NEWSLETTER

European Union Reference Laboratory for *Salmonella*

Vol. 24 No. 4
December 2018

ISSN 2211-6877

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

Produced by

European Union Reference Laboratory for *Salmonella*

National Institute of Public Health and the Environment
P.O. Box 1, 3720 BA Bilthoven, The Netherlands

phone: +31 30 274 3537 (Kirsten Mooijman)
+31 30 274 4290 (Wilma Jacobs)
e-mail: kirsten.mooijman@rivm.nl
wilma.jacobs@rivm.nl

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Editorial Note

Bilthoven, 4 January 2019

Dear colleague,

I hope you all have had a very nice Christmas break and a good start of the New Year. On behalf of the EURL-*Salmonella* staff, I would like to wish you all a **very good and healthy 2019!**

Since the last Newsletter of September 2018, we organised two Proficiency Tests (PTs), and started the preparation of a third PT.

In October 2018 the **study on detection of *Salmonella* in samples from the primary production stage** (boot socks with chicken faeces) was organised. The results of this PT (interim summary and NRLs' own results) were reported to the participants shortly before the Christmas break.

In November 2018 the **study on typing of *Salmonella*** was organised. Before the start of this PT, the NRLs-*Salmonella* were asked for their interest in including MLVA typing in the study. However, the number of applications remained below 7, which was the critical number we had set for organising a PT with MLVA. Hence, MLVA typing was not included in this study. For PFGE typing, on the other hand, still more than 10 applications were received; so PFGE was again (optional) part of this typing PT, in addition to (obliged) serotyping. The deadline for reporting the results of this PT was shortly before the Christmas break, meaning that the analysis of the data will soon be started.

In the last quarter of 2018, we also started the preparation of a **PT on detection of *Salmonella* in food or animal feed samples (Spring 2019)**. You may remember that we were facing some problems during the last interlaboratory comparison study on the detection of *Salmonella* in animal feed (Spring 2018). The concentration of *Salmonella* in the final animal feed samples of the study of 2018 was unexpectedly low, so many samples were tested negative. This resulted in the fact that we were not able to determine the performance of the NRLs-*Salmonella* in this study. For that reason we would like to repeat a PT for detection of *Salmonella* in animal feed. On the other hand, we considered it also important to organise again a PT for detection of *Salmonella* in a food product as the last PT with a 'real' food matrix was organised in 2016. Therefore, we have been searching for a matrix which could be considered as a food product as well as an animal feed product and we think we have found a satisfactory product. By mid-December 2018 you were informed by Robin Diddens that the study will be organised with flaxseed (linseed) in March 2019. Flaxseed can be considered as a food product as well as an ingredient of animal feed. NRLs-*Salmonella* which analyse food (products), as well as NRLs-*Salmonella* which analyse animal feed are invited to participate in this Proficiency Test. Do not forget to express your interest by filling the registration form, following the link in the e-mail of Robin. The time table of this PT is included in this Newsletter.

By mid-December 2018 we have submitted the **work program and budget forecast for the activities of the EURL-*Salmonella* for 2019-2020**. The request for submission was late this time. On the other hand, a work program and budget for 2 years instead of 1 year had to be submitted, saving a submission round in fall 2019. Depending when the decision for the work programs and budgets of all EURLs is taken, the final work program of the activities of the EURL-*Salmonella* will be published in the next Newsletter, or in one of the following Newsletters.

In the last quarter of 2018, we started with the preparation of the **EURL-*Salmonella* workshop of 2019**. We decided to stay in the Netherlands for this workshop, at a new location in the center of the Netherlands (Amersfoort). In December 2018 we have sent you the first announcement of the workshop, which is planned on 28 and 29 May 2019. More information, as well as a registration form will be sent to the NRLs-*Salmonella* later in January.

By the end of November 2018, EFSA informed us that the report of the INNUENDO project (EFSA Co-funded) was published. You can find this report at the following link:
https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/sp.efsa.2018.EN-1498

In December 2018, the following reports of EURL-*Salmonella* interlaboratory comparison studies were published:

Jacobs-Reitsma, W.F., Verbruggen, A., Bouw, E., Mooijman, K.A. 22nd EURL-*Salmonella* interlaboratory comparison study (2017) on typing of *Salmonella* spp. RIVM report no.: 2018-0022.
https://www.rivm.nl/bibliotheek/rapporten/2018-0022.pdf

Pol-Hofstad, I.E. and Mooijman, K.A. The combined EURL-*Salmonella* interlaboratory comparison study for Food and Primary Production (2017) - Detection of *Salmonella* in hygiene swabs. National Institute for Public Health and the Environment, Bilthoven, the Netherlands. RIVM Report no.: 2018-0021.
https://www.rivm.nl/bibliotheek/rapporten/2018-0021.pdf

In the EURL-*Salmonella* Newsletter of June 2018 we informed you that the publication of the article on the validation of EN ISO 6579-1 was accepted and available on line ('in press'). I can now inform you that this article is published as part of a special issue of the January version of the International Journal of Food Microbiology, publishing the (15) validation studies performed under the CEN Mandate M/381:

Mooijman, K.A., Pielaat, A. and Kuijpers, F. A. Validation of EN ISO 6579-1 - Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of *Salmonella* - Part 1 Detection of *Salmonella* spp. International Journal of Food Microbiology, 288 (2019), 3-12. Paper accepted and on line since 12-05-2018: https://doi.org/10.1016/j.ijfoodmicro.2018.03.022

Best wishes,
Kirsten Mooijman
Coordinator EURL-*Salmonella*

Contribution of EURL-*Salmonella*

Timetable EURL-*Salmonella* Proficiency Test Food-Feed (March 2019)
Detection of *Salmonella* in flaxseed

Week (2019)	Dates	Subject
11	11 – 15 March	Mailing of the protocol and instructions for the electronic result form to the NRLs by email. Sending the link for the electronic result form to the participants by email.
12	18 March	Mailing of parcels to the NRLs as Biological Substance Cat. B (UN3373) by door-to-door courier service. Preparation of media by the NRLs.
13	25 March	Performance of the study
16	Before 17 April	Deadline for completing the electronic submission of results: **16 April 2019** (23:59h CET) After this deadline the electronic result form will be closed.

From the Literature

Salmonella-related Literature from Scopus: October – December 2018

Methner, U., Merbach, S., Peters, M.
Salmonella enterica subspecies enterica serovar Choleraesuis in a German wild boar population: Occurrence and characterisation
(2018) Acta Veterinaria Scandinavica, 60 (1), art. no. 65, .
ABSTRACT: Background: The swine-adapted serovar Choleraesuis of Salmonella enterica subspecies enterica is found rarely in domestic pigs in Germany. However, a considerable and increasing number of S. Choleraesuis organisms have been isolated from wild boars in Germany in recent years. To investigate a possible epidemiological context, S.Choleraesuis strains from a regional German wild boar population and other hosts were characterised by genotyping methods. Results: Macrorestriction analysis, biochemical differentiation and antimicrobial susceptibility typing enabled the identification of several clusters of S.Choleraesuis. Some clusters occurred almost permanently in a certain district, whereas other groups circulated among different wild boar herds in larger regions. Non-porcine hosts were infected with the same cluster as the wild boars. Conclusions: The emergence of S.Choleraesuis in wild boars might be caused by a higher prevalence in the wild boar population, but also the higher awareness to infections with African swine fever may have resulted in a higher number of examined animals. Separation of wild boar populations and, as a result, also the diverse S.Choleraesuis organisms might be due to natural barriers and artificial barriers like arterial roads. The findings of S.Choleraesuis in domestic pigs emphasize the importance of strict biosecurity measures to prevent transmission from wild boars of this but also other pathogens. To avoid risks for humans by zoonotic pathogens regular inspections of meat from wildlife need to be conducted. ISSN: 0044605X

Hyeon, J.-Y., Mann, D.A., Townsend, A.M., Deng, X.
Quasi-metagenomic Analysis of Salmonella from Food and Environmental Samples
(2018) Journal of visualized experiments : JoVE, (140), .
ABSTRACT: Quasi-metagenomics sequencing refers to the sequencing-based analysis of modified microbiomes of food and environmental samples. In this protocol, microbiome modification is designed to concentrate genomic DNA of a target foodborne pathogen contaminant to facilitate the detection and subtyping of the pathogen in a single workflow. Here, we explain and demonstrate the sample preparation steps for the quasi-metagenomics analysis of Salmonella enterica from representative food and environmental samples including alfalfa sprouts, ground black pepper, ground beef, chicken breast and environmental swabs. Samples are first subjected to the culture enrichment of Salmonella for a shortened and adjustable duration (4-24 h). Salmonella cells are then selectively captured from the enrichment culture by immunomagnetic separation (IMS). Finally, multiple displacement amplification (MDA) is performed to amplify DNA from IMS-captured cells. The DNA output of this protocol can be sequenced by high throughput sequencing platforms. An optional quantitative PCR analysis can be performed to replace sequencing for Salmonella detection or assess the concentration of Salmonella DNA before sequencing. ISSN: 1940087X

Dang-Xuan, S., Nguyen-Viet, H., Pham-Duc, P., Grace, D., Unger, F., Nguyen-Hai, N., Nguyen-Tien, T., Makita, K.
Simulating cross-contamination of cooked pork with salmonella enterica from raw pork through home kitchen preparation in Vietnam
(2018) International Journal of Environmental Research and Public Health, 15 (10), art. no. 2324, .
ABSTRACT: Pork is the most commonly consumed meat in Vietnam, and Salmonella enterica is a common contaminant. This study aimed to assess potential S. enterica cross-contamination between raw and cooked pork in Vietnamese households. Different scenarios for cross-contamination were constructed based on a household survey of pork handling practices (416 households). Overall, 71% of people used the same knife and cutting board for both raw and cooked pork; however, all washed their hands and utensils between handling raw and cooked pork. The different scenarios were experimentally tested. First, S. enterica was inoculated on raw pork and surfaces (hands, knives and cutting boards); next, water used for washing and pork were sampled to identify the presence and concentration of S. enterica during different scenarios of food preparation. Bootstrapping techniques were applied to simulate transfer rates of S. enterica cross-

contamination. No cross-contamination to cooked pork was observed in the scenario of using the same hands with new cutting boards and knives. The probability of re-contamination in the scenarios involving re-using the cutting board after washing was significantly higher compared to the scenarios which used a new cutting board. Stochastic simulation found a high risk of cross-contamination from raw to cooked pork when the same hands, knives and cutting boards were used for handling raw and cooked pork (78%); when the same cutting board but a different knife was used, cross-contamination was still high (67%). Cross-contamination between was not seen when different cutting boards and knives were used for cutting raw and cooked pork. This study provided an insight into cross-contamination of S. enterica, given common food handling practices in Vietnamese households and can be used for risk assessment of pork consumption.
ISSN: 16617827

Huang, R., de Vries, D., Chen, H.
Strategies to enhance fresh produce decontamination using combined treatments of ultraviolet, washing and disinfectants
(2018) International Journal of Food Microbiology, 283, pp. 37-44.
ABSTRACT: This study investigated the effect of a water-assisted ultraviolet system (WUV; samples were treated by UV while being immersed in agitated water) on the inactivation of Salmonella on baby spinach, iceberg lettuce, blueberry, grape tomato, and baby-cut carrot. The Salmonella inactivation effect of the WUV system was tested in two scales, and three disinfectants, chlorine, peroxyacetic acid (PAA) and hydrogen peroxide (H2O2), were tested in combination with the system to see whether the Salmonella inactivation effect could be enhanced. The fresh produce samples were dip-inoculated with a Salmonella cocktail to final concentrations of 4.6–7.6 log CFU/g. To simulate the washing process in the industry, fresh produce extracts and/or silicon dioxide were added in the wash water to adjust chemical oxygen demand to ~2000 mg/L and turbidity to >60 NTU. In general, the decontamination efficacy of WUV treatments followed this order: Tomato > Carrot > Lettuce ≈ Blueberry > Spinach. In the small-scale study, WUV alone was able to achieve 0.9, 2.6, >3.6, 1.7, and 2.0 log CFU/g reductions of Salmonella on fresh produce for spinach, lettuce, tomato, blueberry, and carrot, respectively. For all fresh produce items, WUV combined with PAA could achieve significantly (P < 0.05) higher Salmonella reduction on fresh produce than chlorine wash and PAA wash. The WUV treatments combined with chlorine or PAA were able to keep residual Salmonella in wash water below the detection limit (2 CFU/mL) for almost all the replicates. Similar Salmonella reductions on fresh produce and in wash water were found in the large-scale study. Considering the decontamination efficacy on fresh produce, the ability to disinfect the wash water, and the cost, we recommend chlorine wash for baby spinach, WUV alone for grape tomato and WUV combined with PAA for iceberg lettuce, blueberry and baby-cut carrot.
ISSN: 01681605

Gu, G., Strawn, L.K., Oryang, D.O., Zheng, J., Reed, E.A., Ottesen, A.R., Bell, R.L., Chen, Y., Duret, S., Ingram, D.T., Reiter, M.S., Pfuntner, R., Brown, E.W., Rideout, S.L.
Agricultural practices influence salmonella contamination and survival in pre-harvest tomato production
(2018) Frontiers in Microbiology, 9 (OCT), art. no. 2451, .
ABSTRACT: Between 2000 and 2010 the Eastern Shore of Virginia was implicated in four Salmonella outbreaks associated with tomato. Therefore, a multi-year study (2012-2015) was performed to investigate presumptive factors associated with the contamination of Salmonella within tomato fields at Virginia Tech's Eastern Shore Agricultural Research and Extension Center. Factors including irrigation water sources (pond and well), type of soil amendment: fresh poultry litter (PL), PL ash, and a conventional fertilizer (triple superphosphate - TSP), and production practices: staked with plastic mulch (SP), staked without plastic mulch (SW), and non-staked without plastic mulch (NW), were evaluated by split-plot or complete-block design. All field experiments relied on naturally occurring Salmonella contamination, except one follow up experiment (worst-case scenario) which examined the potential for contamination in tomato fruits when Salmonella was applied through drip irrigation. Samples were collected from pond and well water; PL, PL ash, and TSP; and the rhizosphere, leaves, and fruits of tomato plants. Salmonella was quantified using a most probable number method and contamination ratios were calculated for each treatment. Salmonella serovar was determined by molecular serotyping. Salmonella populations varied significantly by year; however, similar trends were evident each year. Findings showed use of untreated pond water and raw PL amendment increased the likelihood of Salmonella detection in tomato plots. Salmonella Newport and Typhimurium were the most frequently detected serovars in pond water and PL amendment samples,

respectively. Interestingly, while these factors increased the likelihood of Salmonella detection in tomato plots (rhizosphere and leaves), all tomato fruits sampled (n = 4800) from these plots were Salmonella negative. Contamination of tomato fruits was extremely low (< 1%) even when tomato plots were artificially inoculated with an attenuated Salmonella Newport strain (104 CFU/mL). Furthermore, Salmonella was not detected in tomato plots irrigated using well water and amended with PL ash or TSP. Production practices also influenced the likelihood of Salmonella detection in tomato plots. Salmonella detection was higher in tomato leaf samples for NW plots, compared to SP and SW plots. This study provides evidence that attention to agricultural inputs and production practices may help reduce the likelihood of Salmonella contamination in tomato fields.
ISSN: 1664302X

Xu, X., Gong, L., Wang, B., Wu, Y., Wang, Y., Mei, X., Xu, H., Tang, L., Liu, R., Zeng, Z., Mao, Y., Li, W.
Glycyrrhizin attenuates Salmonella enteric serovar Typhimurium infection: New insights into its protective mechanism
(2018) Frontiers in Immunology, 9 (OCT), art. no. 02321, .
ABSTRACT: Glycyrrhizin (GL), a triterpenoid glycoside, serves important functions in various biological activities, including antiviral and antitumor immune responses. However, the anti-inflammatory effects of GL on Salmonella enterica serovar Typhimurium (ST)-induced injury in mice and the mechanisms underlying the protection of GL are poorly understood. Here, we investigated the effects of GL on host immune responses against ST infection in mice. A phenotypic analysis using hematoxylin and eosin (H&E) staining and transmission electron microscopy showed that GL relieved ST-induced weight loss and intestinal mucosal injury. A colonization assay showed that GL significantly reduced ST colonization in the ileum and colon and translocation to the liver and spleen. An antibacterial activity assay and real-time PCR revealed that GL had no direct inhibitory impact on ST growth or virulence gene expression. ELISA showed that GL pretreatment significantly decreased proinflammatory cytokine (IFN-γ, TNF-α, IL-6) secretion and increased anti-inflammatory cytokine (IL-10) secretion in the ileum, colon and serum of ST-infected mice. Moreover, flora analysis showed that GL reduced Akkermansia, Sutterella, Prevotella and Coprococcus but enriched Parabacteroides and Anaerotruncus in the cecum of ST-infected mice. These results suggest that GL promotes the secretion of immune factors and modulates intestinal flora to prevent further ST infection. We also analyzed the effect of GL on immunocytes and found that GL promoted the phenotypic and functional maturation of murine bone marrow-derived dendritic cells (BMDCs). Flow cytometry and western blotting demonstrated that NF-κB, ERK, and p38 MAPK were required for GL-induced BMDC maturation. The above findings indicate that GL attenuates ST infection by modulating immune function and intestinal flora. This study enriches our current knowledge of GL-mediated immunological function and provides a new perspective on the prevention of Salmonella infection in animals and humans. ISSN: 16643224

Aik, J., Heywood, A.E., Newall, A.T., Ng, L.-C., Kirk, M.D., Turner, R.
Climate variability and salmonellosis in Singapore – A time series analysis
(2018) Science of the Total Environment, 639, pp. 1261-1267.
ABSTRACT: Climate change is expected to bring about global warming and an increase in the frequency of extreme weather events. This may consequently influence the transmission of food-borne diseases. The short term associations between climatic conditions and Salmonella infections are well documented in temperate climates but not in the tropics. We conducted an ecological time series analysis to estimate the short term associations between non-outbreak, non-travel associated reports of Salmonella infections and observed climatic conditions from 2005 to 2015 for Singapore. We used a negative binomial time series regression model to analyse the associations on a weekly scale, controlling for season, long term trend, delayed weather effects, autocorrelation and the period where Salmonella was made legally notifiable. There were a total of 11,324 Salmonella infections reported during our study period. A 1 °C increase in mean ambient air temperature was associated with a 4.3% increase (Incidence Rate Ratio [IRR]: 1.043, 95% confidence interval [CI] = 1.003, 1.084) in reported Salmonella infections in the same week and a 6.3% increase (IRR: 1.063, 95% CI = 1.022, 1.105) three weeks later. A 1% increase in the mean relative humidity was associated with a 1.3% decrease (IRR: 0.987, 95% CI = 0.981, 0.994) in cases six weeks later, while a 10 mm increase in weekly cumulative rainfall was associated with a 0.8% increase (IRR: 1.008, 95% CI = 1.002, 1.015) in cases 2 weeks later but a 0.9% decrease (IRR: 0.991, 95% CI = 0.984, 0.998) in cases 5 weeks later. No thresholds for these weather effects were detected. This study confirms the short-term influence of climatic conditions on Salmonella infections in

Singapore and the potential impact of climate change on Salmonellosis in the tropics. ISSN: 00489697

