

# NEWSLETTER

European Union Reference Laboratory for *Salmonella*

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## Editorial Note

Bilthoven, 1 April 2022

Dear colleague,

I hope you are all doing well. I am not sure about the current situation in your country concerning the **SARS-CoV-2** virus, but in the Netherlands most restrictions have been phased out and we are more or less back to the 'old normal' situation. Although the number of infections are still quite high, the severity of the illness of the current virus variant is much lower than the former variants for most people. Unfortunately we are now facing a terrible other crisis in Ukraine which also may influence the daily lives of many of us. I do hope that this horrible war ends soon and we will have peace again in Europe.

In January/February 2022, the evaluation of the serotyping results of the **PT on typing of *Salmonella* 2021** was performed. Before the end of February 2022, the participants received their own results as well as the interim summary report containing the results of all participants. The interim summary report is also available at the EURL-*Salmonella* website:

<https://www.eurlsalmonella.eu/documenten/interim-summary-report-eurl-salmonella-pt-serotyping-2021> Only one participant did not meet the level of good performance and a follow-up study has recently been organised. In addition to the obligatory serotyping part, this study included also a part on cluster analysis. The results on this (optional) cluster analysis part, using MLVA and/or WGS data, are under evaluation and will be reported separately in the coming months.

Contrary to former years, we do not organise a PT on detection of *Salmonella* in a food sample this spring. This is because we will organise the **interlaboratory study (ILS) for determination of the performance characteristics** of draft ISO/DTS 6579-4 (identification of monophasic *Salmonella* Typhimurium by PCR) in May-July 2022. By the end of February a call for participants for the ILS was sent to the members of ISO/TC34/SC9, to the members of ISO-WG10 and to the NRLs-*Salmonella*. The deadline for subscribing to the ILS for analysing one or more PCR protocols of draft ISO/DTS 6579-4 was 31 March 2022. We are very happy to notice that the number of registrations fulfill very well the minimum number of collaborators (we will need at least 10 valid data sets per PCR protocol). In April 2022 we will inform all subscribers with more details about the ILS.

Instead of organising the **PT on detection of *Salmonella* in Food** in spring, it will be organised as a **combined PT** with the one on **detection of *Salmonella* in samples from the primary production stage (PPS)** in September 2022. The timetable of this combined PT is included in this Newsletter.

Earlier this year we started with the organisation of the **EURL-*Salmonella* workshop 2022**. We have investigated the possibility of organising the workshop as an hybrid meeting. However, only a very limited number of NRLs registered an in person participant, so that we finally decided to change the workshop from an hybrid meeting to (again) an online meeting. The dates remain unchanged: Monday 23 May and Tuesday 24 May 2022. It is a pity that we cannot meet in person yet, but in this time of crisis it is also fully understandable that people prefer not to travel. We still hope for better times next year.

In February 2022, the following EURL-*Salmonella* report was published:  
Mooijman, K.A. The 26<sup>th</sup> EURL-*Salmonella* workshop 28 May 2021, Online.  
National Institute for Public Health and the Environment, Bilthoven, the Netherlands. RIVM Report no.: 2021-0130.  
<https://www.rivm.nl/bibliotheek/rapporten/2021-0130.pdf>

Best wishes,  
Kirsten Mooijman  
Coordinator EURL-*Salmonella*

## Contribution of the EURL-*Salmonella*

### Timetable EURL-*Salmonella* combined Proficiency Test Primary Production Stage-Food 2022 Detection of *Salmonella* in hygiene sponges

Week (2022)	Date	Subject
27-35		E-mailing the link to the registration form for the detection study. Please <b>register by 31 August 2022</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 26 September 2022	Shipment of the parcels to the participants as Biological Substance Category B (UN 3373).
40	Monday 3 October 2022	Performance of the Proficiency Test.
44	4 November 2022 at the latest	Deadline for completing the result form: <b>4 November 2022</b> (23:59h CET) After this deadline the result form will be closed.
	December 2022	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: January – March 2022

**Tanui, C.K., Karanth, S., Njage, P.M.K., Meng, J., Pradhan, A.K.**

*Machine learning-based predictive modeling to identify genotypic traits associated with Salmonella enterica disease endpoints in isolates from ground chicken (2022) LWT, 154, art. no. 112701, .*

ABSTRACT: As the cost of genome sequencing of foodborne pathogens decreases, it has become possible to sequence a large number of isolates and evaluate those using machine learning algorithms. This study aimed to utilize machine learning algorithms to predict the disease endpoints in untagged *Salmonella* genome sequences isolated from ground chicken. Our models recognized genetic patterns in the test dataset based on our training dataset obtained from an extensive literature review, using a semi-supervised approach. Using known genotypes as input features, the semi-supervised random forest model showed the highest overall accuracy of 0.94 (95% confidence interval: 0.85–0.99), and a Kappa value of 0.82, and predicted 87% of the disease endpoints. The model predicted genes associated with specific disease endpoints that were associated with virulence, which could be used as features in predictive modeling endeavors in the future. Our machine learning approach would be useful in different areas of food safety, including identifying pathogen sources, predicting antibiotic resistance, and risk assessment of foodborne pathogens. ISSN: 00236438

**Vázquez, X., García, V., Fernández, J., Bances, M., de Toro, M., Ladero, V., Rodicio, R., Rodicio, M.R.**

*Colistin Resistance in Monophasic Isolates of Salmonella enterica ST34 Collected From Meat-Derived Products in Spain, With or Without CMY-2 Co-production (2022) Frontiers in Microbiology, 12, art. no. 735364, .*

ABSTRACT: Colistin is a last-resort antibiotic in fighting severe infections caused by multidrug resistant Gram negative pathogens in hospitals. Zoonotic bacteria acquire colistin resistance in animal reservoirs and mediate its spread along the food chain. This is the case of non-typhoid serovars of *Salmonella enterica*. Colistin-resistant *S. enterica* in foods represents a threat to human health. Here, we assessed the prevalence of colistin-resistance in food-borne isolates of *S. enterica* (2014–2019; Asturias, Spain), and established the genetic basis and transferability of this resistance. Five out of 231 isolates tested (2.2%) were resistant to colistin. Four of them, belonging to the European monophasic ST34 clone of *S. Typhimurium*, were characterized in the present study. They were collected from pork or pork and beef meat-derived products, either in 2015 (three isolates) or 2019 (one isolate). Molecular typing with XbaI-PFGE and plasmid profiling revealed distinct patterns for each isolate, even though two of the 2015 isolates derived from the same sample. The MICs of colistin ranged from 8 to 16 mg/L. All isolates carried the *mcr-1.1* gene located on conjugative plasmids of the incompatibility groups IncX4 (2015 isolates) or IncHI2 (2019 isolate). Apart from colistin resistance, the four isolates carried chromosomal genes conferring resistance to ampicillin, streptomycin, sulfonamides and tetracycline [*bla*TEM-1, *strA-strB*, *sul2*, and *tet(B)*] and heavy metals, including copper and silver (*silESRCFBAGP* and *pcoGE1ABCDRSE2*), arsenic (*arsRSD2A2BCA1D1*) ± mercury (*merEDACPTR*), which are characteristically associated with the European ST34 monophasic clone. The 2019 isolate was also resistant to other antibiotics, comprising third generation cephalosporins and cephamycins. The latter phenotype was conferred by the *bla*CMY-2 gene located on an Inc11-I(α)-ST2 plasmid. Results in the present study identified meat-derived products as a reservoir of a highly successful clone harboring transferable plasmids which confer resistance to colistin and other clinically important antibiotics. An important reduction in the number of food-borne *S. enterica* detected during the period of the study, together with the low frequency of colistin resistance, underlines the success of One Health initiatives, such as those implemented at the UE, to control zoonotic bacteria along the food chain and to halt the spread of antimicrobial resistance. ISSN: 1664302X

