

are to be applied at 10 lb of end-use product (EP) (equivalent to the low rate of less than 0.01 lb active ingredient (ai) per acre). Within 3 days of application of the pesticide, the fields are furrow irrigated to promote germination of AF36, which apparently colonizes the cotton crop and soil, before the aflatoxin-producing strains of *A. flavus* proliferate. This competitive exclusion of the aflatoxin-producing strains does not increase the total *Aspergillus* population in the environment above background levels as demonstrated in soil and air monitoring studies. [MRIDs 45307201, 45307202; BPPD Review of Soil and Air Monitoring Studies and Product Performance Testing (Efficacy), dated May 15, 2003 (hereinafter referred to as "BPPD Review - May 15, 2003")]. The displacement of the toxigenic strain of *Aspergillus flavus* by AF36 may reduce aflatoxin contamination of cotton seed.

The toxicology and pathogenicity data generated by the petitioner in support of this tolerance exemption, and reviewed by the Agency, are summarized below. The following discussion of the evaluations of the submitted studies and information indicates that exposure to the pesticide is not likely to be greater than that which occurs normally to other ubiquitous *A. flavus* strains. Submitted data also indicate no toxicity or infectivity of AF36 in test mammalian systems. More detailed analyses of these studies can be found in the specific Agency reviews of the studies that are cited below.

1. *Acute oral toxicity/pathogenicity (OPPTS Harmonized Guideline 885.3050; MRID 43972403)*. Agency evaluation of submitted acute oral study indicates no toxicity/infectivity effects of the pesticide. Five male, and five female Sprague Dawley rats were treated orally with the microbial pesticide (500 milligrams/milliliter (mg/mL) or 6.3×10^3 cfu/mL) by gavage. No clinical signs or abnormalities were noted during the study, and the pesticide was considered to be neither toxic nor infective following oral administration of a single dose. The acute oral test resulted in a Toxicity Category IV classification with a lethal dose (LD)₅₀ greater than 5,000 milligrams/kilogram (mg/kg) body weight [MRID 43972403; BPPD Data Evaluation Report, Acute Oral Toxicity Study in Rats, dated April 23, 1996 (hereinafter referred to as "BPPD Review - April 23, 1996")].

2. *Acute pulmonary toxicity/pathogenicity (OPPTS Harmonized Guideline 885.3150; MRID 45798201)*. The Agency required an intratracheal pulmonary infectivity/pathogenicity study. This test involves intratracheal

instillation of the test material and post mortem examination of lungs and other organs for clearance.

Three studies were submitted in support of the mammalian acute infectivity/pathogenicity pulmonary guideline: A range finding study and two complete acute pulmonary studies. The dose-range study concluded that 10^8 cfu/rat would be a suitable test dose level for the acute pulmonary studies [MRID 45739101; BPPD Data Evaluation Report, dated April 02, 2003a (hereinafter referred to as "BPPD Review - April 02, 2003a")]. In the first acute pulmonary study, conducted with Tween 80 as a surfactant in the test material, 26 male and 26 female Sprague Dawley rats (approximately 8 to 10 weeks old) each were dosed with a single intratracheal dose of 1.2 mL/kg at 5.30×10^8 cfu/mL (or 1.28 to 1.63×10^8 cfu/animal). Results from this study indicated that the test organism was neither infective nor pathogenic, in spite of rat mortality, which is believed to have been due to a severe acute inflammatory response to the Tween 80 [MRID 45798101; BPPD Data Evaluation Report, dated April 02, 2003a (hereinafter referred to as "BPPD Review - April 02, 2003b")].

In the second acute pulmonary study, which was a repetition of the first acute pulmonary test, but was conducted without Tween 80, 25 male and 25 female Sprague Dawley rats (approximately 8 to 10 weeks old) each received a single intratracheal dose of approximately 1.2 mL/kg. Mortality of 4 rats by day 2 appeared to be attributable to an initial dosing effect. The rest of the test animals showed an initial response, followed by a rapid recovery indicating no toxicity. Although some surviving rats lost weight intermittently, all surviving rats gained weight prior to scheduled sacrifice. No clinical signs that were considered to be due to the test organism were observed in the test rats. Organs were examined *post mortem* as previously described. *Aspergillus flavus* AF36 was detected in the lungs with clearance by day 8 after dosing. No test organisms were detected in any samples from the shelf control or inactivated test organism treated rats. Based on the presented/submitted data, including the clearance data, the test organism, *Aspergillus flavus* AF36, was considered not toxic, infective, or pathogenic to the rat pulmonary system. The study is acceptable.

3. *Acute inhalation (OPPTS Harmonized Guideline 152-32)*. The inert is sterilized wheat seeds, comprising approximately 99% of this pesticidal product. It acts as a matrix and nutrient source for the germinating

AF36. Because this constitutes the majority of the pesticide and does not contain respirable particles of less than 10 microns, an inhalation study was not required pursuant to 40 CFR 158.740(c). In addition, based on the results obtained through the acute pulmonary toxicity/pathogenicity studies summarized immediately above, AF36 is considered not toxic, infective, or pathogenic to the rat pulmonary system. On the basis of this study and the nature of the inert ingredients present, the pesticide was considered Toxicity Category III for acute inhalation effects. [MRID 45798201; BPPD Data Evaluation Report, dated April 02, 2003c (hereinafter referred to as "BPPD Review - April 02, 2003c")].

4. *Hypersensitivity incidents (OPPTS Harmonized Guideline 152-37; MRID 45739104)*. The registrant submitted information (MRID 45739104) to demonstrate the lack of hypersensitivity to workers who have been exposed during the manufacture, application, and use of the pesticide in the research and experimental phases. No adverse hypersensitivity reaction to AF36 was recorded or reported by a state council or six companies during use for 3 or 6 years [MRID 45739104; BPPD Data Evaluation Report, dated April 02, 2003d (hereinafter referred to as "BPPD Review - April 02, 2003d")]. However, to comply with the Agency's requirements under section 6(a)(2), any incident of hypersensitivity associated with the use of this pesticide must be reported to the Agency.

5. *Data waivers*. Data waivers were requested for the following studies:

- i. Acute dermal toxicity/pathogenicity (OPPTS Harmonized Guideline 885.3100)
- ii. Primary dermal irritation (OPPTS Harmonized Guideline 870.2500)
- iii. Primary eye irritation (OPPTS Harmonized Guideline 870.2400)
- iv. Intravenous, intracerebral, intraperitoneal injection (OPPTS Harmonized Guideline 885.3200)
- v. Hypersensitivity study (40 CFR 152-36)
- vi. Immune response (40 CFR 152-38)

With regards to the dermal and eye irritation guideline tests, it was impractical to apply the end-use product, sterilized wheat seeds inoculated with *Aspergillus flavus* AF36, as test material. Furthermore, non-occupational dermal and eye exposures, or exposures via any of the routes in Unit III.5.i.—vi., are not likely to be above naturally-occurring background levels for the following reasons.

First, *Aspergillus flavus*, a saprophytic fungus, is a normal