

April 2023

Food Safety Briefs



Join Us!

June 13th & 14th
National Press Club • Washington, DC

2023 Annual Summer Science Symposium

A Gathering of Scientific and Regulatory Experts

Register Today



Risk Assessment

Toxicity Prediction Using Target, Interactome and Pathway Profiles as Descriptors

Barbara Füzi, Neann Mathai, Johannes Kirchmair, Gerhard F Ecker. *Toxicol Lett.* 2023 Apr 13;381:20-26. doi: 10.1016/j.toxlet.2023.04.005. [Article link](#)

Significance: In choosing a suitable method for toxicity prediction, the available data and desired toxicity endpoint are two essential factors to consider.

In silico methods are essential to the safety evaluation of chemicals. Computational risk assessment offers several approaches, with data science and knowledge-based methods becoming an increasingly important sub-group. One of the substantial attributes of data science is that it allows using existing data to find correlations, build strong hypotheses, and create new, valuable knowledge that may help to reduce the number of resource intensive experiments. In choosing a suitable method for toxicity prediction, the available data and desired toxicity endpoint are two essential factors to consider. The complexity of the endpoint can impact the success rate of the *in silico* models. For highly complex endpoints such as hepatotoxicity, it can be beneficial to decipher the toxic event from a more systemic point of view. We propose a data science-based modelling pipeline that uses compounds' connections to tissue-specific biological targets, interactome, and biological pathways as descriptors of compounds. Models trained on different combinations of the collected, compound-target, compound-interactor, and compound-pathway profiles, were used to predict the hepatotoxicity of drug-like compounds. Several tree-based models were trained, utilizing separate and combined target, interactome and pathway level variables. The model using combined descriptors of all levels and the random forest algorithm was further optimized. Descriptor importance for model performance was addressed and examined for a biological explanation to define which targets or pathways can have a crucial role in toxicity. Descriptors connected to cytochromes P450 enzymes, heme degradation and biological oxidation received high weights. Furthermore, the involvement of other, less discussed processes in connection with toxicity, such as the involvement of RHO GTPase effectors in hepatotoxicity, were marked as fundamental. The optimized combined model using only the selected descriptors yielded the best performance with an accuracy of 0.766. The same dataset using classical Morgan fingerprints for compound representation yielded models with similar performance measures, as well as the combination of systems biology-based descriptors and Morgan fingerprints. Consequently, adding the structural information of compounds did not enhance the predictive value of the models. The developed systems biology-based pipeline comprises a valuable tool in predicting toxicity, while providing novel insights about the possible mechanisms of the unwanted events.



Foodborne Pathogens

Formation of *Listeria monocytogenes* Persister Cells in the Produce-Processing Environment

Xiran Li, Xavier F Hospital, Eva Hierro, Manuela Fernández, Lina Sheng, Luxin Wang. *Int J Food Microbiol.* 2023 Apr 2;390:110106. doi: 10.1016/j.ijfoodmicro.2023.110106. [Article link](#)

Significance: Chlorine treatment could be a viable sanitation technique to reduce the prevalence of *Listeria* persister cells.

Persisters are a subpopulation of growth-arrested cells that possess transient tolerance to lethal doses of antibiotics and can revert to an active state under the right conditions. Persister cells are considered as a public health concern. This study examined the formation of persisters by *Listeria monocytogenes* (LM) in an environment simulating a processing plant for leafy green production. Three LM strains isolated from California produce-processing plants and packinghouses with the strongest adherence abilities were used for this study. The impact of the cells' physiological status, density, and nutrient availability on the formation of persisters was also determined. Gentamicin at a dose of 100 mg/L was used for the isolation and screening of LM persisters. Results showed that the physiological status differences brought by culture preparation methods (plate-grown vs. broth-grown) did not impact persister formation ($P > 0.05$). Instead, higher persister ratios were found when cell density increased ($P < 0.05$). The formation of LM persister cells under simulated packinghouse conditions was tested by artificially inoculating stainless steel coupons with LM suspending in media with decreasing nutrient levels: brain heart infusion broth (1366 mg/L O₂), produce-washing water with various organic loads (1332 mg/L O₂ and 652 mg/L O₂, respectively), as well as sterile Milli-Q water. LM survived in all suspensions at 4 °C with 85 % relative humidity for 7 days, with strain 483 producing the most persister cells (4.36 ± 0.294 Log CFU/coupon) on average. Although persister cell levels of LM 480 and 485 were reasonably steady in nutrient-rich media (i.e., BHI and HCOD), they declined in nutrient-poor media (i.e., LCOD and sterile Milli-Q water) over time. Persister populations decreased along with total viable cells, demonstrating the impact of available nutrients on the formation of persisters. The chlorine sensitivity of LM persister cells was evaluated and compared with regular LM cells. Results showed that, despite their increased tolerance to the antibiotic gentamicin, LM persisters were more susceptible to chlorine treatments (100 mg/L for 2 min) than regular cells.

