Maize (Zea mays) is a widely cultivated cereal that has been safely consumed by humans and animals for centuries. Transgenic or genetically engineered insect-resistant and herbicide-tolerant maize, are commercially grown on a broad scale. Event TC1507 (OECD unique identifier: DAS-01507-1) or the Herculex® I trait, an insect-resistant and herbicide-tolerant maize expressing Cry1F and PAT proteins, has been registered for commercial cultivation in the US since 2001. A science-based safety assessment was conducted on TC1507 prior to commercialization. The safety assessment addressed allergenicity; acute oral toxicity; subchronic toxicity; substantial equivalence with conventional comparators, as well as environmental impact. Results from biochemical, physicochemical, and in silico investigations supported the conclusion that Cry1F and PAT proteins are unlikely to be either allergenic or toxic to humans. Also, findings from toxicological and animal feeding studies supported
that maize with TC1507 is as safe and nutritious as conventional maize. Maize with TC1507 is not
expected to behave differently than conventional maize in terms of its potential for invasiveness,
gene flow to wild and weedy relatives, or impact on non-target organisms. These safety conclusions
regarding TC1507 were acknowledged by over 20 regulatory agencies including United States
Environment Protection Agency (US EPA), US Department of Agriculture (USDA), Canadian Food
Inspection Agency (CFIA), and European Food Safety Authority (EFSA) before authorizing
cultivation and/or food and feed uses. A comprehensive review of the safety studies on TC1507, as
well as some benefits, are presented here to serve as a reference for regulatory agencies and decision
makers in other countries where authorization of TC1507 is or will be pursued.

KEYWORDS. TC1507, Cry1F, GE maize, environmental safety, food and feed safety, global
authorizations

ABBREVIATIONS. aa, amino acid; Bt, Bacillus thuringiensis; CFIA, Canadian Food Inspection
Agency; Cry, crystalline; CTNBio, Comissão Técnica Nacional de Biossegurança; DA-BPI,
Department of Agriculture-Bureau of Plant Industry; DNA, deoxyribonucleic acid; EFSA, European
Food Safety Authority; ELISA, enzyme-linked immunosorbent assay; ERA, environmental risk
assessment; EU, European Union; FAO, Food and Agriculture Organization of the United Nations;
FDA, Food and Drug Administration; FFP, food, feed, and processing; FSANZ, Food Standards
Australia New Zealand; GAIN, Global Agricultural Information Network; GE, genetically
engineered; HGT, horizontal gene transfer; ISAAA, International Service for the Acquisition of Agri-
biotech Applications; LD50, median lethal dose; NCGA, National Corn Growers Association; NTOs,
non-target organisms; nptII, neomycin phosphotransferase II; OECD, Organisation for Economic Co-
operation and Development; PAT, phosphinothricin-N-acetyltransferase; PCR, polymerase chain
reaction; SDS-PAGE, sodium dodecyl sulfate polyacrylamide gel electrophoresis; SE, Substantial
Equivalence; SGF, simulated gastric fluid; US EPA, United States Environment Protection Agency;
WHO, World Health Organization

INTRODUCTION

Maize (Zea mays) is a widely grown cereal that has been safely consumed by humans and
animals for millennia. Currently maize is predominantly used to feed livestock or as raw
material for industrial products, while only 21% is consumed as human food (OECD, 2003).
Between 1996 and 2013, transgenic or genetically engineered (GE) maize was grown on a
cumulative 460 million hectares (based on data derived from ISAAA [2014]) and in 2013 alone
GE maize occupied over 32% or 57 million hectares of maize area. GE maize was grown in 17
countries and the greatest hectarage (in millions of hectares) was in the US (35.6), Brazil (12.9),
Argentina (3.2), South Africa (2.4), and Canada (1.7) at that time (James, 2013).

In the US, GE maize products were developed targeting lepidopteran insect pests due to the
potential for substantial economic damage as a result of significant yield losses. These GE maize
products were first commercialized in 1996. With the success of lepidopteran-resistant maize, GE
maize products that protect against subterranean corn rootworm followed (Castle et al., 2006).
McLaren and Copping (2011) have summarized the global registration status of different commer-
cially available GE maize lines. Transgenic maize hybrids expressing 2 or more traits and
combined through conventional breeding techniques, commonly referred to as breeding stacks,
have been available since 2000. Insect-resistant and herbicide-tolerant traits enable farmers to use
simplified crop management practices (Que et al., 2010); therefore, such breeding stacks occupied
almost 73% of all GE maize hectares planted in 2012 (ISAAA, 2014).

The insect-resistant GE maize currently in the market expresses genes derived from
Bacillus thuringiensis (Bt), and these transgenic products are commonly referred to as Bt maize. Bt is a ubiquitous soil bacterium that has proven to be a rich source of insecticidal proteins, which are considered to be selective and generally active against insects within a specific taxonomic insect order (Van Frankenhuyzen, 2009). All of the commercially available Bt maize products express one or more crystalline (Cry) insecticidal or vegetative insecticidal proteins (Que et al., 2010). The mode of action of Cry proteins is well understood (Estruch et al., 1996; Whalon and Wingerd, 2003; OECD, 2007) though scientific studies continue to further elucidate these mechanisms (Vachon et al., 2012). In general, Cry proteins are ingested, processed by intestinal proteases, and converted to active toxin in the insect alkaline midgut. The active toxins bind to specific receptors present in the gut of susceptible insects. The bound toxin-receptor complex leads to gut membrane pore formation, subsequent septicemia, and ultimately death. Humans and other mammals lack the alkaline gut as well as Cry protein-binding receptors and are therefore not vulnerable to toxicity from corresponding Cry proteins. For over 50 years, several varieties of Bt containing Cry proteins have been safely used as insecticidal sprays in commercial agriculture (Siegel, 2001) and, for the past 14 years, products derived from GE crops expressing Bt proteins have been safely consumed as food and feed (Hammond and Koch, 2012). The Organisation for Economic Co-operation and Development (OECD) summarized the safety information regarding Bt proteins derived from GE crops, with a focus on human health assessment and impact on non-target species (OECD, 2007).

