ CFU/cm^2

[oo] Samples were contaminated using feces. Final level of *Salmonella* contamination before treatment not reported.

[pp] 5 expts were conducted. It is unclear if the 3 samples taken per carcass were pooled.

[qq] Mean rinse-off ($\log_{10}/cm^2$ $\pm$ SD)

Morild et al. (20)	15 s water rinse (control)	Swab	ND	3[ss]	6.0 ± 0.25[uu]	See text
	15 s water rinse (control)	Skin	ND	3	6.1 ± 0.16	
	15 s water rinse (control)	Swab + skin	ND	3	6.4 ± 0.19	
	80°C water rinse 5 or 15 s[rr]	Swab	ND	6[tt]	6.8 ± 0.05	
	80°C water rinse 5 or 15 s	Skin	ND	6	6.5 ± 0.06	
	80°C water rinse 5 or 15 s	Swab + skin	ND	6	6.99 ± 0.04	
	55°C 1% LA rinse 5 or 15 s	Swab	ND	6	6.8 ± 0.16	
	55°C 1% LA rinse 5 or 15 s	Skin	ND	6	6.2 ± 0.17	
	55°C 1% LA rinse 5 or 15 s	Swab + skin	ND	6	6.9 ± 0.16	

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[rr] Each treatment was followed 2 min later by inoculation with bacteria followed 2 min later by a 15-s water rinse.

[ss] Number of replicates.

[tt] 6 replicates, since the 5-s (3 replicates) and 15-s (3 replicates) treatment results were pooled because they weren't significantly different.

[uu] Number of *S.* Typhimurium (log_{10} CFU/cm^2) ± SEM remaining after treatment.

Figures

FIG 1 PRISMA (Preferred Reporting Items or Systematic Reviews and Meta-Analyses) flow diagram for a systematic review of pathogen reduction sprays/rinses/washes for pork carcasses and carcass parts (template from Mohr et al. (24))

FIG 2 Risk-of–bias-summary graph for a systematic review of pathogen reduction treatments against *Salmonella* in pork carcasses. Red circles refer to a high risk of bias, green circles to a low risk of bias, and yellow circles to an unclear risk of bias.

FIG 3 Forest plot showing measures of *Salmonella* concentration from intervention studies that assessed lactic acid washes in commercial abattoirs. Standardized mean difference is used as the summary effect measure as the metrics for *Salmonella* were not consistent across studies. These data represent all possible comparisons, so control groups appear multiple times and summary effects are invalid. "?C" indicates that the temperature of the solution used to wash the pork was not reported.

FIG 4 Forest plot showing prevalence of *Salmonella* for interventions that compared variations of water/steam with standard/controls. These data represent all possible comparisons so control groups appear multiple times and summary effects are invalid.

FIG 5 Forest plot showing the prevalence of *Salmonella* for interventions that compared variations of acidic interventions with standard/controls. These data represent all possible comparisons**,** so control groups appear multiple times and summary effects are invalid (and therefore not shown in the figure). LA stands for lactic acid. The Machado et al. (8) study reported the outcome as the percentage of samples that showed reduction in the count of the Most Probable Number of *Salmonella* after treatment.

Figures

2

3**PRISMA 2009 Flow Diagram**

4

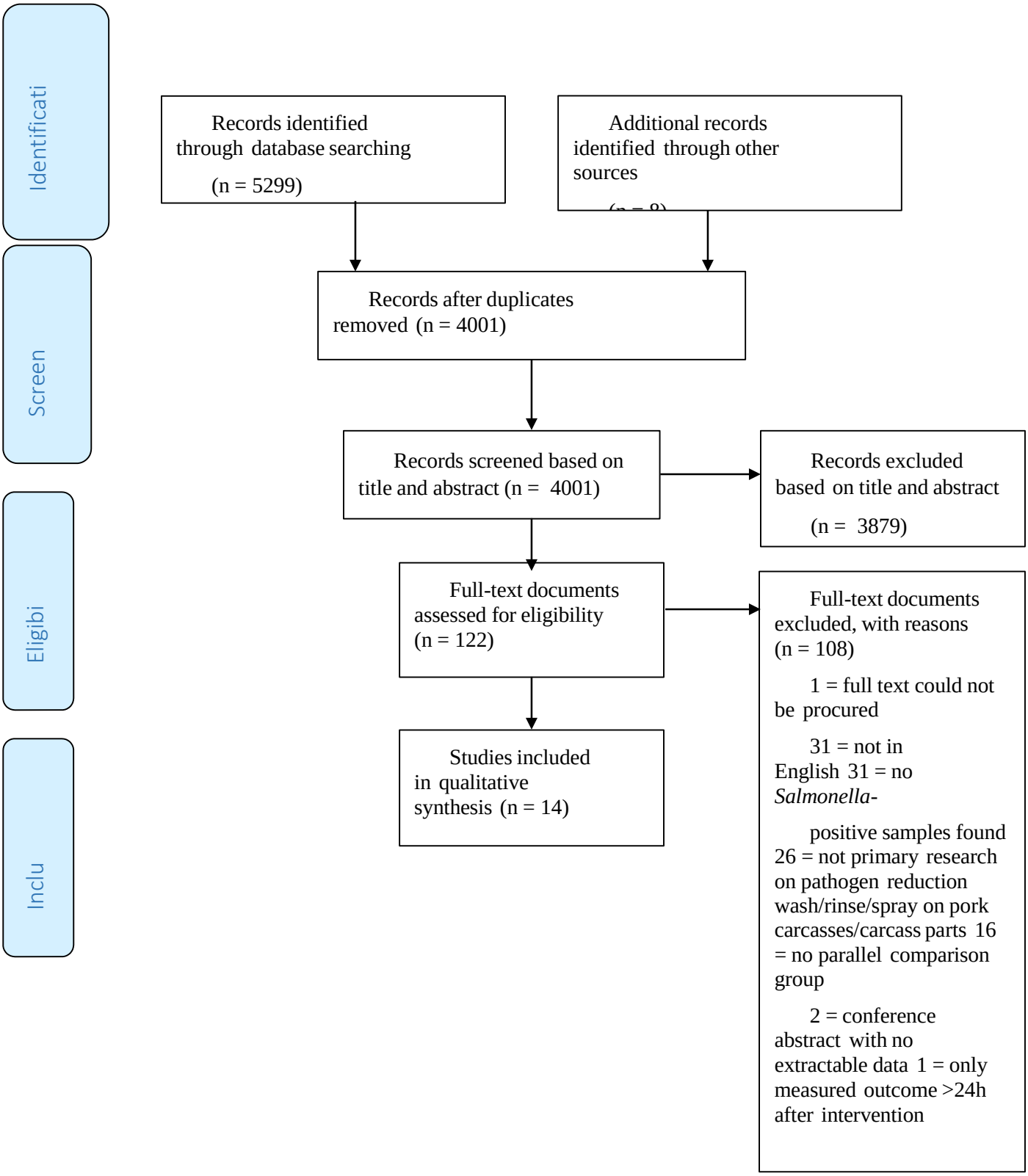


FIG 1 PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flow diagram for a systematic review of pathogen reduction sprays/rinses/washes for pork carcasses and carcass parts (template from Mohr et al. (22))

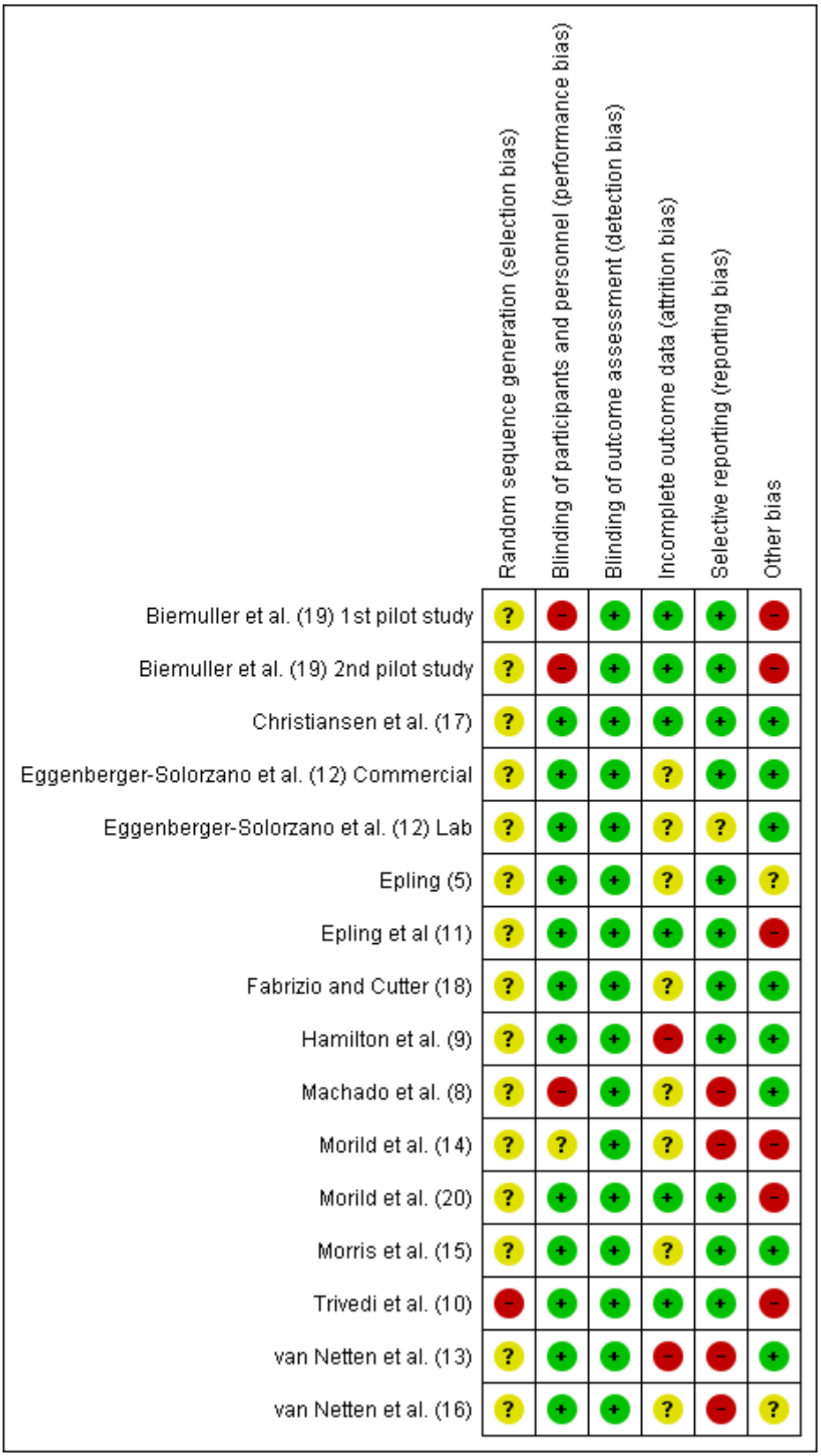
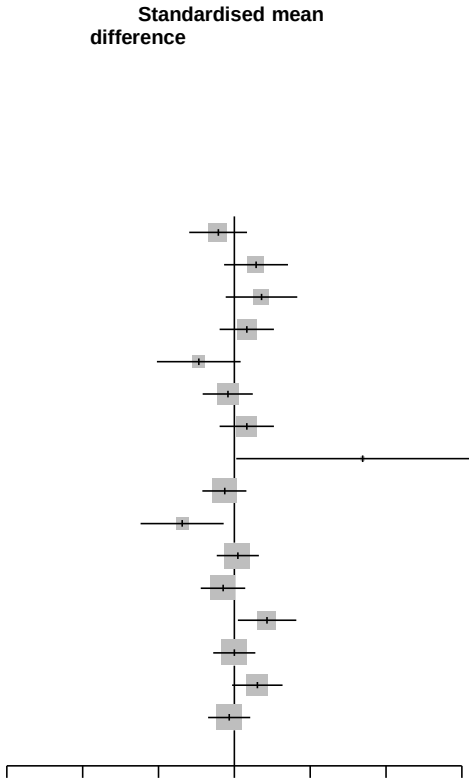

