Developments and applications of food antimicrobials, cleaning and sanitation

Summary prepared by M. Ellin Doyle, PhD

FRI’s second Focus on Food Safety Series meeting was held on September 28-29, 2005 with a series of in-depth presentations on specific topics of interest to food producers and processors. Speakers from FRI, other universities, industry, and government discussed pathogens of concern, cleaning and decontamination of produce, animal carcasses, and surfaces in food processing plants, and commercial antimicrobials used in food production. Over 60 persons, representing 28 companies, attended the meeting.

**Pathogens of Concern**

*Bacteria* (Jean Schoeni, Trac Microbiology) Most of the bacteria commonly associated with foodborne illness, *Clostridium sp.*, *Bacillus cereus*, *Staphylococcus aureus*, *Listeria monocytogenes*, *Campylobacter sp.*, *E. coli* 0157:H7, *Salmonella sp.*, *Shigella sp.*, *Vibrio sp.*, and *Yersinia enterocolitica* are well known pathogens. However, some aspects of bacterial foodborne illness are changing: (a) Instead of localized clusters of cases, some outbreaks are diffuse and widespread due to distribution of contaminated commercial products. An example is the 1994 outbreak of *S. enteritidis* associated with ice cream that affected 224,000 people nationwide. (b) Some bacteria are appearing as contaminants in new or unexpected foods such as *E. coli* 0157: H7 contaminating cheese. An employee who took on a second job milking cows apparently brought this bacterium into the cheese plant. (c) Clinical strains of bacteria, for example methicillin-resistant *S. aureus*, have been detected in some foods. These bacteria apparently originated with an ill employee whose wife was a nurse. (d) Infectious bacteria normally present in an area may become more widely distributed following a disaster. This is exemplified by an increase in infections caused by *Vibrio sp.* following hurricane Katrina.

**Fungi** (Jaehyuk Yu, FRI) Fungal spores are ubiquitous—it has been estimated that we inhale 100-200 spores per minute. This can result in rapid contamination of foods exposed to air. Some foods may be contaminated in the field or during storage with *Fusarium*, *Aspergillus*, or *Penicillium* that produce mycotoxins and with spoilage fungi and yeasts that may damage foods. Factors affecting fungal growth include presence of oxygen, unbound water, nitrogen and other nutrients, and temperature (0-58°C). Strategies to control

Editor’s Note: One of the more egregious sins a Medical Editor can commit is to disappear out of town without ensuring there is appropriate material available for upcoming themed issues. This is the sin of omission that I committed when I went seeking sun and warmth during the winter months. The penalty on return is to find that there had been no manuscripts submitted dealing with food safety and that it was too late to impose on anyone to prepare an “instant” review of the current knowledge and dogma concerning food safety suitable for clinicians. The inquiries I made led me to a veritable gold mine of information: the Food Research Institute (FRI) located on the UW campus. I had been completely unaware of its existence and the interesting work they are doing and I pass on their Web site as a valuable resource: www.wisc.edu/fri. Click on “communications” and then “briefings.” The FRI Briefings sent me to the featured article, a July 2005 literature review on Bovine Spongiform Encephalopathy and Chronic Wasting Disease, the History of “Mad Cow Disease” in Great Britain, Foodborne Parasites (mainly Cryptosporidium), “Alternatives to Antibiotic Use for Growth Promotion in Animal Husbandry,” and more—all providing enough information to satisfy curiosity and with many references to delve into. Also of great interest were the links to other food safety Web sites—CDC, FDA, USDA, FNIC (Food and Nutrition Information Center), FSRIO (Food Safety Research Information Office)—which appealed to me as resources that could be useful in the face of a difficult digestive problem. In addition there are brief but interesting research reports on subjects as eclectic as the identification of the Botox receptor (with possible implications as a biological threat); allergic reactions to tree nuts (versus peanuts); a nutritional solution for cat hairballs (!) and others.

—Thomas C. Meyer, MD, Medical Editor
fungi include the use of organic acids, chelating compounds, modified atmospheres, and some naturally occurring compounds such as vanillin, chitosan, and limonene. New approaches are being developed to detect mold contamination using PCR, DNA chips, electronic nose, and biosensors, and new molecular technologies are also being developed to kill or inhibit growth of molds.

**Noroviruses**
*(Syed Sattar, Univ. Ottawa)*
These viruses are part of the calicivirus group and consist of single-stranded RNA and a protein coat with no lipid envelope. They are the most common enteric viral pathogens worldwide, causing millions of cases of chronic and acute diarrhea, with an infectious dose as low as 10-100 virus particles. As yet, these viruses cannot be cultured in the laboratory, resulting in limited information about efficacy of control strategies. Related caliciviruses survive on metal surfaces for at least a week and on hands longer than many bacteria. Domestic bleach can be used to disinfect surfaces. Washing hands in water can remove 90% of virus particles while washing in 60-80% ethanol results in a 95% reduction.

