

# NEWSLETTER

European Union Reference Laboratory for *Salmonella*

**Vol. 29 No. 2**  
**June 2023**

ISSN 2211-6877



Continuation of Newsletter Community Reference Laboratory for *Salmonella*  
ISSN 1572-3836

Produced by

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## Editorial Note

Bilthoven, 4 July 2023

Dear colleagues,

It was very nice that many of you participated in the 28<sup>th</sup> **EURL-*Salmonella* workshop**, which we organised, for the 4<sup>th</sup> time remotely, on 22 and 23 May 2023. We had several interesting presentations and despite the fact that we met online again, we also had some nice discussions. When looking at the evaluation of the workshop, I am happy to notice that the majority of participants was also satisfied with the workshop. The presentations and group picture of the workshop were published at our website shortly after the meeting and can be found at the following link: <https://www.euralsalmonella.eu/workshop-2023>.

An update on our **Proficiency Tests** (PTs):

Shortly before the workshop, the individual laboratory results of the cluster analysis part within the **PT on typing of *Salmonella* 2022** were shared with the NRLs. The results of all participants in the optional part of this PT were presented at the workshop in May 2023 and will be summarised in an interim summary report. This report is almost finalised and will soon be sent to the participants.

In March 2023, the **combined PT for Food-Feed on detection of *Salmonella* in seeds (2023)** was organised. The matrix under analysis was flaxseed, which is used as a food product as well as an ingredient of animal feed. NRLs-*Salmonella* which analyse food samples and NRLs-*Salmonella* which analyse animal feed products were invited to participate in this PT, resulting in a total of 51 participants. We were happy to notice that all participants scored a good performance in this PT. The individual laboratory results of this PT were shared with the participants by mid-May 2023 and the overall results were presented at the workshop. The interim summary of this PT was sent to the NRLs early June 2023 and can be found at our website at the following link: <https://www.euralsalmonella.eu/documenten/interim-summary-report-eurl-salmonella-combined-pt-food-feed-2023>

Currently we are busy with the preparations of the **PT on detection of *Salmonella* in samples from the primary production stage 2023**. This PT will focus on the detection of *Salmonella* in chicken faeces and the performance is planned in October 2023. The time table for this PT is included in this Newsletter.

In November, the **2023 PT on typing of *Salmonella*** will be organised. Like former years, this study will contain an obligatory part on serotyping of *Salmonella*, and a voluntary part on cluster analysis (NGS only). The timetable for this PT is also included in this Newsletter.

On 20 and 21 June 2023, the **Inter-EURLs working group on NGS**, organised a joint training course on NGS for 24 participants of 8 EURL/NRL networks at the premises of the EURL-*Salmonella* (Bilthoven, the Netherlands). The training was well received and it will be investigated if presentations given at the training can be shared at the websites of the EURLs.

In earlier e-mails we have informed you that EFSA and the Inter-EURLs working group on NGS are jointly organising a second edition of the **Science meets policy conference**. The event will focus on EU initiatives towards the large-scale use of Next Generation Sequencing (NGS) to tackle foodborne threats. The event will take place at the EFSA premises in Parma, Italy as well as online on 5 and 6 September 2023. The meeting is open to all interested parties.

It is particularly relevant for food safety risk assessment competent authorities and policymakers in the EU and beyond, as well as food business operators, academics and stakeholders with an interest in the topic. The registration for physical participation was closed by 31 May 2023, but the registration for online participation is still open until 1 September 2023. For more information and for online registration, please use the following link:

<https://www.efsa.europa.eu/en/events/science-meets-policy-conference-using-next-generation-sequencing-tackle-foodborne-threats>

From 26 until 30 June 2023, the **annual meetings of CEN/TC463 and ISO/TC34/SC9 (Microbiology of the Food chain)** were organised. After 3 years of online meetings, this year's meeting was organised as a hybrid meeting in Stockholm, Sweden. As convenor of an ISO Ad hoc group (AHG 'Guidance document for drafting ISO/CEN standards') and two *Salmonella* ISO working groups for detection (WG9) and typing of *Salmonella* (WG10), the EURL-*Salmonella* participated in these annual meetings and presented the progress of the AHG and WGs. A more detailed report on the relevant CEN/ISO subjects will be included in the next Newsletter. For now, only a short summary of the state of play of the (draft) ISO/TS 6579-4 ('Identification of monophasic *Salmonella* Typhimurium by PCR') is given. At the annual SC9 meeting, the validation study results of this ISO document were presented, existing of a method(s) evaluation study and an interlaboratory study. The performance characteristics (inclusivity and exclusivity) calculated from these studies have been summarised in Tables in Annex E of draft ISO/TS 6579-4 and the sixth draft ISO/DTS 6579-4 version was shared for comments with the SC9 members and with the NRLs-*Salmonella* from 1 June until 12 July 2023. All information was well received at the annual meeting of SC9 and it was questioned why we are planning to publish this ISO document as a Technical Specification (TS) instead of a full ISO. Generally a standard is published as an ISO/TS when no validation data are available. However, as for this ISO document the validation data are available, it was suggested to still try to publish the document as a full ISO instead of a TS. A request for this was sent to ISO central secretariat by the end of June 2023 and if accepted, the document will be launched for DIS voting (Draft International Standard) in the second half of 2023. If not accepted, the document will be forwarded for ISO/DTS voting instead (as planned before). We will keep you updated on the next steps in this standardization process.

In June 2023, the following EURL-*Salmonella* report was published: Jacobs-Reitsma, W.F., Verbruggen, A., Diddens, R.E., van Hoek, A.H.A.M., Mooijman, K.A., 2023. EURL-*Salmonella* Proficiency Test Typing 2021. National Institute for Public Health and the Environment, Bilthoven, the Netherlands. RIVM report no.: 2022-0105. <https://www.rivm.nl/bibliotheek/rapporten/2022-0105.pdf>

Best wishes,  
Kirsten Mooijman  
Coordinator EURL-*Salmonella*

## Contribution of the EURL-*Salmonella*

### Timetable EURL- *Salmonella* Proficiency Test Primary Production Stage 2023. Detection of *Salmonella* in chicken faeces.

Week	Date	Subject
27-35		E-mailing the link to the registration form for the Proficiency Test. Please <b>register by 31 August 2023</b> at the latest.
39		E-mailing the link for the result form to the participants. E-mailing the protocol and instructions for the result form to the NRLs. Preparation of media by the NRLs.
39	Monday 25 September 2023	Shipment of the parcels to the participants as Biological Substance Category B (UN 3373).
40	<b>Monday 2 October 2023</b>	<b>Start performance of the Proficiency Test.</b>
43	27 October at the latest	Deadline for completing the result form: <b>27 October 2023</b> (23:59h CET) After this deadline the result form will be closed.
	December 2023	Interim summary report

If you have questions or remarks about this Proficiency Test, please contact:

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## Timetable EURL- *Salmonella* Proficiency Test Typing 2023 Serotyping and optional part NGS Cluster analysis

Week	Date	Subject
39	Week of 25 September	Emailing of the link to the registration form for the typing study. Please <b>register by 20 October 2023</b> at the latest.
43	Week of 23 October	Emailing of the protocol 2023.
45	Monday 6 November	Shipment of the parcels to the participants as Biological Substance Category B (UN 3373).
45	Week of 6 November	<i>Upon receipt:</i> Starting the identification of the strains, according to the usual practice of the laboratory. Sending the link for the result form on Serotyping to the participants. Sending the link for the result form on NGS Cluster Analysis to the participants in a separate email.
50	15 December 2023 at the latest	Deadline for completing the electronic submission of <b>Serotyping</b> results: <b>15 December 2023</b> . After this deadline, the result form for serotyping will be closed.
5	31 January 2024 at the latest	Deadline for completing the electronic submission of <b>NGS Cluster Analysis</b> results: <b>31 January 2024</b> .
	February 2024	Serotyping: Evaluation of individual laboratory results and Interim summary report.
	April/May 2024	NGS Cluster Analysis: Evaluation of individual laboratory results and Interim summary report.