Random sequence generation (selection bias)
Blinding of participants and personnel (performance bias)
Blinding of outcome assessment (detection bias)
Incomplete outcome data (attrition bias)
Selective reporting (reporting bias)
Other bias
Biemuller et al. (19) 1st pilot study
Biemuller et al. (19) 2nd pilot study
Christiansen et al. (17)
Eggenberger-Solorzano et al. (12) Commercial
Eggenberger-Solorzano et al. (12) Lab
Epling (5)
Epling et al (11)
Fabrizio and Cutter (18)
Hamilton et al. (9)
Machado et al. (8)
Morild et al. (14)
Morild et al. (20)
Morris et al. (15)
Trivedi et al. (10)
van Netten et al. (13)
van Netten et al. (16)

FIG 2 Risk-of–bias-summary graph for a systematic review of pathogen reduction treatments against *Salmonella* in pork carcasses. Red circles refer to a high risk of bias, green circles to a low risk of bias, and yellow circles to an unclear risk of bias.

Name	Outcome	Lactic.Acid	Water	SMD 95%-CI
Christiansen (17)	Mean log10 reduction	1% -80C-15 sec	80C 15 sec	-1.06 [-2.96; 0.84]
Christiansen (17)	Mean log10 reduction	1%-55C-15sec	80C 15 sec	1.44 [-0.68; 3.55]
Christiansen (17)	Mean log10 reduction	1%-55C-5sec	80C 5 sec	1.79 [-0.56; 4.15]
Christiansen (17)	Mean log10 reduction	1%-80C- 5 sec	80C 5 sec	0.83 [-0.96; 2.61]
Christiansen (17)	Mean log10 reduction	2.5% -80C-15 sec	80C 15 sec	-2.33 [-5.10; 0.43]
Christiansen (17)	Mean log10 reduction	2.5%- 80C-5 sec	80C 5 sec	-0.43 [-2.08; 1.22]
Christiansen (17)	Mean log10 reduction	2.5%-55C -15sec	80C 15 sec	0.83 [-0.96; 2.62]
Christiansen (17)	Mean log10 reduction	2.5%-55C -5sec	80C 5 sec	8.46 [0.14; 16.78]
Fabrizio (18)	Mean log10 CFU/cm2	2%-?C-15sec	?C-15sec	-0.64 [-2.10; 0.81]
Fabrizio (18)	Mean log10 CFU/cm2	2%-?C-15sec	Untreated	-3.44 [-6.18; -0.70]
Fabrizio (18)	Mean log10 CFU/cm2 (skin)	1%-55C-5 or 15 sec	?C-15 sec	0.23 [-1.16; 1.63]
Morild (20)	Mean log10 CFU/cm2 (skin)	1%-55C-5 or 15 sec	80C-5 or 15 sec	-0.74 [-2.20; 0.72]
Morild (20)	Morild (20)	Mean log10 CFU/cm2 (swab)		Mean log10 CFU/cm2 (swab)



	1%-55C-5 or 15 sec	?C-15 sec	2.16 [0.24; 4.08]			
	1%-55C-5 or 15 sec	80C-5 or 15 sec	0.00 [-1.39; 1.39]			
Morild (20)	Mean log10 CFU/cm2 (skin + swab)	1%-55C-5 or 15 sec	?C-15 sec			1.52 [-0.15; 3.19]
Morild (20)	Mean log10 CFU/cm2 (skin + swab)	1%-55C-5 or 15 sec	80C-5 or 15 sec			-0.34 [-1.74; 1.06]

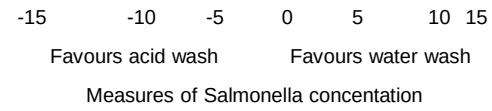


FIG 3 Forest plot showing measures of *Salmonella* concentration from intervention studies that assessed lactic acid washes in commercial abattoirs. Standardized mean difference is used as the summary effect measure as the metrics for *Salmonella* were not consistent across studies. These data represent all possible comparisons, so control groups appear multiple times and summary effects are invalid. "?C" indicates that the temperature of the solution used to wash the pork was not reported.

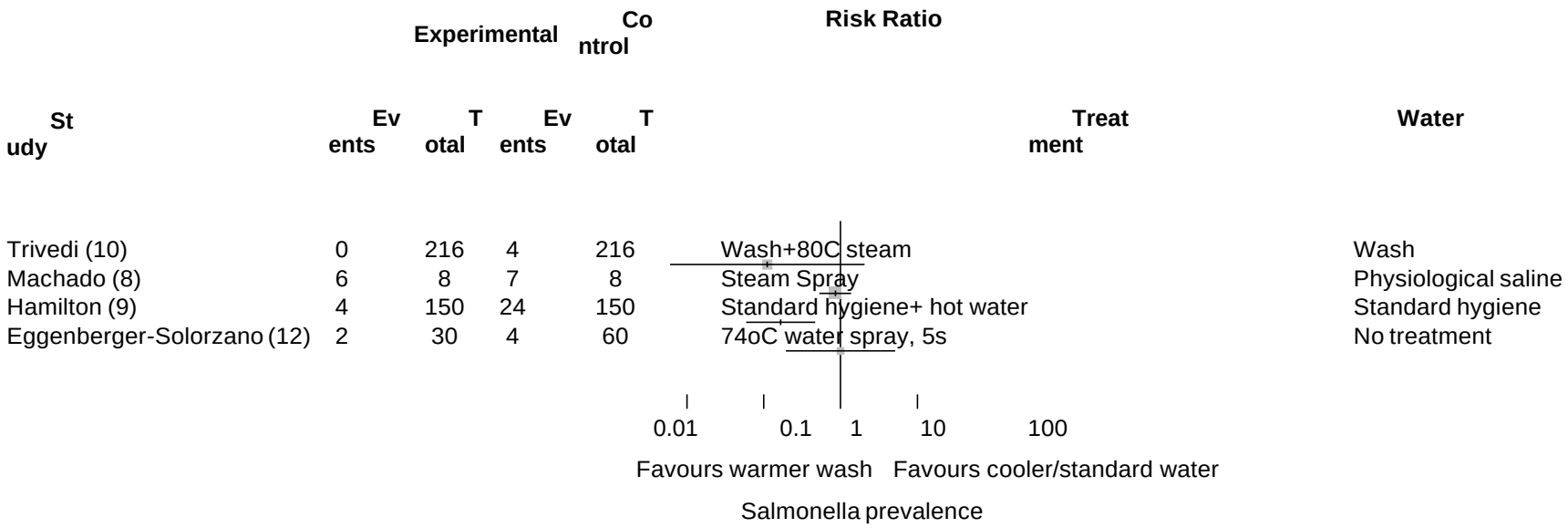
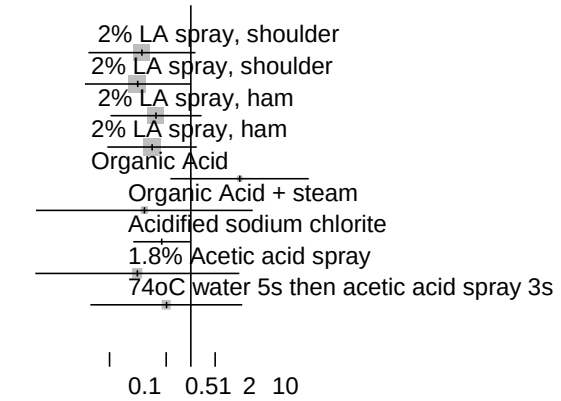

Experimental
Control
Risk Ratio
Study
Events
Total
Events
Total
Treatment
Water
Trivedi (10) 0 216 4 216 Wash+80C steam Wash
Machado (8) 6 8 7 8 Steam Spray Physiological saline
Hamilton (9) 4 150 24 150 Standard hygiene+ hot water Standard hygiene
Eggenberger-Solorzano (12) 2 30 4 60 74oC water spray, 5s No treatment
0.01
0.1
1
10
100
Favours warmer wash
Favours cooler/standard water
Salmonella prevalence

FIG 4 Forest plot showing prevalence of *Salmonella* for interventions that compared variations of water/steam with standard/controls. These data represent all possible comparisons so control groups appear multiple times and summary effects are invalid.

