Novel Carcass Rinse Solutions

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ASL-R1609

Summary and Implications

An exploratory project ("Discovery" category, Iowa State University Food Safety Consortium) was initiated to evaluate different compounds. These compounds were intended to interfere with the initial stages of bacterial attachment. These compounds included mannose and a variety of different detergents. The rinse solutions were applied to pork muscle tissue prior to inoculation with strains of *Escherichia coli* and *Salmonella typhimurium*. Although there was some slight numerical reductions in attachment, these reductions were not statistically significant and did not exceed a 50% reduction.

Introduction:

Extensive research has been conducted to identify procedures for reduction of the microbial burden on animal carcasses. Many species of animal carcasses are exposed to wash or rinse solutions prior to evisceration (scalding in swine and poultry, preevisceration washing in beef), and this offers an opportunity to reduce the bacterial populations by applying antimicrobial compound (2). Several antimicrobial agents have been evaluated, including trisodium phosphate organic acids (3) and hot water (1).

Studies examining mechanisms of attachment of bacteria to meat surfaces have been extensive, but comparisons between methods and species are often difficult because of the physiochemical differences between meat surfaces. One aspect that has been explored is the role of fimbriae or pili (6). Many members of the family Enterobacteriaceae, including members of the genera *Salmonella, Escherichia, Shigella* and *Vibrio*, posses external cell structures termed common pili or type 1 fimbriae (5). These are composed of 127 to 21 kilodalton repeating subunits with minor proteins at the tip, or inserted periodically along the entire length. These proteins are termed adhesions, and these adhesions function to bind D-mannose residues on eucaryotic cells. This binding is instrumental in the initial attachment of cells to eucaryotic cells, and is often a first step in the invasion process.

Animal carcasses are currently rinsed at various stages in processing, including prior to evisceration (previsceration washing–beef; scalding–swine and poultry). It may be possible to influence the level of bacterial contamination on animal carcasses by altering the composition of these rinse solutions and interfering with bacterial attachment to the carcasses. If possible, this would result in carcasses with lower overall levels of bacterial contamination and potentially lower risk of containing bacterial pathogens. If successful, this process could potentially be applied at all levels of processing plants, from the very large to the very small. This exploratory project will determine the feasibility of the concept and provide the basis for an extramural grant application.

Given the function and significance of mannose receptors in initial attachment, a possible approach is to block the mannose receptors on pili with mannose containing solutions. If successful, this might interfere with the initial stages of bacterial attachment to animal tissue surfaces and facilitate the removal and destruction of contaminating bacteria on carcasses. Preliminary experiments have indicated that mannose treated meat contained 2.5 to 3 times fewer bacteria when inoculated in a similar manner to meat that was not treated with mannose (Grant, personal communication).

Materials and Methods:

Marker strains of naladixic acid-resistant *Salmonella typhimurium* and rifampicin-resistant *Escherichia coli* were grown in trypic soy broth under both aerobic and anaerobic conditions. Pork skin and muscle tissue were contaminated using modified versions of the skin or muscle attachment models that have been elaborated for multiple food animal species (pork, poultry, fish). Briefly, defined areas of skin or muscle tissue were subjected to low dose irradiation to eliminate most of the Gram negative microflora. These surfaces are then inoculated with the target microorganism by immersion for 1 minute by using either broth or buffer. The sessile and planktonic populations of both the target microorganism and the total aerobic populations were determined using selective and nonselective media. The total populations of these bacteria were determined, as well as the ratio of sessile to planktonic bacteria and sessile to total bacterial populations (the "SR" value described by Dickson and Crouse [4]) to establish baseline data for growth conditions and inoculum material.

The experiments were repeated, applying mannose containing rinse solutions (5% wt:vol) prior to inoculation. Briefly, the tissue samples were immersed in the rinse solution for 5 minutes, then immersed for 30 seconds in the inoculating bacterium. The tissue was removed from the inoculum, and then rinsed in 100-ml bottles of buffered peptone water by gently inverting the bottles 10 times within 15 seconds. The buffer was decanted, and bacteria remaining on the tissue were considered to be attached. The tissue samples were stomached for 2 minutes in buffered peptone water or alternately homogenized in a Waring Blendor for 30 seconds. The samples were serially diluted an spiral plated on tryptic soy agar and tryptic soy agar containing 200 ppm of naladixic acid (Salmonella) or 200 ppm of rifampicin (*E. coli*). The plates were incubated aerobically at 37^{o}C for 24 to 48 hours.

The experiments were repeated substituting a variety of cationic, anionic, and non-ionic detergents in place of the mannose-containing rinse solutions.

Results and Discussion:

Irrespective of the type of solution, a significant reduction in attached bacteria was not seen. At most, a threefold reduction was seen in bacteria attached to muscle tissue. Because this did not amount to more than a 0.5 $\log_{10}$ reduction in population, it was concluded that the solutions, as evaluated in these preliminary experiments, were of little use in reducing contamination of pork tissue.

References:

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