**Shelton, C.D., Yoo, W., Shealy, N.G., Torres, T.P., Zieba, J.K., Calcutt, M.W., Foegeding, N.J., Kim, D., Kim, J., Ryu, S., Byndloss, M.X.**

*Salmonella enterica* serovar Typhimurium uses anaerobic respiration to overcome propionate-mediated colonization resistance  
(2022) *Cell Reports*, 38 (1), art. no. 110180, .

**ABSTRACT:** The gut microbiota benefits the host by limiting enteric pathogen expansion (colonization resistance), partially via the production of inhibitory metabolites. Propionate, a short-chain fatty acid produced by microbiota members, is proposed to mediate colonization resistance against *Salmonella enterica* serovar Typhimurium (S. Tm). Here, we show that S. Tm overcomes the inhibitory effects of propionate by using it as a carbon source for anaerobic respiration. We determine that propionate metabolism provides an inflammation-dependent colonization advantage to S. Tm during infection. Such benefit is abolished in the intestinal lumen of *Salmonella*-infected germ-free mice. Interestingly, S. Tm propionate-mediated intestinal expansion is restored when germ-free mice are monocolonized with *Bacteroides thetaiotaomicron* (B. theta), a prominent propionate producer in the gut, but not when mice are monocolonized with a propionate-production-deficient B. theta strain. Taken together, our results reveal a strategy used by S. Tm to mitigate colonization resistance by metabolizing microbiota-derived propionate.  
ISSN: 22111247

**Guo, D., Bai, Y., Fei, S., Yang, Y., Li, J., Yang, B., Lü, X., Xia, X., Shi, C.**

*Effects of 405 ± 5-nm LED Illumination on Environmental Stress Tolerance of Salmonella Typhimurium in Sliced Beef*  
(2022) *Foods*, 11 (2), art. no. 136, .

**ABSTRACT:** *Salmonella Typhimurium* is a widely distributed foodborne pathogen and is tolerant of various environmental conditions. It can cause intestinal fever, gastroenteritis and bacteremia. The aim of this research was to explore the effect of illumination with 405 nm light-emitting diodes (LEDs) on the resistance of S. Typhimurium to environmental stress. Beef slices contaminated with S. Typhimurium were illuminated by 405 nm LEDs ( $18.9 \pm 1.4$  mW/cm<sup>2</sup>) for 8 h at 4° C; controls were incubated in darkness at 7° C. Then, the illuminated or non-illuminated (control) cells were exposed to thermal stress (50, 55, 60 or 65° C); oxidative stress (0.01% H<sub>2</sub>O<sub>2</sub> [v/v]); acid stress (simulated gastric fluid [SGF] at pH 2 or 3); or bile salts (1%, 2%, or 3% [w/v]). S. Typhimurium treated by 405 nm LED irradiation showed decreased resistance to thermal stress, osmotic pressure, oxidation, SGF and bile salts. The transcription of eight environmental tolerance-related genes were downregulated by the illumination. Our findings suggest the potential of applying 405 nm LED-illumination technology in the control of pathogens in food processing, production and storage, and in decreasing infection and disease related to S. Typhimurium. ISSN: 23048158

**Bashir, A., Lambert, P.A., Stedman, Y., Hilton, A.C.**

*Combined Effect of Temperature and Relative Humidity on the Survival of Salmonella Isolates on Stainless Steel Coupons*  
(2022) *International Journal of Environmental Research and Public Health*, 19 (2), art. no. 909, .

**ABSTRACT:** The survival on stainless steel of ten *Salmonella* isolates from food factory, clinical and veterinary sources was investigated. Stainless steel coupons inoculated with *Salmonella* were dried and stored at a range of temperatures and relative humidity (RH) levels representing factory conditions. Viability was determined from 1 to 22 days. Survival curves obtained for most isolates and storage conditions displayed exponential inactivation described by a log-linear model. Survival was affected by environmental temperatures and RH with decimal reduction times (DRTs) ranging from <math>\lt; 1</math> day to 18 days. At 25° C/15% RH, all isolates survived at levels of 10<sup>3</sup> to 10<sup>5</sup> cfu for >22 days. Furthermore, temperatures and RH independently influenced survival on stainless steel; increasing temperatures between 10° C and 37° C and increasing RH levels from 30–70% both decreased the DRT values. Survival curves displaying a shoulder followed by exponential death were obtained for three isolates at 10° C/70% RH. Inactivation kinetics for these were described by modified Weibull models, suggesting that cumulative injury occurs before cellular inactivation. This study highlights the need to control temperature and RH to limit microbial persistence in the food manufacturing environment, particularly during the factory shut-down period for cleaning when higher temperature/humidity levels could be introduced. ISSN: 16617827

**Bertoldi, B., Bardsley, C.A., Pabst, C.R., Baker, C.A., Gutierrez, A., De, J., Luo, Y., Schneider, K.R.**



*Influence of Free Chlorine and Contact Time on the Reduction of Salmonella Cross-Contamination of Tomatoes in a Model Flume System*  
(2022) *Journal of Food Protection*, 85 (1), pp. 22-26.

