Risk Assessment

Validation of Quantitative Microbial Risk Assessment Using Epidemiological Data From Outbreaks of Waterborne Gastrointestinal Disease

Burch T. *Risk Anal.* 2019 Mar;39(3):599-615. doi: 10.1111/risa.13189. Article Link

Significance: This study validated quantitative microbial risk assessment for predicting disease rates due to *Cryptosporidium* and *Giardia*.

The assumptions underlying quantitative microbial risk assessment (QMRA) are simple and biologically plausible, but QMRA predictions have never been validated for many pathogens. The objective of this study was to validate QMRA predictions against epidemiological measurements from outbreaks of waterborne gastrointestinal disease. I screened 2,000 papers and identified 12 outbreaks with the necessary data: disease rates measured using epidemiological methods and pathogen concentrations measured in the source water. Eight of the 12 outbreaks were caused by *Cryptosporidium*, three by *Giardia*, and one by noro-virus. Disease rates varied from 5.5×10^{-6} to 1.1×10^{-2} cases/person-day, and reported pathogen concentrations varied from 1.2×10^{-4} to 8.6×10^{2} per liter. I used these concentrations with single-hit dose-response models for all three pathogens to conduct QMRA, producing both point and interval predictions of disease rates for each outbreak. Comparison of QMRA predictions to epidemiological measurements showed good agreement; interval predictions contained measured disease rates for 9 of 12 outbreaks, with point predictions off by factors of 1.0-120 (median = 4.8). Furthermore, 11 outbreaks occurred at mean doses of less than 1 pathogen per exposure. Measured disease rates for these outbreaks were clearly consistent with a single-hit model, and not with a "two-hit" threshold model. These results demonstrate the validity of QMRA for predicting disease rates due to *Cryptosporidium* and *Giardia*.

Improving Hazard Characterization in Microbial Risk Assessment Using Next Generation Sequencing Data and Machine Learning: Predicting Clinical Outcomes in Shigatoxigenic *Escherichia coli*

Njage PMK, Leekitcharoenphon P, Hald T. *Int J Food Microbiol.* 2019 Mar 2;292:72-82. doi: 10.1016/j.ijfoodmicro.2018.11.016. Article Link

Significance: Machine learning may be applied for real-time online analysis of whole genome sequence data to estimate the human health risk at the population or strain level.

The ever decreasing cost and increase in throughput of next generation sequencing (NGS) techniques have resulted in a rapid increase in availability of NGS data. Such data have the potential for rapid, reproducible and highly discriminative characterization of pathogens. This provides an opportunity in microbial risk assessment to account for variations in survivability and virulence among strains. A major challenge towards such attempts remains the highly dimensional nature of genomic data versus the number of isolates. Machine learning-based (ML) predictive risk modelling provides a solution to this "curse of dimensionality" while accounting for individual effects that are dependent on interactions with other genetic and environmental factors. This pilot study explores the potential of ML in the prediction of health endpoints resulting from shigatoxigenic *E. coli* (STEC) infection. Accessory genes in amino acid sequences were used as model input to predict and differentiate health outcomes in STEC infections including diarrhea, bloody diarrhea, hemolytic uremic syndrome and their combinations. Outcomes severity was also distinguished by hospitalization. A matrix of percent similarity between accessory genes and the *E. coli* genomes was generated and subsequently used as input for ML. The performances of ML algorithms random forest, support vector machine (radial and linear kernel), gradient boosting, and logit boost were compared. Logit boost was the best model showing an outcome prediction accuracy of 0.75 (95% CI: 0.60, 0.86), an excellent or substantial performance (Kappa = 0.72). Important genetic predictors of riskier STEC clinical

outcomes included proteins involved in initial attachment to the host cell, persistence of plasmids or genomic islands, conjugative plasmid transfer and formation of sex pili, regulation of locus of enterocyte effacement expression, post-translational acetylation of proteins, facilitation of the rearrangement or deletion of sections within the pathogenic islands and transport macromolecules across the cell envelope. We propose further studies are proposed on the proteins with undefined or unclear functionality. One protein family in particular predicted HUS outcome. Toxin-antitoxin systems are potential stress adaptation markers which may mediate environmental persistence of strains in diverse sources. We foresee the application of ML approach to the set-up of real-time online analysis of whole genome sequence data to estimate the human health risk at the population or strain level. The ML approach is envisaged to support the prediction of more specific STEC clinical endpoints type by inputting isolate sequence data.