If you have questions or remarks about this Proficiency Test, please contact:

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## From the Literature

### Salmonella-related Literature from Scopus: April – June 2023

**Nickodem C., Arnold A.N., Gehring K.B., Gill J.J., Richeson J.T., Samuelson K.L., Morgan Scott H., Smith J.K., Matthew Taylor T., Vinasco J., Norman K.N.**

*A Longitudinal Study on the Dynamics of Salmonella enterica Prevalence and Serovar Composition in Beef Cattle Feces and Lymph Nodes and Potential Contributing Sources from the Feedlot Environment*

(2023) *Applied and Environmental Microbiology*, 89 (4)

ABSTRACT: Salmonella can persist in the feedlot pen environment, acting as a source of transmission among beef cattle. Concurrently, cattle that are colonized with Salmonella can perpetuate contamination of the pen environment through fecal shedding. To study these cyclical dynamics, pen environment and bovine samples were collected for a 7-month longitudinal comparison of Salmonella prevalence, serovar, and antimicrobial resistance profiles. These samples included composite environment, water, and feed from the feedlot pens (n = 30) and cattle (n = 282) feces and subiliac lymph nodes. Salmonella prevalence across all sample types was 57.7%, with the highest prevalence in the pen environment (76.0%) and feces (70.9%). Salmonella was identified in 42.3% of the subiliac lymph nodes. Based on a multilevel mixed-effects logistic regression model, Salmonella prevalence varied significantly ( $P < 0.05$ ) by collection month for most sample types. Eight Salmonella serovars were identified, and most isolates were pansusceptible, except for a point mutation in the parC gene, associated with fluoroquinolone resistance. There was a proportional difference in serovars Montevideo, Anatum, and Lubbock comparing the environment (37.2, 15.9, and 11.0%, respectively), fecal (27.5, 22.2, and 14.6%, respectively), and lymph node (15.6, 30.2, and 17.7%, respectively) samples. This suggests that the ability of Salmonella to migrate from the pen environment to the cattle host-or vice versa-is serovar specific. The presence of certain serovars also varied by season. Our results provide evidence that Salmonella serovar dynamics differ when comparing environment and host; therefore, developing serovar-specific preharvest environmental Salmonella mitigation strategies should be considered. **IMPORTANCE** Salmonella contamination of beef products, specifically from the incorporation of bovine lymph nodes into ground beef, remains a food safety concern. Current postharvest Salmonella mitigation techniques do not address Salmonella bacteria that are harbored in the lymph nodes, nor is it well understood how Salmonella invades the lymph nodes. Alternatively, preharvest mitigation techniques that can be applied to the feedlot environment, such as moisture applications, probiotics, or bacteriophage, may reduce Salmonella before dissemination into cattle lymph nodes. However, previous research conducted in cattle feedlots includes study designs that are cross-sectional, are limited to point-in-time sampling, or are limited to sampling of the cattle host, making it difficult to assess the Salmonella interactions between environment and hosts. This longitudinal analysis of the cattle feedlot explores the Salmonella dynamics between the feedlot environment and beef cattle over time to determine the applicability of preharvest environmental treatments. ISSN: 00992240

**Łuczyńska A., Dreesman J., Mertens E., Wollenweber M., Perriat D., Rosner B.M.**

*Recruiting controls from an online panel for a case-control study enabled a timely and reliable foodborne Salmonella outbreak investigation, Germany 2021*

(2023) *Epidemiology and Infection*, 151, art. no. e70

ABSTRACT: We explored the feasibility, suitability, and reliability of using controls recruited among members of a non-probabilistic online panel ('panel controls') in a case-control study (CCS) to investigate a Salmonella Braenderup outbreak in Germany. For comparison, another control group was recruited via random digit dialling ('classical controls'). Panel members received questionnaires by email; classical controls were interviewed by phone. Both control groups were frequency-matched to cases by age and sex; the classical controls also by federal state. Cases and controls were queried mainly about fruit consumption since melons were the suspected infection vehicle. We calculated adjusted odds ratios (aOR) and 95% confidence intervals (CIs) using single-variable and multivariable logistic regression. The study included 32 cases, 81 panel controls and 110 classical controls. Analyses identified melons, particularly Galia melons, as the most likely infection vehicle using either control group (panel controls - aOR 12, CI 2.7-66; classical controls - aOR 55, CI 8-1100). Recruitment of panel versus classical controls required



substantially less person-time (8 vs. 111 hours) and was about 10 times less expensive. We recommend this timely and reliable control recruitment method when investigating diffuse foodborne outbreaks with CCS. ISSN: 09502688

**Obe T., Siceloff A.T., Crowe M.G., Morgan Scott H., Shariat N.W.**

*Combined Quantification and Deep Serotyping for Salmonella Risk Profiling in Broiler Flocks (2023) Applied and Environmental Microbiology, 89 (4)*

ABSTRACT: Despite a reduction of *Salmonella* contamination on final poultry products, the level of human salmonellosis cases attributed to poultry has remained unchanged over the last few years. There needs to be improved effort to target serovars which may survive antimicrobial interventions and cause illness, as well as to focus on lessening the amount of contamination entering the processing plant. Advances in molecular enumeration approaches allow for the rapid detection and quantification of *Salmonella* in pre- and postharvest samples, which can be combined with deep serotyping to properly assess the risk affiliated with a poultry flock. In this study, we collected a total of 160 boot sock samples from 20 broiler farms across four different integrators with different antibiotic management programs. Overall, *Salmonella* was found in 85% (68/80) of the houses, with each farm having at least one *Salmonella*-positive house. The average *Salmonella* quantity across all four complexes was 3.6 log<sub>10</sub> CFU/sample. Eleven different serovars were identified through deep serotyping, including all three key performance indicators (KPIs; serovars Enteritidis, Infantis, and Typhimurium) defined by the U.S. Department of Agriculture-Food Safety and Inspection Service (USDA-FSIS). There were eight multidrug resistant isolates identified in this study, and seven which were serovar Infantis. We generated risk scores for each flock based on the presence or absence of KPIs, the relative abundance of each serovar as calculated with CRISPR-SeroSeq (serotyping by sequencing the clustered regularly interspaced palindromic repeats), and the quantity of *Salmonella* organisms detected. The work presented here provides a framework to develop directed processing approaches and highlights the limitations of conventional *Salmonella* sampling and culturing methods. ISSN: 00992240

**Burgess B.A.**

*Salmonella in Horses*

(2023) *Veterinary Clinics of North America - Equine Practice, 39 (1), pp. 25 - 35*

**Nuchchanart W., Pikoolkhao P., Saengthongpinit C.**

*Development of a lateral flow dipstick test for the detection of 4 strains of Salmonella spp. in animal products and animal production environmental samples based on loop-mediated isothermal amplification*

