

NEPA Decision Summary for Permit #08-057-109r

Dr. Kan Wang of Iowa State University has requested a permit for a confined field release of 0.25 acres of genetically engineered corn plants at a site in Marshall County, Iowa.

Based on a review of Permit #08-057-109r, the following determinations were made:

1. The gene construct proposed for the confined field release is expected to result in corn that produces heat labile *E. coli* enterotoxin (LT-B) in the endosperm of transgenic corn. The construct consists of the gamma zein (seed-specific) promoter from *Zea mays*, the LT-B gene (B unit of the *E. coli* enterotoxin) and the vegetative storage protein terminator from *Glycine max*. The construct also includes the selectable marker gene, *bar*, which codes the enzyme phosphinothricin acetyl transferase, from *Streptomyces hygroscopicus*. The promoter for the selectable marker is the ubiquitin promoter from *Zea mays* and the nos terminator from *Agrobacterium tumefaciens*. This construct containing the LT-B gene has been previously used under permits 07-052-106r, 06-061-01r, 05-069-01r, and 04-131-01r. The gene conferring Basta resistance (*bar*) has been previously approved by APHIS for 2500+ applications in multiple plant species over a period of 15 years. The gene construct contains non-coding regions derived from plant pests (the terminator from *Agrobacterium tumefaciens*) that have been safely used to regulate the expression of transgenes in corn and other plants. None of the genes encoding the desired traits or the selectable marker, nor the regulatory elements controlling their expression, have any inherent plant pest characteristics, and they are not likely to pose a plant pest risk.
2. Based on biolistic transformation, only the genetic construct that is designed to be expressed in the genetically engineered corn lines is expected to be efficiently and stably inserted into the corn genome. No plant pest vectors are expected to be associated with the transformed corn lines as a result of the transformation process.
3. The intent of this field release is to further develop a male-sterile (pollenless) corn line as a proof-of-concept project to express desired pharmaceutical proteins. The applicant has no intention of commercializing future production of the LT-B protein. The resulting seeds from this permit will be used for breeding purposes, and for biochemical and immunological analyses of the LT-B protein under laboratory conditions.
4. The LT-B protein produced is non-toxic and is responsible for the immunological recognition of the toxin by the Gm1 receptors underlying the epithelium cells of the gastrointestinal tract of a host. LT-A, the subunit responsible for the toxic effects of the *E. coli* enterotoxin, is not produced in the transgenic corn. Thus, the protein produced during this pharmaceutical field trial is not toxic.
5. The proposed field site has a fully fenced perimeter. An electrical fence will be placed four feet outside the chain-link fence on the perimeter of the field site. A gate to the plot will also include electric fence hardware. Adequate security is in place for this field trial.

6. The proposed 0.25 acre field release site is limited to 3000 transgenic corn plants and 3000 nontransgenic corn plants. Given the small size of the field trial, it is anticipated that adequate resources will be available to manage the trial effectively.
7. The pollen donor plants are not transgenic. The female plants are male sterile (cytoplasmic male sterility, Texas strain [cms-T]). These transgenic plants are sterile and will not produce pollen. The cms-T system was used extensively in commercial production in the 1960's and 1970's. Among cms types, T is considered relatively stable (V.E. Gracen, 1982, Types and Availability of Male Sterile Cytoplasm) and highly stable to reversion (meaning that it tends not to spontaneously mutate back to fertility -- J.R. Laughnan and S. Gabay-Laughnan, 1982, Nuclear Control over Reversions to Male Fertility in S Male-sterile Maize"). No leakage has been observed in the lines used by the researchers (personal communication). [References found in "Maize for Biologic Research", edited by W.F. Sheridan.] Additionally, the applicant will survey the plants daily during tassel emergence, and tassels extruding anthers will be removed by pulling them by hand out of the whorl. Thus it is highly unlikely that any transgenic pollen will be produced during the field trial.
8. The isolation distance between the transgenic corn and the nearest non-transgenic corn is greater than 0.5 miles. The planting of this field site will occur 28 days after non-transgenic corn fields are planted between 0.5 and 1 mile of the field site. This isolation distance and temporal isolation meets the APHIS guidelines for a pharmaceutical trial for corn that uses reproductive isolation methods.
9. There is a 50 foot buffer surrounding the field test site. No food or feed will be harvested from the buffer zone and the plants within the buffer zone are not sexually compatible with corn.
10. All pollinated ears will be hand harvested at maturity with the husks on to further minimize any accidental spillage of seed. After hand harvesting the ears, stalks will be tilled or mixed with the soil at the site, buried at the site or removed from the site and incinerated or autoclaved.
11. The field plots and isolation distance (0.5 mile) will be monitored for volunteers every 4 weeks during the following (2009) growing season (May to August). All volunteer corn plants found will be removed chemically, manually, or mechanically.
12. During the 2009 growing season, if the field is not used again for another pharmaceutical field trial, the area will be monitored the following year for volunteer corn plants. The site will remain fenced. The soil inside the fence will be sprayed with herbicide in the spring.
13. The confinement measures described in the application and supplemental permit conditions should be sufficient to prevent any unplanned releases of the transgenic plant material or transgenic seed; or the persistence of the transgenic material or its progeny in the environment.

14. The only federal listed TES species in Marshall County (using Iowa Natural Areas Inventory http://csbweb.igsb.uiowa.edu/ims gate/maps/natural_areas.asp Accessed 04/08/08) is the Topeka shiner. Critical habitat for the Topeka shiner is not found in Marshall County. There is a small stream within 0.5 miles of the field trial, however, the stream is not within the action area. There are 15 other animal species listed as TES for the state of Iowa (www.ecos.fws.gov; accessed 04/08/08). None use corn as a food plant. The range of the Indiana bat is not known to include Marshall county, IA (draft recovery plan, accessed 04/09/08). The Higgins eye pearly mussel is found on the main stem of the Mississippi, and other major rivers (recovery plan, accessed 04/09/08). There are no major rivers in the action area. Piping plovers and least terns are known in the far western region of Iowa and not in Marshall County (recovery plans, accessed 04/09/08). The Iowa Pleistocene snail and the Pallid sturgeon are aquatic species. This action area of this field trial does not include rivers or streams. No toxins are produced by the regulated article. No transgenic protein is expressed in pollen. Only the grain produced during this field trial expresses the LT-B protein, and the grain will be collected and not available for other species as a food resource. Furthermore, in the unlikely event of accidental consumption, the pharmaceutical protein produced during this field trial is non-toxic and is not expected to harm animals feeding on this grain. There are 5 plant species listed by FWS as TES in Iowa. None are sexually-compatible with corn. Therefore these field trials will have no effect on threatened or endangered species.
15. According to <http://crithab.fws.gov/>, accessed 04/09/08, there is no designated critical habitat or proposed designated critical habitat found in this county.
16. The gene products used in this field trial are not known to be toxic. Based on the above, APHIS is confident that these field trials will not harm or have adverse or other significant effects on threatened or endangered species either by direct or indirect exposure.
17. Regulated materials in this field trial are not intended for food and/or feed. 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].

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 state is required.

Signed: _____
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Date: _____
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