A Modular Bayesian Salmonella Source Attribution Model for Sparse Data

Mikkelä A, Ranta J, Tuominen P. *Risk Anal.* 2019 Mar 20. doi: 10.1111/risa.13310. Article Link

Significance: The Bayesian salmonella source attribution model presented in this study can be applied to small and sparse annual data sets.

Several statistical models for salmonella source attribution have been presented in the literature. However, these models have often been found to be sensitive to the model parameterization, as well as the specifics of the data set used. The Bayesian salmonella source attribution model presented here was developed to be generally applicable with small and sparse annual data sets obtained over several years. The full Bayesian model was modularized into three parts (an exposure model, a subtype distribution model, and an epidemiological model) in order to separately estimate unknown parameters in each module. The proposed model takes advantage of the consumption and overall salmonella prevalence of the studied sources, as well as bacteria typing results from adjacent years. The latter were used for a smoothed estimation of the annual relative proportions of different salmonella subtypes in each of the sources. The source-specific effects and the salmonella subtype-specific effects were included in the epidemiological model to describe the differences between sources and between subtypes in their ability to infect humans. The estimation of these parameters was based on data from multiple years. Finally, the model combines the total evidence from different modules to proportion human salmonellosis cases according to their sources. The model was applied to allocate reported human salmonellosis cases from the years 2008 to 2015 to eight food sources.

Foodborne Pathogens

Microbiological Safety of Dried Spices

Gurtler JB, Keller SE. *Annu Rev Food Sci Technol.* 2019 Mar 25;10:409-427. doi: 10.1146/annurev-food-030216-030000. Article Link

Significance: Emerging detection and inactivation methods applicable to foodborne pathogens in spices are examined and discussed.

Spices in the desiccated state provide an environment that allows the survival of many foodborne pathogens. Currently, the incidence of pathogen-positive spices imported into the United States is 1.9 times higher than for any other imported food. Correspondingly, imported spices have been associated with numerous foodborne outbreaks and multiple product recalls. Despite the association with recalls and outbreaks, the actual pathogen populations in spices, when found, are frequently extremely small. In addition to pathogenic bacterial species, toxigenic molds have been frequently recovered from spices, and aflatoxins have been found in as many as 58% of the spices sampled. The presence of toxigenic molds is especially problematic to the immunocompromised or those on immunosuppressive therapy and has been linked to gut aspergillosis. Numerous detection methods, including both traditional and advanced DNA regimes, are being tested to optimize recovery of pathogens from spices. Further, a number of new inactivation intervention methods to decontaminate spices are examined and discussed.

Mycotoxins

Dietary Exposure Assessment to Mycotoxins Through Total Diet Studies. A Review

Carballo D, Tolosa J, Ferrer E, Berrada H. *Food Chem Toxicol.* 2019 Mar 23;128:8-20. doi: 10.1016/j.fct.2019.03.033. Article Link

Significance: This review explores human mycotoxin exposure and risk assessment through total diet studies.