Foodborne Illness

An Overview of Foodborne Sample-Initiated Retrospective Outbreak Investigations and Inter-agency Collaboration in the United States

Allison Wellman, Michael C Bazaco, Tyann Blessington, Arthur Pightling, Asha Dwarka, Leslie Hintz, Matthew E Wise, et. al. *J Food Prot.* 2023 Apr 5;86(6):100089. doi: 10.1016/j.jfp.2023.100089. [Article link](#)

Significance: The ability to share whole genome sequencing subtyping data of foodborne pathogens can enhance food safety efforts by identifying links between illnesses and their potential sources earlier.

Foodborne outbreak investigations have traditionally included the detection of a cluster of illnesses first, followed by an epidemiologic investigation to identify a food of interest. The increasing use of whole genome sequencing (WGS) subtyping technology for clinical, environmental, and food isolates of foodborne pathogens, and the ability to share and compare the data on public platforms, present new opportunities to identify earlier links between illnesses and their potential sources. We describe a process called sample-initiated retrospective outbreak investigations (SIROIs) used by federal public health and regulatory partners in the United States. SIROIs begin with an evaluation of the genomic similarity between bacterial isolates recovered from food or environmental samples and clusters of clinical isolates while subsequent and parallel epi-demiologic and traceback investigations are initiated to corroborate their connection. SIROIs allow for earlier hypothesis generation, followed by targeted collection of information about food exposures and the foods and manufacturer of interest, to confirm a link between the illnesses and their source. This often leads to earlier action that could reduce the breadth and burden of foodborne illness outbreaks. We describe two case studies of recent SIROIs and present the benefits and challenges. Benefits include insight into foodborne illness attribution, international collaboration, and opportunities for enhanced food safety efforts in the food industry. Challenges include resource intensiveness, variability of epidemiologic and traceback data, and an increasingly complex food supply chain. SIROIs are valuable in identifying connections among small numbers of illnesses that may span significant time periods; detecting early signals for larger outbreaks or food safety issues associated with manufacturers; improving our understanding of the scope of contamination of foods; and identifying novel pathogen/commodity pairs.

Mycotoxins

Mycotoxins: Emerging Toxic Mechanisms and Unanswered Research Questions

Qinghua Wu, Li You, Wenda Wu, Miao Long, Kamil Kuca. *Food Chem Toxicol.* 2023 Apr;174:113673. doi: 10.1016/j.fct.2023.113673. [Article link](#)

Significance: Key scientific questions need to be answered on the toxic mechanisms of mycotoxins including a possible link to human neurodegenerative diseases.

Recently, a series of toxic mechanisms have been explored in mycotoxins. Emerging evidence show that mycotoxins may induce human neurodegenerative diseases (ND); however, this idea is still unproven. Besides identifying this hypothesis, some questions, for example, how the mycotoxins induce this disease and what the molecular mechanism is, as well as whether the brain-gut axis is involved in this context, should be answered. Very recent studies further reported an “immune evasion” mechanism in trichothecenes; moreover, hypoxia seems to play important function in this process; nevertheless, whether this “immune evasion” process is present in other mycotoxins, especially in aflatoxins, should be tested. In this work, we mainly discussed some key scientific questions that need to be answered in the toxic mechanisms of mycotoxins. We especially focused on the research questions in the key signaling pathways, balance mechanism of immunostimulatory and immunosuppressive effects, and the relationship between autophagy and apoptosis. Interesting topics such as mycotoxins and aging, cytoskeleton and immunotoxicity are also discussed.