Study	Experimental Events	Experimental Total	Control Events	Control Total	Risk Ratio	Treatment	Water
Epling (11)	2	75	8	75		2% LA spray, shoulder	Washed sholder
Epling (11)	2	75	9	75		2% LA spray, shoulder	Washed ham
Epling (11)	3	75	8	75		2% LA spray, ham	Washed sholder
Epling (11)	3	75	9	75		2% LA spray, ham	Washed ham
Machado (8)	4	8	1	8		Organic Acid	Physiological saline
Machado (8)	0	10	1	8		Organic Acid + steam	Physiological saline
Hamilton (9)	7	100	24	150		Acidified sodium chlorite	Standard hygiene
Eggenberger-Solorzano (12)	0	30	4	60		1.8% Acetic acid spray	No treatment
Eggenberger-Solorzano (12)	1	30	4	60		74oC water 5s then acetic acid spray 3s	No treatment



Salmonella prevalence

FIG 5 Forest plot showing the prevalence of *Salmonella* for interventions that compared variations of acidic interventions with standard/controls. These data represent all possible comparisons**,** so control groups appear multiple times and summary effects are invalid (and therefore not shown in the figure). LA stands for lactic acid. The Machado et al. (8) study reported the outcome as the percentage of samples that showed reduction in the count of the Most Probable Number of *Salmonella* after treatment.

Supplemental Material

TABLE S1 Initial (not final) search in CABI Abstracts for a systematic review of pathogen reduction treatments against *Salmonella* on pork carcasses (additional terms highlighted in green)
[sss]

Search no.	No. hits	Search string
#1	25,552	TS=((pork or swine or pig or pigs or hog or hogs or boar or boars or sow or sows) near/7 (carcass* OR slaughter* or abattoir* or bellies)) Index=CAB Abstracts Timespan=All years
#2	7614	TS=(pathogen near/4 reduc*) OR TS=prt Index=CAB Abstracts Timespan=All years
#3	57,209	TS=(wash or washes or washing or washed or rinse or rinses or rinsing or rinsed) Index CAB Abstracts Timespan=All years
#4	192,274	TS=(spray or sprays or spraying or sprayed) Index=CAB Abstracts Timespan=All years
#5	157	TS=(Organic NEAR/5 (decontaminat* or saniti*)) Index=CAB Abstracts Timespan=All years
#6	46,191	TS=(PEROXYACETIC OR LACTIC) Index=CAB Abstracts Timespan=All years

[sss] Initially, a more detailed search strategy was used to search CABI Abstracts, which included the additional search terms: Topic field tag (TS) = contaminat*, decontaminat*, "food sanitation", antibacterial$ or disinfect*, "heat treatment" and peracetic. This search produced 3842 records. An abbreviated search that did not use those terms returned 1943 records. The additional records found using the full search but not by the abbreviated search were sorted by relevance and the first 300 of these were screened. None of these records was found to be relevant, so it was decided to use the abbreviated search in CAB Abstracts as the final search strategy.

#7	57,823	TS=(ACETIC OR hypobromous or citric or "mineral acid$" or peracetic)
#8	6609	TS=((HYDROCHLORIC OR NITRIC OR PHOSPHORIC OR ACID) NEAR/5 (spray* or decontaminat* or saniti* or wash*)) Index=CAB Abstracts Timespan=All years
#9	134	TS=NONACID Index=CAB Abstracts Timespan=All years
#10	50,030	TS=((hot or cold or electrolyzed or electrolysed or warm) NEAR/3 water) OR TS=("heat treatment") Index=CAB Abstracts Timespan=All years
#11	70,878	ts="water treatment$" Index=CAB Abstracts Timespan=All years
#12	24,497	TS=steam Index=CAB Abstracts Timespan=All years
#13	70	TS="AQUEOUS OZONE" Index=CAB Abstracts Timespan=All years
#14	2975	TS=("POTASSIUM HYDROXIDE" OR "POTASSIUM SORBATE") Index=CAB Abstracts Timespan=All years
#15	10,343	ts=("sodium hypochlorite" OR NaClO or "sodium acetate" or "sodium citrate" or "sodium chlorite" or "sodium lactate") Index=CAB Abstracts Timespan=All years
#16	232,715	ts=(TSP or phosphate$ or antibacterial$ or disinfect*) Index=CAB Abstracts Timespan=All years
#17	139,833	TS=(CHLORINE OR ALCIDE OR ULTRAVIOLET OR UV OR IRRADIAT* OR "DRY HEAT" OR ULTRASOUND) Index=CAB Abstracts Timespan=All years
#18	264,693	TS=(contaminat* or decontaminat*) OR TS=("food sanitation") Index=CAB Abstracts Timespan=All years
#19	34,856	TS=(Chilling or "freezing air" or "high air velocity" or blasting) Index=CAB Abstracts Timespan=All years
#20	1,050,577	#19 OR #18 OR #17 OR #16 OR #15 OR #14 OR #13 OR #12 OR #11 OR #10 OR #9 OR #8 OR #7 OR #6 OR #5 OR #4 OR #3 OR #2 Index=CAB Abstracts Timespan=All years
#21	3476	#20 AND #1

		Index=CAB Abstracts Timespan=All years[ttt]
#22	473	TI=((DECONTAMINAT* OR CONTAMINAT*) AND CARCASS*) Index=CAB Abstracts Timespan=All years[uuu]
#23	3842	#22 OR #21[vvv] Index=CAB Abstracts Timespan=All years

15
16

[ttt] Pork carcasses, etc. AND decontamination techniques (title or abstract)

[uuu] Title words only decontamination of carcasses

[vvv] Both options OR'd

TABLE S2 Search conducted in SCI-EXPANDED and CPCI-S on 25 January 2015 for a systematic review of pathogen reduction treatments of pork carcasses against *Salmonella* (Timespan=1900-2015, searched on Web of Science)

Search no.	No. hits	Search string
# 1	9258	TS=((pork or swine or pig or pigs or hog or hogs or boar or boars or sow or sows) near/7 (carcass* OR slaughter* or abattoir* or bellies)) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 2	5338	TS=(pathogen near/4 reduc*) OR TS=prt Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 3	79,602	TS=(wash or washes or washing or washed or rinse or rinses or rinsing or rinsed) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 4	100,629	TS=(spray or sprays or spraying or sprayed) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 5	230	TS=(Organic NEAR/5 (decontaminat* or saniti*)) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 6	64,083	TS=(PEROXYACETIC OR LACTIC) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 7	96,531	TS=(ACETIC OR hypobromous or citric or "mineral acid$") Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 8	5351	TS=((HYDROCHLORIC OR NITRIC OR PHOSPHORIC OR ACID) NEAR/5 (spray* or decontaminat* or saniti* or wash*)) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 9	461	TS=NONACID Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 10	37,518	TS=((hot or cold or electrolyzed or electrolysed or warm) NEAR/3 water) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 11	34,935	ts="water treatment$" Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 12	61,104	TS=steam Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015

# 13	237	TS="AQUEOUS OZONE" Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 14	5538	TS=("POTASSIUM HYDROXIDE" OR "POTASSIUM SORBATE") Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 15	16,014	ts=("sodium hypochlorite" OR NaClO or "sodium acetate" or "sodium citrate" or "sodium chlorite" or "sodium lactate") Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 16	320,726	ts=(TSP or phosphate$) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 17	1,004,162	TS=(CHLORINE OR ALCIDE OR ULTRAVIOLET OR UV OR IRRADIAT* OR "DRY HEAT" OR ULTRASOUND) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 18	26,255	TS=((Prevent* or reduc*) near/4 contaminat*) or TS=decontaminat* Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 19	78,332	TS=(Chilling or "freezing air" or "high air velocity" or blasting) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 20	1,849,801	#19 OR #18 OR #17 OR #16 OR #15 OR #14 OR #13 OR #12 OR #11 OR #10 OR #9 OR #8 OR #7 OR #6 OR #5 OR #4 OR #3 OR #2 Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 21	780	#20 AND #1 Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 22	359	TI=((DECONTAMINAT* OR CONTAMINAT*) AND CARCASS*) Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015
# 23	1099	#22 OR #21 Indexes=SCI-EXPANDED, CPCI-S Timespan=1900-2015

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TABLE S3 Search strategy in Ovid MEDLINE® In-Process & other non-Indexed citations and Ovid MEDLINE® (1946 to Present) on Ovid conducted on 25 January 2015 for a systematic review of pathogen reduction treatments of pork carcasses against *Salmonella*

Search no.	No. hits	Search string
#1	179,296	exp Swine/
#2	4465	Abattoirs/
#3	59,989	Meat-Packing Industry/ or Food Contamination/ or Food-Processing Industry/ or Meat/ or Meat Products/
#4	8060	1 and (2 or 3)
#5	4254	((pork or swine or pig or pigs or hog or hogs or boar or boars or sow or sows) adj7 (carcass* or slaughter* or abattoir* or bellies)).ti,ab,kf.
#6	10,631	or/4-5
#7	2163	((pathogen adj4 reduc*) or prt).ti,ab,kf.
#8	65,754	(wash or washes or washing or washed or rinse or rinses or rinsing or rinsed).ti,ab,kf.
#9	9755	Disinfectants/
#10	25,086	(spray or sprays or spraying or sprayed).ti,ab,kf.
#11	45	(Organic adj5 (decontaminat* or saniti*)).ti,ab,kf.
#12	3523	Decontamination/
#13	33,240	(PEROXYACETIC or LACTIC).ti,ab,kf.
#14	8964	exp acetic acid/ or peracetic acid/
#15	32,199	lactic acid/ or sodium lactate/
#16	42,343	(ACETIC or hypobromous or citric or "mineral acid$").ti,ab,kf.
#17	9656	exp bromates/ or exp hydrobromic acid/
#18	7862	exp Citric Acid/
#19	100,388	hydrochloric acid/ or nitric acid/ or exp phosphorus acids/