Benoun, J.M., Peres, N.G., Wang, N., Pham, O.H., Rudisill, V.L., Fogassy, Z.N., Whitney, P.G., Fernandez-Ruiz, D., Gebhardt, T., Pham, Q.-M., Puddington, L., Bedoui, S., Strugnell, R.A., McSorley, S.J.
Optimal protection against Salmonella infection requires noncirculating memory (2018) Proceedings of the National Academy of Sciences of the United States of America, 115 (41), pp. 10416-10421.
ABSTRACT: While CD4 Th1 cells are required for resistance to intramacrophage infections, adoptive transfer of Th1 cells is insufficient to protect against Salmonella infection. Using an epitope-tagged vaccine strain of Salmonella, we found that effective protection correlated with expanded Salmonella-specific memory CD4 T cells in circulation and nonlymphoid tissues. However, naive mice that previously shared a blood supply with vaccinated partners lacked T cell memory with characteristics of tissue residence and did not acquire robust protective immunity. Using a YFP-IFN-γ reporter system, we identified Th1 cells in the liver of immunized mice that displayed markers of tissue residence, including P2X7, ARTC2, LFA-1, and CD101. Adoptive transfer of liver memory cells after ARTC2 blockade increased protection against highly virulent bacteria. Taken together, these data demonstrate that noncirculating memory Th1 cells are a vital component of immunity to Salmonella infection and should be the focus of vaccine strategies. ISSN: 00278424

Birhanu, B.T., Park, N.-H., Lee, S.-J., Hossain, M.A., Park, S.-C.
Inhibition of Salmonella Typhimurium adhesion, invasion, and intracellular survival via treatment with methyl gallate alone and in combination with marbofloxacin (2018) Veterinary Research, 49 (1), art. no. 49, .
ABSTRACT: Salmonella enterica serovar Typhimurium infects intestinal epithelia and macrophages, which is prevented by inhibiting adhesion and cell invasion. This study aimed to investigate the role of methyl gallate (MG) in adhesion, invasion, and intracellular survival of Salmonella Typhimurium in Caco-2 and RAW 264.7 cells via a gentamicin protection assay, confocal microscopy, and quantitative reverse-transcription polymerase chain reaction. MG (30 μg/mL) inhibited adhesion and invasion of Salmonella Typhimurium by 54.01% and 60.5% in RAW 264.7 cells, respectively. The combination of MG with sub-minimum inhibitory concentration (MIC) of marbofloxacin (MRB) inhibited the adhesion, invasion, and intracellular survival by 70.49%, 67.36%, and 74%, respectively. Confocal microscopy further revealed reductions in bacterial count in Caco-2 cells treated with MG alone or with sub-MIC of MRB. Furthermore, MG alone or in combination with sub-MIC of MRB decreased the motility of Salmonella Typhimurium. Quorum sensing genes including sdiA, srgE, and rck were downregulated by 52.8%, 61.7%, and 22.2%, respectively. Moreover, rac-1 was downregulated by 56.9% and 71.9% for MG alone and combined with sub-MIC of MRB, respectively, in mammalian cells. Furthermore, MG downregulated virulence genes of Salmonella Typhimurium including cheY, ompD, sipB, lexA, and ompF by 59.6%, 60.2%, 20.5%, 31.4%, and 16.2%, respectively. Together, the present results indicate that MG alone or in combination with a sub-MIC of MRB effectively inhibited the adhesion, invasion, and intracellular survival of Salmonella Typhimurium in vitro by downregulating quorum sensing and virulence genes. ISSN: 09284249

Uğur, S., Akçelik, N., Yüksel, F.N., Taşkale Karatuğ, N., Akçelik, M.
Effects of dam and seqA genes on biofilm and pellicle formation in Salmonella (2018) Pathogens and Global Health, 112 (7), pp. 368-377.
ABSTRACT: In this study, the effects of dam and seqA genes on the formation of pellicle and biofilm was determined using five different Salmonella serovars S. Group C1 (DMC2 encoded), S. Typhimurium (DMC4 encoded), S. Virchow (DMC11 encoded), S. Enteritidis (DMC22 encoded), and S. Montevideo (DMC89 encoded). dam and seqA mutants in Salmonella serovars were performed by the single step lambda red recombination method. The mutants obtained were examined according to the properties of biofilm on the polystyrene surfaces and the pellicle formation on the liquid medium. As a result of these investigations, it was determined that the biofilm formation properties on polystyrene surfaces decreased significantly ($p < 0.05$) in all tested dam and seqA mutants, while the pellicle formation properties were lost in the liquid medium. When pBAD24 vector, containing the dam and seqA genes cloned behind the inducible arabinose promoter, transduced into dam and seqA mutant strains, it was determined that the biofilm formation properties on the polystyrene surfaces reached to the natural strains' level in all mutant strains. Also, the pellicle formation ability was regained in the liquid media. All these data

demonstrate that dam and seqA genes play an important role in the formation of biofilm and pellicle structures in Salmonella serovars. ISSN: 20477724

Yin, B., Zhu, L., Zhang, Y., Dong, P., Mao, Y., Liang, R., Niu, L., Luo, X.
The Characterization of Biofilm Formation and Detection of Biofilm-Related Genes in Salmonella Isolated from Beef Processing Plants
(2018) Foodborne Pathogens and Disease, 15 (10), pp. 660-667.
ABSTRACT: The biofilm formation behavior of Salmonella isolated from beef processing plants was investigated under varying temperatures (4°C, 10°C, 25°C, 37°C, and 42°C) and pH (4.5, 5.0, 5.5, 6.0, 7.0, and 8.0). The relationships between the presence of biofilm-related genes and the biofilm formation capacity were evaluated. A total of 77 Salmonella strains in 8 different serotypes were assessed: Salmonella Agona (n = 43), Salmonella Senftenberg (n = 13), Salmonella Meleagridis (n = 8), Salmonella Derby (n = 7), Salmonella Kottbus (n = 2), Salmonella Calabar (n = 2), Salmonella Kingston (n = 1), and Salmonella Typhimurium (n = 1). The results showed that all tested Salmonella strains produced biofilm at 25°C and 37°C after 3 d, and Salmonella Kingston and Salmonella Senftenberg had higher biofilm production than other strains under test conditions. Serotype, incubation temperature, pH, and their interactions had significant effects on biofilm formation for Salmonella. The strongest biofilm formation capacity of Salmonella (serovar Agona, Senftenberg, Kottbus, Calabar, Kingston, and Typhimurium) occurred at 25°C and at pH 7.0. Biofilm formation was significantly inhibited for all Salmonella strains incubated at 4°C. The detection rates of genes rpoS, fliC, wcaA, and invA were 100%, and the rates of genes csgB, csgD, csrA, sirA, adrA, gly, fimH, sdiA, ompR, sipB, sipC, luxS, and pfs exceeded 75% among all biofilm producer strains. The detection rate of igaA was significantly different between different biofilm producers. Based on the findings in this study, useful information on biofilm formation of Salmonella isolated from beef processing plants in China is provided, which could help clear the technological hurdle in delaying biofilm production to deal with risks from Salmonella biofilms in the beef industry. ISSN: 15353141

Ainslie-Garcia, M.H., Farzan, A., Newman, J.E., Friendship, R.M., Lillie, B.N.
Salmonella fecal shedding in pigs from birth to market and its association with the presence of salmonella in palatine tonsils and submandibular lymph nodes at slaughter
(2018) Canadian Journal of Veterinary Research, 82 (4), pp. 249-255.
ABSTRACT: Salmonella is an important cause of foodborne illnesses in humans. Food-producing animals, including swine, are a major source of Salmonella in food products. This study investigated on farm Salmonella fecal shedding in pigs at different production stages — from weaning to marketing — and its association with the presence of Salmonella in tissues at slaughter. Fourteen groups from 8 commercial farrowing sources (N = 809 pigs) were monitored 5 times from birth to slaughter. Fecal and tissue samples were collected from pigs and cultured for Salmonella. A survey was conducted to collect farm management information. A multi-level mixed-effects logistic regression modelling method was used to analyze Salmonella shedding over time and the association between Salmonella shedding and the presence of Salmonella in tissue samples. Salmonella was recovered from 13% (421/3339) of fecal samples collected from 809 pigs over the course of the study. Overall, 35% (284) of pigs shed Salmonella at least once, while 12% (99) shed more than once. Salmonella shedding increased as pigs aged ($P = 0.01$) and increased in the summer months ($P < 0.01$). Salmonella was isolated from tissue samples collected from 23% (134/580) of pigs; however, the presence of Salmonella at slaughter was not associated with on farm shedding. The seasonal trend in Salmonella shedding and its association with age may be used to identify high-risk groups and implement more effective control measures accordingly. The identification of repeat shedders warrants interventions that target this source of infection on swine farms. ISSN: 08309000

Morales-Partera, A.M., Cardoso-Toset, F., Luque, I., Astorga, R.J., Maldonado, A., Herrera-León, S., Hernández, M., Gómez-Laguna, J., Tarradas, C.
Prevalence and diversity of Salmonella spp., Campylobacter spp., and Listeria monocytogenes in two free-range pig slaughterhouses
(2018) Food Control, 92, pp. 208-215.
ABSTRACT: Salmonella spp., Campylobacter spp. and Listeria monocytogenes have a significant impact on public health with slaughterhouses providing many opportunities for the proliferation of pathogenic bacteria. To evaluate the prevalence and diversity of these microorganisms along the free-range pork production chain, a total of 750 samples (5 samples/animal; 15 animals/farm; 5 farms/slaughterhouse) were collected from two slaughterhouses and analysed by specific ISO methodologies. Salmonella spp. (12.93%, CI95 10.72–15.52%), Campylobacter spp. (17.17%, CI95 13.00–21.74%) and L.

monocytogenes (9.07%, CI95 7.21–11.33%) were recovered at different stages of the production chain, with the highest prevalence detected in tonsils for Salmonella spp. (30.67%, CI95 23.85–38.44%) and L. monocytogenes (39.33%, CI95 31.87–47.32%) and in faeces for Campylobacter spp. (57.33%, CI95 49.33–64.96%). Thirteen different Salmonella serotypes were detected with monophasic Salmonella Typhimurium as the most frequent one. C. coli, C. jejuni and L. monocytogenes serotype 4b and 1/2a were also identified. A significant higher prevalence of Salmonella spp. in total and from skin samples in slaughterhouse B than in slaughterhouse A was detected. In addition, a higher, although not significant, prevalence of the selected pathogens was observed in meat samples from slaughterhouse B with respect to slaughterhouse A (10.67% vs 0% for Campylobacter spp.; and 4% vs 0% for Salmonella spp. and L. monocytogenes). Our results highlight the risk of contamination of pork meat by the microorganisms under study and point out the importance of implementing specific control measures. ISSN: 09567135

Heymans, R., Vila, A., van Heerwaarden, C.A.M., Jansen, C.C.C., Castelijn, G.A.A., van der Voort, M., Biesta-Peters, E.G.
Rapid detection and differentiation of Salmonella species, Salmonella Typhimurium and Salmonella Enteritidis by multiplex quantitative PCR
(2018) PLoS ONE, 13 (10), art. no. e0206316, .
ABSTRACT: A multiplex quantitative PCR (qPCR) was developed and evaluated for the simultaneous detection of Salmonella spp., S. enterica serovar Typhimurium and S. enterica serovar Enteritidis in various (food) matrices. Early and fast detection of these pathogens facilitates effective intervention and prevents further distribution of contaminated food products on the market. Three primer and probe sets were designed to target the invA gene, the STM4200 gene, and the SEN1392 gene to detect and differentiate Salmonella spp., S. Typhimurium, and S. Enteritidis, respectively. The multiplex qPCR targeting these three genes was optimized for efficiency and linearity. By testing 225 Salmonella isolates and 34 non-Salmonella isolates from various sources the inclusivity and exclusivity were determined. The inclusivity of the multiplex qPCR was 100% for all Salmonella isolates, including 72 S. Typhimurium isolates, and 53 S. Enteritidis isolates. The exclusivity for Salmonella spp., S. Typhimurium, and S. Enteritidis was 100%, 94.6%, and 100%, respectively. No positive results were reported for non-Salmonella isolates. The limit of detection (LOD) for the qPCR was determined for the matrices poultry, minced meat, egg, herbs/spices, powdered milk, fish, animal feed, bootsocks with chicken feces and chicken down. LOD values for qPCR and the conventional culture methods were similar, except for the matrix boot-socks and down, for which the LOD for the conventional culture methods performed better than the qPCR method. In conclusion, the multiplex qPCR assay developed allows for rapid screening of Salmonella spp., S. Typhimurium, and S. Enteritidis in various (food) matrices. ISSN: 19326203

Minor Fernandes Inagaki, J., Jagnow Sereno, M., Pegoraro, K., Zanatta Waz, M., Mendonça Soares, V., Gonçalves Pereira, J., Bersot, L.D.S.
Effect of organic matter and pH on the resistance of Salmonella Typhimurium and Salmonella Derby in scalding water from pig slaughter
(2018) Journal of Food Safety, 38 (5), art. no. e12504, .
ABSTRACT: The thermal resistance of Salmonella Typhimurium and Derby was evaluated simulating the pig slaughter scalding process in water with different pH values and organic matter concentrations. In samples of scalding water tanks collected at 0, 2, and 4 hr during the slaughter, Salmonella serotypes were quantified at 0, 1, 3, and 5 min of heating at 62 °C. Variations of water with different pH levels and organic matter contents were also analyzed. A general reduction of 3.19 log CFU/mL in the scalding water and a D value of 1.65 min was observed. Salmonella was more sensitive to hot water in an alkaline pH, and the organic matter was not able to interfere with the survival of Salmonella. We conclude that the accumulation of organic matter in the scalding process was not relevant for the survival of Salmonella and that the water alkalinization was important in improving the elimination of this microorganism. Practical applications: To know the behavior of Salmonella in different stages of pig slaughter is important for establishing parameters of process, limits, or actions in the control of the contamination program. This study may contribute novel predictions about Salmonella elimination in the scalding stage and provide evidence about factors that can modify the kinetics of reduction, such as the accumulation of organic matter in the scalding tank during the slaughter of pigs and the different pH levels of the water used in this process. This article demonstrated that the addiction of calcium oxide to the scalding water may contribute to the control of Salmonella in the scalding process by reducing the destruction time. ISSN: 01496085

McLauchlin, J., Aird, H., Charlett, A., Chattaway, M., Elviss, N., Hartman, H., Jenkins, C., Jørgensen, F., Larkin, L., Sadler-Reeves, L., Willis, C.
Imported edible leaves collected at retail sale in England during 2017 with an emphasis on betel and curry leaves: microbiological quality with respect to Salmonella, Shiga-toxin-producing E. coli (STEC) and levels of Escherichia coli
(2018) Journal of Applied Microbiology, 125 (4), pp. 1175-1185.
ABSTRACT: Aims: To investigate the microbiological quality of imported fresh leaves on retail sale during 2017 with respect to Salmonella, Shiga-toxin-producing Escherichia coli (STEC) and levels of E. coli. Methods and Results: Two hundred and seventy-nine samples of imported edible leaves (69 banana, 77 betel, 118 curry and 15 other types) were tested. Salmonella spp. were confirmed by whole-genome sequencing and isolated from 44 samples, 26% from curry leaves, 14% from betel and 2·4% from all other leaf types: 80% of all samples contained ≥102, 44% ≥103 and 22% ≥104 CFU of E. coli CFU per g. All samples where Salmonella were detected also yielded ≥20 CFU of E. coli/g. 54 samples were tested for STEC which was detected in six samples and isolated from three: one was identified as STEC O157:H7. Conclusions: This report further highlights an ongoing problem of Salmonella contamination of imported fresh edible leaves. Significance and Impact of the Study: Among all food tested by Public Health England (approximately 11 000 per annum), curry leaves were the herb most commonly contaminated with Salmonella, and betel leaves were the most commonly contaminated ready-to-eat food. The high proportion with unsatisfactory E. coli levels and the detection of STEC suggests risks of contamination by multiple enteric pathogens. ISSN: 13645072

Shi, H., Zhou, X., Zou, W., Wang, Y., Lei, C., Xiang, R., Zhou, L., Liu, B., Zhang, A., Wang, H.
Co-occurrence of biofilm formation and quinolone resistance in Salmonella enterica serotype typhimurium carrying an IncHI2-type oqxAB-positive plasmid
(2018) Microbial Pathogenesis, 123, pp. 68-73.
ABSTRACT: The objective of this study was to investigate the co-occurrence of biofilms and quinolone resistance in Salmonella enterica serotype Typhimurium mediated by IncHI2-type oqxAB-positive plasmids. Among the 40 Salmonella strains, we found that 27 isolates formed biofilms and displayed identical multidrug-resistance profiles to ciprofloxacin, doxycycline, sulfamethoxazole-trimethoprim, ampicillin and streptomycin, based on biofilm formation assays and antimicrobial susceptibility testing. In particular, a single S. Typhimurium isolate named SC523 produced the thickest biofilms and exhibited the highest-level resistance (MIC = 8 μg/mL) to ciprofloxacin compared to those of the other isolates. The detection of known plasmid-mediated quinolone resistance (PMQR) genes and point mutations in the quinolone resistance-determining region (QRDR) by PCR assay showed that oqxAB genes were present in 27 biofilm-positive isolates. Conjugation experiments, S1-pulse-field gel electrophoresis and biofilm formation assays demonstrated that the conjugative plasmid that encoded biofilms and quinolone resistance in Salmonella SC523 could be transferred to a recipient with a frequency of 4.7 × 10−3 per recipient cell. The results of PCR-based replicon typing (PBRT) showed that the IncHI2-type plasmids accounted for 100% of the biofilm-oqxAB-positive isolates and transconjugants. The sequence analysis of Salmonella SC523 confirmed that the oqxAB cassette and fourteen DNA transfer genes in the IncHI2-type oqxAB-positive conjugative plasmid were genetically responsible for the phenotypic quinolone resistance and biofilm formation. The conclusion is that the IncHI2-type plasmid in S. Typhimurium isolate from chicken farm was identified and sequenced, which contained oqxAB and tra/trh and encoded quinolone resistance and biofilms, and could be transferred to recipients through conjugation. Notably, the prevalence of IncHI2-type biofilm-oqxAB-positive plasmids in animal-origin Salmonella poses a threat to public health, as these Salmonella from poultry farms show a decreased susceptibility to quinolones and could spread to humans. ISSN: 08824010

Duffy, L.L., Osmond-McLeod, M.J., Judy, J., King, T.
Investigation into the antibacterial activity of silver, zinc oxide and copper oxide nanoparticles against poultry-relevant isolates of Salmonella and Campylobacter
(2018) Food Control, 92, pp. 293-300.
ABSTRACT: Nanotechnology is currently contributing substantially to the development of a broad range of innovative technologies in the agricultural, feed and food sector. To date, very little work has been done to investigate the efficacy of antimicrobial nanoparticles against the pathogens of highest concern to the poultry industry. This study is the first to report on the effectiveness of CuO nanoparticles against Salmonella and on the effectiveness of Ag and CuO nanoparticles against Campylobacter. The aim of this study was to assess the in vitro activity of silver (Ag), zinc oxide (ZnO) and copper oxide (CuO) nanoparticles against Salmonella and Campylobacter isolated from poultry. The minimum

inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were determined by a broth microdilution method in 96 well plates. The MIC for all Campylobacter strains was in the order of Au ≥ CuO ≥ ZnO nanoparticles. Ag nanoparticles were the most effective against Salmonella. The growth kinetics of the Salmonella strains were also assessed during exposure to nanoparticles in liquid media. Growth was dependant on NP concentration with some differences noted between strains. While these specific size nanoparticles are effective against pure cultures of bacteria the antimicrobial effectiveness of the nanoparticles should be further examined under industry-relevant conditions. ISSN: 09567135