Heavy Metals

Cognitive Outcomes Caused by Low-Level Lead, Cadmium and Mercury Mixture Exposure at Distinct Phases of Brain Development

Lu Ouyang, Qi Li, Shaoqi Rao, Rui Su, Yanhui Zhu, Guihua Du, Jie Xie, Fankun Zhou et. al. *Food Chem Toxicol.* 2023 May;175:113707. doi: 10.1016/j.fct.2023.113707. [Article link](#)

Significance: The effects of lead, cadmium and methylmercury on cognition vary depending on the stage of brain development.

Contaminated water and food are the main sources of lead, cadmium, and mercury in the human body. Long-term and low-level ingestion of these toxic heavy metals may affect brain development and cognition. However, the neurotoxic effects of exposure to lead, cadmium, and mercury mixture (Pb + Cd + Hg) at different stages of brain development are rarely elucidated. In this study, different doses of low-level Pb + Cd + Hg were administered to Sprague-Dawley rats via drinking water during the critical stage of brain development, late stage, and after maturation, respectively. Our findings showed that Pb + Cd + Hg exposure decreased the density of memory- and learning-related dendritic spines in the hippocampus during the critical period of brain development, resulting in hippocampus-dependent spatial memory deficits. Only the density of learning-related dendritic spines was reduced during the late phase of brain development and a higher-dose of Pb + Cd + Hg exposure was required, which led to hippocampus-independent spatial memory abnormalities. Exposure to Pb + Cd + Hg after brain maturation revealed no significant change in dendritic spines or cognitive function. Further molecular analysis indicated that morphological and functional changes caused by Pb + Cd + Hg exposure during the critical phase were associated with PSD95 and GluA1 dysregulation. Collectively, the effects of Pb + Cd + Hg on cognition varied depending on the brain development stages.

Food Packaging

Antimicrobial Properties of Poly (vinyl alcohol Films with Zeolitic Imidazolate Framework (ZIF-8) Nanoparticles for Food Packaging

Yutaro Hara, Maria Elena Castell-Perez, Rosana G Moreira. *J Food Sci.* 2023 Apr 17. doi: 10.1111/1750-3841.16572. [Article link](#)

Significance: This study demonstrates the efficacy of biopolymeric films embedded with nanoparticles to entrap natural antimicrobials and inhibit the growth of *Escherichia coli*.

Antimicrobial packaging films are of interest to the fresh produce industry due to the high number of foodborne illness outbreaks associated with these products. This study evaluated the antimicrobial effectiveness of poly (vinyl alcohol) (PVA) films with embedded zeolitic imidazolate framework (ZIF-8) nanoparticles carrying trans-cinnamaldehyde (TC)

against *Escherichia coli* MG1655 in spinach leaves. PVA films were synthesized using distilled water and mixed at 90°C for 1 h. The synthesized nanoparticles were introduced at various mass ratio concentrations (0%-5% weight of ZIF-8@TC nanoparticles to PVA), namely PVA-Z8-0 (control) through PVA-Z8-5. The PVA and ZIF-8@TC solution was mixed for 24 h until it seemed homogenous, cast, and dried in a ventilated oven at 35°C for 24 h. The release rate of TC from the PVA/ZIF-8@TC into both ethanol and methanol was characterized using HPLC methods. Disk diffusion and growth studies were performed to quantify the films antimicrobial effectiveness. Disk diffusion test showed that antimicrobial activity against *E. coli* MG1655 increased ($p < 0.05$) with increased nanoparticles concentration. Growth characteristics were described by the Baranyi model with some variations. About 0.26, 0.73, and 1.65 log reductions were achieved with the PVA-Z8-1 to PVA-Z8-3 films, respectively, while total inactivation was achieved with both the PVA-Z8-4 and PVA-Z8-5 films ($p < 0.05$). Similarly, the Baranyi model described the inhibitory profiles of the different films. This study contributes to the overall food safety body of knowledge regarding fresh produce and other packaged foods through the development of biopolymeric films with embedded nanoparticles to entrap natural antimicrobials.