#20	2128	((HYDROCHLORIC or NITRIC or PHOSPHORIC or ACID) adj5 (spray* or decontaminat* or saniti* or wash*)).ti,ab,kf.
#21	278	nonacid.ti,ab,kf.
#22	9550	((hot or cold or electrolyzed or electrolysed or warm) adj3 water).ti,ab,kf.
#23	10208	disinfection/
#24	5765	"water treatment$".ti,ab,kf.
#25	5427	steam.ti,ab,kf.
#26	1737	steam/
#27	53	"aqueous ozone".ti,ab,kf.
#28	1730	("POTASSIUM HYDROXIDE" or "POTASSIUM SORBATE").ti,ab,kf.
#29	9964	("sodium hypochlorite" or NaClO or "sodium acetate" or "sodium citrate" or "sodium chlorite" or "sodium lactate").ti,ab,kf.
#30	3520	Sodium Hypochlorite/
#31	492	Sodium Acetate/
#32	212,849	(TSP or phosphate$).ti,ab,kf.
#33	85,548	exp Phosphates/
#34	473,536	(CHLORINE or ALCIDE or ULTRAVIOLET or UV or IRRADIAT* or "DRY HEAT" or ULTRASOUND).ti,ab,kf.
#35	9187	Chlorine/
#36	64,614	Ultraviolet Rays/
#37	1604	Food Irradiation/
#38	13,907	(((Prevent* or reduc*) adj4 contaminat*) or decontaminat*).ti,ab,kf.
#39	3279	(Chilling or "freezing air" or "high air velocity" or blasting).ti,ab,kf.
#40	20,190	Freezing/

#41	60,900	exp Cold Temperature/
#42	1,059,030	or/7-41
#43	1265	6 and 42
#44	197	((DECONTAMINAT* or CONTAMINAT*) and CARCASS*).ti.
#45	1440	43 or 44

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TABLE S4 Search run in Science.gov (http://www.science.gov/scigov/) on 30 January 2015 for a systematic review of pathogen reduction treatments of pork carcasses against *Salmonella*

Sources searched within Science.gov	Search strategy
Science.gov websites	Searched all fields: (pork or swine or pig or pigs or hog or hogs or boar or boars or sow or sows) AND (wash or washing or washed or washes or rinse or rinsing or rinses or rinsed or spray or sprays or spraying or sprayed or acid or acids or decontamination or decontaminated or decontaminate or contaminants or contamination or contaminated or sanitised or sanitized or nonacid or water or steam or ozone or potassium or sodium or phosphate or chlorine or alcide or ultraviolet or irradiation or irradiated or heat or ultrasound or chilling or freezing or blasting) AND (carcass or carcasses or abattoir or abattoirs or slaughter or slaughterhouse or slaughterhouses)
AGRICOLA	
Center for Food Safety and Applied Nutrition	
Technology Transfer Automated Retrieval System	
USDA Food and Nutrition Center	
Applied Science and Technologies databases	
Biology and nature databases	
General science databases	

TABLE S5 Search strategy used for the International Conference on the Epidemiology and Control of Biological, Chemical and Physical Hazards in Pigs and Pork (1996–2012) (http://lib.dr.iastate.edu/safepork/) on 31 January 2015 for a systematic review of pathogen reduction treatments of pork carcasses against *Salmonella*

No. hits	Search string
651	Using the 'All fields' option and no date limits: wash* OR rinse* OR rinsing OR spray* OR acid OR acids Or decontaminat* OR contamina* OR saniti* OR nonacid OR water OR steam OR ozone OR potassium OR sodium OR phosphate OR chlorine OR alcide OR ultraviolet OR irradiat* OR radiation OR heat OR ultrasound OR chilling OR freezing OR blasting AND carcass* OR abattoir* OR slaughter* OR pork

TABLE S6 Study-level information form used in the data extraction phase of a systematic review of pathogen reduction treatments of pork carcasses against *Salmonella*

Study-Level Information Form		
Question	Style	Response
Q1. In what year(s) was the data collected?	Radio	Specify year or Not Reported. Do not use publication date to answer this question.
Q2. In what country was the study carried out?	Checkbox	(Select all that apply) List of countries with option for the reviewer to add to the list as needed. Not Reported
Q3. In what setting was the study conducted?	Radio	Commercial Abattoir University/Research Slaughter Plant Laboratory Small Holder Slaughter (Exclude) Not Reported
Q4. If the study was conducted in a commercial abattoir, was the slaughter capacity reported?	Radio (only visible if Q3 = commercial)	Yes: Enter number No
Q5. If the slaughter capacity was reported, what were the units?	Text (only visible if Q3 = commercial)	Specify the units
Q6. If the study was conducted in a commercial abattoir, what was the total number of slaughter plants?	Radio (only visible if Q3 = commercial	Specify number or Not Reported
Q7. What was the experimental unit in this study (i.e. the unit to which the intervention was assigned)?	Checkbox	Select all that apply Carcass Pork belly Skin (Option for reviewer to add items to this list as needed)
Q8. What was the weight of the experimental units in this study?	Radio	Specify weight in kg Not Reported
Q9. What was the descriptor the weight of the experimental units?	Radio	Mean Median Range

		Not Reported Not Applicable
Q10. What was the dispersion of the experimental unit weight?	Radio	Specify a number or Not Reported
Q11. What was the dispersion descriptor for the experimental unit's weight?	Radio	SD SEM Not Reported Not Applicable
Q12. Did the investigators artificially contaminate the experimental units?	Radio	Yes No Can't Tell
Q13. What was the concentration of *Salmonella* applied to the experimental unit?	Radio (only visible if Q12 = yes)	Specify number Not Reported
Q14. What were the units of the concentration of *Salmonella* applied to each experimental unit?	Radio (only visible if Q12 = yes)	Specify units Not Reported
Q15. What was the contact time (i.e. the number of seconds that the *Salmonella* was kept on the experimental unit)?	Radio (only visible if Q12 = yes)	Specify time (in seconds) Not Reported
Q16. What serotype of *Salmonella* was inoculated onto the experimental unit?	Radio (only visible if Q12 = yes)	Specify serotype(s) Not Reported
Q17. What was the method used to inoculate the experimental unit?	Radio (only visible if Q12 = yes)	Sprayed Injected Brushed on (Option for reviewer to add different methods as necessary) Not Reported
Q18. At what locations on the carcass/what parts of the experimental unit was *Salmonella* inoculated?	Checkbox (only visible if Q12 = yes)	Check all that apply Jowls Pork Belly (Option for reviewer to add new locations as necessary) Not Reported

Q19. How many times was *Salmonella* applied to the experimental unit?	Radio (only visible if Q12 = yes)	Specify number of times Not Reported
Q20. What was the time interval between inoculation and application of the intervention?	Radio (only visible if Q12 = yes)	Specify time interval (seconds) Not Reported Not Applicable (this is the control group, i.e. it didn't receive an intervention)
Q21. Which diagnostic method(s) were used to determine the prevalence or level of the bacteria?	Checkbox	Select all that apply: Bacterial culture PCR ELISA Other (specify) Not Reported
Q22. If bacterial culture was used, what was the pre-enrichment media?	Radio (only visible if Q21 = Bacterial culture)	Specify Not Reported Not Applicable (pre-enrichment not done)
Q23. If bacterial culture was used, what was the pre-enrichment incubation time?	Radio (only visible if Q21 = Bacterial culture)	Specify time (h) Not Reported Not Applicable (pre-enrichment not done)
Q24. If bacterial culture was used, what was the pre-enrichment incubation temperature?	Radio (only visible if Q21 = Bacterial culture))	Specify temperature (^{o}C) Not Reported Not Applicable (pre-enrichment not done)
Q25. If bacterial culture was used, specify the enrichment media.	Radio (only visible if Q21 = Bacterial culture)	Specify Not Reported Not Applicable (enrichment not done)
Q26. If bacterial culture was used, specify the enrichment incubation time.	Radio (only visible if Q21 = Bacterial culture)	Specify (h) Not Reported Not Applicable (enrichment not done)
Q27. If bacterial culture was used, specify the enrichment incubation temperature.	Radio (only visible if Q21 = Bacterial culture)	Specify temperature (^{o}C) Not Reported Not Applicable (enrichment not done)

Q28. If bacterial culture was used, specify the culture media.	Radio (only visible if Q21 = Bacterial culture)	Specify Not Reported
Q29. If bacterial culture was used, specify the culture incubation time.	Radio (only visible if Q21 = Bacterial culture)	Specify (h) Not Reported
Q30. If bacterial culture was used, specify the culture incubation temperature.	Radio (only visible if Q21 = Bacterial culture)	Specify (^{o}C) Not Reported
Q31. If bacterial culture was used, was a confirmation method also used?	Radio (only visible if Q21 = Bacterial culture)	Yes (specify method) No Confirmation done, but method not reported
Q32. If bacterial culture was used and concentration of bacteria was determined, what was the enumeration method used?	Radio (only visible if Q21 = Bacterial culture)	Specify method Not Reported Not Applicable (enumeration of bacteria not done)
Q33. If PCR was used to detect the bacteria, please give details of the methods (i.e. PCR type, conditions, primer name, target sequence) (copy-paste from text)	Text (only visible if Q21 = PCR)	Give details or Not Reported
Q34. If ELISA was used to detect the bacteria, please give details of the methods used. (copy-paste from text)	Text (only visible if Q21 = ELISA)	Give details or Not Reported
Q35. Are there any additional comments?	Text	Please add any additional information about the study not captured by the previous questions that you think is relevant to this review.
Q36. Specify the ARM.	Radio	Option for reviewer to name the arm (e.g. Ref ID# 40004 Pilot Slaughter Plant)

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TABLE S7 Intervention-Outcome information form used in the data extraction phase of a systematic review of pathogen reduction treatments of pork carcasses against *Salmonella*

Intervention/Outcome Form

Question	Style	Response
Q1. Description of ARM / Methods as described by authors (copy/paste from text)	Radio	Describe the intervention used and outcome(s) measured (Option for reviewer to add new ARM descriptions as needed.)
Q2. Which treatment (intervention) was applied to the experimental unit? (check all that apply for this particular ARM)	Checkbox	List of interventions with option for the reviewer to add more as necessary No treatment (control group)
Q3. Is there any additional descriptive information about this intervention (not captured by the other questions on this form) that you would like to add?	Text	
Q4. What was the pH of the intervention as it was applied to the experimental units?	Radio	Specify pH Not Reported/Not Discernible Not Applicable (if it's a no-intervention control group)
Q5. What was the concentration of the intervention?	Radio	Specify concentration (number only) Not Reported/Not Discernible Not Applicable (e.g. if it's a no-intervention control group or if it's pure water)
Q6. What were the units of the intervention concentration?	Radio	Specify units (e.g. %) Not Reported Not Applicable
Q7. What was the temperature of the intervention?	Radio	Specify temperature (number only) Not Reported Not Applicable