**Decontamination of Produce and Raw Meat**

**Fresh Produce**
*(Larry Beuchat, Univ. Georgia)*
Numerous outbreaks of foodborne disease have been traced to fresh fruits and vegetables contaminated with bacteria, viruses, and parasitic protozoa. Plants can be very difficult to clean because of irregular surfaces and waxes covering some structures. Pathogens may find safe havens in cracks in seeds, under trichomes (hairlike projections) on leaves, in lenticels (pores) on stems, in stem and blossom ends of fruits, and in areas where there has been tissue damage. Washing in water to remove dirt should be the first step but the wash water should be warmer than the food to prevent infiltration of microbes. Chlorine is widely used for sanitation and electrolyzed water and organic acids are useful for some foods. The efficacy of particular sanitizers and their effect on sensory qualities varies with different foods. Irradiation, UV light, high pressure, sonication, and packaging technologies are also being investigated to reduce pathogen numbers and improve shelf life.

**Animal Carcasses**
*(Kere Kemp, Ecolab)*
Cleaning of animal carcasses is accomplished using immersion systems or spray cabinets for whole carcasses or other spray systems for parts and trim. Among the most widely used chemical sanitizers are chlorine dioxide, acidified sodium chlorite, and peroxy acids. Trisodium phosphate (TSP) is extensively used in poultry operations and cetylpyridinium chloride is used in some plants. Several issues impact the choice of a sanitizer including the cleaning system used, worker safety, risk of residues and organoleptic changes in the meat, effects on the environment and waste stream, and, of course, regulatory approval.

**Antimicrobial Food Preservatives**

**Perspectives on Antimicrobial Food Preservatives**
*(Michael Davidson, Univ. Tennessee)*
The history of antimicrobial preservatives began very early with the use of salt, smoke and fermentation to preserve foods. Numerous antimicrobials have been developed with the goal of inhibiting or killing spoilage organisms and pathogens. These compounds vary in effectiveness with characteristics of foods such as pH and polarity and with other factors such as temperature and processing conditions. Recently, there has been a great deal of interest in natural antimicrobials such as spices (cinnamon, cloves, thyme, oregano) and animal products (lysozyme, lactoperoxidase, chitosan, lactoferrin). Other plant and microbial products are being investigated but a great deal of information on activity, structures, toxicology, sensory effects, and compatibility is needed before regulatory approval and applications in different foods.

**Commercial Antimicrobials**
*(George Weber, Danisco)*
Sales of antimicrobials for the food industry exceed $300 million yearly and over half of that is spent on sorbates. When choosing an antimicrobial, one should first define the goal: Is there a particular pathogen of concern, e.g. Listeria, or is shelf life (spoilage) of greater concern? Then, consider what is approved and legal (CFR21, FSIS directives), cost, and the food matrix. Available antimicrobial strategies include chemicals such as benzoate, peroxyacetate, organic acids, extracts, processes such as irradiation, high pressure, encapsulation, and packaging including different types of plastics and modified atmospheres.

**Mechanisms of Action of Natural Food Preservatives**
*(Michael Tchikindas, Rutgers Univ.)*
Consumers are demanding more fresh, minimally processed, and “natural” foods. This has stimulated research into naturally produced antimicrobials such as bacteriocins, lysozyme and lactoferrin. Nisin, produced by Lactococcus
lactis, is heat stable and kills vegetative cells and prevents spore outgrowth. It has been approved for use in cheese and, in some countries, is permitted in canned vegetables. It interacts with lipids in cell membranes, causing holes in the membranes. Nisin is often used as part of a multiple hurdle strategy, e.g. nisin plus cold storage plus modified atmosphere or nisin plus sublethal heat.

**Antimicrobial Properties of a Glucose By-Product (Charles Kaspar, FRI)**
Some current research is focused on “designer antimicrobials” that target key cell structures or processes and may include antisense RNA to inhibit toxin production and alternate sigma factors and other compounds that disrupt cell regulation. A furan-type compound has been produced by heating glucose or other reducing sugars in the presence of phosphate and was found to damage DNA. This compound is effective against Gram-positive and -negative bacteria and even has bactericidal effects against some spore formers. Studies are underway to further purify this compound and determine more specifically its activities.

**New Antimicrobials and FDA Regulations (Michael Pariza, FRI)**
Safety of new antimicrobials is determined by FDA by evaluating data on chemical nature of the active ingredient, data on the manufacture of the antimicrobial, a standard battery of toxicology tests including rodent assays, and all reports on adverse effects. GRAS status is reviewed by experts. FDA no longer “affirms” GRAS status but rather issues a “no objection” letter or else informs the petitioner that more information is required.