**ABSTRACT:** The process of washing tomatoes in dump (flume) tanks has been identified as a potential source of cross-contamination. This study's objective was to assess the potential for *Salmonella enterica* cross-contamination at various inoculation levels in the presence of free chlorine (HOCl) and organic matter. Uninoculated tomatoes were introduced into a laboratory-based model flume containing tomatoes inoculated with a cocktail of five rifampin-resistant *S. enterica* serovars at 104, 106, or 108 CFU per tomato in water containing 0 or 25 mg/L HOCl and 0 or 300 mg/L chemical oxygen demand (COD). Uninoculated tomatoes exposed to the inoculated tomatoes were removed from the water after 5, 30, 60, and 120 s and placed in bags containing tryptic soy broth supplemented with rifampin and 0.1% sodium thiosulfate. Following incubation, enrichment cultures were plated on tryptic soy agar supplemented with rifampin and xylose lysine deoxycholate agar to determine the presence of *Salmonella*. HOCl and pH were measured before and after each trial. The HOCl in water containing 300 mg/L COD significantly declined ( $P \leq 0.05$ ) by the end of each 120-s trial, most likely due to the increased demand for the oxidant. Higher inoculum levels and lower HOCl concentrations were significant factors ( $P \leq 0.05$ ) that contributed to increased cross-contamination. At 25 mg/L HOCl, no *Salmonella* was recovered under all conditions from uninoculated tomatoes exposed to tomatoes inoculated at 104 CFU per tomato. When the inoculum was increased to 106 and 108 CFU per tomato, cross-contamination was observed, independent of COD levels. The results from this study indicate that the currently required sanitizer concentration (e.g., 100 or 150 mg/L) for flume water may be higher than necessary and warrants reevaluation. ISSN: 0362028X

**Wiser, B., Niebuhr, S.E., Dickson, J.S.**

*Impact of Interventions on the Survival of Salmonella enterica I 4,[5],12:i:- in Pork*  
(2022) *Journal of Food Protection*, 85 (1), pp. 27-30.

**ABSTRACT:** A mixed culture of *Salmonella enterica* serovar I 4,[5],12:i:- isolates was compared with a mixed culture of reference *Salmonella* serovars and nonpathogenic *Escherichia coli* surrogates. The two groups of *Salmonella* were compared for their resistance to commonly used pork carcass interventions, survival in ground pork, and thermal resistance in ground pork. No differences in responses were observed between the two groups of *Salmonella* serovars and the nonpathogenic *E. coli* surrogates within intervention type. No differences in recovery and survival or in heat resistance were observed between the two groups of *Salmonella* serovars in pork that had been treated, ground, and stored at 58C for 2 weeks. However, the heat resistance of both groups of *Salmonella* serovars decreased after refrigerated storage. Because no differences were observed between *Salmonella* serovar I 4,[5],12:i:- and the reference *Salmonella* serovars in response to interventions commonly used in the pork industry, *Salmonella* I 4,[5],12:i:- does not present a unique challenge to the pork industry. ISSN: 0362028X

**Lozano-León, A., García-Omil, C., Rodríguez-Souto, R.R., Lamas, A., Garrido-Maestu, A.**

*An Evaluation of the Pathogenic Potential, and the Antimicrobial Resistance, of Salmonella Strains Isolated from Mussels*  
(2022) *Microorganisms*, 10 (1), art. no. 126, .

**ABSTRACT:** *Salmonella* spp. and antimicrobial resistant microorganisms are two of the most important health issues worldwide. In the present study, strains naturally isolated from mussels harvested in Galicia (one of the main production areas in the world), were genetically characterized attending to the presence of virulence and antimicrobial resistance genes. Additionally, the antimicrobial profile was also determined phenotypically. Strains presenting several virulence genes were isolated but lacked all the antimicrobial resistance genes analyzed. The fact that some of these strains presented multidrug resistance, highlighted the possibility of bearing different genes than those analyzed, or resistance based on completely different mechanisms. The current study highlights the importance of constant surveillance in order to improve the safety of foods. ISSN: 20762607

**Schilling, T., Hoelzle, K., Philipp, W., Hoelzle, L.E.**

*Survival of Salmonella Typhimurium, Listeria monocytogenes, and ESBL Carrying Escherichia coli in Stored Anaerobic Biogas Digestates in Relation to Different Biogas Input Materials and Storage Temperatures*  
(2022) *Agriculture (Switzerland)*, 12 (1), art. no. 67, .

**ABSTRACT:** Anaerobic digestates derived from agricultural mesophilic biogas plants are mainly used as organic fertilizers. However, animal derived pathogens could persist in the anaerobic digestates (ADs) posing an epidemiological risk. The present study investigated whether storage of ADs could reduce *Salmonella* Typhimurium, *Listeria monocytogenes*, and ESBL carrying *Escherichia coli* and whether reduction rates are dependent on temperature and substrate. Quantified bacterial suspensions were used to inoculate ADs derived from five biogas plants using different input materials to investigate the substrate dependence of the pathogen reduction. ADs were stored over six months with four different temperature profiles each representing six consecutive months, and, thus, the four seasons. Pathogen reduction during storage was shown to be strongly dependent on the temperature but also on the type of AD. This influence was higher at low temperatures. At higher temperatures (spring and summer profiles), a 5-log reduction was achieved after twelve weeks for *S. Typhimurium*, after twenty weeks for *E. coli* (ESBL) and after twenty-four weeks for *L. monocytogenes* in all ADs, respectively. In contrast at lower temperatures (autumn and winter profiles), a 5- log reduction was reached after twenty-four weeks for *S. Typhimurium* and not reached for ESBL- *E. coli* and *L. monocytogenes*. In conclusion, storing the ADs after the biogas process improves the hygienic quality and reduce the risk of introducing pathogens to the environment, but each case should be evaluated individually considering the composition of the ADs and the storage temperatures. ISSN: 20770472

**Amagliani, G., La Guardia, M.E., Dominici, S., Brandi, G., Omiccioli, E.**

*Salmonella Abortusovis: An Epidemiologically Relevant Pathogen*  
(2022) *Current Microbiology*, 79 (1), art. no. 3, .

**ABSTRACT:** The ovine pathogen *Salmonella enterica* serovar *Abortusovis* (SAO), a pathogen strictly adapted to ovine hosts, is endemic in several European and Asian countries, where it causes significant economic losses due to the high rates of abortion in infected flocks. In some countries (i.e. Switzerland and Croatia), re-emergence of infection by SAO occurred after decades during which the disease has not been reported. The introduction of (SAO) epidemic strains in new areas is difficult to control due to the asymptomatic behaviors in infected adult lambs, rams, and nonpregnant ewes. Culture-based diagnosis may provide false-negative results. Moreover, the retrospective identification of *Salmonella* infection in ewes is challenging as excretion of the causative agent is transient and the serum antibodies fall to low titres soon after the abortion. Therefore, regular monitoring of pathogen exposure, mainly through seroconversion assessment, is advisable to prevent disease introduction and spread in SAO-free areas, especially in case of animal export, and to reduce abortion risk. ISSN: 03438651

**Parker, E.M., Parker, A.J., Short, G., O'Connor, A.M., Wittum, T.E.**

*Salmonella detection in commercially prepared livestock feed and the raw ingredients and equipment used to manufacture the feed: A systematic review and meta-analysis*  
(2022) *Preventive Veterinary Medicine*, 198, art. no. 105546, .