Mycotoxins are secondary metabolites of fungi that contaminate food in several stages and their increasing presence in food chain demand further control. Assessment of mycotoxins human exposure through processed diet is an important component of food safety strategies. The present review explores and summarises total diet studies (TDS) carried out in different countries focusing on mycotoxins determination. TDS were classified by samples preparation, mycotoxins analysis and dietary exposure

evaluation. Most of reviewed TDS performed multi-mycotoxins analysis in composite samples mainly, prepared taking into account local culinary habits. High performance liquid chromatography coupled with fluorescence detector was the predominant and the most sensitive technique used for determination. Ochratoxin A was the most analyzed mycotoxin, followed by trichothecenes, aflatoxins, zearalenone, fumonisins, patulin, enniatins, and beauvericin respectively. Alternaria toxins and ergot alkaloids were also included. Food commonly analyzed were cereals, meat, vegetables, fruits, nuts and beverages. The findings in food were in below the current European legislation, except for some sporadic samples of wine and milk meaning less than 1% of total analyzed samples. Dietary exposure was evaluated, through the estimated daily intake mycotoxin evaluation and risk assessment concluded that relatively scarce toxicological concern was associated to mycotoxins exposure. However, a special attention should be paid to meat and cereal products high percentile consumers.

Numerical Optimization of Temperature-Time Degradation of Multiple Mycotoxins

Gbashi S, Madala NE, De Saeger S, De Boevre M, Njobeh PB. *Food Chem Toxicol.* 2019 Mar;125:289-304. doi: 10.1016/j.fct.2019.01.009. Article Link

Significance: Thermal degradation patterns of mycotoxins as a function of temperature and time are presented.

Mycotoxins are potent food contaminants that exert significant deleterious effects on human and animal health, yet, there is limited and often conflicting data on their thermal stability. The present study systematically investigated the thermal degradation patterns of multiple mycotoxins as a function of temperature and time, in pure form and spiked into a food matrix (maize flour), using a numerical modelling approach. Mycotoxins under investigation included aflatoxins (AFs), fumonisins (FBs), zearalenone and its analogue α and β epimers (ZEAs), ochratoxins (OTs), T-2 toxin (T-2), alternariol monomethyl ether (AME) and sterigmatocystin (STEG). A set of statistically-designed experiments were conducted, and a second-order optimization function fitted to the experimental data. The resultant models were well fit with R^2 values ranging from 0.87 to 0.99 and 0.89 to 0.99, for pure mycotoxin standards and spiked maize flour, respectively. It was also possible to statistically determine the optimum degradation conditions which were 216.57 °C/63.28 min and 210.85 °C/54.71 min for pure mycotoxins and spiked into maize flour, respectively. Our observations herein could be critical for food safety applications targeted at reducing or at best eliminating completely multi-mycotoxins in food using heat processing while limiting the destruction of food quality factors.

Chemical Contaminants

Cold Plasma as a Tool for the Elimination of Food Contaminants: Recent Advances and Future Trends

Gavahian M, Khaneghah AM. *Crit Rev Food Sci Nutr.* 2019 Mar 29:1-12. doi: 10.1080/10408398.2019.1584600. Article Link

Significance: The benefits and limitations for cold plasma-induced degradation of food contaminants are discussed.

Food contaminants are challenging the food industry due to the inefficiency of conventional decontamination techniques. Cold plasma as an emerging technique for the degradation of food contaminants attracted notable attention. The current study overviews the plasma-induced degradation of food contaminants, discusses the mechanisms involved, points its benefits and drawbacks out, highlights the research needed in this area, and explores future trends. According to the literature, cold plasma efficiently degraded many common pesticides (e.g. parathion, paraoxon, omethoate, dichlorvos, malathion, azoxystrobin, cyprodinil, fludioxonil, cypermethrin, and chlorpyrifos) and food allergens (e.g. tropomyosin, b-conglycinin, glycinin, trypsin inhibitor, and Kunitztype trypsin inhibitor). These degradations occurred primarily due to the presence of reactive oxygen species (ROS) and reactive nitrogen species (RNS) in the plasma that attack the chemical bonds of food contaminants. The type of pesticide degrades are highly dependent on the concentrations of plasma-generated ROS and RNS. Research showed that several parameters, such as plasma generation device, plasma exposure time, plasma power, and the carrier gas composition, influence the type and concentration of reactive species (e.g. ROS and RNS) and the overall efficiency of cold plasma degradation for a specific pesticide or allergen. Highlights Cold plasma can be used for degradation of many types of pesticides and allergens. Plasma-generated reactive species and UV can interact with pesticides and allergens. The scaled up removal of pesticides and allergens by plasma can be challenging.