(2023) *Animal Bioscience, 36 (4), pp. 654 - 670*

ABSTRACT: Objective: This study aimed to develop loop-mediated isothermal amplification (LAMP) combined with lateral flow dipstick (LFD) and compare it with LAMP-AGE, polymerase chain reaction (PCR), and standard *Salmonella* culture as reference methods for detecting *Salmonella* contamination in animal products and animal production environmental samples. Methods: The SalInvA01 primer, derived from the InvA gene and designed as a new probe for LFD detection, was used in developing this study. Adjusting for optimal conditions by temperature, time, and reagent concentration includes evaluating the specificity and limit of detection. The sampling of 120 animal product samples and 350 animal production environmental samples was determined by LAMP-LFD, comparing LAMP-AGE, PCR, and the culture method. Results: *Salmonella* was amplified using optimal conditions for the LAMP reaction and a DNA probe for LFD at 63°C for 60 minutes. The specificity test revealed no cross-reactivity with other microorganisms. The limit of detection of LAMP-LFD in pure culture was 3×10<sup>2</sup> CFU/mL (6 CFU/reaction) and 9.01 pg/μL in genomic DNA. The limit of detection of the LAMP-LFD using artificially inoculated in minced chicken samples with 5 hours of pre-enrichment was 3.4×10<sup>4</sup> CFU/mL (680 CFU/reaction). For 120 animal product samples, *Salmonella* was detected by the culture method, LAMP-LFD, LAMP-AGE, and PCR in 10/120 (8.3%). In three hundred fifty animal production environmental samples, *Salmonella* was detected in 91/350 (26%) by the culture method, equivalent to the detection rates of LAMP-LFD and LAMP-AGE, while PCR achieved 86/350 (24.6%). When comparing sensitivity, specificity, positive predictive value, and accuracy, LAMP-LFD showed the best results at 100%, 95.7%, 86.3%, and 96.6%, respectively. For Kappa index of LAMP-LFD, indicated nearly perfect agreement with culture method. Conclusion: The LAMP-LFD *Salmonella* detection, which used InvA gene, was highly specific, sensitive, and convenient for identifying *Salmonella*. Furthermore, this method could be used for *Salmonella* monitoring and primary screening in animal products and animal production environmental samples. ISSN: 27650189

**Ferdous J., Khan M.N.A., Rahman M.K., Kamal M., Reza M.S.**

*Effect of three commonly used aquaculture chemicals against enteropathogenic Escherichia coli and Salmonella spp.*

(2023) *Applied Water Science*, 13 (4), art. no. 96

ABSTRACT: Enteric bacteria such as *Escherichia coli* and *Salmonella* spp. are significant fish pathogens and related to thousands of cases of food-borne diseases every year in human. Since aquatic environments are reservoirs of these pathogens, they may contaminate the food fish and result in outbreaks. Therefore, it is crucial to reduce or eliminate these pathogens from aquaculture facilities. We tested effectiveness of three commonly used aquaculture chemicals, viz., lime, hydrogen peroxide and zeolite on bacterial load, *Escherichia coli* and *Salmonella* spp. under laboratory and earthen pond conditions where they were applied at a dose recommended for freshwater aquaculture. Results of the bacteriological study showed that lime had a significant role in reducing bacterial abundance from an initial value of  $1.5 \times 10^3$  to  $1.9 \times 10^3$  cfu/ml and  $3.9 \times 10^3$  to  $6.3 \times 10^3$  to a final value of  $1.2 \times 10^3$  to  $1.5 \times 10^3$  cfu/ml and  $1.9 \times 10^3$  to  $6.3 \times 10^3$  cfu/ml within 24 h under aquaria and pond condition, respectively. A complete inactivation of *E. coli* was observed in lime treated aquaria and ponds 24 h post-treatment, whereas *Salmonella* spp. remained unaffected in pond water but inactivated under aquarium condition after same period. However, neither hydrogen peroxide nor zeolite treatment reduced total bacterial count as well as *E. coli* and *Salmonella* even after 1-week post-treatment. It is, therefore, recommended that the water treatment chemicals should be applied in aquaculture ponds at appropriate dose, and farmers need to adopt biosecurity measures to ensure food safety. ISSN: 21905487

**Tommasoni C., Schiavon E., Lisuzzo A., Gianesella M., Merenda M., Coin P., Patregnani T., Tola S., Catania S., Barberio A.**

*Salmonella enterica serovar Dublin infection in dairy cattle: a case study on the management of an outbreak in Italy*

(2023) *Large Animal Review*, 29 (2), pp. 99 - 103

ABSTRACT: *Salmonella enterica* subsp. *enterica* serovar Dublin (S. Dublin), is a serovar adapted to cattle, causing both intestinal and systemic infections. The introduction of the bacterium leads to serious economic losses due to abortions, high mortality in calves and per-sistent infections, also representing a major health problem as zoonotic agent. The aim of this study was to describe an outbreak of S. Dublin on an Italian dairy cattle farm and to assess the effectiveness of the management protocol prepared by the Istituto Zooprofilattico Sperimentale delle Venezie (IZSVe). S. Dublin was diagnosed on a cattle farm in Northeastern Italy following the conferral at the IZSVe of a newborn calf that died from enteric syndrome. At the autoptic exam pathological findings were observed in gut, liver, pericardium, lungs, joints, lymph nodes and abomasum. Considering the pathogenesis of S. Dublin, authors decided to apply a protocol prepared by the IZSVe based both on direct and indirect prophylaxis. Particularly, an autogenous vaccine against S. Dublin prepared by the Istituto Zooprofilattico Sperimentale della Sardegna (IZS Sardegna) was administered. Screening tests were performed on fecal and milk samples (bulk tank milk) and on environmental swabs from lactating and dry cows' boxes. A pre and post-vaccination screening in 3-times (T0, T1, T2) was performed on serum, feces and milk to assess the immunization of cows and the effectiveness of the protocol itself. The first sampling took place 1-day prior immunization, the second and the third 2 and 11 months later respectively. Serological examination identified 25%, 100% and 73% positive animals at T0, T1 and T2 respectively. No fecal sample in all time-points was found positive. After vaccination only 1 milk sample turned out positive. Considering the pathogenesis of S. Dublin, the negativity of the bacteriological exams suggests a positive effect of the protocol in the reduction of clinical cases, circulation of the etiological agent and biocontainment of the infection. ISSN: 11244593

**Zarei M., Rahimi S., Fazlara A., Anvari S.E.**

*High biofilm-forming Pseudomonas strains isolated from poultry slaughterhouse surfaces: Their importance in the persistence of Salmonella enteritidis in slaughterhouses*