**ABSTRACT:** *Salmonella* contamination of livestock feed is a serious veterinary and public health issue. In this study we used a systematic review to assess the prevalence and characterization of *Salmonella* isolates detected in raw feed components, feed milling equipment and finished feed from 97 studies published from 1955 to 2020 across seven global regions. Eighty-five studies were included in a meta-analysis to estimate the combined prevalence of *Salmonella* detection and to compare the risk of contamination associated with different sample types. We found the overall combined prevalence estimate of *Salmonella* detection was 0.14 with a prevalence of 0.18 in raw feed components, 0.09 in finished feed and 0.08 in feed milling equipment. Animal based raw feed components were 3.9 times more likely to be contaminated with *Salmonella* than plant based raw feed components. Differences between studies accounted for 99 % of the variance in the prevalence estimate for all sample types and there was no effect of region on the prevalence estimates. The combined prevalence of *Salmonella* detection in raw feed components decreased from 0.25 in 1955 to 0.11 in 2019. The proportion of *Salmonella* isolates that were resistant to antimicrobials was largest for amikacin (0.20), tetracycline (0.18) streptomycin (0.17), cefotaxime (0.14) and sulfisoxazole (0.11). The prevalence of *Salmonella* contamination of animal feed varies widely between individual studies and is an ongoing challenge for the commercial feed industry. Control relies on the vigilant monitoring and control of *Salmonella* in each individual mill. ISSN: 01675877

**Alban, L., Poulsen, M.K., Petersen, J.V., Lindegaard, L.L., Meinert, L., Koch, A.G., Møgelmose, V.**

*Assessment of risk to humans related to Salmonella from bile on pig carcasses*

(2022) *Food Control*, 131, art. no. 108415, .

**ABSTRACT:** In the European Union (EU), *Salmonella* is the main zoonotic hazard of interest in pig meat. Contamination occurs during slaughter mainly due to spread of faecal material. In 2020, the Danish competent authorities (CA) raised the question of the risk to humans of *Salmonella* resulting from bile contamination of pig carcasses. To address this, a study was undertaken involving 1) a pilot study to develop an aseptic way of collecting bile and 2) 299 gall bladder samples collected from finishing pigs from 28 pig herds. The samples were subjected to standard laboratory analysis and none were positive for *Salmonella*. A simulation model was set up using the collected data, plus data from the Danish meat inspection database as well as expert opinion, retrieved from the CA and the food business operator (FBO). The objective was to estimate the number of carcasses contaminated with *Salmonella* from bile that could be overlooked if responsibility for handling bile contamination were to rest solely with the FBO. The basic scenario showed that a median of nine (90% C.I.: 0–53) carcasses would be overlooked in a production of 16 million finishing pigs in one year, whereas 103 carcasses (90% C.I.: 7–544) would be overlooked in the worst-case scenario. Compared to the current *Salmonella* programme, the median relative efficacy of focusing on bile-contamination to detect *Salmonella* was 0.008% (basic scenario) or 0.087% (worst-case scenario). In conclusion, the risk to human health associated with *Salmonella* in bile on finishing pig carcasses was calculated to be negligible. Moreover, the FBO's handling of bile contamination prevents bile-contaminated carcasses from leaving the abattoir. ISSN: 09567135

**Muñoz-Vargas, L., Pempek, J.A., Proudfoot, K., Eastridge, M.L., Rajala-Schultz, P.J., Wittum, T., Habing, G.**

*The Impact of Overstocking and Negative Energy Balance on Quantitative Measurement of Non-typhoidal Salmonella in Periparturient Dairy Cattle*

(2022) *Frontiers in Veterinary Science*, 9, art. no. 779900, .

**ABSTRACT:** Stressful conditions in animal production facilities may exacerbate the fecal shedding and foodborne transmission of non-typhoidal *Salmonella enterica* subsp. *enterica*. Dairy cows are reservoirs of this zoonotic microorganism, and its prevalence has roughly doubled in the past decade on dairy farms in United States. Dairy cows are commonly overstocked at the feed bunk, and stressors placed on the animal prior to parturition may exacerbate *Salmonella* shedding. However, no studies have evaluated the impact of overstocking and metabolic stress on fecal concentrations of the pathogen. Therefore, we conducted a field trial with 120 multiparous dairy cows randomized into one of four treatment groups with different stocking densities at the feed bunk during the periparturient period as follows: US, understocked from –60 to –1 DRC; OS, overstocked from –60 to –1 DRC; USOS, understocked from –60 to –26 DRC/overstocked from –25 to –1 DRC; and OSUS, overstocked from –60 to –26 DRC/ understocked from –25 to –1 DRC. Fecal and blood samples were collected at four time points relative to calving. qPCR assays were used to quantify *Salmonella invA* gene and total bacterial community from fecal samples, and a subset of isolates recovered from fecal bacterial culture were characterized using pulsed field gel electrophoresis and serotyping. Serum non-esterified fatty acids (NEFA) were measured as a metabolic stress indicator using an immunoassay. Multivariable analyses were performed to test if changes in *Salmonella* concentrations were associated with stocking density, energy balance, or days relative to calving. From fecal isolates, three *Salmonella* serovars were identified, S. Cerro; Kentucky; Meleagridis. Concentrations of *Salmonella* increased as cows approached calving. Higher stocking densities at the feed bunk did not impact total bacterial community or NEFA; however, cows in the overstocked groups had higher *Salmonella* fecal concentrations. Further, cows with higher NEFA concentrations after calving had a higher likelihood of detection of *Salmonella*. Future farm interventions should aim to reduce environmental and metabolic stress during the periparturient period to decrease the dissemination of *Salmonella* to cattle, the environment, and humans. ISSN: 22971769

**Kim, U., Moon, Y.-J., Kim, J.-H., Lee, S.-Y., Oh, S.-W.**

*Development of modified enrichment broth for short enrichment and recovery of filter-injured Salmonella Typhimurium*

(2022) *International Journal of Food Microbiology*, 362, art. no. 109497, .

**ABSTRACT:** The filter concentration method facilitates the rapid detection of foodborne pathogens. The filter concentration method lowered the limit of detection (LOD) of artificially inoculated cabbage with *Salmonella Typhimurium*; however, the procedure injured foodborne pathogens during filtering procedure. Thus, to detect injured pathogens under the detection limit, an enrichment broth promoting pathogen resuscitation and growth is required. To rapidly recover, cultivate and lower the time to result (TTR) of *S. Typhimurium* detection after filter concentration method, a brain heart infusion (BHI)

broth-based modified enrichment broth (MEB) was developed. The MEB was developed by fitting growth curves to a modified Gompertz model; 1.00 g/L of sodium pyruvate, 0.20 g/L proline and 2.0 g/L magnesium sulphate additives were optimized as additional components to rapidly grow filter-injured *S. Typhimurium*. As a result, the rate of filter-injured *S. Typhimurium* went from 100% to 0.0% using MEB within 3.5 h. In contrast, BHI required 4 h and buffered peptone water (BPW) required more than 4 h to decrease the injury rate to 0.0%. Using MEB, BHI and BPW, filter-injured *S. Typhimurium* in cabbages were enriched to  $4.056 \pm 0.026$  Log CFU/25 g,  $3.571 \pm 0.187$  Log CFU/25 g and  $3.708 \pm 0.156$  Log CFU/25 g, respectively. Additionally, 1–9 CFU/mL *S. Typhimurium* in cabbage was detected within 3.0 h, including 1 h enrichment with MEB, whereas 5.0 h was required for BHI and BPW. Thus, the MEB developed in this study showed great potential as a short enrichment broth for the rapid detection of filter-injured *S. Typhimurium*. ISSN: 01681605

**Michael, M., Acuff, J.C., Vega, D., Sekhon, A.S., Channaiah, L.H., Phebus, R.K.**

*Survivability and thermal resistance of Salmonella and Escherichia coli O121 in wheat flour during extended storage of 360 days*

(2022) *International Journal of Food Microbiology*, 362, art. no. 109495, .

