NEPA Decision Summary for Permit #**12-276-102r**

Dr. Lloyd Yu, the Director Operations for Planet Biotechnology Inc., has requested a permit for a small confined field release of genetically engineered tobacco (*Nicotiana tabacum*) with pharmaceutical intent at a paved and fenced in site in Alameda County, California.

Based on a review of Permit **12-276-102r**, the following determinations were made:

1. The gene constructs proposed for the confined field release are expected to result in tobacco that produces an immunoadhesin against the protective antigen of *Bacillus anhracis* (PBI-220). The protein of interest is a fusion protein comprised of the extracellular portion of the Capillary Morphogenesis Protein 2 and the hinge, CH2 and CH3 domains from the IgG1 heavy chain. The purposes of this field trial are to a) Prove the suitability of recombinant male-sterile plants for later commercial release, b). Quantify the presence of the pharmaceutical protein in flowers and other biomass and, c). Produce seeds for future trials.

2. All constructs was developed through Agrobacterium-mediated transformation. Disarmed *Agrobacterium tumefaciens* is incapable of inducing tumor formation. No plant pest vectors or vector agents are expected to be associated with the transformed lines and the genetic constructs are expected to be efficiently and stably inserted into the tobacco genome.

3. APHIS has authorized numerous field releases of genetically modified tobacco. APHIS also has issued more than 15 release permits for tobacco expressing pharmaceutical proteins. An EA was prepared and a FONSI published for tobacco producing an antibody against dental caries in response to permit 05-354-03r submitted by Planet Biotechnologies, Inc. The date of the FONSI is 2/4/2008**.**

   The structure of tobacco flower is to favor self-fertilization and often, anthers dehisce or release the pollen while the corolla is still closed. The frequency of cross-pollination is less than 5%. Lines developed for this field test will be in fertile (*N. tabacum* Wisconsin 38, parental) or male-sterile (*N. tabacum* KT209LC) backgrounds.

4. In this 0.05 acre field release, the applicant will use male-sterile and fertile lines of recombinant *N. tabacum* expressing PBI-220, an immunoadhesin against anthrax. All lines carry the same expression construct for the anthrax anti-toxin, which contains no toxin or *B. anthracis* components. Plants derived from the specified expression plasmid were previously used under permit 11-074-103rm. The gene product was field released and does not have characteristics of known toxins or allergens. No foreseeable effects on other organisms are expected. Furthermore, due to the presence of nicotine in the tobacco plant, few organisms consume tobacco. The release site (see Figure1) was previously used under permit 05-354-04r, which was fenced, secured and paved. Only very small animals and insects can get into the release site. Because PBI-220 is non-toxic and ubiquitous, consumption of tobacco containing these recombinant proteins is an unlikely event and no significant negative effect should occur.

5. GE tobacco seeds will be germinated in a greenhouse and plantlets will be transplanted into pots and then moved to the field release site. This reduces the possibility of seeds being released into the environment. The field release site is paved, security controlled and surrounded by industrial settings (no farmlands within 1 mile radius) and no sexually-compatible relatives of tobacco are known to exist in the area where the trial will be performed. About 10-20 plants are allowed to set seeds for this field trial and only a limited number of flowers will be bagged for each fertile plant. The bagged fertile flowers will not allow dehiscing from the plants until the last stage of seed maturation. From flower emergence, it will take more than 3 weeks to produce mature seeds. In the last stage of the seedpod maturation, the applicant will detach bagged bracts and bring them into the greenhouse. The viable seed will be matured and produced in on-site greenhouse. This strategy is expected to eliminate concern that the GE seeds will shattered and persistent in release site. The applicant also will monitor the plant daily during the period when buds and flowers are present. The compost will be recycled back into greenhouse and not be sold or otherwise disposed.

   Biomass from leaves and stems will be harvested and extracted for PBI-220 protein in the lab. The rest of the non-viable biomass (without seeds) from the release site and greenhouse will be composted in designated secured bins on-site. Seed germination and seedling growth will take place in the secured greenhouses. Compost will be monitored for seedlings weekly, and there will be no post-harvest monitoring period for the release site because the location is a paved surface, the regulated articles will be placed in containers and all seeds will be matured in the greenhouse.

   Applicant provided confinement measures described in the application and the proposed Experimental and /or Production Design should be sufficient to prevent any unplanned releases of transgenic tobacco in the environment.

6. Personnel who handle the regulated materials will receive training. This training will be documented. The Standard Operating Procedures and their logs will be made available to APHIS inspectors.

7. APHIS analyzed the potential effects on federally-listed threatened or endangered species and their critical habitat.

   Threatened and Endangered species listed for California include: 123 animals and 180 plants (http://ecos.fws.gov/tess_public/StateListingAndOccurrence.do?state=all, accessed 11/23/2012). There are no threatened or endangered plants listed that are sexually compatible with tobacco. Of the 123 listed threatened and endangered animal species, some insects, birds and small mammals can get in the fence and feed on those tobacco plants in this confined trial. As noted in previous Field Test Reports, Dr.Yu reported hummingbirds and bees visited the tobacco plants in the same release site. However, the PBI-220 is non-toxic and ubiquitous, consumption of tobacco containing these recombinant proteins is an unlikely event and no significant negative effect should occur.

The applicant and APHIS also searched the US FWS web site (http://crithab.fws.gov) and relevant Federal Register Notices relating to listed endangered and threatened species. The applicant has done a critical habitat analysis of the action area. From the proposed field release site, the nearest designated critical habitat in Alameda County, CA which is more than 10 miles away at tidal area on the San Francisco Bay.

Based on the APHIS analysis and the applicant's "Threatened and Endangered Species Analysis", APHIS has determined that there is no effect to any of these species or that the activities authorized in this permit would not result in the alteration of any designated critical habitat and will not harm or have any adverse or other significant effects on threatened and endangered species by direct or indirect exposure.

8. None of the regulated biomass from this field trial is allowed to be sold or used in humans or animals. Any use of these products for food or feed must be in compliance with the guidelines published in the Federal Register by the United States Food and Drug Administration-57 FR 22984, May 29, 1992.

The proposed 0.05 acre release site is surrounded by paved surface with the industrial setting. It was approved for Field Releases of regulated Tobacco in the past. The field release is not expected to significantly alter the agro-ecosystem of the release area.

For the above reasons, and those documented on the NEPA/ESA decision document, APHIS has determined that this permit involves a confined field trial of genetically engineered organisms or products that do NOT involve a new species or organism or novel modification that raises new issues. Issuance of this permit qualifies for categorical exclusion status under 7 CFR § 372.5(c)(3)(ii), and none of the exceptions for categorically excluded actions under 7 CFR § 372.5(d) apply to this action because APHIS has determined that all environmental impacts resulting from the issuance of this permit will be insignificant. APHIS has determined that this action does NOT have the potential to significantly affect the quality of the human environment, and neither an environmental assessment nor an environmental impact statement is required.