Safety of GE crops and the food/feed derived from them is evaluated through various laboratory and field experiments and toxicological studies with non-target organisms (e.g., arthropods, aquatic organisms, birds, rodents, large mammals) (Craig et al., 2008). Such regulatory studies, performed to investigate the safety of a GE product prior to commercialization, can cost up to US $13 – 18 million and take an average of 3 to 4 years to conduct (McDougall, 2011). The safety data required for product authorizations are generated from research in the discovery phase through product development and regulatory studies. The cost and time to conduct these studies can vary significantly depending on the crop (food or non-food crop), nature of the introduced trait(s), country requirements, etc. Moreover, the estimated total cost for biotechnology trait development from discovery to commercial entry is US $136 million with regulatory costs including authorizations projected at 26% of that total cost (McDougall, 2011).
The purpose of this paper is to review the robust food, feed, and environmental safety information that served as the basis for securing regulatory authorization of maize event TC1507 in more than 20 countries, as well as some beneficial aspects from TC1507 commercialization. Information sources reviewed herein include dossiers presented to regulatory agencies in several countries, regulator safety assessment reports, peer-reviewed literature, online databases, and technology developer product materials. This comprehensive review is assembled to aid authorities making regulatory decisions in countries where registrations are being pursued for maize event TC1507, as a single event product and in breeding stack products, and for others interested in the safety of TC1507.

Event TC1507 and Background

Insect-resistant transgenic maize event TC1507 (OECD identifier: DAS-Ø15Ø7–1), also referred to as 1507 in the regulatory context and the Herculex® I trait commercially, was jointly developed by Pioneer Hi-Bred International, Inc. (DuPont Pioneer) and Dow AgroSciences LLC. TC1507 maize was developed to provide farmers a simple and highly effective tool to control certain key lepidopteran larval pests while tolerating glufosinate herbicidal active ingredients.

The process used to assess the safety of TC1507 summarized here is consistent with the recommended international guidelines as reviewed by Delaney (2009). During the assessment process, a variety of data were considered including the history of safe use of maize; source of the introduced genes (donor organisms); molecular characterization of the event; genetic stability; inheritance pattern; protein expression; protein specificity and efficacy; protein biochemistry and bioinformatics; toxicology; substantial equivalence with conventional comparators; impact on non-target organisms; and fate in the environment. Safety data, as required, were submitted to multiple regulatory agencies including the US, Canada, EU, Japan, Australia, Brazil, Argentina, Philippines, and South Africa, before obtaining authorizations for cultivation, food, and/or feed use.

Genes, Donor Organisms and Their Safety

TC1507 maize was designed to express sufficient levels of Cry1F and phosphinothricin-N-acetyltransferase (PAT) proteins, encoded by cry1F and pat genes, respectively, to achieve efficacious insect resistance and herbicide tolerance. The cry1F gene was derived from Bacillus thuringiensis var. aizawai. A modified cry1F gene that was codon-optimized for more efficient in planta expression was used to produce TC1507 maize. The plant-expressed cry1F gene encodes a protein of 68 kDa that is a truncated version of the native protein with a single amino acid substitution (USDA APHIS, 2000). The Cry1F protein is active against certain lepidopterans including key pests such as European corn borer (Ostrinia nubilalis), fall armyworm (Spodoptera frugiperda), corn earworm (Helicoverpa zea), and black cutworm (Agrotis ipsilon). It is worth referring to Wolt (2011) for a detailed list of lepidopteran species and observed susceptibilities to Cry1F protein in laboratory studies.

The pat gene was derived from the aerobic, non-pathogenic, naturally occurring soil actinomycete Streptomyces viridochromogenes. The PAT protein acetylates phosphinothricin, the active isomer present in the non-selective glufosinate-ammonium herbicide, to a metabolite, N-acetyl phosphinothricin, that is non-phytotoxic (OECD, 2002). In this way, expression of the PAT protein in TC1507 maize confers tolerance to glufosinate-ammonium herbicidal active ingredient and serves as a marker to select transformed maize in the laboratory. Separate reviews demonstrate the safety of PAT to human health (Hérouet et al., 2005) and the environment (CERA, 2011).

The pat gene in event TC1507 is a modified version of the native bacterial gene that was codon-optimized for improved in planta expression. The amino acid sequence of the plant-derived PAT protein is identical to the native PAT protein (Meyer, 1999). Refer to OECD (1999), for a general review on the pat genes and their enzymatic proteins.
**Transformation and Event Development**

PHI8999A, a linear DNA fragment containing the cry1F gene and the pat selectable marker gene, was obtained from plasmid PHP8999. The cry1F and pat gene coding sequences were driven by regulatory sequences enabling constitutive expression of the Cry1F and PAT proteins throughout the plant. An inbred maize line was transformed with PHI8999A by a micro-projectile bombardment (biolistic) method. Positively transformed plants containing both the cry1F and pat genes were evaluated in greenhouse testing and in the field. Of these, line 1507, which would later be designated as event TC1507, was selected for its good agronomic characteristics and efficacy against target insects.