Q8. What were the units of the temperature of the intervention?	Radio	Specify units (^{o}C, ^{o}F) Not Reported Not Applicable
Q9. How was the intervention applied?	Radio	Spray Rinse Immersion Other (Specify) Not Reported/Not Discernible Not Applicable
Q10. At what pressure was the intervention applied?	Radio	Specify pressure (number only) Not Reported/Not Discernible Not Applicable
Q11. What units were used to describe the pressure the intervention was applied at?	Radio	Specify units Not Reported Not Applicable
Q12. At what point(s) in the processing chain was the intervention applied? Regardless of how the investigators described the data, report the data collection as occurring after the processing point. For example, if the original author described the sample as being collected pre-kill, refer to such a sample as a bleed sample, meaning the post-bleeding but pre-killing sample point.	Checkbox	Final wash, Immediately after chill, 18–48 h after chilling Stun Bleed Kill Scald Dehair Singe Polish Bung removal Evisceration Split Stamp Not Reported Not Applicable (no-treatment control group) Other (specify)
Q13. What was the total duration that the intervention was applied?	Radio	Specify the total duration (s) Not Reported Not Applicable
Q14. What was the number of times that the intervention was applied?	Radio	Specify number of times Not Reported Not Applicable

Q15. If the intervention was applied more than once, what was the time interval between the applications?	Radio	Specify time (s) Not Reported/Not Discernible Not Applicable
Q16. In which type of sample was the outcome measured?	Radio	Carcass Rinse Carcass Swab Excised Skin Sample Not Reported (Option for reviewer to add to this list as needed)
Q17. Which bacteria were measured as the outcome?	Radio	*Salmonella* (specify serotype, strain, etc., if reported)
Q18. N (number of samples analyzed for prevalence) for this ARM	Radio	Report number analysed Not Reported Not Applicable (prevalence not estimated)
Q19. R (if prevalence data are described) (Note that we are only interested in results ≤24h after application of the intervention.)	Radio	Report number of positive samples Not Reported Not Applicable (prevalence not estimated)
Q20. N (number of samples analyzed for concentration estimate)	Radio	Report number analyzed Not Reported Not Applicable (concentration not estimated)
Q21. What was the concentration of the bacteria after the intervention? (Note that we are only interested in results ≤ 24h after application of the intervention.)	Radio	Specify concentration (no units) Not Reported Not Applicable
Q22. What are the units of the reported concentration?	Radio	Specify units Not Reported Not Applicable
Q23. What is the precision of the concentration estimate?	Radio	Specify number only Not Reported Not Applicable (concentration not estimated)
Q24. What was the descriptor of the precision of the concentration estimate?	Radio	SD SEM

		95% Confidence Interval Not Reported Not Applicable Other (specify)
Q25. Did the investigators report an effect estimate?	Radio	Yes No
Q26. What was the comparison or control group?	Radio	Describe control group Not Applicable (This was the control group or the authors did not report an effect size or a p-value)
Q27. What was the Effect Estimate?	Checkbox (only visible if reviewer answered Yes to Q25)	Odds Ratio (specify) Risk Ratio (specify) Mean Difference (specify) Not Reported (Option for reviewer to add another selection to the list)
Q28. What was the dispersion of the effect estimate?	Checkbox (only visible if the reviewer answered Yes to Q25)	SD (specify) SEM (specify) 95% Confidence Interval (specify) Not Reported (Option for reviewer to add another selection to the list)
Q29. What was the P-value for the comparison?	Radio	Specify Not Reported Not Applicable (no statistical tests performed or this was a control group)
Q30. Were the outcome assessors blinded to the intervention groups?	Radio	Yes No Not Reported
Q31. Additional Comments	Text	Please add any additional information about the intervention that you feel is relevant that wasn't captured by the other questions in this form.

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TABLE S8 Risk-of-Bias tool used in a systematic review of *Salmonella* reduction treatments on pig carcasses (modified from The Cochrane Collaboration's Risk-of-Bias Tool (1))

Question	Style	Response
Selection Bias		
Q1. Was allocation to treatment group randomized?	Radio	Yes (method of randomization reported) Reported random, but method of randomization not disclosed No (method of allocation reported, but was not random) Method of allocation not reported
Q2. What was the risk of bias due to allocation method? (If the authors did not describe the method used to randomize allocation, choose "Unclear". If the authors described the method used to achieve randomization, choose "Low", if the authors did not randomize allocation, choose, "High".)	Radio	Low High Unclear
Q3. What was the rationale for risk of bias due to allocation method	Text	
Performance Bias		
Q4. Were measures to blind owners/personnel described?	Radio	Yes No
Q5. What was the risk of bias due to knowledge of the allocated interventions by owners/handlers/personnel during the study? Answer "High" if the method of sampling was swabbing but the method used to swab was not described or was obviously subject to individual variation. Answer "Low" if the method of sampling was	Radio	Low High Unclear

tissue excision, carcass rinse or an objective method of swabbing (e.g. FSIS method).		
Q6. What was the rationale for risk of bias due to blinding of owners/personnel (e.g. personnel collecting the swab samples could tell what treatment the experimental unit received and might therefore have swabbed more or less vigorously based on whether they expected to find *Salmonella*)	Text	
Detection Bias		
Q7. Do they describe measures to blind outcome assessors?	Radio	Yes No
Q8. What was the risk of bias due to knowledge of the allocated interventions by outcome assessors?	Radio	Low High Unclear
Q9. What was the rationale for risk of bias due to blinding of outcome assessors?	Text	
Attrition Bias		
Q10. Were there incomplete outcome data in the study? (If this was not reported, choose "unable to assess")	Radio	No loss to follow-up Loss to follow-up present but explained Loss to follow-up present but not explained Unable to assess (numbers not reported comprehensively)
Q11. What was the risk of bias due to amount, nature, or handling of incomplete outcome data? (If the authors performed a sensitivity analysis to see how the missing or lost data would have affected the effect measure, then select "Low". If data/animals are missing and the authors do nothing to address this, the risk of bias is High).	Radio	Low High Unclear

Q12. What is the rationale for risk of bias due to incomplete outcome data?	Text	

Reporting Bias

Q13. Was there selective reporting of outcomes?	Radio	Yes No Unable to discern
Q14. What was the risk of bias due to selective outcome reporting? (Might the funding source for the study affect the authors' motivation to report all results?) Answer "High" if it looks the authors were "data-mining" in order to find any kind of significant difference between intervention and control groups or if the authors reported results in something other than the standard "prevalence" or "concentration" of *Salmonella*.	Radio	Low High Unclear
Q15. What was the rationale for risk of bias due to selective reporting of outcomes?	Text	

Other Bias

Q16. Other potential sources of bias identified: Did the analyses fail to take into account pseudo replication? We acknowledge that this bias in truth affects precision, rather than a systematic direction bias.	Text	
Q17. Are there concerns about multiplicity? (e.g. If the authors did an ANOVA then did an F-test and it's significant and then the authors look at all the comparisons within the ANOVA and did a Bonferroni correction within the test, but not correct for multiple comparisons across the study (just within the ANOVA), there are still multiplicity problems if you do, say, 20 ANOVAs, there's still a problem with multiplicity.	Radio	Yes No Unclear
Q18. What was the risk of bias due to other potential sources of bias not identified in the preceding questions?	Radio	Low High Unclear

Q19. What was the rationale for risk of bias due to other sources of bias?	Text	
Q20. Additional Comments (any additional information you feel is relevant to the assessment of risk of bias that was not captured by the previous questions)	Text	
Q21. Give a description of the ARM.	Radio	Describe the ARM (e.g. Ref ID, setting, etc.). Option for reviewer to add new ARM

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TABLE S9 List of documents excluded at Level 2 (full-text assessment) with reasons for exclusions in a systematic review of *Salmonella* reduction treatments on pig carcasses

Reference information	Reason for exclusion
Delchev H, Savov D. 1967. Investigations into the source of bacterial contamination of pork. I. In skinning. Veterinarnomeditsinski Nauki **4:**19–25.	Non-English language (Bulgarian)
Gerats GE, Snijders JMA, van Logtestijn JG. 1981. Slaughter methods and contamination of pig carcasses Slachttechniek en contaminatie van varkenskarkassen. Vleesdistributie en Vleestechnologie **16:**31–33.	Non-English language (Dutch)
de Kruijf JM. 1979. Bacteriological quality of pig liver Bacteriologische kwaliteit van varkenslever. Ph.D. thesis. University of Utrecht, The Netherlands.	Non-English language (Dutch)
Snijders JMA. 1976. Pig slaughtering hygiene Hygiene bij het slachten van varkens Ph.D. thesis. University of Utrecht, The Netherlands.	Non-English language (Dutch)
le Roux A, Minvielle B, Gault E. 2007. Control of carcase contamination levels at line end: utility of lactic acid Maitrise du niveau de contamination des carcasses en fin de chaine: interet de l'acide lactique. Techni-Porc **30:**29–33, 2.	Non-English language (French)
Boudry C, Korsak N, Jacob B, Etienne G, Thewis A, Daube G. 2002. Ecology of *Salmonella* in slaughter pigs digestive tract and study of the contamination of carcasses Ecologie de *Salmonella* dans le tube digestif du porc a l'abattage et etude de la contamination des carcasses. Ann Med Vet **146:**353–360.	Non-English language (French)
Rheault N, Quessy S. 1999. Comparison of hot water wash and trimming of pork carcasses for reducing the level of bacterial contamination Comparaison de l'effet du parage et du lavage a l'eau chaude des carcasses de porcs afin de reduire le niveau de la contamination microbienne. Can Vet J **40:**792–795.	Non-English language (French)
Schertenleib TI, Stephan R, Scheeder M, Zweifel C. 2011. Visual and microbiological process analysis of pig slaughtering in a small-scale abattoir Visuelle und mikrobiologische Prozessanalyse der	Non-English language (German)

Schweineschlachtung in einem Kleinbetrieb. Arch Lebensmittelhyg **62:**52–57.