ABSTRACT: Foodborne pathogens like *Salmonella* and *Escherichia coli* O121 can endure the harsh low water activity (aw) environment of wheat flour for elongated periods of time and can proliferate when hydrated for baking or other purposes. This study determined the survivability and thermal tolerance (D- and z-values) of *Salmonella* and *Escherichia coli* O121 in wheat flour and muffin batter (prepared from inoculated flour on the days of analyses) during the storage period of 360 days. The *Salmonella* and *E. coli* O121 studies were conducted as two independent experiments. Both studies were designed as randomized complete block with three replications as blocks. All experimental data were analyzed using one-way ANOVA and Tukey's test in Minitab® software, and  $P \leq 0.05$  was considered significant. The wheat flour was spray inoculated individually with 7-isolate *Salmonella* or 3-isolate *E. coli* O121 cocktail and then dried back to the original aw levels. On each analysis day, inoculated wheat flour (~5 g) or muffin batter (~2.5 g) was placed inside the TDT disks, heat treated at set temperatures in hot water baths, and sampled at predetermined time intervals for determining the survival microbial population. The population of *E. coli* O121 and *Salmonella* cocktails in wheat flour at day 1 were  $7.6 \pm 0.18$  and  $7.8 \pm 0.07$  log CFU/g, respectively, which decreased to  $2.0 \pm 0.40$  and  $2.8 \pm 0.59$  log CFU/g on day 360, respectively. The D-values of *Salmonella* and *E. coli* O121 cocktails in inoculated flour and muffin batter prepared from inoculated flour (on the day of analysis) were determined on days 1, 30, 90, 180, 270, and 360 [given enough surviving bacterial population (~3 to 4 log CFU/g) was present in the flour]. The population of *Salmonella* and *E. coli* O121 in wheat flour decreased by 5.0 and 5.6 log CFU/g, respectively, during the storage period of 360 days. The D70°C, D75°C, and D80°C values of *Salmonella* in wheat flour remained similar during the storage period. Whereas, for *E. coli* O157:H7 in wheat flour, the D70°C value decreased from  $20.3 \pm 2.82$  to  $7.1 \pm 2.82$  min, and D75°C decreased from  $10.2 \pm 2.14$  to  $2.7 \pm 0.27$  min, during the storage period of 180 days. The z-values of *Salmonella* or *E. coli* O157:H7 remained similar during the storage period. The D- and z-values from this research can be employed for validation of thermal process to ensure safety of wheat flour. ISSN: 01681605

**Bertelloni, F., Cagnoli, G., Turchi, B., Ebani, V.V.**

*Low Level of Colistin Resistance and mcr Genes Presence in Salmonella spp.: Evaluation of Isolates Collected between 2000 and 2020 from Animals and Environment*

(2022) *Antibiotics*, 11 (2), art. no. 272, .

ABSTRACT: Salmonellosis is one of the most important zoonoses in Europe and the world. Human infection may evolve in severe clinical diseases, with the need for hospitalization and antimicrobial treatment. Colistin is now considered an important antimicrobial to treat infections from multidrug-resistant Gram-negative bacteria, but the spreading of mobile colistin-resistance (mcr) genes has limited this option. We aimed to evaluate colistin minimum inhibitory concentration and the presence of mcr (mcr-1 to mcr-9) genes in 236 *Salmonella* isolates previously collected from different animals and the environment between 2000 and 2020. Overall, 17.79% of isolates were resistant to colistin; no differences were observed in relation to years of isolation (2000–2005, 2009–2014, and 2015–2020), *Salmonella enterica* subspecies (*enterica*, *salamae*, *diarizonae*, and *houtenae*), origin of samples (domestic animals, wildlife, and environment), or animal category (birds, mammals, and reptiles); only recently isolated strains from houseflies showed the most resistance. Few isolates (5.93%) scored positive for mcr genes, in particular for mcr-1, mcr-2, mcr-4, mcr-6, and mcr-8; furthermore, only 2.54% of isolates were mcr-positive and colistin-resistant. Detected resistance to colistin was equally

distributed among all examined *Salmonella* isolates and not always related to the presence of *mcr* genes. ISSN: 20796382

**Patarata, L., Fernandes, L., Silva, J.A., Fraqueza, M.J.**

*The Risk of Salt Reduction in Dry-Cured Sausage Assessed by the Influence on Water Activity and the Survival of Salmonella*  
(2022) *Foods*, 11 (3), art. no. 444, .

ABSTRACT: Water activity (*aw*) is the main hurdle for microbial control in dry-cured sausages. The *aw* can be influenced by drying or adding electrolytes or humectants. Dry-cured meat products are partially dried, which, together with added salt, results in safe *aw* values. Currently, there is a trend to reduce salt in meat products, which can compromise the preservation process. The present work aims to evaluate the influences of added salt levels (1% or 3%) and the use or omission of phosphates and wine on the *aw* of a dry-cured sausage, and to evaluate the possibility of estimating the *aw* from the moisture loss and the behavior of *Salmonella* during dry-cured sausage (*chouriço*) processing. There was a strong relationship between moisture and *aw*, regardless of the salt level and the presence of phosphates or wine. Predicting *aw* from moisture loss is possible using the Boltzmann sigmoid function. The salt level strongly influences *Salmonella* behavior, mainly through *aw* reduction. An increase in *aw* by 0.01 units reduced the odds of achieving a 5-log reduction in *Salmonella* counts to half. Increasing added salt from 1% to 3% increased the odds of achieving a 5-log *Salmonella* reduction 7.5-fold. The current trend to reduce salt in foods must be carefully approached if applied to cured meat products, as it has substantial consequences on *aw* evolution and *Salmonella* survival. ISSN: 23048158

**Mitchell, P.K., Wang, L., Stanhope, B.J., Cronk, B.D., Anderson, R., Mohan, S., Zhou, L., Sanchez, S., Bartlett, P., Maddox, C., DeShambo, V., Mani, R., Hengesbach, L.M., Gresch, S., Wright, K., Mor, S., Zhang, S., Shen, Z., Yan, L., Mackey, R., Franklin-Guild, R., Zhang, Y., Prarat, M., Shiplett, K., Ramachandran, A., Narayanan, S., Sanders, J., Hunkapiller, A.A., Lahmers, K., Carbonello, A.A., Aulik, N., Lim, A., Cooper, J., Jones, A., Guag, J., Nemser, S.M., Tyson, G.H., Timme, R., Strain, E., Reimschuessel, R., Ceric, O., Goodman, L.B.**

*Multi-laboratory evaluation of the Illumina iSeq platform for whole genome sequencing of Salmonella, Escherichia coli and Listeria*  
(2022) *Microbial genomics*, 8 (2), .