Occurrence of Environmental Pollutants in Foodstuffs: A Review of Organic vs. Conventional Food

González N, Marquès M, Nadal M, Domingo JL. *Food Chem Toxicol.* 2019 Mar;125:370-375. doi: 10.1016/j.fct.2019.01.021. Article Link

Significance: This study recommends monitoring for environmental contaminants in both conventional and organic foods.

In recent years, there is an increasing trend to consume organic foods instead of the conventional foodstuffs. This tendency is mainly due to the concern raised by the potential adverse health effects derived from the intake of pesticides, fertilizers, hormones and antibiotics, which are widely used in regular food production. Although organic label forbids the use of these

products, environmental contamination is likely to occur in both, conventional and organic foodstuffs. The main purpose of this review was to compare the levels of a number of environmental pollutants such as polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDD/Fs), polychlorinated biphenyls (PCBs), mycotoxins, trace elements, polycyclic aromatic hydrocarbons (PAHs), etc., in organic and conventional food items. The results show that, in general, the presence of nearby anthropogenic sources of pollution is the key issue influencing the occurrence of environmental pollutants in foodstuffs, regardless their organic or conventional origin. Based on this, we suggest that environmental contaminants should be monitored in both conventional and organic foods. Finally, the safety feature, which has been globally attributed to organic foods, might be questionable depending on the potential environmental contamination of these foods.

Heavy Metals

Early-Life Cadmium Exposure and Bone-Related Biomarkers: A Longitudinal Study in Children

Malin Igra A, Vahter M, Raqib R, Kippler M. *Environ Health Perspect.* 2019 Mar;127(3):37003. doi: 10.1289/EHP3655. Article Link

Significance: This longitudinal study found that childhood cadmium exposure was associated with several bone-related biomarkers at nine years of age.

Background: Chronic cadmium exposure has been associated with osteotoxicity in adults, but little is known concerning its effects on early growth, which has been shown to be impaired by cadmium. *Objectives:* Our objective was to assess the impact of early-life cadmium exposure on bone-related biomarkers and anthropometry at 9 y of age. *Methods:* For 504 children in a mother–child cohort in Bangladesh, cadmium exposure was assessed by concentrations in urine (U-Cd, long-term exposure) and erythrocytes (Ery-Cd, ongoing exposure) at 9 and 4.5 y of age, and in their mothers during pregnancy. Biomarkers of bone remodeling [urinary deoxypyridinoline (DPD), urinary calcium, plasma parathyroid hormone, osteocalcin, vitamin D3, insulin-like growth factor (IGF) 1, IGF binding protein 3, thyroid stimulating hormone] were measured at 9 y of age. *Results:* In multivariable-adjusted linear models, a doubling of concurrent U-Cd was associated with a mean increase in osteocalcin of 2.7 ng/mL (95% CI: 0.042, 5.9) and in urinary DPD of 22 nmol/L (95% CI: 12, 32). In a combined exposure model, a doubling of maternal Ery-Cd was associated with a mean increase in urinary DPD of 15 nmol/L (95% CI: −0.047, 30). Stratifying the osteocalcin model by gender ($p_{interaction}$ = 0.001), a doubling of concurrent U-Cd was associated with a mean decrease in osteocalcin of −4.3 ng/mL (95% CI: −8.5, −0.080) in boys and a mean increase of 9.4 ng/mL (95% CI: 5.4, 13) in girls. The same pattern was seen with U-Cd at 4.5 y of age ($p_{interaction}$ = 0.016). Children's U-Cd and Ery-Cd, concurrent and at 4.5 y of age, were inversely associated with vitamin D3. *Conclusions:* Childhood cadmium exposure was associated with several bone-related biomarkers and some of the associations differed by gender. https://doi.org/10.1289/EHP3655