(2023) *International Journal of Food Microbiology*, 390, art. no. 110126

ABSTRACT: The surfaces of poultry slaughterhouse equipment are significant sources of contamination with *Pseudomonas* strains, which leads to spoilage of poultry meat during subsequent refrigerated storage. In this study, *Pseudomonas* strains with high biofilm-forming ability were isolated from different surfaces of the poultry slaughterhouse equipment, identified based on molecular data, and characterized their biofilm-forming ability. After 24 h of incubation at 25 °C, 54 out of 58 *Pseudomonas* strains produced biofilm in vitro on polystyrene microplates. Seven isolates with high-ability to produce biofilm were identified as *P. fragi* (three strains), *P. fluorescens* (two strains), *P. lundensis*

and *P. cedrina*. Despite their differences, these strains produced high amounts of biofilm in pure- and dual-species cultures with *S. enteritidis* on stainless steel surfaces. However, their ability to produce dual-species biofilms with *S. enteritidis* depends on whether *S. enteritidis* form the biofilm simultaneously with the *Pseudomonas* strains or whether *Pseudomonas* strains have already formed a biofilm. In concurrent inoculation, *S. enteritidis* participated in biofilm formation with all seven *Pseudomonas* strains with varying percent contributions. However, in delayed inoculation, *S. enteritidis* did not contribute in the biofilm formed by *P. lundensis* R26, *P. fragi* R39, and *P. fluorescens* R47. In addition to highlighting the complexity of bacterial interactions associated with *Pseudomonas* strains, these results showed that *Pseudomonas* strains can be implicated in *Salmonella* persistence in poultry slaughterhouses. ISSN: 01681605

**Chan S.H., Liao S.H., Low Y.J., Chng K.R., Wu Y., Chan J.S.H., Tan L.K.**

*A Real-Time PCR Approach for Rapid Detection of Viable Salmonella Enteritidis in Shell Eggs*

(2023) *Microorganisms*, 11 (4), art. no. 844

ABSTRACT: Rapid and robust detection assays for *Salmonella Enteritidis* (SE) in shell eggs are essential to enable a quick testing turnaround time (TAT) at the earliest checkpoint and to ensure effective food safety control. Real-time polymerase chain reaction (qPCR) assays provide a workaround for the protracted lead times associated with conventional *Salmonella* diagnostic testing. However, DNA-based analysis cannot reliably discriminate between signals from viable and dead bacteria. We developed a strategy based on an SE qPCR assay that can be integrated into system testing to accelerate the detection of viable SE in egg-enriched cultures and verify the yielded SE isolates. The specificity of the assay was evaluated against 89 *Salmonella* strains, and SE was accurately identified in every instance. To define the indicator for a viable bacteria readout, viable or heat-inactivated SE were spiked into shell egg contents to generate post-enriched, artificially contaminated cultures to establish the quantification cycle (Cq) for viable SE. Our study has demonstrated that this technique could potentially be applied to accurately identify viable SE during the screening stage of naturally contaminated shell eggs following enrichment to provide an early alert, and that it consistently identified the serotypes of SE isolates in a shorter time than conventional testing. ISSN: 20762607

**Haque M., Wang B., Mvuyekure A.L., Chaves B.D.**

*Growth behavior of Shiga toxin-producing Escherichia coli, Salmonella, and generic E. coli in raw pork considering background microbiota at 10, 25, and 40 °C*

(2023) *International Journal of Food Microbiology*, 391-393, art. no. 110134

ABSTRACT: Recent epidemiological evidence suggests that pork products may be vehicles for the transmission of Shiga toxin-producing *Escherichia coli* (STEC) to humans. The severe morbidity associated with STEC infections highlights the need for research to understand the growth behavior of these bacteria in pork products. Classical predictive models can estimate pathogen growth in sterile meat. However, competition models considering background microbiota reflect a more realistic scenario for raw meat products. The objective of this study was to estimate the growth kinetics of clinically significant STEC (O157, non-O157, and O91), *Salmonella*, and generic *E. coli* in raw ground pork using competition primary growth models at temperature abuse (10 and 25 °C) and sublethal temperature (40 °C). A competition model incorporating the No lag Buchanan model was validated using the acceptable prediction zone (APZ) method where >92 % (1498/1620) of the residual errors fell within the APZ (pAPZ > 0.70). The background microbiota (mesophilic aerobic plate counts, APC) inhibited the growth of STEC and *Salmonella* indicating a simple one-directional competitive interaction between pathogens and the mesophilic microbiota of ground pork. The maximum specific growth rate ( $\mu_{max}$ ) of all the bacterial groups was not significantly different ( $p > 0.05$ ) based on fat content (5 vs 25 %) except for generic *E. coli* at 10 °C. *E. coli* O157 and non-O157 behaved similarly in terms of  $\mu_{max}$  and maximum population density (MPD). *Salmonella* showed a similar ( $p > 0.05$ )  $\mu_{max}$  to *E. coli* O157 and non-O157 at 10 and 40 °C but a significantly higher rate ( $p < 0.05$ ) at 25 °C. STEC were more prone to be inhibited by APC than *Salmonella* at 10 and 25 °C. The  $\mu_{max}$  of O91 was lower ( $p < 0.05$ ) than other STEC and *Salmonella* at 10 and 25 °C but similar ( $p > 0.05$ ) at 40 °C. Generic *E. coli* showed a two- to five-times higher ( $p < 0.05$ )  $\mu_{max}$  ( $0.028 \pm 0.011 \log_{10} \text{CFU/h}$ ) than other bacterial groups ( $0.006 \pm 0.004$  to  $0.012 \pm 0.003 \log_{10} \text{CFU/h}$ ) at 10 °C making it a potential indicator bacteria for process control. Industry and regulators can use competitive models to develop appropriate risk assessment and mitigation strategies to improve the microbiological safety of raw pork products. ISSN: 01681605

**Morgan G., Saal M., Corr A., Jenkins C., Chattaway M.A., Pinchbeck G., Williams N.**  
*Isolation of Salmonella species of public health concern from commonly fed dried meat dog treats*

(2023) *Veterinary Record*, 192 (7), pp. no

ABSTRACT: Background: Dried non-heat-treated meat treats, such as ears, skin and tails, are popular supplementary dog foods. Previous studies have demonstrated Salmonella spp. contamination on treats, particularly in pig ears and chicken products. This small, exploratory, cross-sectional study investigated Salmonella spp. presence in dried treats available in the UK. Methods: A selection of dried treats from local pet shops and online retailers underwent bacterial culture for Salmonella spp. and subsequent antimicrobial susceptibility testing, with Salmonella serotype determined by whole genome sequencing. Results: Eighty-four samples were tested, with 16% being Salmonella spp. positive. Five Salmonella serotypes were identified, each associated with specific treat types. An antimicrobial-resistant phenotype was identified in 39% of isolates. All serotypes identified are known to cause human infection. Limitations: This study was limited by a small sample size and limited number of retail sources. Conclusion: Salmonella spp. of public health concern were present in some dried dog treats in this study. Dog owners, pet food retailers and veterinary professionals should be aware of the potential zoonotic disease risk associated with these treats, and appropriate hygiene measures, including thorough hand washing, should be utilised if they are fed. ISSN: 00424900

**Sandrasaigaran P., Kuan C.H., Son R., Gobal D., Abidin U.F.U.Z., Rukayadi Y., Hasan H.**

*Multiplex touchdown Polymerase Chain Reaction for rapid detection of Salmonella enterica subsp. enterica serovars Enteritidis and Typhimurium in food*  
(2023) *Food Research*, 7 (2), pp. 129 - 136