Zweifel C, Spescha C, Stephan R. 2007. Process stages in pig slaughter: influence on the microbiological contamination of carcasses in two abattoirs Prozessstufen in der Schweineschlachtung: Einfluss auf den Oberflachenkeimgehalt von Schlachttierkorpern am Beispiel zweier Betriebe. Arch Lebensmittelhyg **58:**7–12. Non-English language (German)

Troeger K. 1993. Influence of scalding and dehairing technique on the bacterial contamination of pig carcasses Bruh- und Enthaarungstechnik. Einfluss auf den Keimgehalt von Schweineschlachtkorperchen. Fleischwirtschaft **73:**128–133, 171. Non-English language (German)

Troeger K. 1993. Changes in the microbial count of scalding water during a pig slaughter session, and its effect on surface contamination of the carcass Keimzahlentwicklung im Bruhwasser im Schlachtverlauf. Auswirkung auf die Oberflachenkeimgehalte der Schweineschlachttierkorper. Fleischwirtschaft **73:**816–819. Non-English language (German)

Jones B, Nilsson T, Sorqvist S. 1984. Contamination of pig carcasses with scalding water. Continued studies with radiolabelled solutes and particles. Fleischwirtschaft **64:**1226–1228, 1243–1246. Non-English language (German)

Jones B, Nilsson T, Ekman L, Ostlund K. 1979. Contamination of pig carcasses with scalding water studied with a radiolabelled colloid Nachweis von Bruhwasser in Schlachtschweinen mit einem radioaktiven Kolloid. Fleischwirtschaft **59:**1524–1526. Non-English language (German)

Snijders JMA, Gerats GE, Corstiaensen GP. 1977. Hygiene in pig slaughtering. V. Chlorinated water to clean carcasses Hygiene bei der Schlachtung von Schweinen. V. Verwendung chlorierten Wassers bei der Reinigung der Tierkorper-Oberflachen. Fleischwirtschaft **57:**2212–2215. Non-English language (German)

Snijders JMA. 1975. Hygiene in the slaughter of pigs. I. Scalding Hygiene bei der Schlachtung von Non-English language (German)

Schweinen. I. Das Bruhen der Schlachtschweine. Fleischwirtschaft **55:**836-840.

Troeger K. 1992. Extent of an internal contamination of carcasses of slaughter pigs by microorganisms in the scalding water. Arch Lebensmittelhyg **43:**11–13. Non-English language (German)

Woltersdorf W, Mintzlaff HJ. 1995. Pig scalding using a condensation method – is it a practicable one I Scalding effect and surface bacterial content. Fleischwirtschaft **75:**1077–1081. Non-English (German)

Takacs I. 1985. Hygiene in swine slaughter technology. I. Hygiene of scalding and singeing A sertesvagas technologiai higieniaja. I. Magy Allatorvosok Lapja **40:**407–412. Non-English language (Hungarian)

Bersani C, Fava M. 2004. Enforcement of the decision 2001/471/CE in a pig slaughterhouse Applicazione della Decisione 2001/471/CE in un macello suino. Industrie Alimentari **43:**376–381. Non-English language (Italian)

Hara K, Watanabe M, Yosizaki S, Endou T, Yokota T. 1998. Contamination of hog carcasses skinned by standing-type skin stripper. Journal of the Japan Veterinary Medical Association **51:**687–691. http://doi.org/10.12935/jvma1951.51.687. Non-English language (Japanese)

Takeshige K, Iida T, Takagi H, Kurihara S, Ogawa J, Tensho T, Maruyama T. 1995. Epidemiological studies of *Listeria monocytogenes* from dressed carcasses at a slaughter house. Nippon Juishikai Zasshi **48:**131–135. Non-English language (Japanese)

Akashi K, Kuroki H, Ebara S. 1972. Studies on microorganisms contaminating carcasses. IV. Microbial contamination of porcine carcasses in the scalding tank and subsequent processes and the protease activity of the bacteria isolated. Nippon Juishikai Zasshi **25:**70–76. Non-English language (Japanese)

Yang HS, Jeong JY, Moon SH, Park GB, Joo ST. 2007. Establishment of an optimal washing condition of a high temperature steaming system for the production of high quality pork. J Anim Sci Technol **49:**121–128. Non-English language (Korean)

Non-English language (Korean)

Kim IS, Kim DH, Hwang SK, Shin DK, Lee M. 1999. Assessment of microbial contamination of pork carcasses during the slaughtering process. Korean J Anim Sci **41:**199–206.

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Gill CO, McGinnis DS, Bryant J, Chabot B. 1995. Decontamination of commercial, polished pig carcasses with hot water. Food Microbiol **12:**143–149. doi:10.1016/S0740-0020(95)80090-5.	*Salmonella* not found in any samples or not tested for.
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Troeger K. 1994. Development of bacterial count in scalding water during slaughter – effect on surface bacterial counts on pig carcasses. Fleischwirtschaft **74:**518–520.	Not primary research on pathogen reduction wash/rinse/spray on pork carcasses or pork carcass parts.
Van Netten P, Valentijn A, Mossel DAA, Huis in't Velt JHJ. 1998. The survival and growth of acid-adapted mesophilic pathogens that contaminate meat	Not primary research on pathogen reduction wash/rinse/spray on pork carcasses or pork carcass parts.

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Greer G, Dilts BD. 1995. Lactic acid inhibition of the growth of spoilage bacteria and cold tolerant pathogens on pork. Int J Food Microbiol **25:**141–151. doi:10.1016/0168-1605(94)00088-N.	Not primary research on pathogen reduction wash/rinse/spray on pork carcasses or pork carcass parts.
King AM, Miller RK, Castillo A, Griffin DB, Hardin MD. 2012. Effects of lactic acid and commercial chilling processes on survival of *Salmonella, Yersinia enterocolitica,* and *Campylobacter coli* in pork variety meats. J Food Prot **75:**1589–1594. doi:10.4315/0362-028X.JFP-12-004.	Not primary research on pathogen reduction wash/rinse/spray on pork carcasses or pork carcass parts.
Sanguankiat A. 2013. Molecular epidemiology and serodiversity of *Salmonella enterica* in a pork chain "From Farm to Fork" in Northern Thailand. Dissertation. Freie Universität Berlin, Berlin, Germany.	No parallel comparison group
Cocora AM, Dobai G, Lazarescu C, Tibru I. 2013. The impact of the slaughtering technological's flow stages on the *Salmonella* spp. prevalence. Lucrări	No parallel comparison group

Ştiinţifice - Universitatea de Stiinte Agricole a Banatului Timişoara, Medicină Veterinară **46:**37–42.

Mannion C, Fanning J, McLernon J, Lendrum L, Gutierrez M, Duggan S, Egan J. 2012. The role of transport, lairage and slaughter processes in the dissemination of *Salmonella* spp. in pigs in Ireland. Food Res Int **45:**871–879. doi:10.1016/j.foodres.2011.02.001.

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Tibru I, Cerna D, Barna A. 2009. Hog carcass decontamination with lactic acid. Lucrări Ştiinţifice - Medicină Veterinară, Universitatea de Ştiinţe Agricole şi Medicină Veterinară "Ion Ionescu de la Brad" Iasi **52:**1196–1199.

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Chau PY, Shortridge KF, Huang CT. 1977. *Salmonella* in pig carcasses for human consumption in Hong Kong: a study on the mode of contamination. J Hyg (Lond) **78:**253–260.	No parallel comparison group
Alban L, Sørensen LL. 2009. Prevalence of *Salmonella* spp. after hot-water decontamination of finishers slaughtered in a Danish abattoir during 2004–2008, p 405–408. International Conference on the Epidemiology and Control of Biological, Chemical and Physical Hazards in Pigs and Pork. Iowa State University, Ames, IA, USA.	No parallel comparison group
Brustolin JC, Pisol AD, Steffens J, Toniazo G, Valduga E, Di Luccio M, Cansian RL. 2014. Decontamination of pig carcasses using water pressure and lactic acid. Braz Arch Biol Technol **57:**954–961. http://dx.doi.org/10.1590/S1516-8913201402363.	No parallel comparison group
Saidealbornoz JJ, Knipe CL, Murano EA, Beran GW. 1995. Contamination of pork carcasses during slaughter, fabrication, and chilled storage. J Food Prot **58:**993–997.	No parallel comparison group
Tadee P, Boonkhot P, Patchanee P. 2014. Quantification of contamination levels and particular risk of *Salmonella* spp. in pigs in slaughterhouses in Chiang Mai and Lamphun provinces, Thailand. Jpn J Vet Res **62:**171–179.	No parallel comparison group
Shotts EB jr, Martin WT, Galton MM. 1962. Further studies on *Salmonella* in human and animal foods and in the environment of processing plants, p 309–318. Proceedings of the 65th Annual Meeting of the U.S. Livestock Sanitary Association. MacCrellish and Quigley Co., Minneapolis, MN, USA.	No parallel comparison group
Carpenter CE, Smith JV, Broadbent JR. 2011. Efficacy of washing meat surfaces with 2% levulinic, acetic, or lactic acid for pathogen decontamination and	Outcome measured only >24 h after the intervention was applied

residual growth inhibition. Meat Sci **88:**256–260. doi:10.1016/j.meatsci.2010.12.032.

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PROTOCOL S1 Protocol for the assessment of the magnitude of change in the prevalence of *Salmonella* and quantity of *Salmonella* after administration of pathogen reduction treatments on pork carcasses

SUMMARY

To be completed

RATIONALE

To be completed

OBJECTIVES AND PICO (S) REVIEW QUESTION

The purpose of this project is to describe changes in *Salmonella* prevalence or quantity (most probably number of colony forming units) in pork carcasses after receiving pathogen reduction treatments during processing. The question is a PICO question: What is the change in *Salmonella* prevalence or quantity (O-Outcome) associated with the use of pathogen reduction treatments applied as washes, rinses or sprays (I - interventions) to pork carcasses (P-population)?

PROTOCOL REGISTRATION

The protocol is not registered.