Henry, A.E., Letellier, A., Cŏté, J.-C., Desmarais, G., Lachapelle, V., Bergeron, N., Lewandowsky, S.L., Fravalo, P.
Overlooked sources of Salmonella contamination in the pig production network: Slaughterhouse yard pathways and mudguards and carpets from transport trucks
(2018) Canadian Veterinary Journal, 59 (10), pp. 1105-1108.
ABSTRACT: This report describes various Salmonella serovars which were found on often overlooked locations in a pig farm/slaughterhouse interface. These include slaughterhouse yard pathways and mudguards and carpets of transport trucks arriving at and departing from production sites. ISSN: 00085286

Omer, M.K., Álvarez-Ordoñez, A., Prieto, M., Skjerve, E., Asehun, T., Alvseike, O.A.
A Systematic Review of Bacterial Foodborne Outbreaks Related to Red Meat and Meat Products
(2018) Foodborne Pathogens and Disease, 15 (10), pp. 598-611.
ABSTRACT: Our investigation focused on foodborne outbreaks related to meat and meat products, published in peer-reviewed journals in the period 1980-2015. Most of the outbreaks, investigated in this study, were caused by Escherichia coli and Salmonella, causing 33 and 21 outbreaks, respectively, mostly in Europe and the United States. In the E. Coli outbreaks, the total number of reported cases was 1966, of which 1543 were laboratory confirmed. The number of cases requiring hospitalization was 476, of whom 233 cases had a hemolytic-uremic syndrome (HUS), and the reported deaths were 32. All of the E. Coli outbreaks, except four, were caused by serovar O157:H7. The other four outbreaks were caused by the following serovars: O111:H8, O26:H11, O111, and O103:H25. Fresh processed meat products were the category most frequently implicated. In the Salmonella outbreaks, the total number of all reported cases was 2279, of whom 1891 were laboratory confirmed. The number of reported cases requiring hospitalization was 94, and seven were reported dead. Regarding Salmonella, eight serovars caused those outbreaks. The most common serovar causing Salmonella-related outbreaks was Salmonella Typhimurium. The food category most frequently implicated in those outbreaks was raw-cured fermented sausages. Other organisms linked to meat-associated outbreaks, but less frequently reported, were Staphylococcus aureus, Bacillus cereus, Clostridium perfringens, Clostridium botulinum, and Listeria monocytogenes. Issues of the burden of outbreaks, the challenges of comparing global outbreaks, food attribution, and how the meat industry works to meet consumer demands while maintaining food safety are discussed. ISSN: 15353141

Farahani, R.K., Ehsani, P., Ebrahimi-Rad, M., Khaledi, A.
Molecular detection, virulence genes, biofilm formation, and antibiotic resistance of salmonella enterica serotype enteritidis isolated from poultry and clinical samples
(2018) Jundishapur Journal of Microbiology, 11 (10), art. no. e69504, 9 p.
ABSTRACT: Background: Salmonella spp. is one of the most important zoonotic pathogens transmitting among human and animals. Due to the similarity of antibiotic classes used to treat animals and humans, there is a high risk for emerging the multi-drug resistant (MDR) strains. Objectives: The current study aimed at evaluating molecular detection, virulence genes, biofilm formation, and antibiotic resistance of Salmonella enterica serotype enteritidis recovered from poultry and clinical isolates. Methods: A total of 282 isolates were recovered from chicken meat, live poultry feces, eggs, and human feces in Iran. The presence of virulent factors in the isolates was confirmed using biochemical and microbiological tests. The presence of Salmonella genus was determined using antiserum. Triplex polymerase chain reaction (PCR) was performed to detect Salmonella spp., serogroup D and the discriminate S. enteritidis from other species. Kirby-Bauer disk diffusion method was applied to perform the susceptibility testing. Quantification of biofilm formation was determined in 96-well microtiter plates as recommended by the defined protocol. The data were then analyzed with SPSS using consensus tables and Chi-square test. Results: Based on the results, all the isolates were positive for invA, sdiA, hilA, and ratA. Moreover, spvC had the lowest prevalence (37.6%). Of all strains, 67% were MDR,

51.7% of which were recovered from humans. Furthermore, 34.5% of isolates were strong biofilm producers. There was a significant correlation between the strong biofilm formation and the antibiotic resistance to colistin, ceftazidime, chloramphenicol, gentamicin, trimethoprim, penicillin, and trimethoprim-sulfamethoxazole. Conclusions: The results of the current study showed a significant correlation between the strong biofilm formation and the antibiotic resistance to some antibiotics. ISSN: 20083645

Mokracka, J., Krzymińska, S., Ałtunin, D., Wasyl, D., Koczura, R., Dudek, K., Dudek, M., Chyleńska, Z.A., Ekner-Grzyb, A.
In vitro virulence characteristics of rare serovars of Salmonella enterica isolated from sand lizards (Lacerta agilis L.)
(2018) Antonie van Leeuwenhoek, International Journal of General and Molecular Microbiology, 111 (10), pp. 1863-1870.
ABSTRACT: The aim of this study was to estimate virulence potential of Salmonella enterica strains colonizing the gut of free-living sand lizards (Lacerta agilis L.). The strains belonged to three Salmonella serovars: Abony, Schleissheim, and Telhashomer. Adhesion and invasion abilities of the strains were determined in quantitative assays using the gentamicin protection method. Induction of apoptosis was assessed using HeLa cell monolayers. PCR assays were used for detection of 26 virulence genes localised within mobile elements: pathogenicity islands, virulence plasmids, and prophage sequences. In vitro studies revealed that all strains had adhesion and invasion abilities to human epithelial cells. The isolates were cytotoxic and induced apoptosis of the cells. The serovars differed in the number of virulence-associated genes: up to 18 genes were present in Salmonella Schleissheim, 17 in Salmonella Abony, whereas as few as six genes were found in Salmonella Telhashomer. Generally, Salmonella Abony and Salmonella Schleissheim did not differ much in gene content connected with the presence SPI-1 to -5. All of the strains lacked genes localised within bacteriophages and plasmids. The presence of virulence-associated genes and in vitro pathogenicity assays suggest that Salmonella sp. strains originating from autochthonous, free-living lizards can potentially infect and cause disease in humans. ISSN: 00036072

Esteban-Cuesta, I., Drees, N., Ulrich, S., Stauch, P., Sperner, B., Schwaiger, K., Gareis, M., Gottschalk, C.
Endogenous microbial contamination of melons (Cucumis melo) from international trade: an underestimated risk for the consumer?
(2018) Journal of the Science of Food and Agriculture, 98 (13), pp. 5074-5081.
ABSTRACT: BACKGROUND: Fruits and vegetables have increasingly been related to foodborne outbreaks. Besides surface contamination, a possible internalization of microorganisms into edible parts of plants during growth has already been observed. To examine an actual risk for the consumer, microbial contamination of the rind and pulp of 147 muskmelons from international trade was assessed using cultural and biochemical methods, polymerase chain reaction and matrix-assisted laser desorption/ionization-time of flight mass spectrometry. RESULTS: One hundred percent of the rind samples [3.69–8.92 log colony forming units (CFU) g−1] and 89.8% of the pulp samples (maximum load 3.66 log CFU g−1) were microbiologically contaminated. Among the 432 pulp isolates, opportunistic and potentially pathogenic bacteria were identified, mainly Staphylococcus spp. (48.9%), Clostridium spp. (42.9%) and Enterobacteriaceae (27.9%). Salmonella spp., Escherichia coli and isolates of the Bacillus cereus group were found on the rind (1.4%, 0.7% and 42.9%, respectively) and in the pulp (0.7%, 1.4% and 4.7%). Clostridium perfringens was isolated from the rind of seven melons. CONCLUSION: The present study revealed a regularly occurring internal contamination of melons. Possible health risks for consumers because of an occurrence of microorganisms in melon pulp should be considered in future food safety assessments. ISSN: 00225142

Crim, S.M., Chai, S.J., Karp, B.E., Judd, M.C., Reynolds, J., Swanson, K.C., Nisler, A., McCullough, A., Gould, L.H.
Salmonella enterica Serotype Newport Infections in the United States, 2004-2013: Increased Incidence Investigated Through Four Surveillance Systems
(2018) Foodborne Pathogens and Disease, 15 (10), pp. 612-620.
ABSTRACT: Newport is the third most common Salmonella enterica serotype identified among the estimated 1.2 million human salmonellosis infections occurring annually in the United States. Risk factors for infection and food items implicated in outbreaks vary by antimicrobial resistance pattern. We conducted a descriptive analysis of data from four enteric disease surveillance systems capturing information on incidence, demographics, seasonality, geographic distribution, outbreaks, and antimicrobial resistance of Newport infections over a 10-year period from 2004 through 2013. Incidence increased through

2010, then declined to rates similar to those in the early years of the study. Incidence was highest in the South and among children <5 years old. Among isolates submitted for antimicrobial susceptibility testing, 88% were susceptible to all antimicrobials tested (pansusceptible) and 8% were resistant to at least seven agents, including ceftriaxone. Rates of pansusceptible isolates were also highest in the South and among young children, particularly in 2010. Pansusceptible strains of Newport have been associated with produce items and environmental sources, such as creek water and sediment. However, the role of environmental transmission of Newport in human illness is unclear. Efforts to reduce produce contamination through targeted legislation, as well as collaborative efforts to identify sources of contamination in agricultural regions, are underway. ISSN: 15353141

Peng, M., Salaheen, S., Buchanan, R.L., Biswas, D.
Alterations of Salmonella enterica serovar Typhimurium antibiotic resistance under environmental pressure
(2018) Applied and Environmental Microbiology, 84 (19), art. no. e01173-18, .
ABSTRACT: Microbial horizontal gene transfer is a continuous process that shapes bacterial genomic adaptation to the environment and the composition of concurrent microbial ecology. This includes the potential impact of synthetic antibiotic utilization in farm animal production on overall antibiotic resistance issues; however, the mechanisms behind the evolution of microbial communities are not fully understood. We explored potential mechanisms by experimentally examining the relatedness of phylogenetic inference between multidrug-resistant Salmonella enterica serovar Typhimurium isolates and pathogenic Salmonella Typhimurium strains based on genome-wide single-nucleotide polymorphism (SNP) comparisons. Antibiotic-resistant S. Typhimurium isolates in a simulated farm environment barely lost their resistance, whereas sensitive S. Typhimurium isolates in soils gradually acquired higher tetracycline resistance under antibiotic pressure and manipulated differential expression of antibiotic-resistant genes. The expeditious development of antibiotic resistance and the ensuing genetic alterations in antimicrobial resistance genes in S. Typhimurium warrant effective actions to control the dissemination of Salmonella antibiotic resistance. ISSN: 00992240

Holstege, M.M.C., de Bont-Smolenaars, A.J.G., Santman-Berends, I.M.G.A., van der Linde-Witteveen, G.M., van Schaik, G., Velthuis, A.G.J., Lam, T.J.G.M.
Factors associated with high antimicrobial use in young calves on Dutch dairy farms: A case-control study
(2018) Journal of Dairy Science, 101 (10), pp. 9259-9265.
ABSTRACT: Since 2012, the Dutch Veterinary Medicine Authority reports antimicrobial usage (AMU) in young calves (<56 d) on dairy farms on an annual basis. The AMU distribution in this age group is skewed, with a low AMU in young calves on the majority of dairy farms and a high AMU in a relatively small number of farms. This results in a notable difference between the mean and median AMU. To further reduce the mean AMU, the AMU on the high-AMU farms must be decreased. The objective of this study was to evaluate the association between both young stock management and an indication of the farmers' mindset and AMU in young calves on Dutch dairy farms with a high and low AMU in young calves. This knowledge may be helpful in decreasing AMU in young calves on high-AMU farms. We performed a case-control study in which 200 dairy farms (100 with high AMU and 100 with low AMU in young calves) participated. Case farms were defined as farms with an animal daily-defined dose at the farm level in young calves >28 in 2012 and 2013, based on the 90th percentile of the use of antimicrobials in young calves in 2012. Control farms had an animal daily-defined dose at the farm level in young calves of <0.5 in 2012 and 2013, which was determined to be low use. A questionnaire was conducted about general farm and young stock management, hygiene, housing, vaccination, and calf health. An indication of the farmers' mindset with regard to AMU and treatment of sick calves was determined by including statements (agree/disagree) in the questionnaire. In addition, routinely collected data on herd size, growth in herd size, replacement, and calf mortality were available for analysis. Dairy farmers that immediately started antimicrobial treatment in sick calves had higher odds of being in the high-AMU group than farmers who started treatment of sick calves with supportive nonantimicrobial therapy. Other variables associated with a high AMU in young calves included housing calves on partially slatted floors, a high prevalence of respiratory disease, an unfavorable Salmonella status, and not agreeing with the statement “Young stock need specific management.” Both dairy farm management and opinions of dairy producers regarding AMU, indicative of mindset, are important when distinguishing farms with high and low AMU in young calves. Although the rationale behind mindset warrants more research, likely a change in both aspects seems to be required to reduce the AMU in young calves on dairy farms. ISSN: 00220302

Bu, S., Wang, K., Ju, C., Han, Y., Li, Z., Du, P., Hao, Z., Li, C., Liu, W., Wan, J.
A pregnancy test strip for detection of pathogenic bacteria by using concanavalin A-human chorionic gonadotropin-Cu3(PO4)2 hybrid nanoflowers, magnetic separation, and smartphone readout
(2018) Microchimica Acta, 185 (10), art. no. 464, .
ABSTRACT: Pregnancy test strips are widely used in daily life. A commercial pregnancy test strip was modified to obtain a point-of-care device for the detection of pathogenic bacteria. Hybrid nanoflowers were prepared from concanavalin A, human chorionic gonadotropin, and Cu3(PO4)2 via a one-pot method. They were used as signaling probes in an off-the-shelf pregnancy test strip. This modified lateral flow immunoassay can detect Escherichia coli O157:H7 with a detection limit of 4 CFU·mL−1, and Salmonella typhimurium with a detection limit of 3 CFU·mL−1. Conceivably, the method has high potential as a portable and cost-effective tool for rapid determination of a wide range of analytes, especially in resource-constrained settings. [Figure not available: see fulltext.]. ISSN: 00263672

Prakash, A., Nithyanand, P., Vadivel, V.
In vitro antibacterial activity of nut by-products against foodborne pathogens and their application in fresh-cut fruit model
(2018) Journal of Food Science and Technology, 55 (10), pp. 4304-4310.
ABSTRACT: Aqueous extract of nut by-products (cashewnut shell, coconut shell, and peanut hull) were studied for their physicochemical properties, antibacterial activity and food preservation potential in an artificially inoculated fresh-cut fruit (papaya) model. Physicochemical characteristics revealed the colour, odor, nearly neutral pH (6.67–6.83), high water solubility (69.18–82.63%) and total phenolic content (1130.54–2403.41 mg GAE/100 g) of the extracts. The antibacterial property of the extracts evaluated by zone of inhibition assay revealed that cashew nut shell extract had a strong inhibition effect on Escherichia coli (18 mm), Listeria monocytogenes (18 mm), and Salmonella enterica (16 mm). Food preservative effect of extracts was examined in an artificially inoculated fresh-cut papaya model, and both cashewnut and coconut shell extracts significantly reduced the population of the above mentioned foodborne pathogens. However, when compared to coconut shell extract, the application of cashewnut shell extract was found to affect the sensory property of the fresh-cut fruit as darkening of the cut fruit was observed. So, the coconut shell extract could be considered as a natural source of antibacterial agent for food preservative applications. Phytochemical investigation through LC–MS/MS technique revealed that luteolin as the major constituent of coconut shell extract. ISSN: 00221155

Worley, J., Meng, J., Allard, M.W., Brown, E.W., Timme, R.E.
Salmonella enterica Phylogeny Based on Whole-Genome Sequencing Reveals Two New Clades and Novel Patterns of Horizontally Acquired Genetic Elements
(2018) mBio, 9 (6), .
ABSTRACT: Using whole-genome sequence (WGS) data from the GenomeTrakr network, a globally distributed network of laboratories sequencing foodborne pathogens, we present a new phylogeny of Salmonella enterica comprising 445 isolates from 266 distinct serovars and originating from 52 countries. This phylogeny includes two previously unidentified S. enterica subsp. enterica clades. Serovar Typhi is shown to be nested within clade A. Our findings are supported by both phylogenetic support, based on a core genome alignment, and Bayesian approaches, based on single-nucleotide polymorphisms. Serovar assignments were refined by in silico analysis using SeqSero. More than 10% of serovars were either polyphyletic or paraphyletic. We found variable genetic content in these isolates relating to gene mobilization and virulence factors which have different distributions within clades. Gifsy-1- and Gifsy-2-like phages appear more prevalent in clade A; other viruses are more evenly distributed. Our analyses reveal IncFII is the predominant plasmid replicon in S. enterica Few core or clade-defining virulence genes are observed, and their distributions appear probabilistic in nature. Together, these patterns demonstrate that genetic exchange within S. enterica is more extensive and frequent than previously realized, which significantly alters how we view the genetic structure of the bacterial species.IMPORTANCE Rapid improvements in nucleotide sequencing access and affordability have led to a drastic increase in availability of genetic information. This information will improve the accuracy of molecular descriptions, including serovars, within S. enterica Although the concept of serovars continues to be useful, it may have more significant limitations than previously understood. Furthermore, the discrete absence or presence of specific genes can be an unstable indicator of phylogenetic identity. Whole-genome sequencing provides more rigorous tools for assessing the distributions of these genes. Our phylogenetic and genetic content analyses reveal how active genetic elements

are dynamically distributed within a species, allowing us to better understand genetic reservoirs and underlying bacterial evolution. ISSN: 21507511

Petrosus, E., Silva, E.B., Lay, D., Jr., Eicher, S.D.
Effects of orally administered cortisol and norepinephrine on weanling piglet gut microbial populations and Salmonella passage
(2018) Journal of Animal Science, 96 (11), pp. 4543-4551.
ABSTRACT: Stress and anxiety have been associated with changes in the microbiota of the gut and ultimately diminished resistance to pathogens. The objective of this study was to observe intestinal microbiota and susceptibility to Salmonella associated with stress hormones, cortisol (CORT), and norepinephrine (NE), in piglets. At weaning, 90 piglets (15 for a Salmonella challenge) were trained to take the carrier (apple juice) orally. At 2 wk after weaning, pens of piglets were assigned randomly to 1 of 3 treatments: control (CNT), NE, or CORT. Blood samples were collected prior to treatment, then piglets were dosed orally with treatments twice on day 0; at 0800 and 1600 h. Control piglets were administered 6.1 mL of the carrier only, NE pigs were administered 40 mg/mL of NE-bitartrate salt dissolved in the carrier, and CORT pigs were administered 12 mg/ mL of hydrocortisone acetate dissolved in the carrier. Jugular blood samples were collected prior to necropsies (n = 5/treatment) at 0800 and 1600 h on day 1, and at 0800 h on days 2, 7, and 14 after treatments were started. A subset of pigs were subjected to a 24-h Salmonella challenge. Jejunal and ileal tissues and jejunal, ileal, cecal, and rectal contents were collected and colonies were counted. Microbial data and blood samples were analyzed using mixed models with fixed effects of treatment and day. Cortisol-treated piglets exhibited a spike in plasma CORT concentrations at 0800 h day 1 ($P = 0.001$) accompanied by greater concentrations of cecal Escherichia coli ($P < 0.05$) and a shift in intestinal environment to favor coliforms on day 2 ($P < 0.05$). Salmonella concentrations from rectal contents tended ($P = 0.07$) to be suppressed by CORT. Lactic acid-producing bacteria rectal concentrations were greater ($P = 0.03$) in CORT pigs on 0800 h on day 1 then NE pigs and tended to be greater than CNT ($P = 0.09$) and were greater on day 14 for both CNT ($P = 0.003$) and NE ($P = 0.02$). Norepinephrine spiked in NE piglets at 0800 h on day 1 ($P = 0.001$), 1600 h day 1 ($P = 0.004$), through day 2 ($P = 0.04$). Intestinal environment of NE pigs shifted to favor ileal anaerobes ($P = 0.05$) and facultative anaerobes (E. coli; $P = 0.01$) compared to CNT. However, Salmonella concentrations in rectal contents were suppressed by NE compared to CNT ($P = 0.05$). Oral administration of NE and CORT had the desired effect of increasing concentrations of stress hormones and resulted in microbiome shifts throughout the intestines. ISSN: 00218812