Nanotechnology

Nanotechnology — A Shelf Life Extension Strategy for Fruits and Vegetables

Liu W, Zhang M, Bhandari B. *Crit Rev Food Sci Nutr.* 2019 Mar 21:1-16. doi: 10.1080/10408398.2019.1589415. Article Link

Significance: The advantages of nanotechnology-related shelf life extension strategies over traditional preservative methods recommendations for future nanotechnology shelf life extension research are discussed.

Most harvested fruits and vegetables cannot be stored in natural conditions for a satisfactory shelf life duration due to their perishable nature. There are conventional preservation methods but have the limitations of high production cost and/or unsatisfactory shelf-life and/or undesirable residue, etc. Because of a number of unique properties, nanotechnology-related shelf life extension strategies have the potential to compensate the shortcomings of traditional preservation methods. In this review article, basic principles and recent highly efficient applications of nanotechnology-related shelf life extension strategies in fruits and vegetables are described. Data analysis of these strategies are carried out to make readers more intuitive to the research status. Finally, the recommendations for future research of nanotechnology in fruits and vegetables shelf life extension are discussed. According to statistical data, nano-zinc dioxide is the most mentioned nanomaterials in the references, followed by silver nano particles. Nanotechnology-related combined preservation strategies and nanotechnology-related intelligent labeling system are considered as valuable areas for future researches. In addition, modeling studies, legal aspects, safety concerns, cost-benefit analyses and technical optimization need to be taken into account in future researches.

Food Processing Safety

Biofilms in Food Processing Environments: Challenges and Opportunities

Alvarez-Ordóñez A, Coughlan LM, Briandet R, Cotter PD. *Annu Rev Food Sci Technol.* 2019 Mar 25;10:173-195. doi: 10.1146/annurev-food-032818-121805. Article Link

Significance: Tools and novel strategies for in situ monitoring and characterization of biofilms in food processing facilities are discussed.

This review examines the impact of microbial communities colonizing food processing environments in the form of biofilms on food safety and food quality. The focus is both on biofilms formed by pathogenic and spoilage microorganisms and on those formed by harmless or beneficial microbes, which are of particular relevance in the processing of fermented foods. Information is presented on intraspecies variability in biofilm formation, interspecies relationships of cooperativism or competition within biofilms, the factors influencing biofilm ecology and architecture, and how these factors may influence removal. The effect on the biofilm formation ability of particular food components and different environmental conditions that commonly prevail during food processing is discussed. Available tools for the in situ monitoring and characterization of wild microbial biofilms in food processing facilities are explored. Finally, research on novel agents or strategies for the control of biofilm formation or removal is summarized.

Food Packaging

Intelligent Food Packaging: A Review of Smart Sensing Technologies for Monitoring Food Quality

Yousefi H, Su HM, Imani SM, Alkhaldi K, Filipe CDM, Didar TF. *ACS Sens*. 2019 Mar 25. doi: 10.1021/acssensors.9b00440. Article Link

Significance: This review highlights the applications of intelligent packaging for real-time monitoring of food quality.

Food safety is a major factor affecting public health and the well-being of society. A possible solution to control food-borne illnesses is through real-time monitoring of the food quality throughout the food supply chain. The development of emerging technologies, such as active and intelligent packaging, has been greatly accelerated in recent years, with a focus on informing consumers about food quality. Advances in the fields of sensors and biosensors has enabled the development of new materials, devices, and multifunctional sensing systems to monitor the quality of food. In this Review, we place the focus on an in-depth summary of the recent technological advances that hold the potential for being incorporated into food packaging to ensure food quality, safety, or monitoring of spoilage. These advanced sensing systems usually target monitoring gas production, humidity, temperature, and microorganisms' growth within packaged food. The implementation of portable and simple-to-use hand-held devices is also discussed in this Review. We highlight the mechanical and optical properties of current materials and systems, along with various limitations associated with each device. The technologies discussed here hold great potential for applications in food packaging and bring us one step closer to enable real-time monitoring of food throughout the supply chain.