Chemical Contaminants

In Vitro and In Silico Assessment of Endocrine Disrupting Effects of Food Contaminants through Pregnane X Receptor

Yuan Liang, Qiuyan Jiang, Yiyao Gong, Yifan Yu, Haoyang Zou, Jingqi Zhao, Tiehua Zhang, et. al. *Food Chem Toxicol.* 2023 May;175:113711. doi: 10.1016/j.fct.2023.113711. [Article link](#)

Significance: Food contaminants may have endocrine disrupting effects through specific receptors.

As a promiscuous xenobiotic receptor, pregnane X receptor (PXR) has been confirmed to participate in numerous physiological process. In addition to the conventional estrogen/androgen receptor, PXR also serves as an alternative target for environmental chemical contaminants. In this work, the PXR-mediated endocrine disrupting effects of typical food contaminants were explored. Firstly, the time-resolved fluorescence resonance energy transfer assays confirmed the PXR binding affinities of 2,2',4,4',5,5'-hexachlorobiphenyl, bis(2-ethylhexyl) phthalate, dibutyl phthalate, chlorpyrifos, bisphenol A, and zearalenone, with IC₅₀ values ranging from 1.88 to 4284.00 nM. Then their PXR agonist activities were assessed by PXR-mediated CYP3A4 reporter gene assays. Subsequently, the regulation of gene expressions of PXR and its targets CYP3A4, UGT1A1, and MDR1 by these compounds was further investigated. Intriguingly, all the tested compounds interfered with these gene expressions, confirming their endocrine disrupting effects via PXR-mediated signaling. The compound-PXR-LBD binding interactions were explored by molecular docking and molecular dynamics simulations to unravel the structural basis of their PXR binding capacities. The weak intermolecular interactions are key players in stabilizing these compound-PXR-LBD complexes. During the simulation process, 2,2',4,4',5,5'-hexachlorobiphenyl remained stable while the other 5 compounds underwent relatively severe disturbances. In conclusion, these food contaminants might exhibit endocrine disrupting effects via PXR.

Caffeine

Genetics of Caffeine and Brain-Related Outcomes - A Systematic Review of Observational Studies and Randomized Trials

Angeliki Kapellou, Alexandra King, Catherine A M Graham, Leta Pilic, Yiannis Mavrommatis. *Nutr Rev.* 2023 Apr 8;nuado29. doi: 10.1093/nutrit/nuado29. [Article link](#)

Significance: This review provides evidence that variability in the CYP1A2 and the ADORA2A genes may modulate the association between caffeine and brain-related outcomes.

Context: Although the stimulant and anxiogenic properties of caffeine are widely accepted, research on its specific effects on the brain remains controversial. Growing evidence shows that interindividual differences in caffeine response may be partly due to variations in genes such as CYP1A2 and ADORA2A, which have been used to identify individuals as “fast” or “slow” caffeine metabolizers and as having a “high” or “low” caffeine sensitivity, respectively. Objective: The objective of this review was to identify, evaluate, and discuss current evidence on the associations between common genetic variants, caffeine consumption, and brain-related outcomes in humans. Data sources: PubMed and Embase databases were searched for relevant reports based on a predetermined search strategy. Data extraction: Reports of observational and experimental studies on healthy adults who underwent (a) genetic analysis for polymorphisms in genes associated with caffeine metabolism and effects and (b) measurements of brain-related effects such as anxiety, insomnia, and cognitive performance associated with the consumption of caffeine (habitual intake or supplementation) were included. Data analysis: Of the 22 records included, 15 were randomized controlled trials, 6 were cross-sectional studies, and 1 was a genome-wide association study. The main outcomes identified were cognitive performance (n = 9), anxiety (n = 7), and sleep disturbance/insomnia (n = 6). Polymorphisms in the CYP1A2 gene were associated with cognitive function, while variations in the ADORA2A gene were associated with anxiety and sleep disturbance. Conclusion: The present

review has provided evidence that variability in the CYP1A2 and the ADORA2A genes may modulate the association between caffeine and brain-related outcomes. Future studies are warranted to investigate the specific polymorphisms implicated in each brain outcome, which cognitive functions are particularly related to caffeine (simple vs complex), whether there are gender differences in anxiety effects, and how habitual caffeine intake may influence the acute effects of caffeine.