ABSTRACT: There is a growing need for public health and veterinary laboratories to perform whole genome sequencing (WGS) for monitoring antimicrobial resistance (AMR) and protecting the safety of people and animals. With the availability of smaller and more affordable sequencing platforms coupled with well-defined bioinformatic protocols, the technological capability to incorporate this technique for real-time surveillance and genomic epidemiology has greatly expanded. There is a need, however, to ensure that data are of high quality. The goal of this study was to assess the utility of a small benchtop sequencing platform using a multi-laboratory verification approach. Thirteen laboratories were provided the same equipment, reagents, protocols and bacterial reference strains. The Illumina DNA Prep and Nextera XT library preparation kits were compared, and 2×150 bp iSeq i100 chemistry was used for sequencing. Analyses comparing the sequences produced from this study with closed genomes from the provided strains were performed using open-source programs. A detailed, step-by-step protocol is publicly available via protocols.io (<https://www.protocols.io/view/iseq-bacterial-wgs-protocol-bij8kcrw>). The throughput for this method is approximately 4-6 bacterial isolates per sequencing run (20-26 Mb total load). The Illumina DNA Prep library preparation kit produced high-quality assemblies and nearly complete AMR gene annotations. The Prep method produced more consistent coverage compared to XT, and when coverage benchmarks were met, nearly all AMR, virulence and subtyping gene targets were correctly identified. Because it reduces the technical and financial barriers to generating WGS data, the iSeq platform is a viable option for small laboratories interested in genomic surveillance of microbial pathogens. ISSN: 20575858

**Perilli, M., Scattolini, S., Telera, G.C., Cornacchia, A., Tucci, P., Sacchini, F., Sericola, M., Romantini, R., Marotta, F., Di Provvido, A., Pomilio, F., De Massis, F.**

*Distribution of Salmonella spp. Serotypes Isolated from Poultry in Abruzzo and Molise Regions (Italy) during a 6-Year Period*  
(2022) *Microorganisms*, 10 (2), art. no. 199, .

ABSTRACT: Human salmonellosis incidence is increasing in the European Union (EU). *Salmonella enterica* subsp. *enterica* serovar Enteritidis, *Salmonella enterica* subsp. *enterica* serovar Typhimurium (including its monophasic variant) and *Salmonella enterica* subsp. *enterica* serovar Infantis represent targets in control programs due to their frequent

association with human cases. This study aimed to detect the most prevalent serotypes circulating in Abruzzo and Molise Regions between 2015 and 2020 in the framework of the Italian National Control Program for Salmonellosis in Poultry (PNCS)]. A total of 332 flocks of Abruzzo and Molise Regions were sampled by veterinary services in the period considered, and 2791 samples were taken. Samples were represented by faeces and dust from different categories of poultry flocks: laying hens (n = 284), broilers (n = 998), breeding chickens (n = 1353) and breeding or fattening turkeys (n = 156). Breeding and fattening turkeys had the highest rate of samples positive for Salmonella spp. (52.6%; C.I. 44.8%–60.3%). Faeces recovered through boot socks represented the greatest number of positive samples (18.2%). Salmonella enterica subsp. enterica serovar Infantis was the prevalent serotype in breeding and fattening turkeys (32.7%; C.I. 25.8%–40.4%) and in broiler flocks (16.5%; C.I. 14.4%–19.0%). Salmonella enterica subsp. enterica serovar Typhimurium was detected at low levels in laying hens (0.7%; C.I. 0.2%–2.5%) followed by breeding and fattening turkeys (0.6%; C.I. 0.2%–2.5%). Salmonella enterica subsp. enterica serovar Enteritidis was also detected at low levels in laying hens (2.5%; C.I. 1.2%–5.0%). These findings highlight the role of broilers and breeding/fattening turkeys as reservoirs of Salmonella spp. and, as a consequence, in the diffusion of dangerous serotypes as Salmonella enterica subsp. enterica serovar Infantis. This information could help veterinary services to analyze local trends and to take decisions not only based on indications from national control programs, but also based on real situations at farms in their own competence areas. ISSN: 20762607

**Medina-Santana, J.L., Ortega-Paredes, D., de Janon, S., Burnett, E., Ishida, M., Sauders, B., Stevens, M., Vinueza-Burgos, C.**

*Investigating the dynamics of Salmonella contamination in integrated poultry companies using a whole genome sequencing approach*

(2022) *Poultry Science*, 101 (2), art. no. 101611, .

ABSTRACT: The study of non-typhoid Salmonella in broiler integrations has been limited by the resolution of typing techniques. Although serotyping of Salmonella isolates is used as a traditional approach, it is not of enough resolution to clearly understand the dynamics of this pathogen within poultry companies. The aim of this research was to investigate the epidemiology and population dynamics of Salmonella serotypes in 2 poultry integrations using a whole genome sequencing approach. Two hundred and forty-three Salmonella isolates recovered from the broiler production chain of 2 integrated poultry companies were whole genome sequenced and analyzed with dedicated databases and bioinformatic software. The analyses of sequences revealed that S. Infantis was the most frequent serotype (82.3%). Most isolates showed a potential for resistance against medically important antibiotics and disinfectants. Furthermore, 97.5% of isolates harbored the pESI-like mega plasmid, that plays an important role in the global dissemination of AMR. SNP tree analysis showed that there were clones that are niche-specific while other ones were distributed throughout the broiler production chains. In this study, we demonstrated the potential of whole genome sequencing analysis for a comprehensive understanding of Salmonella distribution in integrated poultry companies. Data obtained with these techniques allow determination of the presence of genetic factors that play an important role in the environmental fitness and pathogenicity of Salmonella. ISSN: 00325791

**Slowey, R., Kim, S.W., Prendergast, D., Madigan, G., Van Kessel, J.A.S., Haley, B.J.**

*Genomic diversity and resistome profiles of Salmonella enterica subsp. enterica serovar Kentucky isolated from food and animal sources in Ireland*

(2022) *Zoonoses and Public Health*, 69 (1), pp. 1-12.