**Antimicrobials for Processed Ready-to-Eat Foods (Joseph Meyer, Kraft Oscar Mayer)**
The ideal antimicrobial for RTE foods should be robust (stable to heat, drying, enzymes and effective even after package is opened), consumer friendly (non-allergenic) and manufacturer friendly (reasonable cost, broad spectrum, no impact on organoleptic properties). RTE beverages often use sorbates and benzoates to retard spoilage; low pH usually inhibits pathogens. Cheese may also contain sorbates and benzoates and in addition, nisin and phosphate salts to inhibit pathogens. Packaged RTE meat is a great culture medium for both spoilage and pathogenic organisms. Nitrite, sodium chloride, lactate and diacetate are used to inhibit bacterial growth.

**Antimicrobials for Meat and Poultry Products—USDA Regulations (Amelia Sharar, FSIS)**
Antimicrobials approved for use in meat and poultry are listed in 21 CFR. An FSIS directive specifies the amounts of antimicrobials that can be used in particular foods and labeling requirements. Applications for new antimicrobials and processes and for new applications for approved antimicrobials are reviewed by FSIS New Technology Staff.

**Validation of Efficacy of Antimicrobials (Kathy Glass, FRI)**
In assessing efficacy of an antimicrobial, food components, processing and packaging conditions, sources of contamination and target organisms should be considered. A literature search can identify possible antimicrobials and their characteristics. Compounds of interest can be screened in laboratory media where the effects of some factors such as pH, fat, and salts can be tested. Further tests can be conducted in model food systems such as milk and meat suspensions, but ultimately the efficacy of an antimicrobial system should be validated using a challenge study in the target food substrate with an appropriate pathogen or surrogate and mimicking usual processing conditions.

**Cleaners and Sanitizers Why Clean and Sanitize? (Bruce Cords, Ecolab)**
Longer food distribution chains, increased at-risk populations, increasing amounts of fresh produce and numbers of in-store delicatessens in grocery stores, and the high percentage of meals eaten or purchased outside the home have made sanitizing of food preparation/storage areas even more important. Nevertheless, surveys show that sanitation is often inadequate, with contaminated surfaces detected at deli departments in retail stores and with poor personal hygiene in restaurants and nursing homes. Lack of sanitation may be a cause of foodborne illness and can also lead to insect problems—in one year, a pair of cockroaches can produce enough offspring to stretch for 7 miles!

**Fundamentals of Cleaning (Mike Coughlin, JohnsonDiversey)**
Important aspects of cleaning strategies can be summarized as TACT (Time, Action, Concentration, Temperature). Concentration of cleaners and times and temperatures used should be tailored for the types and quantities of soil present. Action of cleaning processes is also important with pressures, abrasion, and flow velocities important for dislodging soil, particularly for clean-in-place equipment. Cleaning challenges include...
WINS (Water quality, Individuals, Nature of soil, Surfaces). Hardness of water and presence of chloride and sulfate may impact efficacy of cleaners and their effects on equipment. Some types of soil, including biofilms, and some surfaces can be more difficult to clean. Adequate training and retraining of personnel in charge of the cleaning is also critical.

Advances in Sanitizers
(Kere Kemp, Ecolab)
Numerous compounds have been used for cleaning and sanitizing, including hypochlorites, iodines, quats, acid anionics, acid sanitizers, peracetates, and, more recently, peracid combinations and acidified sodium chlorite. Contact time and temperature, as well as pH and water quality, affect the efficacy of sanitizers and cleaners. In addition, some microbes are resistant to the actions of some compounds.

Verification of Efficacy
(Douglas Anderson, ATS Labs)
Claims of antimicrobial efficacy should be substantiated by use of published standard methods with reference microbes at specific exposures. Dilution tests and germicidal spray tests may be used to test efficacy of 30-second exposures of sanitizers against \textit{S. aureus} or against a combination of \textit{S. aureus}, \textit{Salmonella choleraesuis}, and \textit{Pseudomonas aeruginosa}. Accurate testing is necessary as part of EPA registration and for label claims and directions for use.

Regulatory Issues
(Paul Strege, JohnsonDiversey)
Sanitizers are regulated as pesticides under FIFRA and the Toxic Substances Control Act while antibacterial hand sanitizers are regulated as drugs by FDA. USDA certifies cleaners for meat and poultry plants. State and local regulations may also affect use of sanitizers and Canada and the European Union have their own regulations governing the use of these compounds.

Wm. C. Frazier Memorial Award and Lecture
Martin Cole, National Center for Food Safety and Technology, recipient of the 2006 Win. C. Frazier Memorial Award, delivered a lecture entitled “International Developments in Managing Microbial Food Safety.” A large fraction of world food production moves through trade to distant locations, and processed food trade is growing twice as fast as trade in raw foods. International sourcing leads to more complex food chains. Minimizing pathogens in the final product requires analysis and monitoring of numerous performance criteria and interventions to reduce hazards.
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