Results of the molecular characterization revealed event TC1507 consisted of an insert at a single genetic locus that included the nearly full-length intact copy of the DNA insert, which contained the cry1F and pat genes. In addition, there are a few non-functional rearranged cry1F and pat partial fragments that are interspersed among native maize genomic sequences on both flanking regions, which are commonly observed during genomic integration via micro-projectile bombardment transformation (Pawlowski and Somers, 1996; Makarevitch et al., 2003). The event TC1507 does not contain the antibiotic resistance gene (nptII) that was included in the plasmid backbone, but was not present in the PHI8999A fragment used for maize transformation (USDA APHIS, 2000; US EPA, 2005).

Stability of the inserted genes was studied over multiple generations. Event TC1507 was crossed and backcrossed with an elite inbred to produce hybrids that were tested for glufosinate tolerance and resistance to European corn borer. Southern blot analyses demonstrated the stability of the inserted genes in progenies across at least 6 generations, and inheritance followed a Mendelian segregation pattern for a single dominant gene (USDA APHIS, 2000).

**Protein concentration**

The concentration of the Cry1F and PAT proteins in TC1507 maize has been well characterized. To date, protein expression in TC1507 maize has been characterized in over 20 field studies, spanning multiple geographies (including Brazil, Canada, Chile, Spain, and the US) and years (2005–2013). Concentrations of Cry1F and PAT proteins in representative tissues and developmental growth stages (e.g., leaf, root, pollen, stalk, whole plant, grain) were analyzed by enzyme-linked immunosorbent assay (ELISA) methods (Table 1). The Cry1F protein was detected in leaf, root, pollen, stalk, whole plant, and grain tissues. The PAT protein was detected in leaf, root, stalk, whole

| Tissue (Growth stage)     | Mean Cry1F Protein (Range)* (ng/mg tissue dry weight) | Mean PAT Protein (Range)* (ng/mg tissue dry weight) | Number of Studies** |
|---------------------------|-------------------------------------------------------|---------------------------------------------------|---------------------|
| Leaf (R1)                 | 20 (15 – 31)                                         | 9.0 (5.1 – 16)                                    | 21                  |
| Root (R1)                 | 5.7 (1.5 – 7.9)                                      | 0.51 (0.090 – 1.4)                                | 19                  |
| Pollen (R1)               | 27 (23 – 37)                                         | <0.28                                             | 21                  |
| Stalk (R1)                | 8.8 (4.7 – 13)                                       | 0.16 (0.022 – 0.50)                               | 19                  |
| Whole Plant (R1)          | 13 (8.3 – 21)                                        | 3.3 (1.6 – 6.9)                                   | 18                  |
| Grain (R6)                | 3.7 (2.1 – 5.7)                                      | <0.069 (<0.069 – 0.073)                           | 21                  |

*mean is reported as the overall mean of the reported mean protein concentrations across all studies; range spans the minimum and maximum reported mean protein concentrations across all studies.

R1 – stage of plant development when silks become visible.

R6 – stage of plant development regarded as physiological maturity.

**Field studies conducted in Brazil, Canada, Chile, Spain, and the US.
plant, and grain tissues. The PAT protein was close to the lower limit of the quantitative range of the ELISA in stalk, root, and grain, and was below the lower limit of the quantitative range of the ELISA in pollen (Table 1).

Diagnostic tools like gene- and event-specific polymerase chain reaction (PCR) methods, ELISA, and lateral flow strips are widely available to detect the introduced genes or the expressed Cry1F and PAT proteins in TC1507. These methods are employed for research, adventitious presence testing in seed or grain, inspection of food stuff, environmental monitoring and quarantine at ports (La Paz et al., 2006; Heide et al., 2008; Shrestha et al., 2008; Holck et al., 2009; Kim et al., 2010; Park et al., 2010; Rimachi et al., 2011; Takabatake et al., 2010).

**Agronomic characteristics**

Field trials were conducted in multiple key maize-growing regions in the US to evaluate the agronomic performance of TC1507 compared with an appropriate non-GE counterpart. Data on parameters such as yield, time to pollen shed, time to silking, grain density, plant height, ear height, early stand count, emergence, vigor, stalk lodging, root lodging, dropped ears, and integrity of the stalk were recorded from these field trials. Germination of TC1507 and control maize under cold and warm growing conditions was examined in laboratory studies. Results from these field trials show that the evaluated agronomic parameters were comparable between TC1507 and the non-GE comparator (USDA APHIS, 2000). Multiple additional field studies have been conducted in the US, Canada, Italy, France, Bulgaria, Spain, and Argentina, which all support the results of this field trial (Pioneer Hi-Bred International, Inc. internal unpublished data).

**Safety Assessment of Cry1F and PAT Proteins**

Safety tests are conducted with the introduced protein(s) to evaluate potential risks of a transgenic event. These studies are used to assess the potential allergenicity and toxicity of the introduced proteins and to inform conclusions as to the safety of the proteins in the context of food and feed uses.