Food Allergens

Epidermal Differentiation Complex Genetic Variation in Atopic Dermatitis and Peanut Allergy

Michelle F Huffaker, Kanika Kanchan, Henry T Bahnson, Ingo Ruczinski, Gautam Shankar, Donald Y M Leung, Carolyn Baloh. *J Allergy Clin Immunol.* 2023 Apr;151(4):1137-1142.e4. doi: 10.1016/j.jaci.2022.11.008. [Article link](#)

Significance: Variations in a specific gene play a role in the severity of atopic dermatitis and the development of a peanut allergy.

Background: Deleterious variation in the epidermal differentiation complex (EDC) on chromosome 1 is a well-known genetic determinant of atopic dermatitis (AD) and has been associated with risk of peanut allergy (PA) in population-based studies. **Objective:** Our aim was to determine the effect of genetic variation in the EDC on AD trajectory and risk of PA in early life. **Methods:** Genome sequencing was used to measure genetic variation in the EDC in the Learning Early about Peanut Allergy (LEAP) study participants. Association tests were done to identify gene- and variant-level predicted deleterious variation associated with AD severity by using the Scoring Atopic Dermatitis (SCORAD) tool (n = 559) at baseline and each follow-up visit, as well as PA and food allergy in peanut avoiders (n = 275). Predicted deleterious variants included missense variants that were frameshift insertions, frameshift deletions, stop-gain mutations, or stop-loss mutations. Associations between variant load, SCORAD score, and PA were tested by using linear and generalized linear regression models. **Results:** The genes FLG, FLG2, HRNR, and TCHH1 harbored the most predicted deleterious variation (30, 6, 3, and 1 variant, respectively). FLG variants were associated with SCORAD score at all time points; 4 variants (R1798X, R501X, S126X, and S761fs) drove the association with SCORAD score at each time point, and higher variant load was associated with greater AD severity over time. There was an association between these variants and PA, which remained significant independent of baseline AD severity (odds ratio = 2.63 [95% CI = 1.11-6.01] [P = .02]). **Conclusions:** Variation in FLG predicted to be deleterious is associated with AD severity at baseline and longitudinally and has an association with PA independent of baseline severity.

Recalls Associated with Food Allergens and Gluten in FDA-Regulated Foods from Fiscal Years 2013 to 2019

Girdhari M Sharma, Yinqing Ma, Stefano Luccioli. *J Food Prot.* 2023 Apr;86(4):100069. doi: 10.1016/j.jfp.2023.100069. [Article link](#)

Significance: A majority of major food allergen recalls involved one allergen. Milk, soy and tree nuts were the most common.

Allergens are one of the leading causes of food recalls in the US. The Food and Drug Administration (FDA) enforces requirements relating to major food allergens (MFAs) and gluten-free labeling to ensure food safety for allergic and celiac patients, respectively. Violative foods are subject to recalls. In this study, recall data for FDA-regulated foods were analyzed for fiscal years (FYs) 2013-2019 to identify trends and root causes associated with 1471 food allergen and gluten recalls. Of the 1471 recalls, 1415 recalls were due to MFAs, 34 recalls were due to gluten-free labeling violation and 23 recalls involved other allergens. Recalls due to MFAs overall increased during the study period with a peak incidence in FY 2017. MFA recall health hazard classifications were assessed as Class I (51.2%), Class II (45.5%), and Class III (3.3%). A majority of MFA recalls involved one allergen (78.8%). Milk was the most common MFA involved in MFA recalls (37.5%), followed by soy (22.5%) and tree nuts (21.6%). Almond, anchovy, and shrimp were the most common allergens recalled within the MFA groups of tree nuts, fish, and Crustacean shellfish, respectively. About 97% of MFA recalls involved one product category and among them, the category of 'bakery products, dough, bakery mixes and icings' ranked first (367 recalls), followed by the category of 'chocolate and cocoa products' (120 recalls). Labeling-associated errors accounted for 71.1% of MFA recalls with known root causes (914 out of 1286). It is important for the industry to develop and implement appropriate allergen controls to reduce the number of MFA recalls.

