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Reports on Food Safety and Radiation Effect Projects

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BRUCE APPEGATE, PURDUE UNIVERSITY:

Pathogenic detection using phage display

Salmonella spp. bacteriophage: To facilitate the construction of a modified P22 bacteriophage for *Salmonella spp.*, it was necessary to construct a recombination vector for insertion of a modified tail spike protein. The recombination vector was constructed by removing approximately 1 kb of the P22 genome in the region of orf 201 and orf 80 to allow the insertion of DNA for recombination. A multicloning site and a TA cloning site were inserted to facilitate rapid insertion of modified DNA to construct the appropriate epitope. Primers were utilized to amplify the tailspike protein from P22 with the appropriate His modifications. The preparative host strain repressor gene cassette was constructed for insertion into the preparative host-strain genome. Work continues on inserting the modified tailspike gene in the previously constructed P22 recombination vector. Appropriate promoter and terminator configurations were added to the modified tailspike protein to repress expression of the His-modified tailspike protein in the preparative host strain. Preparative host strain containing the *lacI* repressor gene cassette was constructed.

E. coli O157:H7 bacteriophage:

The apparent host specificity of ϕ V10, its ability to lysogenically convert without transducing host genes, and its lack of identifiable virulence determinants suggest that ϕ V10 should be useful in biotechnology and that its use in biotechnology should not constitute any undue hazard, making it a good candidate for detection assays.

Listeria monocytogenes bacteriophage: An initial screening of a *Listeria monocytogenes* library to isolate a lysogenic bacteriophage producing preliminary results that suggest success and we are currently further evaluating the isolated bacteriophage and continuing to screen the library.

Packaging: The initial product includes a lateral-flow assay for the multiplexed determination of multiple pathogens. We have begun collaboration with Embedded Concepts, which has patented technology allowing the embedding of antibodies within plastics. This innovation greatly enhances the potential for integration of the phage-based assay into food packaging material without the need for lateral flow, thereby simplifying development of stand-alone kits involving sample preparation and detection integrated into one product.

LISA MAUER, PURDUE UNIVERSITY:

FOOD PROCESSING AND PACKAGING

Initial research efforts focused on 1) characterizing effects of space-relevant radiation doses of gamma-radiation on select foods, food ingredients, and dietary

supplements and identifying threshold radiation levels for quality changes; and 2) characterizing the impact of plant cultivars and growth conditions on nutrient profiles, antioxidant capacity, functionality, and acceptability of foods.

Novel Packaging, Processing and Effects of Radiation on Wheat, Fruits and Vegetables.

The intake of fresh food is very important to a crew's welfare, and thus its quality can have a tremendous impact on a crew's morale and the success of a long mission. Radiation affects the nutritional content, stability, and shelf life of foods. The objectives of this study were to 1) investigate the effects of low-dose γ -radiation and time on specific nutrients and quality traits of fresh apples, strawberries, carrots, and tomatoes 2) investigate the effects of low-dose γ -radiation and time on specific nutrients and quality traits of different cultivars of fresh carrots and tomatoes, and 3) investigate the effects of low-dose γ -radiation, time, and temperature on specific nutrients and quality traits of freeze-dried and air-dried apples, strawberries, carrots, and tomatoes. The fresh and processed fruits and vegetables were exposed to levels of γ -radiation ranging from 0Gy to 10000Gy (1Gy = 1Sv) using a Gammacell 220. The fresh fruits and vegetables were stored at 25°C for 1 and 3 days while the processed fruits and vegetables were stored at 25°C and 35°C for 6 months. After the different storage times, changes in the levels of specific macro- and micro-nutrients, quality traits, and sensory evaluations were done. For the fresh fruits and vegetables after day 1, macro-nutrients such as protein, carbohydrate, sugar, calcium, magnesium, sodium, and potassium were relatively constant upon exposure to radiation doses of up to 10kGy. At low radiation doses (1-5Gy) there was no significant effect on the antioxidant capacity, total phenolics, ascorbic acid (vitamin C), carotenoids (α , β , lycopene). However, at higher radiation doses (≥ 10 Gy), strawberries and tomatoes were more vulnerable than carrots and apples (i.e. breakdown of antioxidant capacity, total phenolics, ascorbic acid (vitamin C), and lycopene was observed). Rotting of the fruits and vegetables was observed after day 3.

Radiation Effects on Oils

Radiation can initiate a process known as autoxidation, in which an initiator species reacts with lipid, removes a hydrogen, and produces a lipid free radical. This lipid free radical can then react with other fatty acids and produce an accelerating chain reaction of similar events during propagation. Products of this oxidation of fatty acids include peroxides, alcohols, aldehydes, and carbonyls. Further reactions with these products lead to off-odors and off-flavors characteristic of rancid fats. Studies using soybean and peanut oils (with and without added antioxidant, irradiated at 0, 3, 10, 100, 200, 400, 600, 800, or 1000Gy, and stored at 65°C) found that addition of 0.02% TBHQ (antioxidant) reduces total oxidation and delays oxidation maxima from 7 months to 42 months for soybean oil and from 21 to 49 months for peanut oil.

Radiation Effects on Antioxidants

Radiation doses during transit and on the surface of Mars are expected to be as high as 3 sieverts (approx. equivalent to 3 Gy), excluding solar events. Studies have shown that antioxidants may provide long-term health protection from oxidative stress caused by radiation exposure; therefore, to counteract the impact of elevated radiation exposure on astronaut health, consumption of antioxidants will be important. In addition to vitamin supplements, antioxidants within foods also will provide protection. While radiation dose significantly affected the stability of vitamin C in solution, stability of vitamin C in dry formulations (vitamin C supplements, multivitamins, and ascorbic acid powder) was stable following radiation doses of up to 1000 Gy.

Radiation Effects on Fruits and Vegetables

Determination of appropriate cultivars not only for yield but also food use are important to the success of a sustainable human space habitat. Preliminary studies have shown that cultivar selection and growth conditions impact the composition and acceptability of crops such as sweetpotato and carrot. While moisture, ash, protein, total sugar, water activity, calcium, magnesium, sodium, and potassium were relatively constant upon exposure to all radiation doses, total extractable fat, vitamin C, carotenoids, and lycopene were significantly affected by radiation doses. The finding that radiation exposure may increase the extractability of fats from plant tissues could be utilized for further food processing development and commercial applications.

Radiation Effects on Wheat

Wheat is the most studied candidate crop and has the highest priority. Apogee and Perigee cultivars of wheat were developed at Utah State University to yield high amounts of wheat berries with a minimum amount of inedible crop waste. While growth conditions have been characterized, further analysis of these cultivars must be conducted to characterize the protein, lipid, and starch functionalities related to food quality, as well as antioxidant capacities, and to compare the food-functionality of these cultivars to common cultivars used in the food industry. Because wheat can be shipped as a bulk ingredient and/or grown in space, additional studies investigated the effects of radiation exposure on the quality and functionality of wheat. For a Mars mission, results indicate that Apogee is a more suitable candidate cultivar than Perigee due to its overall stability and nutritional properties.

For the oils and peanut-stability project, the impact of space-relevant radiation doses on peanut and soybean oils (with and without antioxidants) as well as effects of radiation on intact peanuts is being finalized.

A manuscript is being developed on effects of space-relevant doses of gamma radiation on wheat. Gamma-radiation effects have been characterized on wheat macronutrients, antioxidant capacity, functionality in breads, and sensory profiles of breads.