Caffeine

Long-Term Coffee Consumption, Caffeine Metabolism Genetics, and Risk of Cardiovascular Disease: A Prospective Analysis of Up to 347,077 Individuals and 8368 Cases

Zhou A, Hyppönen E. *Am J Clin Nutr*. 2019 Mar 1;109(3):509-516. doi: 10.1093/ajcn/nqy297. Article Link

Significance: This prospective analysis found a modest increase in cardiovascular disease risk with heavy coffee consumption.

Background: Coffee is one of the most widely consumed stimulants worldwide and is generally considered to be safe or even beneficial for health. However, increased risk of myocardial infarction and hypertension has been suggested for individuals who carry a functional variant at cytochrome P450 1A2 (CYP1A2), which makes them less effective at metabolizing caffeine. *Objectives:* The aim of this study was to examine if the CYP1A2 genotype or a genetic score for caffeine metabolism (caffeine-GS) modifies the association between habitual coffee consumption and the risk of cardiovascular disease (CVD). *Methods:* Genetic data and information on habitual coffee intake and relevant covariates were available for 347,077 individuals in the UK Biobank, including 8368 incident CVD cases. We used logistic regression to test for the association between coffee intake and CVD risk, and whether the association varies with CYP1A2 genotype or caffeine-GS. *Results:* The association between habitual coffee intake and CVD risk was nonlinear, and, compared with participants drinking 1–2 cups/day, the risk of CVD was elevated for nondrinkers, drinkers of decaffeinated coffee, and those who reported drinking >6 cups/day (increase in odds by 11%, 7%, and 22%, respectively, P-curvature = 0.013). CYP1A2 genotype and caffeine-GS were not associated with CVD (P ≥ 0.22 for all comparisons). There was no evidence for an interaction between the CYP1A2 genotype or caffeine-GS and coffee intake with respect to risk of CVD (P ≥ 0.53). *Conclusions:* Heavy coffee consumption was associated with a modest increase in CVD risk, but this association was unaffected by genetic variants influencing caffeine metabolism.

Food Allergy

Sensitivity Analysis to Derive a Food Consumption Point Estimate for Deterministic Food Allergy Risk Assessment

Blom WM, Remington BC, Baumert JL, Bucchini L, Crépet A, Crevel RWR, et al. *Food Chem Toxicol*. 2019 Mar;125: 413-421. doi: 10.1016/j.fct.2019.01.025. Article Link

Significance: This sensitivity analysis established an optimal food consumption estimate for use in deterministic food allergy risk assessment

One of the input parameters in food allergy risk assessment is the amount of a given food consumed at an eating occasion. There is no consensus on how to use food consumption data when assessing the risk from unintended allergen presence in food products. A sensitivity analysis was performed to establish the optimal food consumption estimate for a deterministic food allergy risk assessment. Exposure was calculated for consumption percentiles (50th percentile, P50 to maximum) using the iFAAM consumption database in conjunction with an allergen concentration range from 1 to 1000 ppm. The resulting allergen intakes were compared to the allergic population reference doses proposed by Taylor et al. (2014) for 10 major allergenic foods. Optimal consumption percentiles were defined as those which predicted an intake below the relevant reference dose and met the defined acceptable risk level confirmed by probabilistic risk assessments. Analysis showed that, for 99% of the food groups, the P50 consumption met our criteria, while the P75 did so for 100% of the food groups. We suggest that the P75 is the optimal point estimate for use in deterministic food allergy risk assessment. It meets the safety objective and is adequately conservative for a public health context.