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The use of riparian buffer zones to reduce the risk of salmonella transmission from animal operations to fresh produce

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Due to the recent outbreaks of Salmonella spp. in fresh produce in the United States, it is apparent that the transfer of foodborne pathogens between animal feeding operations (AFO) and fresh produce continues to be a considerable risk.

● **Purpose:**
The purpose of this study is to determine if the establishment of a riparian buffer zone (RBZ) by AFOs could prevent the transfer of Salmonella spp. to nearby fresh produce fields in sustainable farming systems.

● **Methods:**
A five-layer RBZ (15 by 30m) consisting of hardwood trees, two rows of evergreen trees and shrubs, a non-manicured grass strip, and a row of pollinator plants was planted between produce fields and dairy or poultry houses.

Samples were collected from manure, air, soil, and fresh produce (romaine lettuce and tomato) sources for a period of 10 months.

Four replicates of soil and fresh produce samples were taken from plots located 32ft, 200ft, and 400ft away from cattle/poultry AFOs. Air and vegetative strip samples were sampled at 15 day intervals.

● **Results:**
A total of 37 presumptive Salmonella spp. isolates were retrieved from the soil, produce, air, and manure samples from both dairy and poultry farms.

Interestingly, 59.5% of presumptive salmonella isolates were from soil samples (n=22).

The bulk of isolates were collected during the spring/winter months (n=25). Only three isolates were recovered from lettuce samples in the month of November. Salmonella isolates from air samples (n=6) were only recovered from the dairy farm at both the 200ft and 400ft plots during the summer months. Surprisingly, salmonella isolates (n=6) were only found in manure samples from dairy cattle

● **Significance:**
The effectiveness of the RBZ cannot yet be determined. However, it seems that distance and air flow does have an effect on the transmission of salmonella. ■

Bio-based sanitiser delivery systems for improved sanitation of bacterial and fungal biofilms

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Biofilms can persist in food processing environments due to their relatively higher tolerance and resistance to antimicrobials including sanitisers.

● **Purpose:**
In this study, a novel bio-based sanitiser composition was developed to effectively target biofilms and deliver chlorine-based sanitisers to inactivate bacterial and fungal biofilms.

● **Methods:**
The bio-based composition was developed by encapsulating chlorine-binding polymer in a bio-based yeast cell wall particle (YCWP) microcarrier.

The effect of this sanitiser composition on the inactivation of bacterial and fungal cells in simulated wash water and inhibition of biofilm formation was evaluated in this study.

● **Results:**
This study demonstrates the high affinity of bio-based compositions to bind target bacterial and fungal cells and inactivate five log of model pathogenic bacteria and fungi in wash water with and without high organic load (COD=2000mgl) in 30 seconds and five minutes, respectively.

For the sanitation of biofilms, this bio-based sanitiser can inactivate seven log of pathogenic bacteria and three log of fungi after one hour of treatment, while the one-hour treatment using conventional chlorine-based sanitiser can only achieve two to three log reduction for bacterial biofilms and one to two log reduction for fungal biofilms, respectively.

The enhanced antimicrobial activity can be attributed to three factors:

● Localised high concentration of chlorine bound on the YCWPs.
● High affinity of YCWPs to bind diverse microbes.
● Improved stability in an organic-rich aqueous environment.

● **Significance:**
In summary, these unique attributes of bio-based carriers will significantly enhance the sanitation efficacy for biofilms, reduce persistence and transmission of antimicrobial resistance microbes, limit the use of antimicrobial chemicals, and improve the cost-effectiveness of sanitisers. ■

Effects of interventions on indicator organism levels in beef slaughter

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Beef slaughter establishments employ many different interventions to help minimise the incidence of pathogens in their products.

● **Purpose:**
This study explored the relative effectiveness of various common interventions using the answers to questions that FSIS asked as part of the Beef Veal Carcass Baseline Study (2014 to 2015).

● **Methods:**
FSIS collected 2,736 swab samples from carcases at 179 establishments. These included 1,368 samples at post-hide removal (before evisceration) and 1,368 at pre-chill (after evisceration). Samples were tested for pathogens (salmonella and STEC) and indicators (generic E. coli, enterobacteriaceae, coliform, and aerobic count) using FSIS-approved methods.

● **Results:**
Pathogen positive rates were too low to establish a direct correlation between interventions and pathogens. However, all pathogens correlated with all indicators, suggesting the use of indicators as surrogates.

For example, salmonella gave an odds ratio indicating a 57% increase in pathogen prevalence for each log increase in aerobic count.

Therefore, we compared indicator levels to assess the effectiveness of interventions. Some interventions, such as chlorine wash, correlated with decreased indicator levels, while others, such as trimming alone, correlated with an increase.

Most comparisons gave significant trends via ANOVA ($P<0.005$), and many pairwise correlations were also significant ($P<0.05$). However, each of the tested intervention strategies generated a wide range of indicator levels.

● **Significance:**
This suggests that how interventions are applied may be as important as which interventions are applied. Although indicator organisms do not provide a comprehensive picture of process control, the correlation between indicators and pathogens provides useful information. Thus, our results can be used by slaughter establishments to help identify the most effective interventions for pathogen reduction. ■

Using predictive pre-processing risk scores to reduce foodborne disease

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Most food processing plants operate statically: pathogen mitigation interventions are run at the same parameters regardless of the risk posed by incoming products. Often, pre-processing pathogen testing alone is insufficient to differentiate risk. However, combinations of pre-processing testing, historical performance data, and location-based metadata can be used in machine learning (ML) algorithms to generate accurate pre-processing risk scores.