**Cry1F protein equivalency**

Safety assessments of GE crops include regulatory studies covering topics such as toxicology and fate of environmental exposure that require large quantities of protein. Low trait protein expression levels render it impractical to extract sufficient quantities of novel protein from GE plants. Hence, alternative systems such as microbes are engineered to express the novel proteins. The suitability of microbial-expressed protein to serve as a surrogate for use in safety studies is determined by demonstrating equivalence of the proteins derived from microbe and plant sources through biochemical and physicochemical evaluations (Evans, 2004; Raybould et al., 2013).

The Cry1F protein used for the safety assessment studies was produced in the bacteria
*Pseudomonas fluorescens*. Full-length Cry1F toxin was extracted, truncated with trypsin, purified by diafiltration, and concentrated by lyophilization. Comparisons were made between the microbial-expressed Cry1F protein and the Cry1F protein isolated from TC1507 maize to evaluate protein equivalency by mass spectrometry, N-terminal sequencing, sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE), and western blot analyses. The amino acid (aa) sequence of the microbial-expressed Cry1F protein contained a single aa substitution and was 27 aa shorter (at the N-terminus) and 7 aa longer (at the C-terminus) in comparison with the plant-encoded Cry1F protein. Additionally, the Cry1F protein from both sources lacked detectable post-translational glycosylation (Evans, 1998). The bioactivity of the maize-expressed and microbial-expressed Cry1F proteins was tested in bioassays with susceptible insects such as European corn borer (*O. nubilalis*), tobacco budworm (*Heliothis virigescens*), and fall armyworm (*S. frugiperda*). The results indicated that the bioactivity of Cry1F protein from plant and microbial sources was comparable (Evans, 1998).

A similar biochemical comparison was made with plant-derived and microbial-expressed PAT proteins. The results of mass spectrometry, SDS-PAGE, and western blot analyses demonstrated the biochemical equivalency of the PAT protein from the plant and microbial sources (CFIA, 2002). Cumulatively, these studies established that Cry1F and PAT proteins isolated from the microbial source are equivalent to the corresponding proteins isolated from TC1507 maize, and these findings support the use of microbial-expressed proteins for regulatory studies.

### Allergenicity Assessment

No single factor has been recognized as the primary indicator for protein allergenicity and no validated animal model predictive of allergenic potential is available. Therefore, allergenic potential of proteins produced from the introduced genes in transgenic events is typically evaluated through a “weight-of-evidence” approach (CAC, 2009; Ladics, 2008; Ladics et al., 2011). The assessment of allergenic potential is based on the existing knowledge about allergens including the history of exposure and safety of the gene(s) source; the amino acid sequence similarity to known human allergens; and the thermolability, pepsin digestibility, and glycosylation status of the proteins (Ladics, 2008).

Bt (the source of the *cry1F* gene) has no history of causing allergy. In over 50 y of commercial use as a microbial pesticide on food crops, there have been no reports of allergenicity to proteins from Bt, including occupational allergy associated with the manufacture of products containing Bt (Hammond and Koch, 2012). These microbial formulations have been used on a wide variety of crops, including fresh vegetables, with no reported allergic concerns. *S. viridochromogenes* (the source of the *pat* gene) occurs widely in nature and is not known to cause allergy (Hérout et al., 2005; OECD, 1999). This history establishes a sound basis for the lack of allergenic potential for the Cry1F and PAT proteins.

### Bioinformatics in allergenicity assessment

Amino acid (aa) sequence similarity and structural comparisons of a novel protein to known allergenic proteins are important endpoints in the evaluation of allergenicity of GE foods. Bioinformatics analyses were conducted to compare whether the aa sequences of Cry1F and PAT proteins are similar to sequences in a database of food, dermal, and respiratory allergenic proteins. Such *in silico* analyses additionally examine the potential for cross-reactivity to known allergens (Ladics et al., 2011). Similarity (>35% shared identity over 80 aa or greater) was not detected and no contiguous sequence matches (8 aa or greater) were identified compared with sequences in the AllergenOnline database (University of Nebraska, Lincoln). However, a single contiguous match over 6 aa was identified between Cry1F and the Der p7 protein of the dust mite, *Dermatophagoides pteronyssinus*, but there was no evidence of cross-reactivity between the Cry1F and human sera reactive to Der p7 protein.
The lack of any significant aa similarity indicates that the potential for cross-reactivity of either Cry1F or PAT proteins with known allergens is extremely low (Meyer, 1999; Ladics et al., 2006).

**Thermolability of Cry1F and PAT proteins**

Thermal stability of novel proteins is assessed based on the premise that proteins that are less stable and denatured by heat are less likely to be allergenic or cause adverse health effects (Craig et al., 2008; Delaney et al., 2008). Aliquots of microbial-expressed Cry1F protein were subjected to different temperature regimens and applied to the surface of an insect diet provided to neonates of tobacco budworm (*H. virescens*). Diminished larval growth inhibition, demonstrating loss of bioactivity, indicated Cry1F protein was labile to heat at and above 75 °C for 30 min (Herman, 2000). When heated at 55 °C for 10 min, the PAT protein was denatured as corroborated by a loss of enzymatic activity (Wehrmann et al., 1996). Notably, the denatured PAT protein did not show any similarities with IgE epitopes of known allergenic proteins (Hérouet et al., 2005).