ABSTRACT: The Salmonella outbreak is one of the leading foodborne diseases in the world with increasing cases being reported annually. However, the current methods for Salmonella detection in foods are outdated, laborious and time-consuming. This necessitated developing a technique that is rapid for Salmonella detection in foods. Thus, the current study aimed to develop a multiplex touchdown PCR (m-TdPCR) protocol for rapid and simultaneous detection of Salmonella enterica subsp. enterica serovars Enteritidis and Typhimurium in foods. A two-phase m-TdPCR protocol was developed and optimized with primer pairs targeting the Salmonella enterica subsp. enterica (ST11/ST15-0.15 µM), serovars Enteritidis (sd*f*I gene-1.2 µM), Typhimurium (fliC gene-1.5 µM) and an internal amplification control (16S rRNA-0.08 µM). It was found that the m-TdPCR protocol is highly sensitive detecting up to 1 ng of Salmonella DNA and its specificity was verified using the in-silico method. Furthermore, the developed m-TdPCR shows no non-specific PCR amplicons and is able to detect both S. enterica ser. Enteritidis and S. enterica ser. Typhimurium in real-time when tested against the artificially contaminated food samples at up to 10<sup>-3</sup> dilutions. Therefore, the validated m-TdPCR protocol in this study can be used as a tool for rapid detection of S. enterica ser. Enteritidis and S. enterica ser. Typhimurium in food samples and this may significantly reduce any related foodborne incidences in future. ISSN: 25502166

**Arnold A.N., Sawyer J.E., Gehring K.B.**

*Longitudinal Evaluation of Salmonella in Environmental Components and Peripheral Lymph Nodes of Fed Cattle From Weaning to Finish in Three Distinct Feeding Locations*  
(2023) *Journal of Food Protection*, 86 (4), art. no. 100062

ABSTRACT: Salmonella prevalence in bovine lymph nodes (LNs) varies due to seasonality, geographic location, and feedyard environment. The objectives of this study were to (1) establish prevalence rates of Salmonella in environmental components (trough water, pen soil, individual feed ingredients, prepared rations, and fecal samples) and LNs from weaning to finish in three feeding locations, and (2) characterize recovered salmonellae. Calves (n = 120) were raised at the Texas A&M University McGregor Research Center; in lieu of beginning the backgrounding/stocker phase, thirty weanling calves were harvested. Of the remaining ninety calves, thirty were retained at McGregor and sixty were transported to commercial feeding operations (Location A or B; thirty calves each). Locations A and B have historically produced cattle with relatively "low" and "high" rates of Salmonella-positive LNs, respectively. Ten calves per location were harvested at the conclusion of (1) the backgrounding/stocker phase, (2) 60 d on feed, and (3) 165 d on feed. On each harvest day, peripheral LNs were excised. Environmental samples were obtained from each location before and after each phase, and every 30 d during the feeding period. In line with previous work, no Salmonella-positive LNs were recovered from cattle managed at Location A. Salmonella-positive LNs (30%) and environmental components (41%) were most commonly recovered from Location B. Of 7 and 36 total

serovars recovered from Salmonella-positive LN and environmental samples, respectively, Anatum was identified most frequently. Data from this study provide insight into Salmonella prevalence differences among feeding locations and the possible influence of environmental and/or management practices at each. Such information can be used to shape industry best practices to reduce Salmonella prevalence in cattle feeding operations, resulting in a decreased prevalence of Salmonella in LNs, and thus, minimizing risks to human health. ISSN: 0362028X

**McClure M., Whitney B., Gardenhire I., Crosby A., Wellman A., Patel K., McCormic Z.D., Gieraltowski L., Gollarza L., Low M.S.F., Adams J., Pightling A., Bell R.L., Nolte K., Tijerina M., Frost J.T., Beix J.A., Boegler K.A., Dow J., Altman S., Wise M.E., Bazaco M.C., Viazis S.**

*An Outbreak Investigation of Salmonella Typhimurium Illnesses in the United States Linked to Packaged Leafy Greens Produced at a Controlled Environment Agriculture Indoor Hydroponic Operation – 2021*

(2023) *Journal of Food Protection*, 86 (5), art. no. 100079

ABSTRACT: In 2021, the U.S. Food and Drug Administration (FDA), the Centers for Disease Control and Prevention (CDC), and state partners investigated a multistate outbreak of Salmonella Typhimurium illnesses linked to packaged leafy greens from a controlled environment agriculture (CEA) operation in Illinois. Thirty-one illnesses and four hospitalizations were reported in four states, with a significant epidemiologic signal for packaged leafy greens from Farm A. A traceback investigation for leafy greens included seven points of service (POS) with food exposure data from eight ill people. Each POS was supplied leafy greens by Farm A. FDA investigators observed operations at Farm A and noted that 1) the firm did not consider their indoor hydroponic pond water as agricultural water, 2) condensate dripping from the chiller water supply line inside the building, and 3) unprotected outdoor storage of packaged soilless growth media and pallets used for finished product. FDA collected 25 product, water, and environmental samples from Farm A. The outbreak strain was recovered from a water sample collected from a stormwater drainage basin located on the property adjacent to Farm A. In addition, an isolate of Salmonella Liverpool was recovered from two indoor growing ponds within the same growing house, but no illnesses were linked to the isolate. Farm A voluntarily recalled all implicated products and provided their root cause analysis (RCA) and return-to-market plan to FDA. While the source and route of the contamination were not determined by the RCA, epidemiologic and traceback evidence confirmed the packaged salads consumed by ill persons were produced by Farm A. This was the first investigation of a multistate foodborne illness outbreak associated with leafy greens grown in a CEA operation. This outbreak demonstrated the need for growers using hydroponic methods to review their practices for potential sources and routes of contamination and to reduce food safety risks when identified. ISSN: 0362028X

**Arnold M., Lawes J., Davies R.H., Evans S.**

*Study of Salmonella detection in laying hens using a Bayesian model*

(2023) *Zoonoses and Public Health*, 70 (3), pp. 248 - 255

ABSTRACT: As part of the measures to reduce the prevalence of Salmonella in poultry in the UK, National Control Programmes (NCPs) have been implemented. These involve regular statutory testing of poultry holdings to monitor and estimate the prevalence of Salmonella in the national flock population and to control Salmonella on holdings with positive flocks, especially those serovars most identified with human illness: Salmonella Enteritidis (SE) and S. Typhimurium (ST). It is very important to ensure that the level of testing is appropriate so that it is sufficiently effective to identify positive flocks and to monitor prevalence, but also efficient in the use of resources. The aim of this study was to estimate the sensitivity of both the Operator and Competent Authority (CA) Official sampling used to detect infected flocks, and to also estimate the true proportion of infected holdings of commercial laying flocks in GB each year of the NCP, along with the trend of any changes in prevalence for both SE/ST and non-SE/ST. A Bayesian model was developed to estimate the sensitivity of both Operator and CA Official sampling from the NCP data 2009–2018, and to estimate the true prevalence of infected holdings. The model estimate for the prevalence of infected holdings for the first complete year of the NCP was 3.9% (95% Credible Interval (CI) 2.8–6.2%) for non-SE/ST and 0.8% (95% CI: 0.4%–1.5%) for SE/ST. Prevalence had reduced to 1.6% (non-SE/ST) (95% CI 1.0%–2.5%) and 0.2% (SE/ST) (95% CI 0.1%–0.4%) in 2018. Results indicated a very low sensitivity of Operator sampling (~9%), but a much higher sensitivity of CA Official sampling (~44%). The true prevalence of Salmonella infected holdings in the UK had a mean average reduction of 10.6% (95% CI: 6.3%–15.1%) per annum (non-SE/ST) and 15.9% (95% CI: 6.0%–19.8%) annual reduction for SE/ST. This has shown the effectiveness of the NCP for