ELIGIBILITY CRITERIA (PRISMA ITEM 6)

Relevant participants

Pork carcasses produced from commercial swine in commercial abattoirs. Smallholder slaughter approaches are not applicable

Intervention and comparators

Pathogen reduction treatments that are applied as a wash/rise/spray to pork carcasses; this includes organic acids such as peroxyacetic acid, lactic acid, non-acid chemical treatments, acetic acid, lactic acid, hypobromous acid, peroxyacetic acid, citric acid, mineral acids, hydrochloric acid, nitric acid, phosphoric acid, and/or hot water and water treatments such as hot or cold water, steam vacuuming and steam pasteurization, and other compounds such as aqueous ozone, electrolyzed water, potassium hydroxide, potassium sorbate, sodium hypochlorite (NaClO), trisodium phosphate (TSP), cholorine and Alcide® . Combinations of these components are also of interest

Outcomes

The outcome of interest are the following measures of *Salmonella* on the carcass: *Salmonella* prevalence measured by culture (using any culture measure including prescreening with ELISA methods or the presence of *Salmonella* antigen measured by PCR or other antigen detection methods), and *Salmonella* quantity measures (either *Salmonella* colony-forming units and most probable number or “quantifiable PCR” if such an approach exists).

We will also identify studies that have *E coli*, coliforms, Enterobacteriae, and Total Plate Counts (TPC) and may include these in a second review.

Relevant study designs

Data may come from comparative experiments. The experiments may use either deliberate contamination or naturally occurring levels of *Salmonella* on the carcasses.

INFORMATION SOURCES (PRISMA ITEM 7)

The electronic indexes searched will be science citation index, Medline search and CABI. We will also hand search the reference lists of relevant reviews identified during the search and the conference proceedings of Safe Pork

SEARCH STRATEGY (PRISMA ITEM 8)

Initially, two search strategies were run in CABI (Web of Science™, Thomson Reuters™), one containing additional indexing terms to maximize the number of hits; the second search (below) did not contain any of those additional terms. The first search resulted in 1899 hits more than the second search. Those additional records were sorted by relevance and the first 300 of those 1899 were screened for relevance using the Level 1 screening question. None of those 300 records were found to be relevant. Therefore, the final search was run using the search without the additional indexing terms. This search was conducted in in CABI on January 21st, 2015. No restrictions were made with respect to language or document type. Timespan=all years.

De-duplication was performed in EndNote and 5 records were removed, resulting in 1938 records being uploaded to Distiller for screening.

Search No	No Hits	Search Terms Used	Comments
# 23	1943	#22 OR #21	Both options ORed
# 22	473	TI=((DECONTAMINAT* OR CONTAMINAT*) AND CARCASS*)	Title words only decontamination of carcasses
# 21	1,516	#20 AND #1	Pork carcasses etc AND decontamination techniques (title or abstract)
# 20	756,777	#19 OR #18 OR #17 OR #16 OR #15 OR #14 OR #13 OR #12 OR #11 OR #10 OR #9 OR #8 OR #7 OR #6 OR #5 OR #4 OR #3 OR #2	
# 19	34,856	TS=(Chilling or "freezing air" or "high air velocity" or blasting)	
# 18	15,540	TS=((Prevent* or reduc*) near/4 contaminat*) or TS=decontaminat*	

# 17	139,833	TS=(CHLORINE OR ALCIDE OR ULTRAVIOLET OR UV OR IRRADIAT* OR "DRY HEAT" OR ULTRASOUND)
# 16	144,840	ts=(TSP or phosphate$)
# 15	10,343	ts=("sodium hypochlorite" OR NaClO or "sodium acetate" or "sodium citrate" or "sodium chlorite" or "sodium lactate")
# 14	2,975	TS=("POTASSIUM HYDROXIDE" OR "POTASSIUM SORBATE")
# 13	70	TS="AQUEOUS OZONE"
# 12	24,497	TS=steam
# 11	70,878	ts="water treatment$"
# 10	26,217	TS=((hot or cold or electrolyzed or electrolysed or warm) NEAR/3 water)
# 9	134	TS=NONACID
# 8	6,609	TS=((HYDROCHLORIC OR NITRIC OR PHOSPHORIC OR ACID) NEAR/5 (spray* or decontaminat* or saniti* or wash*))
# 7	56,989	TS=(ACETIC OR hypobromous or citric or "mineral acid$")
# 6	46,191	TS=(PEROXYACETIC OR LACTIC)
# 5	157	TS=(Organic NEAR/5 (decontaminat* or saniti*))
# 4	192,274	TS=(spray or sprays or spraying or sprayed)
# 3	57,209	TS=(wash or washes or washing or washed or rinse or rinses or rinsing or rinsed)
# 2	7,614	TS=(pathogen near/4 reduc*) OR TS=prt

# 1	25,552	TS=((pork or swine or pig or pigs or hog or hogs or boar or boars or sow or sows) near/7 (carcass* OR slaughter* or abattoir* or bellies))

STUDY SELECTION (PRISMA ITEM 9)

Two reviewers will screen the abstracts. For Level 1 we will include studies for which any reviewer says yes. Exclusion will require both reviewers to say no. For Level 2, if both reviewers agree that at the form level it should be excluded, that study will be excluded. The questions will be sequential so that the reason for exclusion will be the 1st question in the sequence that has a response of no by both reviewers.

Screening for eligibility for the review

The 1st level of screening will be based on the title, abstract, year and language. The Level 1 form will have one question:

"Does the study appear to be primary research on pathogen reduction washes/ rinses/ sprays for pork carcasses or parts of a pork carcass?"

Yes—likely (Include)

No (Exclude)

No details—can't tell (Include)

No, but is a relevant review (Exclude)

The second level of screening based on the full text is provided below.

DATA COLLECTION PROCESS (PRISMA ITEM 10)

For the data collection process two reviewers will extract data independently from all relevant studies into DistillerSR® (Ottawa, ON, Canada).

DATA ITEMS (PRISMA ITEM 11)

The data extraction forms for the study level information and the outcome level information are included in the Protocol Table **P1** and **Error! Reference source not found.**.

1. Assessment of risk of bias in included studies (PRISMA ITEM 12)

As most of the studies are likely to be experimental we will use the Cochrane Risk of Bias Tool modified to include information about the challenge for experimental studies that use deliberate contamination of the carcass.

SUMMARY MEASURES (PRISMA ITEM 13)

The summary measures will be mean differences for continuous outcomes, and summary risk ratio or summary odds ratio for categorical outcomes.

SYNTHESIS OF RESULTS (PRISMA ITEM 14)

Dealing with missing data

We will not contact authors to obtain missing data.

Assessment of heterogeneity

We propose, if the sample size is sufficient, to conduct a meta-regression to determine what factors are associated with the magnitude of the effect size based on the demographic factors collected (see Table 2 and 3) .

Data synthesis

We will attempt to conduct a mixed treatment comparison meta-analysis and ranking the interventions. The potential to do this will depend upon the how comprehensively the results of the studies are reported.

RISK OF BIAS ACROSS STUDIES (PRISMA ITEM 15)

We will assess studies to have a high risk of bias if they have at least one high risk of bias domain. If possible we will also conduct an analysis for small-study effects. However it is unclear if this will be useful as most of the studies will be small and it might not be possible to detect small-study effects. The sample size for small is based on the number of experimental units not the number of pseudo-replicates.

ADDITIONAL ANALYSES (PRISMA ITEM 16)

At this point we do not propose to do any additional analyses however if we do they will be reported here as they are not proposed *a priori*.

Proposed level 2 full text screening form

Q1. Is the full text available in English?
Yes (Include)
No (Exclude)

Q2. Based on the full text, is the study about primary research on pathogen reduction washes/ rinses/ sprays for pork carcasses or parts of a pork carcass?
Yes (Include)
No (Exclude)

Q3. Did the investigators look at *Salmonella*?
Yes (Include)
No, but they did look at *E. coli*, Enterobacteriaciae, coliforms and/or Total Plate Count (TPC) (Exclude)
No, the authors did not look at any of the above (Exclude)

Q4. Based on the full text does the study have a parallel comparative group?
Yes (Include)
No (Exclude)

Q5. Did the investigators measure the outcome *only* > 24h after application of the intervention?
Yes (Exclude)
No (Include)

Q6. What type of study is this?
Challenge (artificial contamination)
Control (natural contamination)

Protocol Table P1 Proposed Study level data extraction form for relevant papers

Study-Level Information Form

Question	Style	Response
Q1. In what year(s) was the data collected?	Radio	Specify year or Not Reported. Do not use publication date to answer this question.
Q2. In what country was the study carried out?	Checkbox	(Select all that apply) List of countries with option for the reviewer to add to the list as needed. Not Reported
Q3. In what setting was the study carried out?	Radio	Commercial Abattoir University/Research Slaughter Plant Laboratory Small Holder Slaughter (Exclude) Not Reported
Q4. If the study was conducted in a commercial abattoir, was the slaughter capacity reported?	Radio (only visible is Q3 = commercial)	Yes: Enter number No
Q5. If the slaughter capacity was reported, what were the units?	Text (only visible is Q3 = commercial)	Specify the units
Q6. If the study was conducted in a commercial abattoir, what was the total number of Slaughter Plants?	Radio (only visible is Q3 = commercial	Specify number or Not Reported
Q7. What was the experimental unit in this study? i.e the unit the intervention was assigned to	Checkbox	Select all that apply Carcass Pork belly Skin (Option for reviewer to add items to this list as needed)
Q8. What was the weight of the experimental units in this study?	Radio	Specify weight in kg Not Reported
Q9. What was the descriptor the weight of the experimental units?	Radio	Mean Median Range

		Not Reported Not Applicable
Q10. What was the dispersion of the experimental unit weight?	Radio	Specify a number or Not Reported
Q11. What was the dispersion descriptor for the experimental unit weight?	Radio	SD SEM Not Reported Not Applicable
Q12. Did the investigators artificially contaminate the experimental units?	Radio	Yes No Can't Tell
Q13. What was the concentration of *Salmonella* applied to the experimental unit?	Radio (only visible if Q12 = yes)	Specify number Not Reported
Q14. What were the units of the concentration of *Salmonella* applied to each experimental unit?	Radio (only visible if Q12 = yes)	Specify units Not Reported
Q15. What was the contact time (i.e. the number of seconds that the *Salmonella* was kept on the experimental unit)?	Radio (only visible if Q12 = yes)	Number or Not Reported
Q16. What serotype of *Salmonella* was inoculated onto the experimental unit?	Radio (only visible if Q12 = yes)	Specify serotypes or Not Reported
Q17. What was the method used to inoculate the experimental unit?	Radio (only visible if Q12 = yes)	Sprayed Injected Brushed On Not Reported
Q18. At what locations on the carcass/what parts of the experimental unit was *Salmonella* inoculated?	Checkbox (only visible if Q12 = yes)	Check all that apply Jowls Pork Belly Not Reported Option for Reviewer to add to this list if needed.

