Regulatory Decision on the Application of Bt Cotton-GFM Cry1A for Commercial Propagation

I. Brief Identification of the Genetically Modified Organism

Designation:	Bt Cotton-GFM Cry1A
Applicant:	Philippine Fiber Industry Development Authority (PhilFIDA)
Host Plant:	Cotton (*Gossypium hirsutum* L.)
Trait Description:	Bt Cotton is genetically modified through the insertion of the synthesized insecticidal gene, cry1Ab-Ac which encodes for the insecticidal Bt-protein that confers resistance to the cotton bollworm.
Trait Introduction Method:	Pollen Tube Pathway
Donor Organism (s):	*Bacillus thuringiensis* subsp. kurstaki – the donor bacterium where the cry1Ab-Ac synthetic fusion gene was derived *Escherichia coli* Tn5 transposon – source of *nptII (Neophosphotransferase II)* used as a marker gene *Escherichia coli* – source of the *uidA* gene encodes beta-D-glucuronidase (GUS) allowing detection of transformed cells in the laboratory
Proposed Use:	For commercial propagation

II. History of Safe Use of the Host Plant

Cotton has been used by humans for thousands of years. Various methods of processing using cotton have been developed leading to its widespread adoption. Cotton is a common source of natural fibers mainly used in manufacturing of textiles. The seeds, on the other hand, are used to extract edible oil utilized for salad dressing, margarines, and vegetable oil. Inactivation or removal of gossypol and cyclopropenoid fatty acids (CPFAs) during processing has enabled use of cottonseed meal for catfish, poultry and swine.

III. Characteristics of Host Plant

Cotton, commonly referred to as the cultivated variety of *Gossypium*, includes two diploids and two tetraploids, with only the *hirsutum* species being grown in the Philippines.

Though the exact origins of the *Gossypium* are uncertain, it is believed that its main diversity centers were in west-central and southern Mexico, northeast Africa, Arabia, and Australia. Domestication is thought to have occurred in Africa and Asia, Mesoamerica, and South America.

Cotton typically thrives in specific environments such as in areas with abundant water and/or nutrients, but it does best in regions with deep surface soil, good drainage, and a pH level ranging from 5.5 to 7.

In the Philippines, medium-staple *hirsutum* varieties are cultivated in regions with Type I or Type III climates, where there is a distinct dry season. The growth cycle lasts 5 to 5 1/2 months, with harvesting timed during the dry season to avoid rain damage.

The extent of outcrossing, influenced by insect pollinators such as bumble bees and honey bees, varies based on environmental factors and pollen viability affected by temperature.

Despite centuries of cultivation, cotton has not been identified as a significant weed pest, with no *Gossypium* species recognized as problematic weeds either agriculturally or environmentally, nor do they have problematic weed relatives.

IV. Characteristics and Safety Assessment of the GM Crop

The Bt cotton-GFM Cry1A contains three genes such as the insecticidal gene, *cry1Ab-Ac,* derived from *Bacillus thuringiensis subsp. kurstaki, nptII* kanamycin resistant which serves as a selectable marker gene from *Escherichia coli* Tn5 transposo, and *uidA* gene derived from E. coli which allows the detection of transformed cells. Bt cotton was transformed using the Pollen Tube Pathway allowing the crop to have built-in resistance to the cotton bollworm, *Helicoverpa armigera* (Hubner).

Bacillus thuringiensis subsp. kurstaki, from which the cry1Ab-Ac synthetic fusion gene was derived, is widely known for its insecticidal properties against Lepidopteran pests. There have been no scientific reports indicating toxicity to humans and other mammals. *Escherichia coli* Tn5 transposon, on the other hand, is the source of the *nptII* gene, which serves as a marker gene. This gene has no pesticidal activity, and not known to be toxic or allergens. Similarly, the *uidA* gene sourced from *Escherichia coli,* also used as a marker gene in GMO plants and products, does not possess pesticidal properties. *Escherichia coli* naturally occurs in the digestive tracts of vertebrates, as well as in soil and water ecosystems. Therefore, the presence of these genes in GM food and feed is unlikely to cause harm to either the environment or consumers.

To evaluate the safety of *Bt* cotton for human and animal consumption, the proteins expressed were subjected to allergenicity and toxicological studies, and compositional analysis.

The susceptibility of the Cry1Ab-Ac, NPTII, and GUS proteins to degradation have been studied and based on the results, these proteins were rapidly degraded fluid

after seconds of exposure to simulated gastric fluid. The results support the conclusion that when ingested, these proteins would be readily degraded.

Bioinformatics studies to evaluate any significant amino sequence similarity of Cry1Ab-Ac, NPTII, and GUS proteins with known allergens were conducted. Identity search showed no biologically relevant identity between query proteins and any known allergenic proteins. Furthermore, the result of the acute oral toxicity tests in mammals provide additional evidence supporting the specificity and safety of the proteins.

Analysis of nutritional data revealed no significant differences between Bt and non-Bt cottonseeds, affirming substantial equivalence in their composition. The results of the compositional analyses showed that both transgenic and non-transgenic cotton varieties are within acceptable ranges.

Feeding trials demonstrated that Bt cottonseed does not adversely affect the health or milk production of lactating cows. Milk analysis showed no significant changes in fat, protein, lactose, total solids, ash, or somatic cell count. No guinea pig dermal desensitization was induced by Bt cottonseeds following the experiment using the Buehler method. There is also no adverse effects observed on goat health.

According to a 2004 study by Nath Seeds India, the novel protein Cry1Ab-Ac found in Bt cotton seeds as part of fish feed did not significantly affect the overall health and behavior of catfish. Similarly, Bt cotton seeds as part of feed did not significantly impact the overall health, behavior, or egg-laying capability of laying hens.

Result of the multi-location field trials showed that there were no notable distinctions in the bacterial and fungal populations in soils from either Bt cotton or non-Bt cotton rhizospheres at the test sites. Additionally, Bt cotton does not exhibit harmful effects on non-target insect pests or beneficial arthropods, including predators, parasites/parasitoids, pollinators, and spiders. This further confirms the safety profile of Bt cotton.

V. Regulatory Decision

After reviewing the scientific data and information relevant to the application of the Philippine Fiber Industry Development Authority (PhilFIDA), the Bureau of Plant Industry (BPI) has approved the commercial propagation for Bt cotton-GFM cry1Ab-Ac. The BPI issued Biosafety Permit for Commercial Propagation of Bt-cotton GFM cry1Ab-Ac with conditions that need to be complied with by PhilFIDA. Copy of the Biosafety Permit issued may be accessed through the BPI Biotechnology website.