Salmonella in commercial laying flocks, with reductions in Salmonella overall more or less equal to the target reduction for regulated serovars of 10% per annum. The true prevalence of SE/ST was estimated to be below the final target of less than 2% in every year and was below 0.5% at the end of the 10 year period. ISSN: 18631959

**Thames H.T., Pokhrel D., Willis E., Rivers O., Dinh T.T.N., Zhang L., Schilling M.W., Ramachandran R., White S., Sukumaran A.T.**

*Salmonella Biofilm Formation under Fluidic Shear Stress on Different Surface Materials (2023) Foods, 12 (9), art. no. 1918*

ABSTRACT: This study characterized biofilm formation of various Salmonella strains on common processing plant surface materials (stainless steel, concrete, rubber, polyethylene) under static and fluidic shear stress conditions. Surface-coupons were immersed in well-plates containing 1 mL of Salmonella (6 log CFU/mL) and incubated aerobically for 48 h at 37 °C in static or shear stress conditions. Biofilm density was determined using crystal violet assay, and biofilm cells were enumerated by plating on tryptic soy agar plates. Biofilms were visualized using scanning electron microscopy. Data were analyzed by SAS 9.4 at a significance level of 0.05. A surface-incubation condition interaction was observed for biofilm density ( $p < 0.001$ ). On stainless steel, the OD600 was higher under shear stress than static incubation; whereas, on polyethylene, the OD600 was higher under static condition. Enumeration revealed surface-incubation condition ( $p = 0.024$ ) and surface-strain ( $p < 0.001$ ) interactions. Among all surface-incubation condition combinations, the biofilm cells were highest on polyethylene under fluidic shear stress (6.4 log/coupon;  $p < 0.001$ ). Biofilms of *S. Kentucky* on polyethylene had the highest number of cells (7.80 log/coupon) compared to all other strain-surface combinations ( $p < 0.001$ ). Electron microscopy revealed morphological and extracellular matrix differences between surfaces. Results indicate that Salmonella biofilm formation is influenced by serotype, surface, and fluidic shear stress. ISSN: 23048158

**Wang J., Vaddu S., Bhumanapalli S., Mishra A., Applegate T., Singh M., Thippareddi H.**

*A systematic review and meta-analysis of the sources of Salmonella in poultry production (pre-harvest) and their relative contributions to the microbial risk of poultry meat (2023) Poultry Science, 102 (5), art. no. 102566*

ABSTRACT: Salmonella is a major foodborne pathogen associated with poultry and poultry products and a leading cause for human salmonellosis. Salmonella is known to transmit in poultry flocks both vertically and horizontally. However, there is a lack of knowledge on relative contribution of the factors on Salmonella prevalence in poultry live production system including hatchery, feed, water, environment-interior, and -exterior. Therefore, a systematic review and meta-analysis was conducted to quantify the potential sources of Salmonella during preharvest and their relative contributions to the microbial risk of poultry meat. A total of 16,800 studies identified from Google Scholar and 37 relevant studies were included in the meta-analysis for relative contributions to Salmonella positivity on broilers after applying exclusion criteria. A generalized linear mixed model approach combined with logit transformation was used in the current study to stabilize the variance. The analysis revealed that the hatchery is the most significant contributor of Salmonella with a prevalence of 48.5%. Litter, feces, and poultry house internal environment were the other 3 major contributing factors with a prevalence of 25.4, 16.3, and 7.9%, respectively. Moreover, poultry house external environment (4.7%), feed (4.8%), chicks (4.7%), and drinker water also contributed to the Salmonella positivity. Results from this meta-analysis informed the urgent need for controls in live production to further reduce Salmonella in fresh, processed poultry. The control strategies can include eliminating the sources of Salmonella and incorporating interventions in live production to reduce Salmonella concentrations in broilers. ISSN: 00325791

**Qiu Y., Ozturk S., Cui X., Qin W., Wu Q., Liu S.**

*Increased heat tolerance and transcriptome analysis of Salmonella enterica Enteritidis PT 30 heat-shocked at 42 °C (2023) Food Research International, 167, art. no. 112636*

ABSTRACT: In this study, we compared the heat tolerance parameter (D65°C) values of Salmonella enterica serovar Enteritidis PT 30 (*S. Enteritidis*) heat adapted at different degrees (at 42 °C for 20–180 min) and cultivated using two methods. The treated group with the highest D65°C value (LP-42 °C-60 min) and the untreated groups (Control-TSB and Control-TSA) were subjected to transcriptome analysis. Heat-adaptation increased the D65°C values of *S. Enteritidis* by 24.5–60.8%. The D65°C values of the LP-42 °C-60 min group ( $1.85 \pm 0.13$  min, 7.7% higher) was comparable to that of the Control-TSA. A total

of 483 up- and 443 downregulated genes of *S. enteritidis* were identified in the LP-42 °C-60 min group (log<sub>2</sub>fold change > 1, adjusted p-value < 0.05). Among these genes, 5 co-expressed and 15 differentially expressed genes in the LP-42 °C-60 min and Control-TSA groups possibly contributed to the high D65°C values of *S. Enteritidis*. The Rpo regulon was involved in the heat adaptation of *S. Enteritidis*, as evidenced by the significant upregulation of rpoS, rpoN, and rpoE. KEGG enrichment pathways, such as biosynthesis of secondary metabolites, tricarboxylic acid, and ribosomes were identified and mapped to reveal the molecular mechanisms of *S. enteritidis* during heat adaptation. This study quantified the enhanced heat tolerance of *S. Enteritidis* heat adapted at different degrees of heat-adaptation. The results of this study may serve as a basis for elucidating the molecular mechanisms underlying the enhanced heat tolerance at the transcriptome level. ISSN: 09639969

**Shen A.Q., Dalen A., Bankers L., Matzinger S.R., Schwensohn C., Patel K., Hise K.B., Pereira E., Cripe J., Jervis R.H.**

*Multistate Outbreak of Salmonella Thompson Infections Linked to Seafood Exposure - United States, 2021*

(2023) *MMWR. Morbidity and mortality weekly report*, 72 (19), pp. 513 - 516

ABSTRACT: In July 2021, the Colorado Department of Public Health and Environment (CDPHE) laboratory identified a cluster of five *Salmonella enterica* serotype Thompson isolates related to one another within one allele difference, using whole genome multilocus sequence typing (wgMLST). These five isolates, submitted to the public health laboratory as is routine process for confirmatory testing of *Salmonella*, were highly related to those identified in a 2020 multistate investigation, during which traceback was conducted for sushi-grade tuna and salmon; a common supplier was not identified. The 2021 investigation commenced on August 5, 2021, with five patients living in Colorado, and one each in Missouri, Washington, and Wisconsin. During August-December 2021, CDC, CDPHE, public health and regulatory officials in several states, and the Food and Drug Administration (FDA) conducted epidemiologic, environmental, and laboratory investigations of this multistate outbreak of *Salmonella Thompson*. Isolates were genetically related to one another and to 2020 isolates within zero to one allele difference. Implicated seafood products were traced to a single seafood distributor, in which the outbreak strain was identified through environmental sampling, and in which inspection identified inadequate sanitization and opportunities for cross-contamination of raw fish. The distributor issued a voluntary recall of 16 seafood items with high potential for contamination and completed remediation actions. This outbreak illustrated the importance of effective cleaning and sanitizing procedures and implementation of controls. When multiple products are recalled during an outbreak investigation, collaboration between public health agencies and implicated facilities can help provide food safety information to restaurants, retailers, and consumers, and to ensure disposal of all recalled products. ISSN: 1545861X

**Yan S., Jiang Z., Zhang W., Liu Z., Dong X., Li D., Liu Z., Li C., Liu X., Zhu L.**

*Genomes-based MLST, cgMLST, wgMLST and SNP analysis of Salmonella Typhimurium from animals and humans*

(2023) *Comparative Immunology, Microbiology and Infectious Diseases*, 96, art. no.