Huang, R., Chen, H.
Evaluation of inactivating Salmonella on iceberg lettuce shreds with washing process in combination with pulsed light, ultrasound and chlorine
(2018) International Journal of Food Microbiology, 285, pp. 144-151.
ABSTRACT: This study was conducted to investigate the Salmonella inactivation effects of washing in combination with pulsed light (PL), ultrasound, and chlorine on lettuce shreds. First, the effect of washing combined with PL and chlorine on the inactivation of Salmonella on lettuce and in wash water was evaluated in a small-scale study with clear tap water and turbid tap water containing lettuce extract and silicon dioxide. In general, water wash combined with PL (PL wash) and chlorine wash combined with PL (PL-Cl) were significantly more effective on killing Salmonella on lettuce than the chlorine wash and water wash regardless the wash water quality and inoculation method. We then tested washing combined with PL, ultrasound and chlorine using a large-scale UV setup with turbid wash water. Increasing the sample size decreased the decontamination efficacy of all the treatments. All the treatments resulted in <2 log reductions of Salmonella on lettuce shreds. For both small- and large-scale studies, treatments involving chlorine could keep the Salmonella population in wash water under the detection limit of 2 CFU/mL for almost all the replicates. Taking everything into consideration, we concluded that the combined PL-Cl treatment could be a better alternative to the chlorine wash for lettuce decontamination since it was in general more effective on inactivating Salmonella on lettuce than chlorine wash and could maintain the Salmonella level in wash water under the detection limit of 2 CFU/mL regardless the inoculation method, water quality and sample size, preventing the potential cross contamination through wash water.
ISSN: 01681605

Kang, I.-B., Kim, D.-H., Jeong, D., Park, J.-H., Seo, K.-H.
Heat resistance of Salmonella Enteritidis under prolonged exposure to acid-salt combined stress and subsequent refrigeration
(2018) International Journal of Food Microbiology, 285, pp. 165-172.

ABSTRACT: Salmonella Enteritidis is a major foodborne pathogen exposed to various environmental and preservation stresses in the food chain. Because adaptive responses of viable bacterial cells in the presence of sublethal stress can induce cross-protection against different stresses, we investigated the heat resistance of Salmonella Enteritidis at 60 °C under prolonged exposure to acid-salt combined stress and subsequent refrigeration. Salmonella Enteritidis was grown in tryptic soy broth at four pH values (4.5, 5.4, 6.4, and 7.3) and four NaCl concentrations (0%, 1%, 2%, and 3%) at 37 °C for 24 h and then incubated at 4 °C for 0, 1, 4, or 7 days. For 0 and 1 day-refrigerated cultures, previous adaptation to single stresses (acid or salt stress) increased the heat resistance of Salmonella Enteritidis, resulting in increased D-values, whereas the combination of acid and salt stress reduced heat tolerance; acid stress played a more critical role in mediating this effect than salt concentration. To elucidate the related mechanisms, the expression levels of heat shock sigma factors (rpoH) and heat shock genes (dnaK and groEL) were analyzed and found to be associated with the heat resistance of Salmonella Enteritidis. The refrigeration period was negatively correlated ($P < 0.01$) with the D-value ($r = -0.505$) and with the transcript levels of rpoH ($r = -0.654$), dnaK ($r = -0.652$), and groEL ($r = -0.645$). Our findings demonstrated that acid-salt combined preservation techniques and subsequent refrigeration may prevent S. Enteritidis survival in heat-pasteurized food products caused by cross-protection of acid or salt adapted cells. ISSN: 01681605

Gavin, C., Simons, R.R.L., Berriman, A.D.C., Moorhouse, D., Snary, E.L., Smith, R.P., Hill, A.A.
A cost-benefit assessment of Salmonella-control strategies in pigs reared in the United Kingdom
(2018) Preventive Veterinary Medicine, 160, pp. 54-62.
ABSTRACT: Pork and pork products are a major source of human salmonellosis in the United Kingdom (UK). Despite a number of surveillance programmes, the prevalence of Salmonella in the UK slaughter pig population remains over 20%. Here, we present the results of a Cost-Benefit Analysis comparing five on-farm control strategies (where the cost is the cost of implementation and the benefits are the financial savings for both the human health and pig industries). The interventions considered were: wet feed, organic acids in feed, vaccination, enhanced cleaning and disinfection and movement of outdoor breeding units. The data originate from published papers and recent UK studies. The effectiveness was assessed by adapting a previous risk assessment, originally developed for the European Food Safety Authority. Using this method, none of the intervention strategies produced a net cost-benefit. Our results suggest that the cost of implementation outweighed the savings for all interventions, even if the effectiveness could be improved. Therefore, to achieve a net cost-benefit it is essential to reduce the cost of interventions. Analyses concluded that large cost reductions (up to 96%) would be required. Use of organic acids required the smallest reduction in cost (22.7%) to achieve a net cost benefit. Uncertainty analysis suggested that a small net gain might be possible, for some of the intervention measures. But this would imply that the model greatly underestimated some key parameters, which was considered unlikely. Areas of key uncertainty were identified as the under-reporting factor (i.e. the proportion of community cases of Salmonella) and the source attribution factor (i.e. the proportion of human Salmonella cases attributable to pork products). ISSN: 01675877

Antunes, P., Campos, J., Mourão, J., Pereira, J., Novais, C., Peixe, L.
Inflow water is a major source of trout farming contamination with Salmonella and multidrug resistant bacteria
(2018) Science of the Total Environment, 642, pp. 1163-1171.
ABSTRACT: The impact of European aquaculture, namely trout farms, in the spread of antibiotic resistance and/or zoonotic pathogens has been scarcely addressed. Moreover, aquaculture contamination sources and bacterial dissemination routes have been barely explored. In this study, we assessed the contribution of Portuguese land-based intensive rainbow trout farms and retailed market trout to the spread of Salmonella and bacteria carrying clinically-relevant antibiotic resistance genes (ARGs) as well as inflow water and feed as possible sources of those contaminants. Cultural and molecular methods were used to analyse 53 fish farm samples (upstream/downstream water and sediments, tanks and trout) and 25 marketed trout. Plasmid-mediated quinolone resistance (PMQR) genes were found in 21% (n = 11/53) of samples (water/sediment/feed/trout), from all collection points (upstream/within/downstream tanks) and seasons, as well as in 12% (n = 3/25) of marketed trout (3 supermarkets). PMQR genes (qnrS1-S2-S3, qnrB7-B19, qnrD1, oqxAB) were detected in Enterobacteriaceae or Aeromonas hydrophila. An E. coli strain producing extended-spectrum-beta-lactamase SHV-12 was detected in all sampled points of a fish farm. Salmonella (4 serotypes, including S. Newport-ST118) was detected in 26% (n =

14/53) of the samples from both farms (water/sediment upstream/within tanks). The clinically-relevant plasmid-mediated colistin resistance mcr genes were not detected. However, colistin resistant S. Abony with new mutations in the chromosomal pmrA and pmrB genes was observed. Identical Salmonella and SHV-12-producing E. coli strains (by PFGE/MLST) in water upstream and within trout tanks points to inflow-water of trout farms as an important source of pathogenic bacteria and ARG contamination. These results highlight the need to define microbiological standards for water supplying fish farms in the EU and to establish surveillance and control strategies to limit bacterial transmission associated with this fastest growing food sector worldwide. ISSN: 00489697

Yang, Z., Chen, X.-B., Tu, C.-N., Su, Y., Wang, H.-B.
A High-throughput Platform for the Screening of Salmonella spp./Shigella spp
(2018) Journal of visualized experiments : JoVE, (141), .
ABSTRACT: Fecal-oral transmission of acute gastroenteritis occurs from time to time, especially when people who handled food and water are infected by Salmonella spp./Shigella spp. The gold standard method for the detection of Salmonella spp./Shigella spp. is direct culture but this is labor-intensive and time-consuming. Here, we describe a high-throughput platform for Salmonella spp./Shigella spp. screening, using real-time polymerase chain reaction (PCR) combined with guided culture. There are two major stages: real-time PCR and the guided culture. For the first stage (real-time PCR), we explain each step of the method: sample collection, pre-enrichment, DNA extraction and real-time PCR. If the real-time PCR result is positive, then the second stage (guided culture) is performed: selective culture, biochemical identification and serological characterization. We also illustrate representative results generated from it. The protocol described here would be a valuable platform for the rapid, specific, sensitive and high-throughput screening of Salmonella spp./Shigella spp. ISSN: 1940087X

Pornsukarom, S., van Vliet, A.H.M., Thakur, S.
Whole genome sequencing analysis of multiple Salmonella serovars provides insights into phylogenetic relatedness, antimicrobial resistance, and virulence markers across humans, food animals and agriculture environmental sources
(2018) BMC genomics, 19 (1), p. 801.
ABSTRACT: BACKGROUND: Salmonella enterica is a significant foodborne pathogen, which can be transmitted via several distinct routes, and reports on acquisition of antimicrobial resistance (AMR) are increasing. To better understand the association between human Salmonella clinical isolates and the potential environmental/animal reservoirs, whole genome sequencing (WGS) was used to investigate the epidemiology and AMR patterns within Salmonella isolates from two adjacent US states. RESULTS: WGS data of 200 S. enterica isolates recovered from human (n = 44), swine (n = 32), poultry (n = 22), and farm environment (n = 102) were used for in silico prediction of serovar, distribution of virulence genes, and phylogenetically clustered using core genome single nucleotide polymorphism (SNP) and feature frequency profiling (FFP). Furthermore, AMR was studied both by genotypic prediction using five curated AMR databases, and compared to phenotypic AMR using broth microdilution. Core genome SNP-based and FFP-based phylogenetic trees showed consistent clustering of isolates into the respective serovars, and suggested clustering of isolates based on the source of isolation. The overall correlation of phenotypic and genotypic AMR was 87.61% and 97.13% for sensitivity and specificity, respectively. AMR and virulence genes clustered with the Salmonella serovars, while there were also associations between the presence of virulence genes in both animal/environmental isolates and human clinical samples. CONCLUSIONS: WGS is a helpful tool for Salmonella phylogenetic analysis, AMR and virulence gene predictions. The clinical isolates clustered closely with animal and environmental isolates, suggesting that animals and environment are potential sources for dissemination of AMR and virulence genes between Salmonella serovars. ISSN: 14712164

Rothrock, M.J., Feye, K.M., Kim, S.A., Park, S.H., Locatelli, A., Hiett, K.L., Gamble, J., Sellers, H., Ricke, S.C.
Semi-quantification of total Campylobacter and Salmonella during egg incubations using a combination of 16S rDNA and specific pathogen primers for qPCR
(2018) Frontiers in Microbiology, 9 (NOV), art. no. 02454, .
ABSTRACT: Rapid molecular techniques that evaluate eggs for the presence of foodborne pathogens is an essential component to poultry food safety monitoring. Interestingly, it is not just table eggs that contribute to outbreaks of foodborne disease. Broiler layer production actively contributes to sustaining of foodborne pathogens within a flock. The surface contamination of production eggs with invasive pathogens such as Salmonella enterica, Campylobacter jejuni, and Listeria monocytogenes during embryogenesis results

in gastrointestinal tract (GIT) colonization. Pathogens that secure a niche within the GIT during embryonic development are nearly impossible to eradicate from the food chain. Therefore, current monitoring paradigms are not comprehensive because they fail to capture the presence of invasive pathogens within the embryonic GIT rapidly. By developing tools to recognize the pathogens' presence in the GIT during embryogenesis, producers are then able to spot evaluate broiler eggs for their potential risk as carriers of foodborne pathogens. In this study a novel qPCR assay was developed to semi-quantify pathogen load relative to total bacterial burden. Eggs sampled from three independent production broiler flocks of different ages were assayed for S. enterica (invA), C. jejuni (HipO), and L. monocytogenes (HlyA) against total microbial load (16s). The eggs were sampled at 1-day post-set within each flock, 2 weeks post-set, after vaccination (at 2.5 weeks) and 1-day post-hatch. The eggs were washed, and the yolk and embryonic chick GIT were collected. The DNA was extracted and subjected to a qPCR assay. The results confirm a novel technique for pathogen monitoring relative to total bacterial load and a unique method for monitoring the dynamics of foodborne pathogen invasion throughout broiler egg production. ISSN: 1664302X

Rauch, H.E., Vosik, D., Kariyawasam, S., M'ikanatha, N., Shariat, N.W.
Prevalence of Group I Salmonella Kentucky in domestic food animals from Pennsylvania and overlap with human clinical CRISPR sequence types
(2018) Zoonoses and Public Health, 65 (7), pp. 831-837.
ABSTRACT: Although infrequently associated with illness in humans, Salmonella enterica, subsp. enterica serovar Kentucky is the most common non-clinical, non-human serovar reported in the United States, being largely found in poultry and poultry products, as well as being associated with cattle. This serovar is polyphyletic and can be separated into two groups, Group I and II, based on CRISPR-typing analysis. In Salmonella Kentucky isolates from human clinical samples in Pennsylvania, both lineages are equally represented. The goal of this study was to determine whether both groups were also represented in domestic food animals in Pennsylvania. We analysed the CRISPR arrays from 67 Salmonella Kentucky isolates used PCR and sequencing of CRISPR arrays or analysis of whole genome sequences to analyse the CRISPR arrays and Across a collection of 67 Salmonella Kentucky isolates that includes those collected from farms, veterinary clinical samples as well as isolates from retail meats, we show that Group I Salmonella Kentucky are the exclusive lineage present. We reveal that the specific subtype of over a quarter of these animal isolates are also found to be responsible for causing human salmonellosis in the same region over the same time period. ISSN: 18631959

Lamas, A., Paz-Mendez, A.M., Regal, P., Vazquez, B., Miranda, J.M., Cepeda, A., Franco, C.M.
Food preservatives influence biofilm formation, gene expression and small RNAs in Salmonella enterica
(2018) LWT, 97, pp. 1-8.
ABSTRACT: Salmonella enterica is major foodborne pathogens. Salmonella persists in the food chain due to its ability to produce biofilms under different conditions. One of the biggest challenges in biofilm research is reproducing real food industry conditions. Also, food microbiologists have the challenge of elucidating the role of small RNAs (sRNAs) in the survival of foodborne pathogens in the food chain. This study evaluated food preservatives (sodium nitrite, sodium sulfite and sodium acetate-citric acid) effect on biofilm formation of ten S. enterica strains on two surfaces (polystyrene and stainless steel). The effects of preservatives on transcription of biofilm- and virulence-related genes and sRNAs were evaluated. All Salmonella strains produced biofilm in all the conditions evaluated. However, sodium sulfite reduced biofilm formation by Salmonella in both surfaces tested. Food preservatives influenced biofilm- and virulence-related genes and sRNAs transcription. This study highlights that Salmonella strains can produce biofilms in the presence of food preservatives, representing a public health problem. Fully understanding what metabolic pathways are modified by the presence of preservatives could allow developing new control strategies to prevent foodborne pathogens persistence in the food chain by using effective combinations of preservatives. ISSN: 00236438

Bergamo, G., Timm, C.D., Carvalho, N.R., Helbig, E., Gandra, E.A.
Comparison between the 3M MDS® method and phenotypic methods to detect Salmonella spp. in foods
(2018) LWT, 97, pp. 693-696.
ABSTRACT: The objective of this study was to compare three methods for detection of Salmonella spp., the first recommended by the Brazilian Ministry of Agriculture, Livestock and Supply - known as MAPA -, the second described by Annex D of ISO 6579 - known as MSRV and the third method based on molecular techniques known as 3M MDS®. The assays were performed using samples of artificially contaminated UHT milk (101–103 CFU - Test), and mixed-meat and pork sausages commercialized in southern Brazil. MAPA, MSRV and 3M MDS® methods present a sensitivity of 100%, 100% and 97.6%, respectively. The specificity was 100% for the three methods. The data shows that 3M MDS® and MSRV are reliable methods because they revealed equivalent sensitivity and specificity to the Brazilian official method (MAPA) and that possibly can be used as a viable alternative for the detection of Salmonella spp. ISSN: 00236438

DE Sales, C.V., DE Melo, A.N.F., Niedzwiedzka, K.M., DE Souza, E.L., Schaffner, D.W., Magnani, M.
Changes of Antibiotic Resistance Phenotype in Outbreak-Linked Salmonella enterica Strains after Exposure to Human Simulated Gastrointestinal Conditions in Chicken Meat
(2018) Journal of food protection, 81 (11), pp. 1844-1850.
ABSTRACT: Fifteen outbreak-linked Salmonella enterica strains in chicken meat were evaluated under simulated human gastrointestinal conditions for their resistance and susceptibility to 11 antibiotics from seven antibiotic classes. The MIC of each antibiotic was determined by microdilution in broth before and after the exposure of each strain to a continuous system simulating the conditions in the human mouth, esophagus-stomach, duodenum, and ileum. Strains were inoculated onto chicken breast (9 g; inoculated at 5 log CFU/g) prior to exposure. Data were interpreted according Clinical and Laboratory Standards Institute breakpoints. After the in vitro digestion, 12 Salmonella strains with reduced susceptibility to ciprofloxacin (CIP) changed to CIP resistant. The ceftriaxone (CTX)-intermediate Salmonella Newport strain changed to CTX resistant. The ampicillin (AMP)-susceptible Salmonella Heidelberg strain changed to AMP resistant, and the sulfamethoxazole-trimethoprim (SXT)-susceptible strains of Salmonella serovars Typhimurium, Agona, Newport, Albany, and Corvallis changed to SXT resistant. The Salmonella Heidelberg, Salmonella Newport, Salmonella Albany, and Salmonella Corvallis strains had the highest frequency of changes in antibiotic susceptibility with new resistant phenotypes to AMP and CIP, CTX and SXT, CIP and SXT, and CIP and SXT, respectively. Conditions imposed by a simulated gastrointestinal environment changed the susceptibility of S. enterica strains to clinically relevant antibiotics and should be considered in the selection of therapies for human salmonellosis. ISSN: 19449097