Emerging Science Areas

Emerging Food Safety Innovation

Electrochemical Degradation of PFOA and its Common Alternatives: Assessment of Key Parameters, Roles of Active Species and Transformation Pathway

Fatemeh Asadi Zeidabadi, Ehsan Banayan Esfahani, Sean T. McBeath, Kristian L. Dubrawski, Madjid Mohseni. *Chemosphere*. Vol. 315, Feb. doi.org/10.1016/j.chemosphere.2023.137743. [Article link](#)

Significance: An electrochemical treatment approach guided by a composite central design was used to study variable conditions impacting degradability of different structures of PFAS in polluted drinking water. Results from CCD indicated the importance of mass transfer related to stirring speed, charge amount passing through the electrodes and solution conditions.

This study investigates an electrochemical approach for the treatment of water polluted with per- and poly-fluoroalkyl substances (PFAS), looking at the impact of different variables, contributions from generated radicals, and degradability of different structures of PFAS. Results obtained from a central composite design (CCD) showed the importance of mass transfer, related to the stirring speed, and the amount of charge passed through the electrodes, related to the current density on decomposition rate of PFOA. The CCD informed optimized operating conditions which we then used to study the impact of solution conditions. Acidic condition, high temperature, and low initial concentration of PFOA accelerated the degradation kinetic, while DO had a negligible effect. The impact of electrolyte concentration depended on the initial concentration of PFOA. At low initial PFOA dosage (0.2 mg L^{-1}), the rate constant increased considerably from 0.079 ± 0.001 to $0.259 \pm 0.019 \text{ min}^{-1}$ when sulfate increased from 0.1% to 10%, likely due to the production of $\text{SO}_4^{\bullet-}$. However, at higher initial PFOA dosage (20 mg L^{-1}), the rate constant decreased slightly from 0.019 ± 0.001 to $0.015 \pm 0.000 \text{ min}^{-1}$, possibly due to the occupation of active anode sites by excess amount of sulfate. $\text{SO}_4^{\bullet-}$ and $\bullet\text{OH}$ played important roles in decomposition and defluorination of PFOA, respectively. PFOA oxidation was initiated by one electron transfer to the anode or $\text{SO}_4^{\bullet-}$, undergoing Kolbe decarboxylation where yielded perfluoroalkyl radical followed three reaction pathways with $\bullet\text{OH}$, O_2 and/or H_2O . PFAS electrooxidation depended on the chemical structures where the decomposition rate constants (min^{-1}) were in the order of 6:2 FTCA (0.031) > PFOA (0.019) > GenX (0.013) > PFBA (0.008). PFBA with a shorter chain length and GenX with $-\text{CF}_3$ branching had slower decomposition than PFOA. While presence of C-H bonds makes 6:2 FTCA susceptible to the attack of $\bullet\text{OH}$ accelerating its decomposition kinetic. Conducting experiments in mixed solution of all studied PFAS and in natural water showed that the co-presence of PFAS and other water constituents (organic and inorganic matters) had adverse effects on PFAS decomposition efficiency.

Engage with IAFNS

Metascience 2023 Conference

May 9, 2023 - May 10, 2023, Washington, DC

The scientific community is making a conscious shift towards transparency in science and is seeking prescriptive ideas on how the research process can be improved. The Metascience 2023 Conference will bring together researchers and stakeholders to share ideas to make meaningful change to the practice of research and foster a culture of continuous experimentation and improvement. [Read More.](#)

Food Classification Workshop

May 16 - 17, 2023, Washington DC

Considerations for formulation and degree of processing in food classification systems that support research. [Read More.](#)

IAFNS 2023 Annual Summer Science Symposium

June 13 – June 14, 2023, at the National Press Club, Washington, DC.

A gathering of scientific and regulatory experts to advance food and nutrition sciences in support of positive change.

[Read More.](#)