RESEARCH ABSTRACT
PORK SAFETY

Title: Use of molecular assays to assess *Toxoplasma gondii* burden in commercial meat samples **NPB #02-101**

Investigator: Joan K. Lunney

Institution: ANRI, BARC, ARS, USDA, B.1040, Beltsville, MD 20705

Date Received: March 29, 2005

Abstract: To improve food safety, our efforts have been aimed at developing diagnostic tests to confirm that pork products that contain foodborne microbial pathogens are reliably identified. Specifically, this grant was aimed at determining whether our parasite DNA test could detect pig products that contain the foodborne parasite, *Toxoplasma gondii* (Tg). Our fluorogenic real-time Tg DNA test, developed with previous NPB funding, is a quantitative molecular technique to detect Tg DNA and was recently patented. In collaboration with USDA IFAFS project "Retail meats survey for *Toxoplasma gondii,"* coordinated by Drs. J.P. Dubey and D. Hill, APDL, BARC, we used our fluorogenic Tg DNA assay to test commercial pork products for parasite contamination. Over a 3 year period commercial meat samples were collected under the IFAFS funded project from market sampling areas (MSA) nationwide using a statistically validated process. Results from that grant, assessing Tg burden using cat bioassays and meat juice analyses, will be reported separately.

Aliquots of the IFAFS pork samples were provided to this NPB project for testing for Tg contamination using parasite DNA assay. First, pretest controls were performed; these confirmed that the Tg DNA assays were positive when used on pig products collected from known Tg infected pigs. Then the IFAFS-associated commercial pork samples were tested. For each MSA, 75 pork products were obtained and processed; aliquots of 8-13 mixed pools of ~5 ground pork samples were provided for molecular testing. Over the course of this NPB project a total of 236 aliquots of pooled pork samples were tested; this represented 1180 individual meat products. For efficient sampling and molecular testing DNA was prepared from only 25mg of each pooled meat sample. None of the 226 pooled pork samples tested has been confirmed as positive for Tg by the parasite DNA assay whereas all 226 samples were clearly positive for 18S DNA, affirming DNA quantity. Therefore, the commercial pork sample molecular testing indicates the possible absence, or low level, of Tg parasite contamination.

More pork samples, and larger aliquots of these samples, [at much higher costs] would need to be tested to absolutely prove the lack of Tg parasite contamination especially since such a small sample was tested for the molecular assays. A follow-up survey of 108 individual samples from 1 MSA was performed and 1 potential Tg+ product identified. This is being tested further. Indeed, the definitive test results will be those collected, and reported separately, with the more detailed bioassay and meat juice antibody tests performed during Drs. Dubey and Hill's USDA NRI IFAFS project. All molecular test results will be compared to those results, expected later in 2005. Hopefully, all of our results will affirm the very low contamination of the commercial US pork supply with the foodborne parasite, Tg.