Ågren, E.C.C., Lewerin, S.S., Frössling, J.
Evaluation of herd-level sampling strategies for control of Salmonella in Swedish cattle
(2018) Journal of Dairy Science, 101 (11), pp. 10177-10190.
ABSTRACT: Based on Swedish legislation, all herds where Salmonella of any serotype is detected are put under restrictions, and measures aiming at eradication are required. Costs for sampling and control have increased in recent years and the aim of this study was to investigate the efficiency of different sampling strategies. We also compiled test results from recent surveillance activities and used these to complement and compare with calculated results. Sensitivities and specificities at group and herd level were calculated for different test strategies. A scenario-tree modeling approach was used to account for the hierarchy of animals within herds, and different relative risk of salmonella in different age groups. Negative and positive predictive values (NPV and PPV), and probability of freedom from Salmonella were calculated to compare the added value of different sampling strategies. Results showed that more fecal samples than serological samples per group are needed to reach a group sensitivity >0.50. This also means that serological testing leads to a higher NPV. For example, with 10 negative test-results from a group of 25 animals in a herd with a suspicion of Salmonella, the NPV based on serology was 0.75 and based on culture was 0.56. For the PPV, testing based on culture from fecal sampling was superior, as specificity of such testing was close to perfect. By changing the threshold for considering a group positive, from 1 test-positive animal to 2, the PPV of serological results could be increased without substantial loss in NPV. The herd sensitivity based on (1) bulk milk sampling, (2) fecal sampling of all animals, and (3) bulk milk sampling and individual sera from 20 animals within each age group was 0.53, 0.88, and 0.95, respectively. In low-prevalence regions, this sensitivity was enough to verify a high probability of freedom (>0.99), as the probability of infection in such Swedish regions has been shown to be

0.01. For herds with a higher prior probability of infection, repeated sampling (2–9 sampling occasions) was needed to reach the same level of confidence. Analysis of surveillance data indicated that boot swabs can be used to replace the standard fecal sampling presently used in Sweden. It was also confirmed that the individual specificity of the tests used for serological testing of Swedish calves is high (0.99). The results can form a basis for fit-for-purpose testing strategies (e.g., surveillance or prepurchase testing). ISSN: 00220302

Calhoun, S., Post, L., Warren, B., Thompson, S., Bontempo, A.R.
Prevalence and Concentration of Salmonella on Raw, Shelled Peanuts in the United States (2018) Journal of food protection, 81 (11), pp. 1755-1760.
ABSTRACT: Recalls and outbreaks associated with Salmonella contamination in peanut-containing products have been reported over the past several years. Very limited data existed on the prevalence and concentration of Salmonella on raw, shelled peanuts in the United States. An initial study was completed in 2012 to estimate the prevalence and concentration of Salmonella on Runner- and Virginia-type raw, shelled peanuts in the United States from the 2008 through 2011 crop years, which were proportionately sampled from each growing region based on 2007 production volume. That study was extended to include samples of Runner- and Virginia-type peanuts from 2013, 2014, and 2015 crop years proportionately sampled from each growing region on the basis of the 2008 through 2010 volumes. Of the total 2,506 raw, shelled peanut samples, 41 (1.63%) were positive for Salmonella by the VIDAS SLM assay. Salmonella serovars identified in this study included Agona, Anatum, Bardo, Braenderup, Cannstatt, Dessau, Gaminara, Litchfield, Hartford, Inverness, Mbandaka, Meleagridis, Muenchen, Newport, Pakistan, Rodepoort, Rubislaw, Tennessee, and Tornow. The concentration levels of Salmonella in positive samples, as determined by most probable number (MPN), ranged from <0.003 to 2.4 MPN/g. These data will be useful when designing and validating processes for the reduction or elimination of Salmonella in peanuts or peanut-containing products or both.
ISSN: 19449097

Thompson, C.P., Doak, A.N., Amirani, N., Schroeder, E.A., Wright, J., Kariyawasam, S., Lamendella, R., Shariat, N.W.
High-resolution identification of multiple Salmonella serovars in a single sample by using CRISPRSeroSeq
(2018) Applied and Environmental Microbiology, 84 (21), art. no. e01859-18, .
ABSTRACT: Salmonella enterica is represented by > 2,600 serovars that can differ in routes of transmission, host colonization, and in resistance to antimicrobials. S. enterica is the leading bacterial cause of foodborne illness in the United States, with well-established detection methodology. Current surveillance protocols rely on the characterization of a few colonies to represent an entire sample; thus, minority serovars remain undetected. Salmonella contains two CRISPR loci, CRISPR1 and CRISPR2, and the spacer contents of these can be considered serovar specific. We exploited this property to develop an amplicon-based and multiplexed sequencing approach, CRISPR-SeroSeq (serotyping by sequencing of the CRISPR loci), to identify multiple serovars present in a single sample. Using mixed genomic DNA from two Salmonella serovars, we were able to confidently detect a serovar that constituted 0.01% of the sample. Poultry is a major reservoir of Salmonella spp., including serovars that are frequently associated with human illness, as well as those that are not. Numerous studies have examined the prevalence and diversity of Salmonella spp. in poultry, though these studies were limited to culture-based approaches and therefore only identified abundant serovars. CRISPR-SeroSeq was used to investigate samples from broiler houses and a processing facility. Ninety-one percent of samples harbored multiple serovars, and there was one sample in which four different serovars were detected. In another sample, reads for the minority serovar comprised 0.003% of the total number of Salmonella spacer reads. The most abundant serovars identified were Salmonella enterica serovars Montevideo, Kentucky, Enteritidis, and Typhimurium. CRISPR-SeroSeq also differentiated between multiple strains of some serovars. This high resolution of serovar populations has the potential to be utilized as a powerful tool in the surveillance of Salmonella species. ISSN: 00992240

Hu, J., Huang, R., Wang, Y., Wei, X., Wang, Z., Geng, Y., Jing, J., Gao, H., Sun, X., Dong, C., Jiang, C.
Development of duplex PCR-ELISA for simultaneous detection of Salmonella spp. and Escherichia coli O157: H7 in food
(2018) Journal of Microbiological Methods, 154, pp. 127-133.
ABSTRACT: In the current study, a duplex PCR-ELISA method was developed targeting the specific genes, invA of Salmonella spp. and rfbE of Escherichia coli O157: H7, to detect one

or both bacteria in food. In brief, PCR product amplified by PCR primer labeled with digoxin at the 5′-end and a probe labeled with biotin at the 3′-end can form dimer by nucleic acid hybridization which can be captured by binding of biotin to streptomycin coated in ELISA plate before using enzyme-labeled anti-digoxin antibody and substrate to develop color. Also, evaluation of the duplex PCR-ELISA method was conducted in different food samples including milk, juice, cabbage, shrimp, chicken, pork and beef. Results indicated that the duplex PCR-ELISA developed here was specific when using 25 non-target bacteria strains as controls and was sensitive with a limit of detection (LOD) of 1 CFU/mL, 1, 000 times higher than that of the duplex PCR method and was repeatable regardless of inter- and intra-batch variations. The duplex PCR-ELISA method established in the present study has proven to be highly specific, sensitive and repeatable. It has the potential to be applied in such fields as clinical diagnosis of food-borne diseases, food hygiene monitoring and pathogen detection in food. ISSN: 01677012

Kingsley, R.A., Langridge, G., Smith, S.E., Makendi, C., Fookes, M., Wileman, T.M., El Ghany, M.A., Keith Turner, A., Dyson, Z.A., Sridhar, S., Pickard, D., Kay, S., Feasey, N., Wong, V., Barquist, L., Dougan, G.
Functional analysis of Salmonella Typhi adaptation to survival in water
(2018) Environmental Microbiology, 20 (11), pp. 4079-4090.
ABSTRACT: Contaminated water is a major risk factor associated with the transmission of Salmonella enterica serovar Typhi (S. Typhi), the aetiological agent of human typhoid. However, little is known about how this pathogen adapts to living in the aqueous environment. We used transcriptome analysis (RNA-seq) and transposon mutagenesis (TraDIS) to characterize these adaptive changes and identify multiple genes that contribute to survival. Over half of the genes in the S. Typhi genome altered expression level within the first 24 h following transfer from broth culture to water, although relatively few did so in the first 30 min. Genes linked to central metabolism, stress associated with arrested proton motive force and respiratory chain factors changed expression levels. Additionally, motility and chemotaxis genes increased expression, consistent with a scavenging lifestyle. The viaB-associated gene tviC encoding a glcNAc epimerase that is required for Vi polysaccharide biosynthesis was, along with several other genes, shown to contribute to survival in water. Thus, we define regulatory adaptation operating in S. Typhi that facilitates survival in water. ISSN: 14622912

Xu, Y., Hu, Y., Guo, Y., Zhou, Z., Xiong, D., Meng, C., Li, Q., Geng, S., Pan, Z., Jiao, X.
A new PCR assay based on the new gene-SPUL_2693 for rapid detection of Salmonella enterica subsp. enterica serovar Gallinarum biovars Gallinarum and Pullorum
(2018) Poultry science, 97 (11), pp. 4000-4007.
ABSTRACT: Salmonella enterica subsp. enterica serovar Gallinarum biovar Gallinarum (S. Gallinarum) and biovar Pullorum (S. Pullorum) are gram-negative bacteria, members of the most important infectious pathogens, and have caused common problems in the poultry industry, especially in the developing countries. O- and H-antigen specific anti-sera are commonly for slide and tube agglutination tests to identify Salmonella serovars. However, it is both labor intensive and time consuming, so there is an urgent need for a new technique for the rapid detection of the major Salmonella serovars. In this study, we developed a 1-step PCR assay to identify the serovar Gallinarum. This PCR-based assay was based on the SPUL_2693 gene, which was located in SPI-19 and found by comparing the genomes of the S. Pullorum and S. Gallinarum in the whole data of NCBI. The specificity of this gene was evaluated by bioinformatics analysis, and the results showed that the SPUL_2693 gene exists in all serovar Gallinarum. The specificity and sensitivity of this PCR assay were evaluated in our study. The developed PCR assay was able to distinguish the serovar Gallinarum from 27 different Salmonella serovars and 5 different non-Salmonella pathogens. The minimum limit of genomic DNA of S. Pullorum for PCR detection was 2.143 pg/μL, and the minimum limit number of cells was 6 CFU. This PCR assay was also applied to analyze Salmonella strains isolated from a chicken farm in this study. The PCR assay properly identified the serovar Gallinarum from other Salmonella serovars, and the results were in agreement with the results of a traditional serotyping assay. In general, the newly developed PCR-based assay can be used to accurately judge the presence of the serovar Gallinarum and can be combined with traditional serotyping assays, especially in the case of large quantities of samples. ISSN: 15253171

Hölzel, C.S., Tetens, J.L., Schwaiger, K.
Unraveling the role of vegetables in spreading antimicrobial-resistant bacteria: A need for quantitative risk assessment
(2018) Foodborne Pathogens and Disease, 15 (11), pp. 671-688.

ABSTRACT: In recent years, vegetables gain consumer attraction due to their reputation of being healthy in combination with low energy density. However, since fresh produce is often eaten raw, it may also be a source for foodborne illness. The presence of antibiotic-resistant bacteria might pose a particular risk to the consumer. Therefore, this review aims to present the current state of knowledge concerning the exposure of humans to antibiotic-resistant bacteria via food of plant origin for quantitative risk assessment purposes. The review provides a critical overview of available information on hazard identification and characterization, exposure assessment, and risk prevention with special respect to potential sources of contamination and infection chains. Several comprehensive studies are accessible regarding major antimicrobial-resistant foodborne pathogens (e.g., Salmonella spp., Listeria spp., Bacillus cereus, Campylobacter spp., Escherichia coli) and other bacteria (e.g., further Enterobacteriaceae, Pseudomonas spp., Gram-positive cocci). These studies revealed vegetables to be a potential - although rare - vector for extended-spectrum beta-lactamase-producing Enterobacteriaceae, mcr1-positive E. coli, colistin- and carbapenem-resistant Pseudomonas aeruginosa, linezolid-resistant enterococci and staphylococci, and vancomycin-resistant enterococci. Even if this provides first clues for assessing the risk related to vegetable-borne antimicrobial-resistant bacteria, the literature research reveals important knowledge gaps affecting almost every part of risk assessment and management. Especially, the need for (comparable) quantitative data as well as data on possible contamination sources other than irrigation water, organic fertilizer, and soil becomes obvious. Most crucially, dose-response studies would be needed to convert a theoretical "risk" (e.g., related to antimicrobial-resistant commensals and opportunistic pathogens) into a quantitative risk estimate. ISSN: 15353141

Trmcic, A., Chen, H., Trząskowska, M., Tamber, S., Wang, S.
Biofilm-Forming Capacity of Five Salmonella Strains and Their Fate on Postharvest Mini Cucumbers
(2018) Journal of food protection, 81 (11), pp. 1871-1879.
ABSTRACT: Salmonella enterica is one of the pathogens that is frequently identified as the cause of fresh produce-related outbreaks. Biofilm formation is a factor that can contribute to pathogen survival on produce surface. The goal of our current research was to investigate the survival of five S. enterica strains representing different serotypes (i.e., Typhimurium, Enteritidis, Daytona, Poona, and Newport) on whole mini cucumbers stored at refrigeration (4°C) and room temperature (22°C). We also determined the strains survival on glass slides and in phosphate-buffered saline at 4 and 22°C, as well as the ability to form biofilms on a solid-liquid interphase. A rapid decrease in cell density (>4-log reduction over 8 days) of all five tested strains was observed on glass slides, while a slower die-off (<1-log reduction in 8 days) was observed in PBS. No significant difference in the die-off rate was observed among the five strains at 4 or 22°C. The die-off rate on the surface of mini cucumbers at 4°C was significantly slower ($P < 0.02$) for Salmonella Enteritidis LMFS-S-JF-005 compared with the remaining four strains. At 22°C, Salmonella Poona S306 was able to grow by more than 1.5 log units on whole mini cucumbers over a period of 8 days, while the cell density of the other four strains remained at the same level compared with day 0. At this temperature, Salmonella Poona S306 was also able to form significantly stronger biofilms on a solid-liquid interphase ($P < 0.01$) and was the only strain that presented a red, dry, and rough morphotype on Congo red agar plates, indicating the formation of both curli fimbriae and cellulose. These results revealed that the fate of Salmonella on mini cucumbers is strain specific, which highlighted the need for tailored mitigation strategies, such as the effective control of temperature and moisture for limiting the survival or growth of high-risk Salmonella strains between harvest and consumption of fresh produce. ISSN: 19449097

Rodríguez, F.I., Procura, F., Bueno, D.J.
Comparison of 7 culture methods for Salmonella serovar Enteritidis and Salmonella serovar Typhimurium isolation in poultry feces
(2018) Poultry science, 97 (11), pp. 3826-3836.
ABSTRACT: The present work compared 7 different culture methods and 3 selective-differential plating media for Salmonella ser. Enteritidis (SE) and S. ser. Typhimurium (ST) isolation using artificially contaminated poultry feces. The sensitivity (Se) and accuracy (AC) values increased when Modified Semisolid Rappaport Vassiliadis (MSRV) methods were used in place of the Tetrathionate (TT) or Tetrathionate Hajna broth (TTH) method in the enrichment step. However, there was no significant difference between the pre-enrichment incubation at 4 to 6 and 18 to 24 h for MSRV5 and MSRV24 methods, respectively. All Salmonella strains were recovered in the lowest dilutions tested for MSRV24 and 3 out of 4 for MSRV5 methods (2 to 10 cfu/25 g). The TT and TTH methods showed a detection limit between 2.2 × 101 and 1.0 × 106 cfu/25 g of fecal sample. The

agreement was variable between the methods. However, there was a very good agreement between the MSRV5 and MSRV24 methods, and between tetrathionate direct (TTD, no pre-enrichment media used) and buffered peptone water 18 to 24 h Tetrathionate broth combination (TT24 method) for Salmonella strains. The 3 selective-differential plating media showed an agreement between fair and excellent. They performed a high Se and AC in the MSRV methods for Salmonella strains. There was a significant difference between center and periphery for MSRV methods, and there was a fair agreement between them for all strains. The MSRV methods are better than TT/TTH methods for the isolation of different strains of SE and ST in poultry fecal samples. The MSRV5 method can be used to reduce the time for the detection of SE and ST in these samples. Furthermore, a loopful of the periphery of the growth should be streaked onto differential-selective plating media, even in the absence of halo, to decrease the number of false negative results. ISSN: 15253171

Dlamini, B.S., Montso, P.K., Kumar, A., Ateba, C.N.
Distribution of virulence factors, determinants of antibiotic resistance and molecular fingerprinting of Salmonella species isolated from cattle and beef samples: suggestive evidence of animal-to-meat contamination
(2018) Environmental Science and Pollution Research, 25 (32), pp. 32694-32708.
ABSTRACT: In this study, three hundred presumptive Salmonella strains isolated from cattle faeces and raw beef samples were subjected to both preliminary and confirmatory tests specific for Salmonella. PCR assays revealed that 100%, 20% and 26.7% of the isolates were positive for 16S rRNA, fliC and fljB gene fragments, respectively. Large proportions (62.4 to 94.3%) of these isolates were multiple antibiotic resistant (MAR) strains that were resistant to three or more antibiotics belonging to different classes. MAR phenotypes Ab1, Ab2, Ab3, Ab7, Ab8, Ab9, Ab26 and Ab27 were dominant among the isolates. Cluster analysis of antibiotic inhibition zone diameter data revealed two major clusters (clusters 1 and 2), and each cluster contained two sub-clusters (1A, 1B, 2A and 2B). PCR data revealed that 27.1% and 30.7% of the isolates possessed the spvC and invA virulent genes, respectively. There was a significant correlation between the possession of MAR phenotypes and virulent gene determinants. Analysis of restriction fragment length polymorphism (RFLP) of 16S rRNA gene fragments using EcoRI and HaeIII showed that large proportions of isolates from beef and cattle faeces produced similar genetic fingerprints. From these results, it is suggested that Salmonella species in cattle are transmitted to beef and, therefore, the consumption of undercooked beef could pose severe health complications on consumers. These findings provide baseline data that could be of great epidemiological importance and, thus, the need to utilise more sensitive typing tools in determining the genetic relatedness of isolates from different sources.
ISSN: 09441344

Williams, M.S., Ebel, E.D., Hretz, S.A., Golden, N.J.
Adoption of Neutralizing Buffered Peptone Water Coincides with Changes in Apparent Prevalence of Salmonella and Campylobacter of Broiler Rinse Samples
(2018) Journal of food protection, 81 (11), pp. 1851-1863.
ABSTRACT: Buffered peptone water is the rinsate commonly used for chicken rinse sampling. A new formulation of buffered peptone water was developed to address concerns about the transfer of antimicrobials, used during poultry slaughter and processing, into the rinsate. This new formulation contains additives to neutralize the antimicrobials, and this neutralizing buffered peptone water replaced the original formulation for all chicken carcass and chicken part sampling programs run by the Food Safety and Inspection Service beginning in July 2016. Our goal was to determine whether the change in rinsate resulted in significant differences in the observed proportion of positive chicken rinse samples for both Salmonella and Campylobacter. This assessment compared sampling results for the 12-month periods before and after implementation. The proportion of carcass samples that tested positive for Salmonella increased from approximately 0.02 to almost 0.06. Concurrently, the proportion of chicken part samples that tested for Campylobacter decreased from 0.15 to 0.04. There were no significant differences associated with neutralizing buffered peptone water for the other two product-pathogen pairs. Further analysis of the effect of the new rinsate on corporations that operate multiple establishments demonstrated that changes in the percent positive rates differed across the corporations, with some corporations being unaffected, while others saw all of the establishments operated by the corporation move from passing to failing the performance standard and vice versa. The results validated earlier concerns that antimicrobial contamination of rinse samples was causing false-negative Salmonella testing results for chicken carcasses. The results also indicate that additional development work

may still be required before the rinsate is sufficiently robust for its use in Campylobacter testing. ISSN: 19449097