**Pepsin digestibility**

Important protein allergens have been shown to be stable to peptic digestion; therefore, protein resistance to pepsin indicates that further testing is required to determine allergenic potential (CAC, 2009; Ladics, 2008). Cry1F and PAT proteins were incubated in vitro with simulated gastric fluid (SGF) containing pepsin for different time periods and the digested products were analyzed using SDS-PAGE and western blot analyses. The results demonstrated that Cry1F protein was degraded in less than 1 minute (Evans, 1998) and PAT protein was digested to non-detectable levels within 5 seconds after addition of SGF containing pepsin (FSANZ, 2003). As the Cry1F and PAT proteins are readily digested by pepsin, there is a lower probability they would cause adverse health effects due to limited persistence in the mammalian digestive environment. The Cry1F and PAT proteins are not from allergenic sources, are heat labile, are rapidly degraded in simulated gastric fluid, and are not glycosylated. This evidence supports the conclusion that the allergenic potential of Cry1F and PAT proteins is low (Hérouet et al., 2005; Ladics et al., 2006).

**Mammalian Toxicology Assessment of Cry1F and PAT Proteins**

**Acute oral toxicity in mice**

To date, no known mammalian health effects have been associated with Cry1 proteins from Bt microbial products (Siegel, 2001; Delaney et al., 2008). Safety of the Cry1F protein was demonstrated in a 14-day acute oral toxicity study in mice (Kuhn, 1998). No mortality or clinical/behavioral signs of pathology were observed during the study, and all mice achieved normal relative weight gain. No treatment-related adverse effects were observed at Cry1F protein levels of >576 mg/kg bodyweight. This is equivalent to a dose of pure Cry1F protein of 34.56 g per person, assuming a body weight of 60 kg. Assuming an expression level observed in TC1507 maize grain (3.7 ng/mg tissue dry weight, Table 1), a person would have to consume 9,341 kg of maize for an equivalent exposure to Cry1F protein as the mice received in the acute oral toxicity study.

Safety of PAT protein was also demonstrated in a 2-week acute oral toxicity study in mice (Brooks, 2000). All mice survived the 2-week study and there were no treatment-related clinical observations of toxicity. All mice, with the exception of one female, gained body weight over the duration of the study and there were no gross pathologic lesions observed upon necropsy for any animal. Under the study conditions, the acute oral LD$_{50}$ (median lethal dose) of the PAT protein in mice was >6,000 mg/kg. These results are consistent with previous findings from acute oral toxicity studies indicating that the PAT protein presents no significant human health risk (US EPA, 1997; Health Canada, 1997). Cry1F and PAT proteins were not found to be acutely toxic to mice. These findings support the conclusion...
that Cry1F and PAT proteins are unlikely to be toxic to humans and other mammals, and can be considered safe for mammalian health through dietary consumption.

Subchronic rodent feeding study

A subchronic (90-day) rodent feeding study conducted with whole grains or processed feed fractions from GE crops is routinely requested by regulatory agencies, such as in the EU (Kuiper et al., 2013). The objective of a subchronic (90-day) rodent feeding study is to detect potential toxicological effects of the test diet compared with a control diet. While this study may be required to assess safety of an introduced protein or to identify unintended changes in metabolic pathways attributable to the genetic modification (EFSA, 2008), recent publications have discussed the limited contribution of such studies in the safety assessment (Kuiper et al., 2013; Herman and Ekmay, 2014). A review of published 90-day subchronic feeding studies demonstrated that GE crops do not pose any health hazard (Snell et al., 2012).

The nutritional performance of rats fed diets containing TC1507 maize grain was evaluated in a 90-day subchronic feeding study, in accordance with OECD guidelines (MacKenzie et al., 2007). Standard toxicological response variables were compared between rats fed diet containing 11% or 33% TC1507 maize grain and those in rats fed diet containing either a near-isoline control or non-GE commercial maize grain. The maize grain from TC1507, control, and non-GE commercial hybrids were comparable with respect to content of proximates, amino acids, minerals, anti-nutrients, and secondary metabolites. Individual diets formulated from the 3 hybrids were found to be nutritionally equivalent. There were no toxicologically significant differences in body weight and feed intake between the treatment groups. Neither mortality nor clinical signs of toxicity were observed in any of the treatment groups. Additionally, there were no toxicologically significant differences identified in ophthalmological and neurobehavioral responses, organ weights, and pathology between the treatment groups. Minor differences in a subset of hematological parameters were observed in females, which were not treatment related or biologically significant (MacKenzie et al., 2007). These findings demonstrated that TC1507 maize grain does not have the potential to cause significant toxicological effects in rodents, and that it is as safe as non-GE maize grain for human and animal consumption.

Nutritional Feeding Studies

Demonstrating the nutritional quality and equivalence of food and feed derived from GE crops is critical to ensure the well-being of humans and animals consuming such products. Livestock feeding studies with food derived from GE crops aim to evaluate the nutritional quality and wholesomeness of the novel food (Delaney, 2009). Such studies are generally recommended when there are substantial compositional changes or improved nutritional characteristics as a result of the modification in a GE crop (EFSA, 2008). Due to the widespread cultivation of TC1507 maize and the resultant abundance of GE grain for use in animal feed, livestock studies with TC1507 were designed to demonstrate nutritional equivalence for the purpose of gaining market acceptance. Parameters such as feed consumption, growth performance, and product quality (e.g. milk, meat, eggs) are quantified in these livestock studies to determine the wholesomeness of grain derived from TC1507 maize relative to non-GE maize.