ABSTRACT: *Salmonella Typhimurium* (*S. Typhimurium*) is an important food-borne and zoonotic pathogen that causes salmonellosis. With the development of whole genome sequencing (WGS), genome-based typing has been widely applied to bacteriology. In this study, we investigated genotyping and phylogenetic clusters of *S. Typhimurium* isolates from humans and animals in different provinces (including Beijing, Shandong, Guangxi, Shaanxi, Henan, and Shanghai) of China during 2009–2018 using multi locus sequence typing (MLST), core genome MLST (cgMLST), whole genome MLST (wgMLST) and single nucleotide polymorphism (SNP) based on WGS. 29 *S. Typhimurium* isolates from chicken (n = 22), sick pigeon (n = 2), patients (n = 4) and sick swine (n = 1) were tested. MLST analysis showed *S. Typhimurium* strains were divided into four STs, namely ST19 (n = 14), ST34 (n = 12), ST128 (n = 2) and ST1544 (n = 1). cgMLST and wgMLST divided 29 strains into 27 cgSTs and 29 wgST, respectively. Phylogenetic clustering showed that all isolates were divided into 4 clusters and 4 singletons. SNP analysis was used to examine MLST, cgMLST, wgMLST analysis. Finally, comparisons of MLST, cgMLST, wgMLST, and SNP were analyzed and the results showed their precision increased in order. In summary, genomic typing and phylogenetic relationships of 29 *S. Typhimurium* strains from different sources in China were analyzed. These findings were beneficial to investigate molecular pathogenesis, bacterial diversity, and traceability analysis of *Salmonella*. ISSN: 01479571

**Gast R.K., Jones D.R., Guraya R., Garcia J.S., Karcher D.M.**

*Applied Research Note: Internal organ colonization by Salmonella Enteritidis in experimentally infected layer pullets after rearing in conventional cage or cage-free housing*

(2023) *Journal of Applied Poultry Research*, 32 (2), art. no. 100334

ABSTRACT: Invasive Salmonella Enteritidis infection involving the reproductive organs of laying hens can result in the production of internally contaminated eggs, which continue to be prominent sources of disease transmission to consumers. The poultry housing environment exerts substantial influences on the dissemination and prevalence of S. Enteritidis in laying flocks, and the ongoing transition of the egg industry toward cage-free housing has raised new questions about the food safety ramifications. The present study assessed internal organ colonization by S. Enteritidis in egg-type pullets reared in 2 different housing systems (conventional cage and cage-free). At 16 wk of age, 4 groups of 72 pullets were moved into isolation rooms simulating commercial cage-free barns; 24 pullets in each of 2 rooms were orally inoculated with S. Enteritidis immediately after transfer and 24 pullets in each of the other 2 rooms were similarly infected at 19 wk. At 6 to 12 d post-inoculation, all pullets were euthanized and samples of liver, spleen, and intestinal tract were removed for bacteriologic culturing. S. Enteritidis was isolated significantly ( $P < 0.05$ ) more frequently from spleens and intestines from infected pullets that had been reared in cages than from those reared in cage-free housing, especially when the birds were infected on the day after transfer from the rearing facility. S. Enteritidis was also found significantly more often in livers from birds infected at 19 wk than at 16 wk, especially among birds reared in cage-free housing. These results reinforce the importance of attentive pathogen risk reduction practices at this critical phase in the productive life of egg-laying flocks. ISSN: 10566171

**Rezende A.C.B., Alegbeleye O., Silva Oliveira S.B., Santos J.S., Granato D., Felix**

*Survival behavior of Salmonella enterica in the peel, stalk, pulp, and tip of green, mature, and ripe tropical fruits: Avocado [Persea americana] and sugar apple [Annona squamosa]*  
(2023) *LWT*, 182, art. no. 114813

ABSTRACT: The aim of this study was to determine the growth potential ( $\delta$ ) of Salmonella enterica in various parts (peel, stalk, pulp, and tip) of avocado and sugar apple at different maturation stages (green, mature, and ripe) over time. In addition, physicochemical parameters of the fruits were monitored using standard methods. There were statistically significant differences in the growth potential of S. enterica in the peel, stalk, pulp, and tip of avocado and sugar apple at different maturation stages (green, mature, and ripe) over time. Amongst the four parts of the fruits, the highest growth potential ( $\delta$  up to  $4.4 \pm 0.01$  in ripe custard apple) was obtained in the pulps. Nonetheless, the growth potential varied with the maturation stage and fruit type. For instance, while the pulp of mature sugar apple supported growth on the 4th day ( $\delta = 0.5 \pm 0.01$ ) only, the pulp of ripe sugar apple supported the growth of S. enterica throughout post-inoculation storage with  $\delta = 2.4 \pm 0.05$ ,  $2.7 \pm 0.06$ ,  $4.2 \pm 0.05$  and  $4.4 \pm 0.01$  on the 2nd, 4th, 6th and 8th day post inoculation storage, respectively. ISSN: 00236438

**Suhalim R., Tay A., Ceylan E.**

*Thermal resistance of shiga toxin-producing Escherichia coli, Salmonella and Enterococcus faecium in peanut butter, oatmeal raisin, and chocolate chip cookies*  
(2023) *LWT*, 182, art. no. 114817

ABSTRACT: Thermal-death-time characteristics, D and z-values of shiga toxin-producing Escherichia coli (STEC), Salmonella, and Enterococcus faecium NRRL B-2354 were investigated in peanut butter, oatmeal, and chocolate chip cookie formulations at three moisture levels. E. faecium was shown to be more resistant than Salmonella, followed by E. coli at all water activity levels. E. faecium was at least 1.5 times more heat resistant compared to Salmonella under the same moisture and temperature conditions for all cookie formulations. The E. faecium to Salmonella ratio was larger at higher moisture levels. E. faecium was found to be suitable as a surrogate for Salmonella in validating soft cookie baking. Among the three formulations, Salmonella was the most heat resistant in peanut butter cookies. At the lowest moisture and water activity level (8% moisture,  $a_w$  0.421) of peanut butter cookies the D-values for E. coli were 14.01, 3.56, 1.73 min at 65, 70, 75 °C, while the D-values for Salmonella were 4.97, 2.59, 1.23 min, and for E. faecium were 15.50, 5.02, 2.02 min at 75, 80, and 85 °C, respectively. The z-values for E. coli, Salmonella, and E. faecium, were 11.02, 16.45 and 11.31 °C, respectively. ISSN: 00236438

**Ehrhardt K., Becker A.-L., Grassl G.A.**

*Determinants of persistent Salmonella infections*



(2023) *Current Opinion in Immunology*, 82, art. no. 102306

ABSTRACT: Persistent bacterial infections constitute an enormous challenge for public health. Amongst infections with other bacteria, infections with typhoidal and nontyphoidal *Salmonella enterica* serovars can result in long-term infections of the human and animal host. Persistent infections that are asymptomatic are difficult to identify and thus can serve as a silent reservoir for transmission. Symptomatic persistent infections are often difficult to treat as they harbor a combination of antibiotic-tolerant and antibiotic-resistant bacteria and boost the spread of genetic antibiotic resistance. In the last couple of years, the field has made some major progress in understanding the role of persisters, their reservoirs as well as their interplay with host factors in persistent *Salmonella* infections.