Wang, Y., Chen, X., Hu, Y., Zhu, G., White, A.P., Köster, W.
Evolution and sequence diversity of FhuA in Salmonella and Escherichia
(2018) Infection and Immunity, 86 (11), art. no. e00573-18, .
ABSTRACT: The fhuACDB operon, present in a number of Enterobacteriaceae, encodes components essential for the uptake of ferric hydroxamate type siderophores. FhuA acts not only as a transporter for physiologically important chelated ferric iron but also as a receptor for various bacteriophages, toxins, and antibiotics, which are pathogenic to bacterial cells. In this research, fhuA gene distribution and sequence diversity were investigated in Enterobacteriaceae, especially Salmonella and Escherichia. Comparative sequence analysis resulted in a fhuA phylogenetic tree that did not match the expected phylogeny of species or trees of the fhuCDB genes. The fhuA sequences showed a unique mosaic clustering pattern. On the other hand, the gene sequences showed high conservation for strains from the same serovar or serotype. In total, six clusters were identified from FhuA proteins in Salmonella and Escherichia, among which typical peptide fragment variations could be defined. Six fragmental insertions/deletions and two substitution fragments were discovered, for which the combination of polymorphism patterns could well classify the different clusters. Structural modeling demonstrated that all the six featured insertions/deletions and one substitution fragment are located at the apexes of the long loops present as part of the FhuA external pocket. These frequently mutated regions are likely under high selection pressure, with bacterial strains balancing escape from phage infection or toxin/antibiotics attack via fhuA gene mutations while maintaining the siderophore uptake activity essential for bacterial survival. The unusual fhuA clustering suggests that high-frequency exchange of fhuA genes has occurred between enterobacterial strains after distinctive species were established. ISSN: 00199567

Gurtler, J.B., Doyle, M.P., Erickson, M.C., Jiang, X., Millner, P., Sharma, M.
Composting To Inactivate Foodborne Pathogens for Crop Soil Application: A Review
(2018) Journal of food protection, 81 (11), pp. 1821-1837.
ABSTRACT: Compost is organic material that has been degraded into a nutrient-stabilized humus-like substance through intense microbial activity, which can provide essential plant nutrients (nitrogen, phosphorus) to aid in the growth of fruits and vegetables. Compost can be generated from animal waste feedstocks; these can contain human pathogens, which can be inactivated through the heat and microbial competition promoted during the composting process. Outbreaks of infections caused by bacterial pathogens such as Escherichia coli O157:H7, Salmonella, and Listeria monocytogenes on fruit and vegetable commodities consumed raw emphasize the importance of minimizing the risk of pathogenic contamination on produce commodities. This review article investigates factors that affect the reduction and survival of bacterial foodborne pathogens during the composting process. Interactions with indigenous microorganisms, carbon:nitrogen ratios, and temperature changes influence pathogen survival, growth, and persistence in finished compost. Understanding the mechanisms of pathogen survival during the composting process and mechanisms that reduce pathogen populations can minimize the risk of pathogen contamination in the cultivation of fruits and vegetables. ISSN: 19449097

Antony, L., Behr, M., Sockett, D., Miskimins, D., Aulik, N., Christopher-Hennings, J., Nelson, E., Allard, M.W., Scaria, J.
Genome divergence and increased virulence of outbreak associated Salmonella enterica subspecies enterica serovar Heidelberg
(2018) Gut Pathogens, 10 (1), art. no. 279, .
ABSTRACT: Salmonella enterica serotype Heidelberg is primarily a poultry adapted serotype of Salmonella that can also colonize other hosts and cause human disease. In this study, we compared the genomes of outbreak associated non-outbreak causing Salmonella ser. Heidelberg strains from diverse hosts and geographical regions. Human outbreak associated strains in this study were from a 2015 multistate outbreak of Salmonella ser. Heidelberg involving 15 states in the United States which originated from bull calves. Our clinicopathologic examination revealed that cases involving Salmonella ser. Heidelberg strains were predominantly young, less than weeks-old, dairy calves. Pre-existing or concurrent disease was found in the majority of the calves. Detection of Salmonella ser. Heidelberg correlated with markedly increased death losses clinically comparable to those seen in herds infected with S. Dublin, a known serious pathogen of cattle. Whole genome based single nucleotide polymorphism based analysis revealed that these calf isolates formed a distinct cluster along with outbreak associated human isolates. The defining feature of the outbreak associated strains, when compared to older isolates of S.

Heidelberg, is that all isolates in this cluster contained Saf fimbrial genes which are generally absent in S. Heidelberg. The acquisition of several single nucleotide polymorphisms and the gain of Saf fimbrial genes may have contributed to the increased disease severity of these Salmonella ser. Heidelberg strains. ISSN: 17574749

Methner, U., Moog, U.
Occurrence and characterisation of Salmonella enterica subspecies diarizonae serovar 61: K: 1, 5, (7) in sheep in the federal state of Thuringia, Germany
(2018) BMC Veterinary Research, 14 (1), art. no. 401, .
ABSTRACT: Background: The occurrence of Salmonella enterica subspecies diarizonae serovar 61: k: 1, 5, (7) (SASd) and other Salmonella organisms in sheep in the German federal state of Thuringia was examined for the first time. Pooled faecal samples from 90 flocks located in this state were monitored. Results: Only SASd was detected in 74 (82.2%) out of the 90 sheep herds, other Salmonella serovars were not identified. A positive correlation was found between the flock size and the detection probability of SASd. Despite the agent's high prevalence, clinical symptoms of a disease exclusively due to SASd have not been observed. The SASd strains were characterised by macrorestriction analysis, antimicrobial testing and the biochemical profile. All strains were sensitive to 13 out of 14 antimicrobial substances and resistant to only sulfamethoxazole. The high number of macrorestriction groups of SASd strains indicated a low clonality of the serovar. Conclusions: Data from sheep derived foods and public health data in Germany strongly suggest that the significance of SASd for public health is considerably lower than that of serovars belonging to Salmonella enterica subspecies enterica. For this reason and because of the low disease-causing potential of SASd in sheep, it is worthwile to consider a reduction in ongoing activities from combating to monitoring serovar 61: k: 1, 5, (7) in the sheep population. ISSN: 17466148

Ibrahim, G.M., Morin, P.M.
Salmonella serotyping using whole genome sequencing
(2018) Frontiers in Microbiology, 9 (DEC), art. no. 2993, .
ABSTRACT: Until recently, traditional serology and the Kauffmann White Scheme (KWS) have been the gold standard for Salmonella serotyping. Whole Genome Sequencing (WGS) has now emerged as an alternative in this field. Serotype information remains a cornerstone in food safety and public health activities to reduce the burden of salmonellosis. At the same time, recent advances in WGS have improved the ability to perform advanced pathogen characterization while improving trace back investigations to determine the source of foodborne illness during outbreaks. Serovar prediction based on WGS can be performed using in silico data analysis tools. Three such tools have been developed: (a). Salmonella in silico Typing Resource (SISTR), (b). SeqSero, and (c). In silico 7-gene MLST ST (Multilocus Sequence Typing Sub-Typing) which was generated using the SISTR platform. Public health officials around the world are diligently working to validate these tools for replacing traditional surveillance methods to provide a more powerful approach for molecular epidemiology in support of public health investigations. In this study, we report a retrospective analysis of our laboratory inventory of 1,041 Salmonella isolates collected between 1999 and 2017. These isolates are of public health significance since they all came from either food, feed or environmental swabs. They were all serotyped by both traditional serology and WGS using an in silico SeqSero tool for serovar prediction. Both predicted identical Salmonella serotypes in 899 isolates (86.4% of the 1,041 Salmonella isolates). SeqSero assignments differed from traditional serological testing in 80 isolates (7.7%) and no serotype prediction was ascertained from 62 isolates (5.9%). This retrospective study is an excellent example of using WGS and SeqSero as a data analysis tool to predict Salmonella serotypes that can provide numerous advantages including molecular and genetic details regarding the characteristics of the Salmonella isolates compared to traditional KWS serotyping. In conclusion, it is evident that using WGS and in silico tools for Salmonella serotyping might someday replace traditional serotyping. ISSN: 1664302X

Nurjayadi, M., Islami, N., Pertiwi, Y.P., Saamia, V., Wirana, I.M.
Evaluation of primer detection capabilities of fimC Salmonella typhi using real time PCR for rapid detection of bacteria causes of food poisoning
(2018) IOP Conference Series: Materials Science and Engineering, 434 (1), art. no. 012097, .
ABSTRACT: Food poisoning is a disease caused by bacterial, viral or parasitic infections, which contaminate food. The purpose of this study was to obtain information about the detection of the primer pair of fimC Salmonella typhi genes using the Real-time PCR methods for the rapid kit development. The evaluation of the primer ability is determined

by the accumulation of fluorescence signal from the amplification curve connecting the number of Cycle threshold (Ct) to the intensity of the amplicon signal that can reach the threshold line. The results showed that the primer fimC gene successfully amplifies the S. typhi DNA target fragment on Ct 14.783. In addition, the Ct data from this study also informed that the primer sensitivity of the fimC gene against the target S. typhi bacteria gave a minimum detection rate of 4,528 pg/µL, and the primer specificity evaluation of that primer to non-targeted bacteria Shigella dysenteriae gave Ct 27,949. Based on the data it can be concluded that fimC S. typhi primers gene can be used as sensitive and fast detection devices, but still require improvement in the specificity. ISSN: 17578981

Adhikari, A., Yemmireddy, V.K., Costello, M.J., Gray, P.M., Salvadalena, R., Rasco, B., Killinger, K.
Effect of storage time and temperature on the viability of E. coli O157:H7, Salmonella spp., Listeria innocua, Staphylococcus aureus, and Clostridium sporogenes vegetative cells and spores in vacuum-packed canned pasteurized milk cheese
(2018) International Journal of Food Microbiology, 286, pp. 148-154.
ABSTRACT: The effect of storage temperature and time on the viability of several foodborne bacterial pathogens inoculated into vacuum-packed canned pasteurized cow's milk cheese was investigated. Three popular cheese styles namely, a semi-soft white Monterey Jack style cheese, and two Cheddar cheeses vacuum packaged in mason jars were inoculated with a 3-strain cocktail of each of the following microbes at the mean concentrations listed: Escherichia coli O157:H7 (6.6 log CFU/g), Salmonella spp. (6.3 log CFU/g), Listeria innocua (6.4 log CFU/g), Staphylococcus aureus (3.6 log CFU/g), and Clostridium sporogenes vegetative cells (6.3 log CFU/g), and spores (6.0 log CFU/g). The effect of storage temperature (at 4.4, 10, and 21.1 °C) and the time (from 0 to 365 days) on the survival of the inoculated organisms was evaluated at different sampling times (0, 30, 60, 120,180, and 365 days). Both storage temperature and the time had a significant effect on the viability of the test organisms. Increasing the storage temperature from 4.4 to 21.1 °C and the storage time for up to 365 days increased pathogen reduction. The type of cheese also had a significant effect on the viability of the test organisms. At the same sampling times, the viability of E. coli O157:H7 and Salmonella spp., were highest in Monterey Jack-style cheese followed by the Cheddar cheeses one to which annatto had been added (Cheddar 1) and the second, a white Cheddar that has an added adjunct flavor culture (Cheddar 2). Similarly, the type of cheese and the time-temperature conditions to which the cheese was exposed had a significant effect on the viability of L. innocua. Among the tested organisms, S. aureus was most susceptible while C. sporogenes (both vegetative cells and spores) were most resistant. The findings of this challenge study indicate that vacuum packed canned cheese is not a favorable environment for the growth of bacterial pathogens. Depending upon the type of canned cheese, appropriate storage times and temperatures are critical to ensure microbiological safety. ISSN: 01681605

De Lucia, A., Rabie, A., Smith, R.P., Davies, R., Ostanello, F., Ajayi, D., Petrovska, L., Martelli, F.
Role of wild birds and environmental contamination in the epidemiology of Salmonella infection in an outdoor pig farm
(2018) Veterinary Microbiology, 227, pp. 148-154.
ABSTRACT: Foodborne outbreaks caused by Salmonella are often attributed to the pork consumption. Salmonella contamination of retail pork is directly linked to the Salmonella prevalence on farm. In UK, approximately 40% of breeding pigs are kept outdoors. Aim of this study was to investigate the role of wild birds in the epidemiology of Salmonella in one outdoor pig farm. Three sampling visits were carried out at monthly intervals to an outdoor farm consisting of two fields, one left empty of pigs for more than 2 years (field A) while the second (field B) was occupied by pigs during the first visit only. Faeces from wild bird droppings, environmental samples and pig faeces were tested for Salmonella. Salmonella spp. was isolated from environmental samples also in field A that had not been occupied by pigs more than 2 years. Interestingly, the wild bird population accessing the fields increased considerably once the pigs had left the farm and the proportion of Salmonella positive wild bird droppings increased over time with 7.4%, 15.8% and 44.3% at the first, second and third visit, respectively. The levels of Salmonella identified in some of the wild bird droppings were unusually high (105–106 CFU/g) suggesting that Salmonella was actively replicating in the gastrointestinal tract of these birds. Monophasic Salmonella Typhimurium DT193 was the predominant serotype isolated in pigs as well as in wild bird droppings and the environment, suggesting that the pigs were the original source of infection, as this serovar is typically associated with pigs. ISSN: 03781135

Jiang, Y., Dennehy, C., Lawlor, P.G., Hu, Z., Yang, Q., McCarthy, G., Tan, S.P., Zhan, X., Gardiner, G.E.
Inactivation of Salmonella during dry co-digestion of food waste and pig manure (2018) Waste Management, 82, pp. 231-240.
ABSTRACT: Extremely high volatile fatty acids (VFAs) and ammonia concentrations can accumulate during dry co-digestion of organic wastes, which may inactivate pathogenic microorganisms. In this study, inactivation of Salmonella during dry co-digestion of pig manure (PM) and food waste (FW), which are both reservoirs of zoonotic pathogens, was examined. The effects of pH, VFAs, ammonia and their interactions were assessed on three inoculated Salmonella serotypes. The results show that dry co-digestion significantly decreased the Salmonella inactivation time from several months (in wet digestion) to as short as 6–7 days. A modified Weibull distribution was proposed to simulate Salmonella reduction and to calculate or predict the minimum inhibitory concentrations (MIC) of VFAs and ammonia. Statistical analysis showed that all the factors (pH, VFA type, VFA/ammonia concentration and Salmonella serotype) significantly impacted Salmonella inactivation ($P < 0.01$). The inhibitory effect sequence was pH > VFA concentration > VFA type > Salmonella serotype in VFA MIC tests, and ammonia concentration > pH > Salmonella serotype in ammonia MIC tests. The toxicity of VFAs was much greater than that of ammonia, and an antagonistic effect was found between VFAs and ammonia on Salmonella inactivation. Apart from the toxicity of free VFAs and free ammonia, the inhibitory effects of pH alone, ionized VFAs and ammonium were also observed. ISSN: 0956053X

Hu, L., Deng, X., Brown, E.W., Hammack, T.S., Ma, L.M., Zhang, G.
Evaluation of Roka Atlas Salmonella method for the detection of Salmonella in egg products in comparison with culture method, real-time PCR and isothermal amplification assays
(2018) Food Control, 94, pp. 123-131.
ABSTRACT: With the increasing focus on the food safety, rapid methods for the detection of Salmonella are crucial for both food industry and regulatory agencies. Recently, many molecular methodologies with diverse technologies have been introduced. Roka Atlas® Salmonella Assay (SEN) is a molecular method that uses ribosomal RNA as target for detection, which is theoretically more sensitive than PCR or isothermal amplification methods that target the DNA sequences of single genes. In this study, SEN assay was compared with four PCR- and isothermal amplification-based assays and a culture method, such as the MicroSEQ® Salmonella spp. Detection kit (MicroSEQ), 3M™ Molecular Detection Assay (MDA) Salmonella, ANSRTM Salmonella Assay (ANSR), and Pro-AmpRTTM SALM spp. Kit (Pro-AmpRT). Food samples were prepared and analyzed according to the current U. S. Food and Drug Administration (FDA) Bacteriological Analytical Manual (BAM) Salmonella culture method. A total of 155 bacterial isolates (121 for Salmonella inclusivity and 34 for Salmeonlla exclusivity) and 200 egg product samples inoculated at a level of 1–5 CFU/25 g were analyzed. The study also estimated the limit of detection of these molecular methods, and illustrated their advantages and disadvantages. For exclusivity, all 34 non-Salmonella isolates were negative by all 5 molecular methods studied. For inclusivity, all 121 Salmonella isolates were positive by MDA, ANSR, and Pro-AmpRT methods. However, the SEN and MicroSEQ results were negative for 9 samples inoculated with Salmonella bongori. The detection limit of the 5 molecular methods ranged from 1.76 to 3.76 log CFU/mL pre-enrichment culture, with the SEN assay being the most sensitive (1.76 – 2.64 log CFU/mL). The results indicated that the SEN assay was as effective and sensitive in detecting Salmonella enterica in egg products as was the FDA BAM culture method and the 4 other isothermal amplification and PCR methods evaluated in the study. ISSN: 09567135

Firestone, M.J., Hedberg, C.W.
Restaurant inspection letter grades and salmonella infections, New York, New York, USA (2018) Emerging Infectious Diseases, 24 (12), pp. 2164-2168.
ABSTRACT: Rates of Salmonella infection in the United States have not changed over the past 20 years. Restaurants are frequent settings for Salmonella outbreaks and sporadic infections. Few studies have examined the effect of posting letter grades for restaurant inspections on the incidence of foodborne illness. We compared Salmonella infection rates in New York, New York, USA (NYC), with those in the rest of New York state before and after implementation of a letter grade system for restaurant inspections in NYC. We calculated a segmented regression model for interrupted time series data. After implementation of letter grading, the rate of Salmonella infections decreased 5.3% per year in NYC versus the rest of New York state during 2011–2015, compared with the period before implementation, 2006–2010. Posting restaurant inspection results as letter

grades at the point of service was associated with a decline in Salmonella infections in NYC and warrants consideration for broader use. ISSN: 10806040

Toro, M., Rivera, D., Toledo, V., Campos-Vargas, R., Allard, M.W., Hamilton-West, C., Moreno-Switt, A.I.
Genomics of Salmonella contaminating backyard production systems reveals persistence and transmission of genetically related Salmonella on a farm basis
(2018) Zoonoses and Public Health, 65 (8), pp. 1008-1014.
ABSTRACT: Animals raised in backyard productive systems (BPS) have been frequently associated with Salmonella outbreaks. Several serovars have caused these events, showing that different BPSs can be contaminated by distinct Salmonella serovars. The aim of this study was to characterize the genomic diversity of Salmonella isolates obtained from BPSs in Central Chile to understand their genomic relatedness. A whole-genome SNP-based phylogenetic analysis of 22 Salmonella isolates from 12 locations revealed that S. Typhimurium isolates clustered based on the BPS that they were originally isolated from, and the same was established for S. Enteritidis isolates. Furthermore, our genomic analysis shows that animals from different species (i.e., a chicken, a duck and a pig) carried genetically related S. Typhimurium strains within the same BPS. Moreover, some of these genetically related isolates were obtained in different years (2013 and 2014), indicating that farm-specific Salmonella can persist in BPSs for multiple years and that interspecies transmission is plausible in this environment. Understanding the dynamics of interspecies transmission of Salmonella serovars within a contaminated BPS is fundamental to the design of mitigation strategies to reduce outbreaks of human Salmonella associated with backyard production systems. ISSN: 18631959