Broiler chickens

Due to their rapid growth, broiler chickens are good animal models for the detection of even small dietary nutritional imbalances (EFSA, 2008). The performance of commercial broiler chickens (Cobb x Cobb strain) was examined by feeding the animals with diet containing TC1507 and non-GE grains for 42 d (McNaughton and Zeph, 2004). The maize grain was incorporated at the rate of 54.2% in starter diets and 57% in grower diets, across all treatments. The TC1507 grain diet treatment group
was compared with treatment groups fed diets containing either grain from a non-GE control maize hybrid or from one of 4 non-GE commercial maize sources. These findings revealed performance, as indicated by mortality, mean body weight, and feed conversion, was statistically similar among treatment groups. Therefore, TC1507 maize grain was nutritionally equivalent to maize grain from commercial hybrids when fed to broiler chickens.

**Laying hens**

In a similar study, grain from TC1507 maize was compared with grain from a near-isoline maize line and 2 non-GE conventional maize lines, incorporated at approximately 60% of diet, in a 16-week feeding trial with laying hens (Bovans White) (Scheideler et al., 2008). Hen performance, as measured by parameters including egg production and production efficiency as well as egg qualities such as albumen and color, was evaluated in the different diet treatment groups. The results demonstrated that performance of hens fed TC1507 grain was comparable to those fed grain from near-isoline maize or non-GE conventional maize.

**Beef heifers**

Sindt et al. (2007) compared the growth performance (daily weight gain, dry matter intake, feed efficiency) and carcass traits (liver abscess score, yield and quality grade) of beef heifers fed diets containing grain from a TC1507 maize hybrid with those fed diets containing grain from a near-isoline or one of 2 non-GE commercial maize hybrids. Diets incorporating steam-flaked maize at approximately 75% were individually fed to 20 beef heifers in each of 4 treatment groups for 118 d. The results indicated that growth performance and carcass characteristics of beef heifers were not significantly altered when provided diet containing TC1507 maize grain.

**Dairy cows**

Faust et al. (2007) evaluated the health and performance of lactating dairy cows fed maize silage containing maize grain, incorporated at a concentration of approximately 30%, derived from either TC1507 or near-isoline maize. Parameters such as milk production, production efficiency, and milk composition were compared in a replicated experiment, where 20 Holstein cows in each of 2 treatment groups were fed the maize diets for 28 d. The results demonstrated that the source of the maize grain and silage did not influence dairy production or health of the cows as assessed by physical characteristics, blood chemistry, and hematological indicators.

**Swine**

Stein et al. (2009) evaluated the growth performance and carcass composition of 24 pigs (offspring of Duroc x Large White sires mated to Yorkshire x Duroc x Landrace dams) in each of 4 treatment groups provided diet containing grain from either TC1507, near-isoline, or one of 2 non-GE commercial maize sources. Ground maize grain was incorporated into diets fed at 3 different growth phases. The concentrations in these successive phases were 65.1, 73.5, and 80.6%, respectively. Diets were formulated by mixing maize grain, soybean meal, soybean oil, vitamins, and minerals. Average daily weight gain, average daily feed intake, and gain/feed ratio were calculated to measure growth performance. Live weights at slaughter and standard carcass measurements were used to calculate dressing and lean meat percentages. There were no significant differences observed in the growth performance and carcass measurements between the 4 dietary treatment groups. These findings indicated that the presence of TC1507 grain did not impact the growth performance or carcass composition of pigs, and that these indices were comparable with those evaluated in pigs fed dietary treatments containing non-GE grain. Taken together, the various animal feeding studies support the conclusion that the TC1507 maize is as safe as, and nutritionally equivalent to, non-GE maize.

**Environmental Risk Assessment**

The environmental risk assessment for TC1507 maize evaluated the potential for
invasiveness (weediness), gene flow to sexually compatible wild relatives, horizontal gene transfer, and ecological effects including the potential impact on non-target organisms. Modern-day maize (\textit{Z. mays}) is highly domesticated and unable to establish self-sustaining populations without human intervention; therefore, cultivation of maize poses negligible risk to the environment as a weed (OECD, 2003; Raybould et al., 2012). It has been established that maize event TC1507 is substantially equivalent when compared with its non-GE counterpart. The Cry1F protein provides protection against insect damage from certain lepidopteran pests, which would not be expected to alter the persistence, invasiveness, or weediness of maize outside managed agriculture. The PAT protein confers tolerance to the glufosinate-ammonium herbicide active ingredient. Since glufosinate-ammonium is a broad-spectrum herbicide that is not routinely broadcast outside agricultural habitats, tolerance to this herbicide does not enhance the potential for persistence, invasiveness, or weediness of TC1507 maize in the environment. Thus, TC1507 is not expected to behave differently than conventional maize in terms of invasiveness potential. A 2-year field experiment in south Texas with 5 GE maize events, including TC1507, non-GE maize (4 near-isoline hybrids and a commercial hybrid), and 3 Mexican landraces demonstrated that the insect-resistant trait does not increase the invasiveness potential of TC1507 maize. Researchers concluded that cultivation of Bt maize, similar to non-GE maize, would pose negligible weediness risk (Raybould et al., 2012).