Q19. How many times was *Salmonella* applied to the experimental unit?	Radio (only visible if Q12 = yes)	Specify number of times Not Reported
Q20. What was the time interval between inoculation and application of the intervention?	Radio (only visible if Q12 = yes)	Specify time interval (s) Not Reported Not Applicable (if this is the control group, i.e. it didn't receive an intervention)
Q21. Which diagnostic method(s) were used to determine the prevalence or level of the bacteria?	Checkbox	Bacterial culture PCR ELISA Other (specify) Not Reported
Q22. If bacterial culture was used, what was the pre-enrichment media?	Radio (only visible if Q21 = Bacterial culture)	Specify Not Reported Not Applicable (pre-enrichment not done)
Q23. If bacterial culture was used, what was the pre-enrichment incubation time?	Radio (only visible if Q21 = Bacterial culture)	Specify time (h) Not Reported Not Applicable (pre-enrichment not done)
Q24. If bacterial culture was used, what was the pre-enrichment incubation temperature?	Radio (only visible if Q21 = Bacterial culture))	Specify temperature (in ^{o}C) Not Reported Not Applicable (pre-enrichment not done)
Q25. If bacterial culture was used, specify the enrichment media.	Radio (only visible if Q21 = Bacterial culture)	Specify Not Reported Not Applicable (enrichment not done)
Q26. If bacterial culture was used, specify the enrichment incubation time.	Radio (only visible if Q21 = Bacterial culture)	Specify (h) Not Reported Not Applicable (enrichment not done)
Q27. If bacterial culture was used, specify the enrichment incubation temperature.	Radio (only visible if Q21 = Bacterial culture)	Specify temperature (^{o}C) Not Reported Not Applicable (enrichment not done)

Q28. If bacterial culture was used, specify the culture media	Radio (only visible if Q21 = Bacterial culture)	Specify Not Reported
Q29. If bacterial culture was used, specify the culture incubation time	Radio (only visible if Q21 = Bacterial culture)	Specify (h) Not Reported
Q30. If bacterial culture was used, specify the culture incubation temperature	Radio (only visible if Q21 = Bacterial culture)	Specify (^{o}C) Not Reported
Q31. If bacterial culture was used, was a confirmation method also used?	Radio (only visible if Q21 = Bacterial culture)	Yes (specify method) No Confirmation done, but method not reported
Q32. If bacterial culture was used and concentration of bacteria was determined, what was the enumeration method used?	Radio (only visible if Q21 = Bacterial culture)	Specify method Not Reported Not Applicable (enumeration of bacteria not done)
Q33. If PCR was used to detect the bacteria, please give details of the methods (i.e. PCR type, conditions, primer name, target sequence) (copy-paste from text)	Text (only visible if Q21 = PCR)	Give details or Not Reported.
Q34. If ELISA was used to detect the bacteria, please give details of the methods used. (copy-paste from text)	Text (only visible if Q21 = ELISA)	Give details or Not Reported
Additional Comments	Text	Please add any additional information about the study not captured by the previous questions that you think is relevant to this review.

1 **Protocol Table P2 : Proposed intervention and outcome level form for relevant studies**

Question	Style	Response
Q1. Description of ARM / Methods (as described by authors -- copy - paste from text)	Radio	Describe the intervention used. (Option for reviewer to add new ARM descriptions as needed.)
Q2. Were the interventions assigned randomly?	Radio	Yes (method of randomization was reported) Reported random but method of randomization not disclosed Not Reported
Q3. Which treatment (intervention) was applied to the experimental unit?	Checkbox	Check all that apply: Acetic Acid Chlorine Citric Acid Electrolyzed Oxidizing Water Hot Water Hydrochloric Acid Hypobromous Acid Lactic Acid Ozone (aqueous) Peroxyacetic Acid Potassium Hydroxide Steam Not Reported (Option for reviewer to add new items to the list as needed)
Q4. Is there any additional descriptive information about this intervention (not captured by the other questions on this form) that you would like to add?	Text	
Q5. What was the pH of the intervention as it was applied to the experimental units?	Radio	Specify pH Not Reported/Not Discernible Not Applicable (control group)
Q6. What was the concentration of the intervention?	Radio	Specify concentration (number only) Not Reported/Not Discernible

		Not Applicable (e.g. if it's a no-intervention control group or if it's pure water)
Q7. What were the units of the intervention concentration?	Radio	Specify units (e.g. %) Not Reported Not Applicable
Q8. What was the temperature of the intervention?	Radio	Specify temperature (number only) Not Reported Not Applicable
Q9. What were the units of the temperature of the intervention?	Radio	Specify units (degrees C, degrees F) Not Reported Not Applicable
Q10. How was the intervention applied?	Radio	Spray Rinse Not Reported/Not Discernible Not Applicable Other (Specify)
Q11. At what pressure was the intervention applied?	Radio	Specify pressure (number only) Not Reported/Not Discernible Not Applicable
Q12. What units were used to describe the pressure the intervention was applied at?	Radio	Specify units Not Reported Not Applicable
Q13. At what point(s) in the processing chain was the intervention applied? Regardless of how the authors described the data, we always reported data as occurring after the processing point. For example, if the original author described the sample as being collected pre-kill, we referred to such a sample as a bleed sample, meaning the post-bleeding but pre-killing sample point.	Radio	final wash, immediately after chill, 18–48 h after chilling Not Reported Not Applicable (control group) stun, bleed, kill, scald, dehair, singe, polish, bung removal, evisceration,

		split, stamp, Other (specify)
Q14. What was the total duration that the intervention was applied?	Radio	Specify the total duration (s) Not Reported Not Applicable
Q15. What was the number of times that the intervention was applied?	Radio	Specify number of times Not Reported Not Applicable
Q16. If the intervention was applied more than once, what was the time interval between the applications?	Radio	Specify time (s) Not Reported/Not Discernible Not Applicable
Q17. In which type of sample was the outcome measured?	Radio	Carcass Rinse Carcass Swab Excised Skin Sample Not Reported (Option for reviewer to add to this list as needed)
Q18. Which bacteria were measured as the outcome?	Radio	*Salmonella* (Option for reviewer to add to this list as needed.)
Q19. N (number of samples analyzed for prevalence) for this arm	Radio	Report number analysed Not Reported Not Applicable (prevalence not estimated)
Q20. R (if prevalence data are described) (Note that we are only interested in results <24h after application of the intervention.)	Radio	Report number positive samples Not Reported Not Applicable (prevalence not estimated)
Q21. N (number of samples analyzed for concentration estimate)	Radio	Report number analyzed Not Reported Not Applicable (concentration not estimated)
Q22. What was the concentration of the bacteria after the intervention? (Note that we	Radio	Specify concentration (no units) Not Reported Not Applicable

are only interested in results <24h after application of the intervention.)		
Q23. What are the units of the concentration?	Radio	Specify units Not Reported Not Applicable
Q24. What is the precision of the concentration estimate?	Radio	Specify number only Not Reported Not Applicable (concentration not estimated)
Q25. What was the descriptor of the precision of the concentration estimate?	Radio	SD SEM 95% Confidence Interval Not Reported Not Applicable Other (specify)
Q26. Did the investigators report an effect estimate?	Radio	Yes No Describe control group
Q27. What was the comparison or control group?	Radio	Not Applicable (control group)
Q28. What was the Effect Estimate?	Checkbox (only visible if reviewer answered Yes to Q26)	Odds Ratio (specify) Risk Ratio (specify) Mean Difference (specify) Not Reported (Option for reviewer to add another selection to the list)
Q29. What was the dispersion of the effect estimate?	Checkbox (only visible if the reviewer answered Yes to Q26)	SD (specify) SEM (specify) 95% Confidence Interval (specify) Not Reported (Option for reviewer to add another selection to the list)
Q30. What was the P-value for the comparison ?	Radio	Specify Not Reported Not Applicable (no statistical tests performed)

Q31. Were the outcome assessors blinded to the intervention groups?	Radio	Yes No Not Reported
Additional Comments	Text	

2

REFERENCES

1. **Higgins JPT, Green S.** Updated March 2011. Cochrane handbook for systematic reviews of interventions version 5.1.0. The Cochrane Collaboration, London, England, UK. Available from: http://handbook.cochrane.org/chapter_8/table_8_5_a_the_cochrane_collaborations_tool_for_assessing.htm. Accessed 2 July 2015.

TABLE S10 List of potentially relevant studies to be evaluated in a future update of a systematic review of *Salmonella* reduction treatments on pig carcasses

Clayton NC. 2002. The efficacy of various *Salmonella* intervention methods applied to pork carcasses during slaughter. M.S. thesis. University of Kentucky, Lexington, KY.
Le Roux A, Minvielle B, Gault E. 2008. Validation of steam-vacuum process as corrective measure for visible faecal contamination on carcasses: preliminary results, p 2A–12. Proceedings of the Fifty-Fourth International Congress of Meat Science and Technology. Cape Town, South Africa. doi:10.13140/2.1.2537.6969.
Reynolds AE. 2005. Utilisation of spray wash with organic acids (peroxyacetic acid and lactic acid) and chlorinated wash in combination, utilizing direct application methods, for pathogen reduction on pork and beef carcasses in small and very small meat processing plants. Research Note: FSIS New Food Safety Technologies Applicable for Small and Very Small Plants. http://www.fsis.usda.gov/PDF/New_Technology_C29_Summary_FY2003.pdf. Accessed 25 September 2015.