Arunrut, N., Kiatpathomchai, W., Ananchaipattana, C.
Development and evaluation of real-time loop mediated isothermal amplification assay for rapid and sensitive detection of Salmonella spp. in chicken meat products
(2018) Journal of Food Safety, 38 (6), art. no. e12564, .
ABSTRACT: Salmonella spp., one of the most prevalent bacterial causes of foodborne disease, is a major public health concern due to its common occurrence in chicken meat products. The detection of Salmonella spp. using conventional culturing methods requires an excessive amount of time and effort. Thus, in response to the need for a rapid, sensitive, and convenient method for Salmonella detection, a quantitative assay using real-time Loop-mediated isothermal amplification (real-time LAMP) was developed using gene62181533 as the target sequence amplified. The real-time LAMP assay does not show cross-reactivity with several other common bacterial pathogens and the detection limit for genomic DNA in pure culture was found to be 1.2 CFU/ml. Testing with spiked samples simultaneously detected 7 CFU/ml of Salmonella pathogen in the artificially inoculated samples after enrichment for 6 hr. In comparison with the standard culture-based methods in the analysis of 120 raw chicken meat samples, the results of the sensitivity, accuracy, and specificity tests of the real-time LAMP assay were 94.02, 90.83, and 86.79%, respectively. These results indicate that this method is not only sensitive and specific but can also be used for rapid detection and differentiation of foodborne disease-causing bacteria. Practical applications: Salmonella is an important bacterial genus which causes most of the common foodborne illnesses worldwide. The disease mainly caused by Salmonella spp. through consumption of contaminated eggs and poultry products is salmonellosis which can manifest as bacterial diarrhea through to septicemia. The result indicates the potential usefulness of real-time LAMP assay based on the gene62181533 for a rapid, specific, sensitive, and quantitative assay for Salmonella detection in food products. This assay offers a low cost quantitative method in the clinical diagnosis of Salmonella in resource-limited health-care facilities and clinical laboratories in developing countries and in field tests. ISSN: 01496085

Lopes, S.M., Fösch Batista, A.C., Tondo, E.C.
Salmonella survival during soft-cooked eggs processing by temperature-controlled water circulator
(2018) Food Control, 94, pp. 249-253.
ABSTRACT: Soft-cooked eggs have been cooked and served worldwide, however concerns frequently raise about the safety of these preparations, assuming the possibility of eggs be contaminated by Salmonella. Temperature-controlled water circulators at low temperature (62 °C–65 °C) for long periods (at least 1 h) has been used to thermally process eggs, aiming to modify its textures. However, time and temperature patterns are not in agreement with some recommendations for processing food preparations at least 70 °C. This study was undertaken to analyze the survival of Salmonella spp. during soft-cooked eggs processing by temperature-controlled water circulator. A pool of Salmonella spp. was

inoculated in egg yolks and were incubated at 37 °C, for 18 h, reaching 7.7 ± 0.1 log10 CFU/g. Contaminated eggs were processed at 62 °C for 60 min and samples were collected in order to investigate Salmonella survival. Results indicated that the egg center temperature reached 61.7 ± 0.4 °C after 30 min, completely inactivating 7.7 log of Salmonella spp. After 30 min of cooking, yolk remained liquid and the egg white slightly opaque, demonstrating that the Salmonella inactivation was not related with the solidification of egg white or yolk. The survival curve did not follow first order kinetic and Double Weibull model was used to estimate inactivation kinetic parameters. In summary, the results of this study can be used by food processors in order to validate soft-cooked eggs processing by temperature-controlled water circulator. ISSN: 09567135

Zhang, P., Zhuang, L., Zhang, D., Xu, J., Dou, X., Wang, C., Gong, J.
Serovar-Specific Polymerase Chain Reaction for Detection of Salmonella enterica Serovar Indiana
(2018) Foodborne Pathogens and Disease, 15 (12), pp. 776-781.
ABSTRACT: Salmonella enterica serovar Indiana (S. Indiana) is a newly emerging pathogen with high levels of drug resistance. It has become one of the most common Salmonella serovars in China with a worldwide distribution, posing significant public health concerns. Detection of S. Indiana by traditional bacteriological methods is time-consuming and laborious, which prevents timely surveillance and effective control of the pathogen. In this study, comparative genomics was used to identify an A7P63-13850 gene that is uniquely present in S. Indiana, but not in other Salmonella serovars or any non-Salmonella bacteria. Then, a polymerase chain reaction (PCR) assay targeting this serovar-specific gene was established for specific detection of S. Indiana. The detection limit of this method is 10 pg per reaction for bacterial genomic DNA, being equivalent to 100 colony-forming units (CFU) per reaction. The established PCR amplifies all S. Indiana strains (n = 56), but none of other Salmonella serovars (n = 146) and non-Salmonella species (n = 14). The assay established in this study was also used to detect clinical samples from poultry, showed a positivity of 14.7% (23/156) for S. Indiana, which were verified by bacteriological methods. The highly sensitive and serovar-specific PCR for S. Indiana established in this study is suitable and convenient for detection of S. Indiana which aids in surveillance and control of the pathogen. ISSN: 15353141

Elder, J.R., Paul, N.C., Burin, R., Guard, J., Shah, D.H.
Genomic organization and role of SPI-13 in nutritional fitness of Salmonella
(2018) International Journal of Medical Microbiology, 308 (8), pp. 1043-1052.
ABSTRACT: Salmonella pathogenicity island 13 (SPI-13) contributes to the virulence of Salmonella. The majority of the SPI-13 genes encode proteins putatively involved in bacterial metabolism, however, their functions largely remain uncharacterized. It is currently unknown if SPI-13 contributes to metabolic fitness of Salmonella and, if so, what are the metabolic substrates for the protein encoded by genes within SPI-13. We employed Phenotype Microarray (Biolog, USA) to compare the metabolic properties of SPI-13 deficient mutant (ΔSPI-13) and the WT parent strain of non-typhoidal Salmonella enterica sub sp. enterica serovar Enteritidis (S. Enteritidis). The results of Phenotype Microarray revealed that SPI-13 is required for efficient utilization of two micronutrients, namely, D-glucuronic acid (DGA) and tyramine (TYR), as sole sources of carbon and/or nitrogen. By systematic deletion of the individual gene(s), we identified specific genes within SPI-13 that are required for efficient utilization of DGA (SEN2977-80) and TYR (SEN2967 and SEN2971-72) as sole nutrient sources. The results show that SPI-13 mediated DGA and TYR metabolic pathways afford nutritional fitness to S. Enteritidis. Comparative genomics analysis of the SPI-13 locus from 247 Salmonella strains belonging to 57 different serovars revealed that SPI-13 genes specifically involved in the metabolism of DGA and TYR are highly conserved in Salmonella enterica. Because DGA and TYR are naturally present as metabolic byproducts in the gastrointestinal tract and other host tissues, we propose a metabolic model that shows that the role of SPI-13 mediated DGA and TYR metabolism in the nutritional fitness of Salmonella is likely linked to nutritional virulence of this pathogen. ISSN: 14384221

Tanner, J.R., Kingsley, R.A.
Evolution of Salmonella within Hosts
(2018) Trends in Microbiology, 26 (12), pp. 986-998.
ABSTRACT: Within-host evolution has resulted in thousands of variants of Salmonella that exhibit remarkable diversity in host range and disease outcome, from broad host range to exquisite host restriction, causing gastroenteritis to disseminated disease such as typhoid fever. Within-host evolution is a continuing process driven by genomic variation that occurs during each infection, potentiating adaptation to a new niche resulting from

changes in animal husbandry, the use of antimicrobials, and emergence of immune compromised populations. We discuss key advances in our understanding of the evolution of Salmonella within the host, inferred from (i) the process of host adaptation of Salmonella pathovars in the past, and (ii) direct observation of the generation of variation and selection of beneficial traits during single infections. ISSN: 0966842X

Hernández, M., Rodríguez-Lázaro, D., Valero, A., Cadavez, V., Gonzales-Barron, U.
Zero-inflated binomial regressions for modelling low prevalence of pathogens in chicken meat as affected by sampling site
(2018) Microbial Risk Analysis, 10, pp. 28-36.
ABSTRACT: Contamination of raw poultry meat with foodborne pathogens could occur because of improper handling at primary production and slaughterhouse levels. Low microbial prevalence data often consists of a high amount of non-detections (zero positives), so a flexible framework is required to characterise the underlying microbial distribution and conduct reliable inferential statistics. Thus, the objective of this work was to evaluate the performance of zero-inflated binomial (ZIB) regression models to describe the effects of sampling site (carcass, thigh, breast, wings) on the measured incidences of Salmonella, Listeria monocytogenes and Staphylococcus aureus on chicken meat. For each pathogen, four regression models based on the zero-inflated binomial ZIB (p, w0) distribution were fitted to the presence/absence data with sampling site as covariate and random-effects due to sampling occasion either in the binomial probability (p) or in the extra-proportion of non-detections (w0). For the three pathogens, the sampling site exerted a greater effect on w0 than on p itself, with breast bearing the lowest prevalence estimates of Salmonella spp. (mean: 0.88%; 95% CI: 0.02–1.95%) and S. aureus (mean 1.48%; 95% CI: 0.01–4.00%). The fitting capacity of the models was further improved when random effects due to sampling occasion were placed in w0 (deviances decreased from 146.7–156.7 to 140.2–140.6). This would imply that, theoretically, the variability in pathogens' occurrence from batch to batch mainly arises from the variability in non-contaminated zones. At any sampling site, the mean prevalence was estimated as 1.35 (95% CI: 0.15 – 2.70) for Salmonella, 2.11 (95% CI: 0.04 – 5.63) for L. monocytogenes and 2.36 (95% CI: 0.04 – 5.12) for S. aureus. Sampling performance analysis showed that wings were mostly suitable to detect Salmonella and S. aureus with higher probability (0.016 and 0.035 respectively), while for L. monocytogenes, sampling of thigh could be more effective (0.032). ISSN: 23523522

Robertson, J., Yoshida, C., Gurnik, S., McGrogan, M., Davis, K., Arya, G., Murphy, S.A., Nichani, A., Nash, J.H.E.
An improved DNA array-based classification method for the identification of Salmonella serotypes shows high concordance between traditional and genotypic testing
(2018) PLoS ONE, 13 (12), art. no. e0207550, .
ABSTRACT: Previously we developed and tested the Salmonella GenoSerotyping Array (SGSA), which utilized oligonucleotide probes for O- and H- antigen biomarkers to perform accurate molecular serotyping of 57 Salmonella serotypes. Here we describe the development and validation of the ISO 17025 accredited second version of the SGSA (SGSA v. 2) with reliable and unambiguous molecular serotyping results for 112 serotypes of Salmonella which were verified both in silico and in vitro. Improvements included an expansion of the probe sets along with a new classifier tool for prediction of individual antigens and overall serotype from the array probe intensity results. The array classifier and probe sequences were validated in silico to high concordance using 36,153 draft genomes of diverse Salmonella serotypes assembled from public repositories. We obtained correct and unambiguous serotype assignments for 31,924 (88.30%) of the tested samples and a further 3,916 (10.83%) had fully concordant antigen predictions but could not be assigned to a single serotype. The SGSA v. 2 can directly use bacterial colonies with a limit of detection of 860 CFU/mL or purified DNA template at a concentration of 1.0 x 10−1 ng/μl. The SGSA v. 2 was also validated in the wet laboratory and certified using panel of 406 samples representing 185 different serotypes with correct antigen and serotype determinations for 60.89% of the panel and 18.31% correctly identified but an ambiguous overall serotype determination. ISSN: 19326203

Henley, S.C., Launchi, N., Quinlan, J.J.
Survival of Salmonella on raw poultry exposed to 10% lemon juice and vinegar washes
(2018) Food Control, 94, pp. 229-232.
ABSTRACT: The widespread practice of washing raw poultry has been the target of multiple consumer education campaigns in recent years. In addition to rinsing with plain water, a subset of consumers report using acidic solutions (diluted lemon/lime juice or vinegar) to wash raw poultry. While studies have demonstrated the ineffectiveness of

acidic marinades to eliminate pathogens from raw meat, the effect of acidic washes on raw poultry has not previously been examined. The research reported here determined the fate of Salmonella enterica 19214 inoculated onto raw poultry and subsequently exposed to acidic washes. Chicken breasts were inoculated with approximately 5 × 108 CFU of Salmonella enterica 19214 (resistant to tetracycline, streptomycin and chloramphenicol). Inoculated breasts were then washed for 10 s, 30 s, 2 min or 5 min in control (tap water) or acidic (10% vinegar or 10% lemon juice) solutions to simulate consumer washing. Following washing, S. enterica 19214 levels were determined both in the wash water and on the chicken using media containing antibiotics. Washing with 10% vinegar (pH 3.1) resulted in the recovery of 7.23–7.46 log CFU/ml S. enterica from the chicken and 6.63 to 6.73 log CFU/ml S. enterica from the vinegar wash solution. Washing with 10% lemon juice (pH 2.6) resulted in the recovery of 7.26–7.42 log CFU/ml from the chicken and 6.28 to 7.06 log CFU/ml from the lemon juice wash. Results indicate that acidic washes result in live Salmonella both in the wash as well as remaining on the chicken. Washing raw poultry in a diluted lemon juice or vinegar solution is an inefficient method for removing pathogens and results in pathogens both in the wash water and on the chicken, increasing the risk for cross contamination and potential foodborne illness. ISSN: 09567135

Bundidamorn, D., Supawasit, W., Trevanich, S.
A new single-tube platform of melting temperature curve analysis based on multiplex real-time PCR using EvaGreen for simultaneous screening detection of Shiga toxin-producing Escherichia coli, Salmonella spp. and Listeria monocytogenes in food
(2018) Food Control, 94, pp. 195-204.
ABSTRACT: Shiga toxin-producing Escherichia coli (STEC), Salmonella spp. and Listeria monocytogenes are continuously reported as causative agents of great concern regarding food safety and widespread contamination in many food varieties. Therefore, their simultaneous detection may be advantageous in terms of cost, time and labor savings and simplicity. This study developed a new, simple platform of multiplex real-time polymerase chain reaction (mRT-PCR) for specific, sensitive and rapid detection of STEC, Salmonella spp. and L. monocytogenes in food. The single-tube mRT-PCR format was developed by combining an 18 h enrichment step in simultaneous enrichment broth, boiling based on DNA extraction assay and a mRT-PCR detection system based on melting curve analysis using a fluorescent dye (EvaGreen) for detection of the presence or absence of the three target bacterial pathogens in food samples. Three specific peaks were clearly detected with average melting temperatures of 84.52 ± 0.90 °C, 87.51 ± 0.54 °C and 79.32 °C ± 0.48 °C for STEC, Salmonella spp. and L. monocytogenes, respectively. The sensitivity and specificity of these newly developed mRT-PCR platforms were further investigated using artificially and naturally contaminated food samples. The relative sensitivity, relative specificity and relative accuracy were all 100%, with a detection limit of 1 cfu for each target pathogen in 25 g of food sample. The developed platform of EvaGreen-based single-tube mRT-PCR for detection of the three target pathogenic bacteria in food samples provided results of absence or presence within 20 h. The newly developed mRT-PCR platform in this study offers a promising approach for simple, rapid, sensitive, specific and accurate detection of the three target bacterial pathogens in food. ISSN: 09567135

Li, K., Khouryieh, H., Jones, L., Etienne, X., Shen, C.
Assessing farmers market produce vendors' handling of containers and evaluation of the survival of Salmonella and Listeria monocytogenes on plastic, pressed-card, and wood container surfaces at refrigerated and room temperature
(2018) Food Control, 94, pp. 116-122.
ABSTRACT: This study aims to assess how small produce growers handle containers and evaluate the survival of Salmonella and Listeria monocytogenes on various produce container surfaces commonly used at farmers markets, under storage conditions both at refrigerated and room temperature. In Study I, an anonymous survey was conducted to assess the practices of handling produce containers from 28 vendors at farmers markets in Morgantown, WV and 141 vendors from farmers markets in Kentucky. In Study II, plastic, pressed-card, and wood containers were trimmed (25 cm2) and inoculated with S. typhimurium and Tennessee, and two strains of L. monocytogenes, stored at 3.2 °C (22.19% RH) and 22.5 °C (50.40% RH), respectively, for 21 days and periodically analyzed for microbial populations on XLT-4 (Salmonella) and Modified-Oxford (L. monocytogenes) agars. The survey results showed that plastic, paper, and wood containers are the top three choices for small produce growers to transport and present produce at farmers markets. The pathogens decreased slower (P < 0.05) at 3.2 °C and on pressed-card and wood surfaces than at 22.5 °C and on a plastic surface. At 3.2 °C, Salmonella counts decreased (P < 0.05) from 5.27 to 5.53 to 2.63–2.84 log CFU/cm2, and L. monocytogenes decreased (P < 0.05) from 6.39 to 6.93 to 4.89–5.46 log

CFU/cm2 on the three material surfaces by the end of the storage period, with the lowest (P < 0.05) survival on a plastic surface. At 22.5 °C, Salmonella populations decreased (P < 0.05) from 4.94 to 5.38 to <1.30 log CFU/cm2 (the detection limit) after 3, 9 and 12 days on plastic, pressed-card and wood surfaces, respectively. L. monocytogenes decreased (P < 0.05) from 6.39 to 6.93 to ≤1.30 log CFU/cm2 after 12, 12, and 21 days on plastic, wood and pressed-card surfaces, respectively. These results were confirmed by different mathematical survival models for analyzing pathogen inactivation rates. Vendors at farmers markets should choose plastic containers to store fresh produce and avoid storing containers at refrigerated temperature. ISSN: 09567135

Gosling, R.J., Mueller-Doblies, D., Martelli, F., Nunez-Garcia, J., Kell, N., Rabie, A., Wales, A.D., Davies, R.H.
Observations on the distribution and persistence of monophasic Salmonella Typhimurium on infected pig and cattle farms
(2018) Veterinary Microbiology, 227, pp. 90-96.
ABSTRACT: Following a rapid rise in cases of monophasic Salmonella Typhimurium DT193 (mST) in humans and pigs since 2007 a detailed study of the prevalence and persistence of mST on pig and cattle farms in Great Britain (GB) was undertaken. Thirteen commercial pig farms and twelve cattle farms, identified as mST-positive from surveillance data, were intensively sampled over a three year period. Five indoor and eight outdoor pig farms and four beef and eight dairy farms were included. Individual and pooled faecal samples were collected from each epidemiological group and environmental samples throughout each farm and the antimicrobial resistance profile determined for a selection of mST-positive isolates. Indoor pig farms had a higher mST prevalence than outdoor pig farms, and across both cattle and pig farms the juvenile animals had a higher mST prevalence than the adult animals. Overall, mST prevalence decreased with time across all pig farms, from 25% to less than 15% of environmental samples and 22% to 15% of pooled faecal samples; only one organic outdoor breeding farm was Salmonella-negative at the end of the study. Across the cattle farms no mST was detected by the end of the study, apart from one persistent farm. Clearance time of mST was between seven and twenty-five months. Farms were selected based on having the antimicrobial resistance profile ampicillin, streptomycin, sulphonamides and tetracycline (A, S, SU, T), although resistance to trimethoprim-potentiated sulphamethoxazole was also identified on five pig farms sampled. This study provided a detailed insight into the distribution and persistence of mST on individual pig and cattle farms in GB. It has identified variation in mST shedding of individual animals, and the data can be applied to the wider livestock industry when considering the distribution of mST once identified on an individual farm. ISSN: 03781135