The potential for gene flow from TC1507 maize to its wild and weedy relatives was evaluated. Biology documents on the potential for gene flow in conventional maize have been published by the OECD (2003). Maize has a high outcrossing rate, and can pollinate sexually compatible varieties and hybrids (e.g., other cultivated maize hybrids, landraces, teosinte). However, gene flow in the environment is limited by environmental barriers (pollen viability, pollen dispersal, proximity, and synchrony of flowering) and genetic barriers (ability to outcross and produce fertile progeny). Outcrossing between domesticated maize and \textit{Tripsacum} species is unlikely under natural field conditions (OECD, 2003; US EPA, 2001). None of the genetic modifications in TC1507 maize were intended to alter the agronomy or composition of the TC1507 maize, relative to non-GE maize. As the agronomic characteristics were comparable between TC1507 maize and non-GE maize, there is no evidence to suggest that TC1507 maize has different reproductive biology or would not be subject to the same environmental and genetic barriers to gene flow as conventional maize.

The potential for horizontal gene transfer (HGT) of GE crop transgenes of microbial origin to human gut has been reviewed by Kleter et al. (2005) to suggest that transfer of a gene from GE plants to intestinal microflora is improbable. Plant DNA traversing the gastrointestinal tract and tolerating digestive enzymes, while maintaining the original coding information, is unlikely. Using a weight-of-evidence approach, Kleter et al. (2005) concluded that even in a rare event, HGT of \textit{cry} genes from GE crops to microbes is unlikely to cause pathogenicity in receiving microbes residing in humans and animals. Moreover, there is no evidence of HGT of \textit{pat} genes from GE crops to microorganisms (Hérouet et al., 2005; Kleter et al., 2005). The US EPA surmised HGT of Bt crop transgenes to soil microflora would be extremely rare and unlikely to increase soil microbial fitness (Mendelsohn et al., 2003). To date, there are no reports in the literature demonstrating that HGT occurs from plants to microorganisms, plants, animals, and humans under typical environmental conditions. Therefore, HGT from GE plants poses negligible risks to animal and human health or the environment (Keese, 2008). The \textit{cry1F} and \textit{pat} genes in TC1507 were derived from naturally occurring soil bacteria and are not pathogenic; therefore, microorganisms, plants, animals, and humans are regularly exposed to these organisms and their components without adverse consequences. Even if HGT were to occur, due to the absence of any selective advantage of any of these transgenes, there would be no increased risk of adverse effects attributed to HGT of transgenes in TC1507 maize (Mendelsohn et al., 2003).
A thorough environmental risk assessment (ERA) for the cultivation of TC1507 maize was conducted for non-target organisms (NTOs) present in the maize agro-ecosystem. Groups of NTOs that could be exposed to the Cry1F protein from a cultivated maize field were identified, and factors that affect the magnitude and duration of exposure in the environment were considered. The potential hazard of the Cry1F protein to NTOs in the environment was assessed using a tiered testing approach (Romeis et al., 2008). If no adverse effects are detected in early tier testing using unrealistically high Cry1F protein concentrations (e.g., 10X higher concentrations than those that would be encountered in the field), it can be concluded that at realistic environmental concentrations the risk to NTOs would be low. Laboratory bioassays were conducted on NTOs at high concentrations of Cry1F protein or using TC1507 maize tissue and no adverse effects were detected. The risk to each group of organisms was assessed by considering both the likelihood of exposure in the environment and the potential hazard caused by the Cry1F protein (Table 2).

There are many factors that mitigate the magnitude and duration of exposure of pollinators and pollen feeders to the Cry1F protein in TC1507 maize pollen. For example, many non-target lepidopterans are known to feed on host plants and are exposed to maize pollen indirectly if pollen is present on the host plant. In this case, exposure to maize pollen is limited to host plants that grow in close proximity to maize fields. There is a relatively short period of time when maize pollen is shed, which limits the duration of exposure. The timing of when maize pollen is shed and when the most sensitive life stages are foraging (generally neonates and early instars) may not overlap, which limits exposure. Pollen deposition rates, Cry protein stability in pollen, host plant density, cropping area, temporal and spatial overlap, and larvae feeding behavior are all important considerations that mitigate the magnitude and duration of exposure of pollinators and pollen feeders to Cry proteins in maize pollen (Sears et al., 2001). Hazard studies on honeybee (Apis mellifera) (Maggi, 1999) and monarch butterfly (Danaus plexippus) (Bystrak, 2000) demonstrate low hazard of the Cry1F protein at concentrations that exceed realistic environmental concentrations. Therefore, the risk of cultivation of TC1507 maize on pollinators and pollen feeders is low. Predators and parasitoids could be exposed to the Cry1F protein through secondary (prey-mediated) transfer. In general, Cry proteins have not been found to bioaccumulate in prey (Romeis and Meissle, 2011), therefore, potential exposure to predators and prey to the Cry1F protein is low. Hazard studies on green lacewing (Chrysoperla carnea) (Hoxter, Porch et al., 1999b), parasitic hymenoptera (Nasonia vitripennis) (Hoxter, Krueger et al., 1999a), and ladybird beetle (Hippodamia convergens) (Hoxter, Krueger et al., 1999b) demonstrate low hazard of the Cry1F protein at concentrations that exceed realistic environmental concentrations. Therefore, the risk of cultivation of TC1507 maize on predators and parasitoids is low. In general, Cry proteins do not persist or accumulate in soil. The environmental fate of a variety of Cry proteins in various soil types has been well characterized (Clark et al., 2005; Icoz and Stotzky, 2008). Like other Cry proteins, the soil dissipation of Cry1F proteins can be characterized as rapid (Herman et al., 2002; Shan et al., 2008), and the magnitude and duration of exposure of aquatic organisms and soil-dwelling organisms to the Cry1F protein is low. Hazard studies on water flea (Daphnia magna) (Drottar and Krueger, 1999), earthworm (Eisenia fetida) (Hoxter, Porch, et al., 1999a), and springtail (Folsomia candida) (Halliday, 1998) demonstrate low hazard of the Cry1F protein at concentrations that exceed realistic environmental concentrations. Therefore, the risk of cultivation of TC1507 maize on aquatic organisms and soil dwellers is low.