ABSTRACT: Salmonella enterica subsp. enterica serovar Kentucky is frequently isolated from poultry, dairy and beef cattle, the environment and people with clinical salmonellosis globally. However, the sources of this serovar and its diversity and antimicrobial resistance capacities remain poorly described in many regions. To further understand the genetic diversity and antimicrobial sensitivity patterns among S. Kentucky strains isolated from non-human sources in Ireland, we sequenced and analysed the genomes of 61 isolates collected from avian, bovine, canine, ovine, piscine, porcine, environmental and vegetation sources between 2000 and 2016. The majority of isolates (n = 57, 93%) were sequence type (ST) 314, while only three isolates were ST198 and one was ST152. Several isolates were multidrug-resistant (MDR) and 14 carried at least one acquired antimicrobial resistance gene. When compared to a database of publicly available ST314, four distinct clades were identified (clades I–IV), with the majority of isolates from Ireland clustering together in Clade I. Two of the three ST198 isolates were characteristic of those originating outside of the Americas (Clade ST198.2), while one was distantly clustered with isolates from South and North America (Clade ST198.1). The genomes of the two clade ST198.2 isolates encoded Salmonella Genomic Island 1 (SGI1), were multidrug-resistant and

encoded polymorphisms in the DNA gyrase (*gyrA*) and DNA topoisomerase (*parC*) known to confer resistance to fluoroquinolones. The single ST152 isolate was from raw beef, clustered with isolates from food and bovine sources in North America and was pan-susceptible. Results of this study indicate that most *S. Kentucky* isolates from non-human sources in Ireland are closely related ST314 and only a few isolates are antimicrobial-resistant. This study also demonstrates the presence of multidrug-resistant ST198 in food sources in Ireland. ISSN: 18631959

**Furtado, R., Coelho, A., Morais, M., Leitão, A.L., Saraiva, M., Correia, C.B., Batista, R.**

*Comparison of ISO 6579–1, VIDAS Easy SLM, and SureFast® Salmonella ONE Real-time PCR, for Salmonella Detection in Different Groups of Foodstuffs (2022) Food Analytical Methods, 15 (2), pp. 276-284.*

ABSTRACT: In the European Union (EU), *Salmonella* was the causative agent responsible for almost one in three (30.7%) of all foodborne outbreaks reported by member states during 2018, causing 11,581 cases of illness, which represented an increase of 20.6% compared to 2017. Considering the importance of this foodborne zoonotic bacterium in food safety and human health, several strategies for the control and consequent detection of *Salmonella* in foodstuffs are continuously being developed. In this study, we have tested 137 food samples (78 potentially naturally contaminated, 21 artificially contaminated with high levels of *Salmonella*, and 38 artificially contaminated with low levels of *Salmonella*) in order to compare the results and performance of three *Salmonella* detection methods: standard conventional culture (ISO 6579–1), SureFast® *Salmonella ONE* real-time PCR, and VIDAS® (Vitek Immunodiagnostic Assay System) Easy SLM, an Enzyme Linked Fluorescent Assay (ELFA). Although SureFast® *Salmonella ONE* real-time PCR was the fastest, it showed more inconclusive results, due to PCR inhibition and false positive results. ISO and VIDAS® protocols gave identical results and proved to be more robust than SureFast® *Salmonella ONE* real-time PCR when testing different food matrices, despite its longer response times. SureFast® *Salmonella ONE* real-time PCR may be appropriate to be used when the objective is to test food matrices that are known not to interfere with PCR and expected to be negative for *Salmonella*. All the analytical tested methods have advantages and limitations and thus, depending on the situation, may be used as the elected method for *Salmonella* detection in foodstuffs in accordance with the purpose of the laboratorial analysis. ISSN: 19369751

**Keerthirathne, T.P., Ross, K., Fallowfield, H., Whiley, H.**

*Examination of Australian backyard poultry for Salmonella, Campylobacter and Shigella spp., and related risk factors (2022) Zoonoses and Public Health, 69 (1), pp. 13-22.*

ABSTRACT: Worldwide, foodborne illness is a significant public health issue in both developed and developing countries. Salmonellosis, campylobacteriosis and shigellosis are common foodborne gastrointestinal illnesses caused by the bacteria *Salmonella* spp., *Campylobacter* spp. and *Shigella* spp. respectively. These zoonotic diseases are frequently linked to eggs and poultry products. The aim of this study was to investigate the presence of these pathogens in Australian backyard poultry flocks and to determine risk factors for these pathogens. Poultry faeces samples were collected from 82 backyards and screened for *Salmonella* spp., *Campylobacter* spp. and *Shigella* spp. using qPCR. A questionnaire was administered to the backyard poultry owners to assess their knowledge regarding management of poultry and eggs and to identify potential risk factors that may contribute to the presence of zoonotic pathogens in the flocks. One composite faecal sample was collected from each backyard (82 samples). Composite sampling here means taking one or more grab samples from a backyard to make up approximately 10 grams. Four per cent of samples, that is 4% backyards tested, were positive for *Salmonella* spp., 10% were positive for *Campylobacter* spp. and none were positive for *Shigella* spp. A higher infection rate was seen in multi-aged flocks (24%) compared with the single-aged flocks (3%). The survey found that many participants were engaging in risky food safety behaviours with 46% of participants responding that they washed their eggs with running water or still water instead of wiping the dirt off with a damp cloth to clean the eggs and 19% stored their eggs at room temperature. This study demonstrated that backyard poultry may pose a potential risk for salmonellosis and campylobacteriosis. Additionally, Australian public health and food safety regulations should be modified and effectively implemented to address the risks associated with backyard poultry husbandry. ISSN: 18631959

**Norberto, A.P., Alvarenga, V.O., Hungaro, H.M., Sant'Ana, A.S.**

*Desiccation resistance of a large set of Salmonella enterica strains and survival on dry- and wet-inoculated soybean meal through storage*

(2022) *LWT*, 158, art. no. 113153, .

**ABSTRACT:** This study determined the desiccation resistance of 37 *Salmonella* strains belonging to 16 serotypes isolated from the soybean meal production chain. Besides, the survival of strains from three *Salmonella enterica* serovars (*S. Typhimurium*, *S. Schwarzengrund*, and *S. Havana*) on dry- and wet-inoculated soybean meal through storage at 25 °C and 37 °C was evaluated. Desiccation resistance varied within strains of the same serotype and amongst strains of different serotypes. On the other hand, the isolation source did not affect desiccation resistance. The inoculation method did not influence the survival of the three *Salmonella enterica* strains in soybean meal, but the effects of serovars and temperature were statistically significant ( $p < 0.05$ ). The Weibull model was fitted to *Salmonella* survival in this matrix data, with the time for the first decimal reduction ( $\delta$ ) ranging from 21.1 to 50.8 days at 25 °C and from 2.7 to 7.9 days at 37 °C, respectively. The increase in storage temperature led to a decrease in survival regardless of the variability among the three isolates. The ability of *Salmonella enterica* to resist desiccation and to survive long-term on soybean meal reinforces the need for strategies to control this pathogen in the soybean production chain. ISSN: 00236438

**Milanov, M.V., Mateva, G.I., Stoyanchev, T.T.**

*SURVIVAL AND GROWTH DYNAMICS OF LISTERIA MONO-CYTOGENES AND SALMONELLA SPP. ON ARTIFICIALLY CONTAMINATED COOKED READY-TO-EAT MEAT PRODUCTS (2022) Bulgarian Journal of Veterinary Medicine, 25 (1), pp. 147-160.*