Gurman, P.M., Ross, T., Kiermeier, A.
Quantitative Microbial Risk Assessment of Salmonellosis from the Consumption of Australian Pork: Minced Meat from Retail to Burgers Prepared and Consumed at Home
(2018) Risk Analysis, 38 (12), pp. 2625-2645.
ABSTRACT: Pork burgers could be expected to have an elevated risk of salmonellosis compared to other pork products due to their comminuted nature. A stochastic risk assessment was performed to estimate the risk of salmonellosis from Australian pork burgers and considered risk-affecting factors in the pork supply chain from retail to consumption at home. Conditions modeled included prevalence and concentration of Salmonella in pork mince, time and temperature effects during retail, consumer transport, and domestic storage and the effect of cooking, with the probability of illness from consumption estimated based on these effects. The model was two-dimensional, allowing for the separation of variability and uncertainty. Potential changes to production practices and consumer behaviors were examined through alternative scenarios. Under current conditions in Australia, the mean risk of salmonellosis from consumption of 100 g pork burgers was estimated to be (Formula presented.) per serving or one illness per 65,000,000 servings consumed. Under a scenario in which all pork mince consumed is served as pork burgers, and with conservative (i.e., worst-case) assumptions, 0.746 cases of salmonellosis per year from pork burgers in Australia were predicted. Despite the adoption of several conservative assumptions to fill data gaps, it is predicted that pork burgers have a low probability of causing salmonellosis in Australia. ISSN: 02724332

Govaert, M., Smet, C., Baka, M., Janssens, T., Impe, J.V.
Influence of incubation conditions on the formation of model biofilms by Listeria monocytogenes and Salmonella Typhimurium on abiotic surfaces
(2018) Journal of Applied Microbiology, 125 (6), pp. 1890-1900.
ABSTRACT: Aims: This research aims to develop strongly adherent and mature model biofilms (on a 20 cm² polystyrene surface) for two pathogenic species, i.e. Listeria

monocytogenes and Salmonella Typhimurium. These model biofilms can be used as standards to study biofilms or to study/compare the influence of different inactivation technologies. Methods and Results: Three influencing factors on the formation of biofilms are investigated, i.e. growth medium, incubation temperature and incubation time, which are three easily controllable environmental factors. Optical density measurement and plate counts were used to evaluate the adherence and the maturity of the biofilms, respectively. Confocal laser scanning microscopy was used to verify most important findings obtained with previously mentioned assays. Results indicated that mature and strongly adherent L. monocytogenes biofilms are obtained following 13 h of incubation at 30°C with BHI as growth medium. For S. Typhimurium, an incubation period of 19 h at 25°C was required with 20-fold diluted TSB as growth medium. Conclusions: Based on previously mentioned assays, a protocol for the formation of reproducible model biofilms was obtained. Significance and Impact of the Study: The developed model biofilms can be applied as a standard to study biofilms (in different research fields) and their subsequent inactivation by different methods. In addition, the results of this study could be used to control biofilm formation (e.g. by setting a maximum allowed surface temperature). ISSN: 13645072

Feng, J., Yao, W., Guo, Y., Cheng, Y., Qian, H., Xie, Y.
Incorporation of Heavy Water for Rapid Detection of Salmonella typhimurium by Raman Microspectroscopy
(2018) Food Analytical Methods, 11 (12), pp. 3551-3557.
ABSTRACT: The presence of foodborne pathogens is one of the leading causes of food safety incidents; hence, early monitoring of the status of microbial contamination is a permanent concern. Here, a rapid and sensitive method for detection of Salmonella typhimurium was firstly developed by combining heavy water (D2O) labeling and Raman microspectroscopy to measure bacterial metabolic activity. It was based on the characteristics of Raman signals of carbon–deuterium (C–D) vibration as cells incorporate deuterium in place of hydrogen (H) during biosynthetic activity, and the live and dead Salmonella typhimurium cells could be distinguished by their metabolic activity within 1 h. The CD / (CD + CH)% of D2O-incorporated Salmonella typhimurium in logarithmic growth phase was linearly related to the log of the initial concentration, and Salmonella typhimurium with the initial concentration of 102–106 colony-forming unit (CFU)/mL could be quantified within 4–8 h. Furthermore, desirable recovery ranging from 95.41 to 106.6% was obtained to assess the practicality and stability of this method for rapid detection of Salmonella typhimurium in milk, indicating promising application for the detection of bacterial contamination in food. ISSN: 19369751

Liu, J., Jasim, I., Abdullah, A., Shen, Z., Zhao, L., El-Dweik, M., Zhang, S., Almasri, M.
An integrated impedance biosensor platform for detection of pathogens in poultry products (2018) Scientific Reports, 8 (1), art. no. 16109, .
ABSTRACT: This paper presents an impedance-based biosensor for rapid and simultaneous detection of Salmonella serotypes B, D, and E with very low concentration. The biosensor consists of a focusing region, and three detection regions. The cells focusing was achieved using a ramp down electroplated vertical electrode pair along with tilted thin film finger pairs that generate p-DEP forces to focus and concentrate the bacterial cells into the center of the microchannel, and direct them toward the detection region. The detection regions consist of three interdigitated electrode arrays (IDEA), each with 20 pairs of finger coated with a mixture of anti-Salmonella antibody and crosslinker to enhance the adhesion to IDEA. The impedance changes as the target Salmonella binds to the antibody. The biosensor has showed excellent performance as proven by the detection of a single Salmonella serotype B, and simultaneous detection of two Salmonella serotypes B and D with a limit of detection (LOD) of 8 Cells/ml in ready-to-eat turkey samples, the addition of focusing capability improved the measured signal by a factor of between 4–4.5, the total detection time of 45 minutes, selectivity of the sensor on different types of bacterial cells, and the ability to distinguish between dead and live cells. ISSN: 20452322

Campioni, F., Cao, G., Kastanis, G., Janies, D.A., Bergamini, A.M.M., Rodrigues, D.D.P., Stones, R., Brown, E., Allard, M.W., Falcão, J.P.
Changing of the Genomic Pattern of Salmonella Enteritidis Strains Isolated in Brazil over a 48 year-period revealed by Whole Genome SNP Analyses
(2018) Scientific Reports, 8 (1), art. no. 10478, .
ABSTRACT: Salmonella Enteritidis became the main serovar isolated from gastroenteritis cases in Brazil after the 90's. In this study we used whole genome sequence analysis to determine the phylogenetic relationships among a collection of strains isolated in Brazil to identify possible genomic differences between the strains isolated in the pre and post-

epidemic period. Also, we compared our data from strains isolated in Brazil to strains available in the public domain from other South American countries. Illumina technology was used to sequence the genome of 256 Salmonella Enteritidis strains isolated over a 48 year-period in Brazil, comprising the pre-and post-epidemic period. Phylogenetic analyses revealed distinct lineages for strains isolated before and after 1994. Moreover, the phage region SE20 that may be related to the emergence of Salmonella Enteritidis worldwide was present only in strains of the post-epidemic cluster. In conclusion, our results showed that the genomic profile of Salmonella Enteritidis strains isolated in Brazil shifted after 1994, replaced by a global epidemic group of strains. It may be hypothesized that the presence of the prophage SE20 might have conferred to these strains a better ability to colonize chicken and consequently to infect and cause disease in humans, which might better explain the increase in the number of S. Enteritidis cases in Brazil and other South American countries. However, to verify this hypothesis further studies are needed. ISSN: 20452322

Wang, Y.U., Pettengill, J.B., Pightling, A., Timme, R., Allard, M., Strain, E., Rand, H.
Genetic Diversity of Salmonella and Listeria Isolates from Food Facilities
(2018) Journal of food protection, 81 (12), pp. 2082-2089.
ABSTRACT: Food production-related facilities (farms, packing houses, etc.) are monitored for foodborne pathogens, and data from these facilities can provide a rich source of information about the population structure and genetic diversity of Salmonella and Listeria. This information is of both academic interest for understanding the evolutionary forces acting on these organisms and of practical interest to those responsible for controlling pathogens in facilities and to those analyzing data from facilities in the context of public health decision making. We have collected information about all positive isolates from facility inspections performed by the U.S. Food and Drug Administration for which whole genome sequencing data are available. The within- and between-facilities observed genetic diversity of isolates was computed and related to the common origin of isolates (as the common collected facility). This relationship provides quantification for assessing the relationship between isolates based on their genetic similarity quantified by single-nucleotide polymorphisms (SNPs). Our results show that if the genetic distance (D) between two isolates is low, then more likely than not they are from the same facility or have some overlap in their supply chain. For example, if the genetic distance is no more than 20 SNPs, the probability (P) that two isolates come from the same facility = 0.66 for Salmonella and 0.70 for Listeria. However, if two isolates come from different facilities, their genetic distance is likely large (for Salmonella, P(D > 20 SNPs) = 0.99982; for Listeria, P(D > 20 SNPs) = 0.99949); even if two isolates come from the same facility, their genetic distance is also very likely large (for Salmonella, P(D > 20 SNPs) = 0.794; for Listeria, P(D > 20 SNPs) = 0.692). These results provide insight into what SNP thresholds might be appropriate when determining whether two isolates are from the same facility and thus would be of interest to those investigating foodborne outbreaks and conducting traceback investigations. ISSN: 19449097

Čučak, D., Babić, O., Tamaš, I., Simeunović, J., Karaman, M., Kovač, D., Novaković, M., Markov, S., Knežević, P., Stojanov, I., Obradović, V., Radnović, D.
Prevalence, Antibiotic Resistance and Diversity of Salmonella Isolates from Soils and Sediments in Serbia
(2018) International Journal of Environmental Research, 12 (6), pp. 829-841.
ABSTRACT: Salmonellae are important bacterial pathogens dispersed in the environment through wastewater and agricultural application of organic fertilizers, while infections are mainly food borne. Although routine monitoring in human, meat production and processing areas and in animal meat is conducted, little is known about Salmonella persistence in environmental samples, even though they are highly important ecological reservoirs for the spreading of pathogen and antibiotic resistance determinants. Here, we present a large-scale survey of Salmonella isolates from a variety of soils and sediments in Serbia. Among 1062 analyzed samples, 67 (6.31%) tested salmonellae positive. A number of isolates persisted in soil and sediment stored at low temperatures for up to 415 days. A third of the tested isolates exhibited atypical biopatterns unusual for salmonellae. Majority of strains (87.5%) were either sensitive or intermediate to all the 20 tested antimicrobials, while only two strains were multidrug resistant (6.25%). Notably, all isolates were sensitive to medically important antibiotics such as fluoroquinolone, ciprofloxacin and beta-lactams. Serotyping identified three subspecies, namely enterica, salamae and diarizonae and 15 serovars, with the most prevalent ones being Brandenburg (n = 9), Enteritidis (n = 8) and Wien (n = 7). With the exception of S. Enteritidis, other detected serovars are rarely present in human and animal samples. To conclude, Salmonella strains isolated from

soil and sediment samples across Serbia exhibited unusual biochemical properties, low level of antibiotic resistance and high serovar diversity. Results indicate serovars uncommon in human and animal meat samples are more persistent in the outside non-host environments. ISSN: 17356865

Erickson, M.C., Liao, J.-Y., Payton, A.S., Cook, P.W., Bautista, J., Díaz-Pérez, J.C.
Disposition of Salmonella and Escherichia coli O157:H7 following Spraying of Contaminated Water on Cucumber Fruit and Flowers in the Field
(2018) Journal of food protection, 81 (12), pp. 2074-2081.
ABSTRACT: Cucumbers are frequently consumed raw and have been implicated in several recent foodborne outbreaks. Because this item may become contaminated at the farm, it is vital to explore the fate of attenuated Salmonella Typhimurium or Escherichia coli O157:H7 sprayed onto foliage, flowers, and fruit in fields and determine whether pre- or postcontamination spray interventions could minimize contamination. After spraying cucumber plants with contaminated irrigation water (3.8 log CFU/mL of Salmonella Typhimurium and E. coli O157:H7), 60 to 78% of cucumber fruit were not contaminated because the plant's canopy likely prevented many of the underlying fruit from being exposed to the water. Subsequent exposure of contaminated cucumber plants to a simulated shower event did not appear to dislodge pathogens from contaminated foliage onto the fruit, nor did it appear to consistently wash either pathogen from the fruit. Spraying flowers and attached ovaries directly with a pathogen inoculum (4.6 log CFU/mL) initially led to 100% and 65 to 90% contamination, respectively. Within 3 days, 30 to 40% of the flowers were still contaminated; however, contamination of ovaries was minimal (≤10%), suggesting it was unlikely that internalization occurred through the flower to the ovary with these pathogen strains. In another study, both pathogens were found on a withered flower but not on the fruit to which the flower was attached, suggesting that this contaminated flower could serve as a source of cross-contamination in a storage bin if harvested with the fruit. Because pre- and postcontamination acetic acid-based spray treatments failed to reduce pathogen prevalence, the probability that fruit initially contaminated at 1.3 to 2.8 log CFU of Salmonella Typhimurium or E. coli O157:H7 per cucumber would be positive by enrichment culture decreased by a factor of 1.6 and 1.9 for Salmonella Typhimurium and E. coli O157:H7, respectively, for every day the fruit was held in the field ($P \leq 0.0001$). Hence, to reduce the prevalence of Salmonella Typhimurium on cucumbers below 5%, more than 1 week would be required.
ISSN: 19449097

Lawson, B., Franklinos, L.H.V., Rodriguez-Ramos Fernandez, J., Wend-Hansen, C., Nair, S., Macgregor, S.K., John, S.K., Pizzi, R., Núñez, A., Ashton, P.M., Cunningham, A.A., De Pinna, E.M.
Salmonella Enteritidis ST183: Emerging and endemic biotypes affecting western European hedgehogs (Erinaceus europaeus) and people in Great Britain
(2018) Scientific Reports, 8 (1), art. no. 2449, .
ABSTRACT: The impacts of hedgehog (Erinaceus europaeus) Salmonella infection on public health and on animal welfare and conservation are unknown. We isolated Salmonella Enteritidis multi-locus sequence-type (ST)183 from 46/170 (27%) hedgehog carcasses (27 S. Enteritidis phage type (PT)11, 18 of a novel PT66 biotype and one with co-infection of these PTs) and from 6/208 (3%) hedgehog faecal samples (4 PT11, 2 PT66) from across Great Britain, 2012-2015. Whole genome phylogenetic analysis of the hedgehog isolates and ST183 from people in England and Wales found that PT11 and PT66 form two divergent clades. Hedgehog and human isolates were interspersed throughout the phylogeny indicating that infections in both species originate from a common population. PT11 was recovered from hedgehogs across England and Scotland, consistent with endemic infection. PT66 was isolated from Scotland only, possibly indicating a recent emergence event. People infected with ST183 were four times more likely to be aged 0-4 years than people infected by the more common ST11 S. Enteritidis. Evidence for human ST183 infection being non-foodborne included stronger correlation between geographic and genetic distance, and significantly increased likelihood of infection in rural areas, than for ST11. These results are consistent with hedgehogs acting as a source of zoonotic infection. ISSN: 20452322

Parker, A.E., Hamilton, M.A., Goeres, D.M.
Reproducibility of antimicrobial test methods
(2018) Scientific Reports, 8 (1), art. no. 12531, .
ABSTRACT: We review reproducibility results for methods that test antimicrobial efficacy against biofilms, spores and bacteria dried onto a surface. Our review, that included test results for Pseudomonas aeruginosa, Salmonella choleraesuis and Bacillus subtilis,

suggests that the level of reproducibility depends on the efficacy of the antimicrobial agent being tested for each microbe and microbial environment. To determine the reproducibility of a method, several laboratories must independently test the same antimicrobial agent using the method. Little variability among the efficacy results suggests good reproducibility. Such reproducibility assessments currently are hampered by the absence of an objective process for deciding whether the variability is sufficiently small. We present a quantitative decision process that objectively determines whether any method that assesses antimicrobial efficacy is reproducible. Because the perception of acceptable reproducibility may differ among stakeholders, the decision process is governed by a stakeholder's specifications that necessarily includes the efficacy of the agents to be tested. ISSN: 20452322

Palma, F., Manfreda, G., Silva, M., Parisi, A., Barker, D.O.R., Taboada, E.N., Pasquali, F., Rossi, M.
Genome-wide identification of geographical segregated genetic markers in Salmonella enterica serovar Typhimurium variant 4,[5],12:i:-
(2018) Scientific Reports, 8 (1), art. no. 15251, .
ABSTRACT: Salmonella enterica ser. Typhimurium monophasic variant 4,[5],12:i:- has been associated with food-borne epidemics worldwide and swine appeared to be the main reservoir in most of the countries of isolation. However, the monomorphic nature of this serovar has, so far, hindered identification of the source due to expansion of clonal lineages in multiple hosts and food producing systems. Since geographically structured genetic signals can shape bacterial populations, identification of biogeographical markers in S. 1,4,[5],12:i:- genomes can contribute to improving source attribution. In this study, the phylogeographical structure of 148 geographically and temporally related Italian S. 1,4,[5],12:i:- has been investigated. The Italian isolates belong to a large population of clonal S. Typhimurium/1,4,[5],12:i:- isolates collected worldwide in two decades showing up to 2.5% of allele differences. Phylogenetic reconstruction revealed that isolates from the same geographical origin form highly supported monophyletic groups, suggesting discrete geographical segregation. These monophyletic groups are characterized by the gene content of a large sopE-containing prophage. Within this prophage, genome-wide comparison identified several genes overrepresented in strains of Italian origin. This suggests that certain lineages may be characterized by the acquisition of specific accessory genetic markers useful for improving identification of the source in ongoing epidemics. ISSN: 20452322

Kim, W.-I., Ryu, S.D., Kim, S.-R., Kim, H.-J., Lee, S., Kim, J.
Population changes and growth modeling of Salmonella enterica during alfalfa seed germination and early sprout development
(2018) Food Science and Biotechnology, 27 (6), pp. 1865-1869.
ABSTRACT: This study examined the effects of alfalfa seed germination on growth of Salmonella enterica. We investigated the population changes of S. enterica during early sprout development. We found that the population density of S. enterica, which was inoculated on alfalfa seeds was increased during sprout development under all experimental temperatures, whereas a significant reduction was observed when S. enterica was inoculated on fully germinated sprouts. To establish a model for predicting S. enterica growth during alfalfa sprout development, the kinetic growth data under isothermal conditions were collected and evaluated based on Baranyi model as a primary model for growth data. To elucidate the influence of temperature on S. enterica growth rates, three secondary models were compared and found that the Arrhenius-type model was more suitable than others. We believe that our model can be utilized to predict S. enterica behavior in alfalfa sprout and to conduct microbial risk assessments. ISSN: 12267708