ISSN: 09527915

**Pye H.V., Thilliez G., Acton L., Kolenda R., Al-Khanaq H., Grove S., Kingsley R.A.**

*Strain and serovar variants of Salmonella enterica exhibit diverse tolerance to food chain-related stress*

(2023) *Food Microbiology*, 112, art. no. 104237

ABSTRACT: Non-Typhoidal *Salmonella* (NTS) continues to be a leading cause of foodborne illness worldwide. Food manufacturers implement hurdle technology by combining more than one approach to control food safety and quality, including preservatives such as organic acids, refrigeration, and heating. We assessed the variation in survival in stresses of genotypically diverse isolates of *Salmonella enterica* to identify genotypes with potential elevated risk to sub-optimal processing or cooking. Sub-lethal heat treatment, survival in desiccated conditions and growth in the presence of NaCl or organic acids were investigated. *S. Gallinarum* strain 287/91 was most sensitive to all stress conditions. While none of the strains replicated in a food matrix at 4 °C, *S. Infantis* strain S1326/28 retained the greatest viability, and six strains exhibited a significantly reduced viability. A *S. Kedougou* strain exhibited the greatest resistance to incubation at 60 °C in a food matrix that was significantly greater than *S. Typhimurium* U288, *S. Heidelberg*, *S. Kentucky*, *S. Schwarzengrund* and *S. Gallinarum* strains. Two isolates of monophasic *S. Typhimurium*, S04698-09 and B54Col9 exhibited the greatest tolerance to desiccation that was significantly more than for the *S. Kentucky* and *S. Typhimurium* U288 strains. In general, the presence of 12 mM acetic acid or 14 mM citric acid resulted in a similar pattern of decreased growth in broth, but this was not observed for *S. Enteritidis*, and *S. Typhimurium* strains ST4/74 and U288 S01960-05. Acetic acid had a moderately greater effect on growth despite the lower concentration tested. A similar pattern of decreased growth was observed in the presence of 6% NaCl, with the notable exception that *S. Typhimurium* strain U288 S01960-05 exhibited enhanced growth in elevated NaCl concentrations. ISSN: 07400020

**Chanamé Pinedo L., Van Goethem N., Mallioris P., Pacholewicz E., Pijnacker R., Franz E., Mughini-Gras L.**

*Assessing potential determinants of the stagnating trend in Salmonella Enteritidis human infections in Europe and options for intervention: A multi-criteria decision analysis*

(2023) *One Health*, 16, art. no. 100535

ABSTRACT: Background: After years of significant decline, the incidence of *Salmonella enterica* serotype Enteritidis (SE) human infections in Europe has started stagnating in recent years. The reasons for this stagnation remain largely unclear and are possibly multifactorial and interconnected in nature. We assessed and ranked several potential determinants of the stagnating SE trend in Europe, as well as different options for intervention at the level of poultry health and production, public health (infra)structure, and pathogen biology. Methods: A Multi-Criteria Decision-Analysis (MCDA) approach based on the Analytical Hierarchy Process was used. Through two separate surveys, a European panel of *Salmonella* experts first provided weights for several pre-defined criteria and subsequently scored different potential determinants and options for intervention (i.e. alternatives) against the criteria, during 2020–21. The weighting and scoring were based on Saaty's pairwise comparisons. The final ranking of the alternatives was derived from the summation of the products of each criterion weight with the score of the corresponding alternative. Sensitivity analyses were performed to assess the impact of different methodological choices, including European regions, and domains of expertise on the ranking of the determinants and options for intervention. Results: The first and second-ranked determinants of the stagnated trend in human SE infections were related to poultry health and production, namely "inadequacies of sampling programmes" and "premature relaxation of control measures". This ranking agreed with the ranking of the options for intervention, which were also those at the poultry health and production level, specifically "stricter biosecurity", "improving sampling", and "better/increased vaccination". Differences in rankings were observed among European regions and domains of expertise.

Conclusions: The rankings of potential determinants and options for intervention for the stagnating SE trend in Europe pointed to the level of poultry health and production. Salmonella-control activities in poultry in Europe are harmonized across countries since many years, but the results of this study suggest that further improvements may be necessary for some countries. A multidisciplinary collaboration among veterinarians, public health professionals, and microbiologists is needed to further understand the origins of the stagnating SE trend and to identify effective interventions in order to reverse the trend, contextually in a given country, following a One Health approach. ISSN: 23527714

**Taboada A.C., Glass K., Chateau D., Pavic A.**

*A systematic review and meta-analysis of the effect of dietary additives, vaccinations, and processing aids as control measures for Salmonella spp. in chicken meat (2023) Applied Food Research, 3 (1), art. no. 100254*

ABSTRACT: Salmonellosis is a leading cause of foodborne disease and is often associated with contaminated chicken meat. Research to reduce the risk of Salmonella contamination of chicken products has led to the development of various through-chain control measures. This review aimed to statistically investigate and evaluate the effects of dietary additives, vaccines, and processing aids on Salmonella contamination in chickens. Eligibility criteria was set to include randomised controlled trials (published 2009-2019 in English) that treated meat chickens or products with a dietary additive, vaccine or processing aid and measured Salmonella in the ceca or product within 24 hours. The confidence in cumulative evidence was evaluated based on the GRADE method. Meta-analyses were performed for the effects of each intervention on Salmonella using standardised mean differences (SMD). Additional sub-categories were also meta-analysed and included in a random effects regression model to investigate interactions between intervention effects and subgroups. Many eligible studies included multi-arm interventions, where a total of 20 dietary additive (from 13 studies), 12 vaccination (from 9 studies) and 70 processing aid (from 27 studies) outcomes were synthesised. Most of the outcomes were judged as having an unclear risk of bias, mostly due to no mention of treatment allocation concealment or blinding. The random effects meta-analysis estimated that the average effect of dietary additives (SMD = -1.99; 95% CI: -2.66 to -1.33; P value < 0.001), vaccinations (SMD = -1.21; 95% CI: -1.74 to -0.69; P value < 0.001) and processing aids (SMD = -1.27; 95% CI: -1.57 to -0.97; P value < 0.001) significantly reduced Salmonella. The Chi2 test for heterogeneity (P value < 0.001 for all three analyses) and the I2 statistic (72.5%, 82.8% and 85.8%, respectively) indicated high levels of heterogeneity across the intervention effects. The certainty of the gathered evidence was very low, mainly due to consistently unclear or high risk of bias assessments, high levels of heterogeneity in treatment effects that could not be explained by subgroup analyses, unit of analysis errors and publication bias identified in funnel plots. Although the meta-analyses found large reductions in Salmonella, it is likely that the true effects were smaller due to limitations present. Further research and transparency in methods is required to identify causes of heterogeneity and provide reliable recommendations to the industry. ISSN: 27725022