**ABSTRACT:** The aim of the study was to evaluate the potential of survival and growth dynamics of *Listeria mono-cytogenes* strains and *Salmonella enterica* subsp. *enterica* serovar *Enteritidis* and *Typhimurium*, inoculated artificially (individually and in mixture) on two ready-to-eat (RTE) cooked smoked meat products. For the purpose of the study 120 slices of cooked smoked sausage and 40 slices of cooked smoked loin were purchased and inoculated with the strains. Two storage temperatures were selected: 60 C and 10o C for 8 days. The study was performed as challenge tests in a secondary contamination scenario to investigate the presence and/or absence of pathogenic bacteria during the shelf life of the products. The inoculum levels at the start of the experiment were 4.46 log<sub>10</sub> CFU/g and 2.88 log<sub>10</sub> CFU/g for the *L. monocytogenes* strains at the surface of the cooked smoked loin and cooked smoked sausage respectively. Using the same inoculation method, but adding *Salmonella enterica* serovars to the mixture, the inoculum levels were 4.15 log<sub>10</sub> CFU/g at the surface of the cooked smoked loin and 2.94 log<sub>10</sub> CFU/g at the surface of the cooked smoked sausage. *L. monocytogenes* was detected at all sampling days on both storage temperatures. It showed an average increase by 0.5–1.0 log<sub>10</sub> CFU/g on the cooked smoked sausage and by 2.0–3.3 log<sub>10</sub> CFU/g for the cooked smoked pork loin for the duration of the study. *Salmonella enterica* serovars were also present at each sampling day on both storage temperatures. Typical colonies were isolated and serotyped, confirming the survival of these pathogenic bacteria. *Salmonella enterica* subsp. *enterica* serovar *Typhimurium* was the predominant serovar at almost every sampling day. The results from our study showed no competitive relationship in the presence of *Salmonella* and *L. monocytogenes* in contaminated meat products. The two types of microorganisms were successfully adapted and developed independently under appropriate conditions, including temperature, humidity, water activity and pH. ISSN: 13111477

**Samper-Cativiela, C., Diéguez-Roda, B., Trigo da Roza, F., Ugarte-Ruiz, M., Elnekave, E., Lim, S., Hernández, M., Abad, D., Collado, S., Sáez, J.L., de Frutos, C., Agüero, M., Moreno, M.Á., Escudero, J.A., Álvarez, J.**

*Genomic characterization of multidrug-resistant Salmonella serovar Kentucky ST198 isolated in poultry flocks in Spain (2011-2017) (2022) Microbial genomics, 8 (3), .*

**ABSTRACT:** *Salmonella Kentucky* is commonly found in poultry and rarely associated with human disease. However, a multidrug-resistant (MDR) *S. Kentucky* clone [sequence type (ST)198] has been increasingly reported globally in humans and animals. Our aim here was to assess if the recently reported increase of *S. Kentucky* in poultry in Spain was associated with the ST198 clone and to characterize this MDR clone and its distribution in Spain. Sixty-six isolates retrieved from turkey, laying hen and broiler in 2011-2017 were subjected to whole-genome sequencing to assess their sequence type, genetic relatedness, and presence of antimicrobial resistance genes (ARGs), plasmid replicons and virulence factors. Thirteen strains were further analysed using long-read sequencing technologies to characterize the genetic background associated with ARGs. All isolates belonged to the ST198 clone and were grouped in three clades associated with the presence of a specific point mutation in the *gyrA* gene, their geographical origin and isolation year. All strains carried between one and 16 ARGs whose presence correlated with the resistance phenotype to between two and eight antimicrobials. The ARGs were located in the



*Salmonella* genomic island (SGI-1) and in some cases (*bla*SHV-12, *catA1*, *cmlA1*, *dfrA* and multiple aminoglycoside-resistance genes) in IncHI2/IncI1 plasmids, some of which were consistently detected in different years/farms in certain regions, suggesting they could persist over time. Our results indicate that the MDR *S. Kentucky* ST198 is present in all investigated poultry hosts in Spain, and that certain strains also carry additional plasmid-mediated ARGs, thus increasing its potential public health significance. ISSN: 20575858

**Obe, T., Richards, A.K., Shariat, N.W.**

*Differences in biofilm formation of Salmonella serovars on two surfaces under two temperature conditions*

(2022) *Journal of Applied Microbiology*, 132 (3), pp. 2410-2420.

**ABSTRACT:** Aims: *Salmonella* is extremely diverse, with >2500 serovars that are genetically and phenotypically diverse. The aim of this study was to build a collection of *Salmonella* isolates that are genetically diverse and to evaluate their ability to form biofilm under different conditions relevant to a processing environment. Methods and Results: Twenty *Salmonella* isolates representative of 10 serovars were subtyped using Clustered regularly interspaced short palindromic repeats (CRISPR)-typing to assess the genetic diversity between isolates of each serovar. Biofilm formation of the isolates on both plastic and stainless-steel surfaces at 25 and 15°C was assessed. At 25°C, 8/20 isolates each produced strong and moderate biofilm on plastic surface compared to stainless-steel (3/20 and 13/20 respectively). At 15°C, 5/20 produced strong biofilm on plastic surface and none on stainless-steel. Several isolates produced weak biofilm on plastic (11/20) and stainless-steel (16/20) surfaces. Serovar Schwarzengrund consistently produced strong biofilm while serovars Heidelberg and Newport produced weak biofilm. Conclusion: These results suggest that *Salmonellae* differ in their attachment depending on the surface and temperature conditions encountered, which may influence persistence in the processing environment. Significance and Impact of Study: These differences in biofilm formation could provide useful information for mitigation of *Salmonella* in processing environments. ISSN: 13645072

**Molenaar, F.M., Silvestre, P.**

*Clinical approach to colic and collapse in an Asian elephant (Elephas maximus) with Salmonella saintpaul septicaemia and subsequent ileus*

(2022) *Veterinary Record Case Reports*, 10 (1), art. no. e214, .

**ABSTRACT:** An adult female Asian elephant (*Elephas maximus*) presented with clinical signs of colic unresponsive to analgesia, which progressed to hypothermia and collapse within 48 hours. Repeated sedations using butorphanol and detomidine were performed for initial diagnostic sampling, first aid and subsequent treatment. Initial haematology showed evidence of septicaemia and disseminated intravascular coagulation; urine analysis was consistent with metabolic acidosis. The initial treatment focused on rectal administration of enrofloxacin, metronidazole and fluids. By Day 7, the immune system was recovering as demonstrated by blood parameters but ileus had developed. Sedation interventions were discontinued and treatment consisted of oral ranitidine, fibre provision and rehydration. *Salmonella saintpaul* was cultured from the faeces and a disease risk analysis identified a possible infection route through food contamination. Serial haematology provided direction in clinical decision making throughout this challenging case. ISSN: 20526121