● **Purpose:**
To reduce the risk of foodborne pathogen transmission by providing risk scores that allow producers to differentiate pathogen risk in incoming products.

● **Methods:**
We develop a suite of ML algorithms that takes available pre-processing data including pathogen tests, historical performance data from growing locations, and location-based metadata, and generates a predictive risk score identifying which product batches are most likely to have positive pathogen tests on a final product.

Risk scores can be used to aid decision-making in processing plants – whether to alter intervention parameters, divert certain product batches (e.g. from raw to cooked), or other available techniques depending on the product, with the goal of reducing the final pathogen prevalence and ultimately foodborne disease.

● **Results:**
Utilising data from over 2,000 commercial broiler chicken placements, and combining live operations salmonella results, house equipment (e.g. types of heaters, feeders, drinkers, etc), and early health metrics with processing plant final product tests, we employ an ensemble ML algorithm to assign risk scores based on processing plant outcomes. We test the algorithm on 20% of the data that was initially set aside; it was able to identify with 80% accuracy which days the processing plant would have a positive salmonella test in the outgoing product.

● **Significance:**
Predictive algorithms can accurately assign risk scores prior to processing, allowing for producers to make intervention-based and diversionary decisions that reduce the prevalence of pathogens on the outgoing product, lowering the public risk of foodborne disease. ■

Influence of the germination time on aflatoxins during malting of wheat for use in craft beer

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Mycotoxins are fungal secondary metabolites with harmful effects on humans and animals. They can be produced in contaminated grains if toxigenic fungi are present and conditions are favourable.

When wheat is malted for use in craft beer, grains are immersed in water and kept moist for extended times before kilning. The ability of fungi to produce mycotoxins during wheat malting is unknown.

● **Purpose:**
This study evaluates the production of aflatoxin B1 (AFB1) and aflatoxin B2 (AFB2) during wheat malting under typical conditions used by craft beer producers.

● **Methods:**
Malting was performed according to Central European Commission for Brewing Analysis procedures. Steeping processes include extended periods of alternating water immersion and aeration.

Aflatoxin free, toxigenic fungi-free wheat grains (600g) were inoculated in the first water immersion step by immersing in a suspension of Aspergillus flavus (WG06-A, five log spores/ml), known to produce AFB1 and AFB2 originally isolated from wheat grains.

Grains were germinated at 15°C for 48, 72, 96 and 120 hours. Germinated grains were kilned (16 hours at 50°C, one hour at 60°C, one hour at 70°C and five hours at 80°C) and the rootlets removed.

Duplicate samples of wheat malt obtained from each time were collected for determination of AFB1 and AFB2 (dry basis) by HPLC (detection limit 0.03µg/kg AFB1; 0.02µg/kg AFB2; quantitation limit 0.09µg/kg AFB1; 0.07µg/kg AFB2).

● **Results:**
The highest levels of both AFB1 (160.07±0.99µg/kg) and AFB2 (4.52±0.21µg/kg) were detected in grains germinated for 96 hours.

The lowest levels of AFB1 (103.67±11.97µg/kg) and AFB2 (0.48±0.21µg/kg) were detected in grains germinated for 48 hours.

● **Significance:**
Malting germination time influences the amounts of AFB1 and AFB2 produced in the wheat malt, and the levels of aflatoxin produced exceed Codex Alimentarius recommendations for a variety of foods.

This should be considered by craft beer manufacturers using malted wheat. ■

The faecal resistome of dairy cattle is associated with diet during nursing and weaning

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Antibiotic-resistant bacteria display an age-dependent distribution in dairy cattle, in which younger calves harbour a higher abundance of resistance. The mechanism responsible for this resistome distribution is yet unclear.

● **Purpose:**
The study aimed to examine the hypothesis that the assembly of gut microbiome of dairy calves promotes the establishment of bovine-associated antimicrobial resistance genes (ARGs) and is driven, in part, by diet.

● **Methods:**
Fresh faeces (n=484) were obtained from 22 dairy calves, from at birth to week 10 between April and September of 2015 at UC Davis Dairy Facility.

Two colostrum samples were also collected per calf prior to feeding (n=44).

DNAs were extracted from all samples for 16S-rRNA sequencing. In addition, selected faecal samples (n=12) from dairy calves (n=3) at four time-points per calf (day two, day five, week three and week seven), as well as the corresponding colostrum samples (n=6; two of each calf), were subjected to shotgun metagenomic sequencing.

● **Results:**
The intestinal microbiome and resistome assembled rapidly in newborn calves. A total of 329 ARGs conferring resistance to 17 classes of antibiotics were observed in dairy calves. The abundance of total ARGs declined markedly during nursing, however, some clinically-relevant ARGs encoding resistance to macrolides-lincosamides-streptogramins and tetracyclines increased throughout this period. ARGs in dairy calves was found to co-occur with antibacterial biocide/metal resistance genes. Colostrum was predicted to be the source of over 90% of ARGs observed in dairy calves on day two. The early succession of resistome in dairy calves is a result of gut microbiome assembly associated with the dietary transition, in particular, from colostrum to milk replacer and increased intake of calf starter.

● **Significance:**
The assembly of gut resistome in dairy cattle is associated with diet during early life. Identifying the drivers influencing resistome assembly may inspire targeted interventions of the gut microbiome through food modifications to reduce the overall prevalence of antibiotic resistance. ■