Subsequent to the early-tier laboratory studies that were conducted as part of the original TC1507 safety assessment, several additional studies have been conducted on NTOs including honeybee (A. mellifera) (Hanley et al., 2003), green lacewing (C. rufilabris) (Tian et al., 2013), larval endoparasitoid (Cotesia marginiventris) (Tian et al.,
| Surrogate Species for Hazard Testing (Common Name) | Exposure to Cry1F Protein from TC1507 Maize | Hazard of Cry1F Protein at Environmentally Relevant Concentrations | Environmental Risk Conclusion |
|--------------------------------------------------|---------------------------------------------|---------------------------------------------------------------|--------------------------------|
| Pollinators and pollen feeders                   |                                             |                                                               |                                |
| *Apis mellifera* *(Honeybee)*                    | Low; there are many mitigating factors that decrease the likelihood of exposure to Cry1F protein in TC1507 maize pollen. | Low; no hazard to *Apis mellifera* in laboratory testing using concentrations that exceed realistic environmental concentrations | Low risk to honeybee           |
| *Danaus plexippus* *(Monarch butterfly)*         |                                             |                                                               |                                |
| Predators and parasitoids                        |                                             |                                                               |                                |
| *Chrysoperla carnea* *(Green lacewing)*          | Low; Cry proteins are not likely to bioaccumulate in prey items. | Low; no hazard to *Chrysoperla carnea* in laboratory testing using concentrations that exceed realistic environmental concentrations | Low risk to predators and parasitoids |
| *Nasonia vitripennis* *(Parasitic hymenoptera)*  |                                             |                                                               |                                |
| *Hippodamia convergens* *(Ladybird beetle)*      |                                             |                                                               |                                |
| Aquatic organisms                                |                                             |                                                               |                                |
| *Daphnia magna* *(Water flea)*                   | Low; the concentration of Cry1F protein in aquatic habitats is low. | Low; no hazard to *Daphnia magna* in laboratory testing using concentrations that exceed realistic environmental concentrations | Low risk to aquatic organisms  |
| Soil-dwelling organisms                          |                                             |                                                               |                                |
| *Eisenia fetida* *(Earthworm)*                   | Low; the concentration of Cry1F protein in soil is low, indicating low magnitude of exposure. The dissipation of the Cry1F protein in soil is rapid, indicating low duration of exposure. | Low; no hazard to *Eisenia fetida* in laboratory testing using concentrations that exceed realistic environmental concentrations | Low risk to soil-dwelling organisms |
| *Folsomia candida* *(Springtail)*                |                                             |                                                               |                                |

*a*Maggi (1999).
*b*Bystrak (2000).
*c*Hoxter, Porch et al. (1999b).
*d*Hoxter, Krueger et al. (1999a).
*e*Hoxter, Krueger et al. (1999b).
*f*Drottar and Krueger (1999).
*g*Hoxter, Porch et al. (1999a).
*h*Halliday (1998).
*i*Romeis and Meissel (2011).
*j*Herman et al. (2002); Shan et al. (2008).
2014), pale grass blue butterfly (*Pseudozizeeria maha*) (Wolt et al., 2005), and bobwhite quail (*Colinus virginianus*) (Gallagher et al., 1999) that support the lack of adverse effects of the Cry1F protein. Furthermore, multiple field studies have been conducted in different global geographies, including Vietnam, the US, Spain, France, Philippines, India, and Indonesia, to support regulatory submissions (Pioneer Hi-Bred International, Inc. internal unpublished data). These additional laboratory and field studies all support the conclusion of low environmental risk associated with the cultivation of TC1507 maize. The environmental fate, specificity to lepidopteran pest species, and lack of effects on NTOs of the Cry1F protein are well-characterized. Based on this characterization, the environmental risk associated with the cultivation of TC1507 maize is low.

**Yield Increase/Economic Benefits**

Invariably, increasing yield is the priority when any new crop technologies or hybrids are developed. In the US, data collected between 1964 and 2010 revealed GE traits, since 1996, have had a significant positive impact on maize yield trends (Xu et al., 2013). To this end, Bt maize offers a highly efficient pest control measure that allows growers to produce high-quality grain with reduced insecticide inputs and farm operations, which can contribute to the reduction of greenhouse gas emissions (Barfoot and Brookes, 2014).

Event TC1507 is a popular component among the many Bt maize breeding stacks planted. In maize yield evaluations held in the US during 2011 and 2012, there were 8,431 and 8,263 entries, respectively, under different categories. Among the entries with the highest yield, 59% of hybrids in 2011 and 56% of hybrids in 2012 contained event TC1507 stacked with other insect-resistant or herbicide-tolerant traits using conventional breeding techniques (NCGA, 2011; NCGA, 2012).

As event TC1507 is predominantly planted as part of a breeding stack (ISAAA, 2015), agronomic studies specifically evaluating the yield performance of TC1507 as a single event product compared with conventional hybrids

| Country      | Food direct use or additive | Feed direct use or additive | Cultivation domestic or non-domestic use |
|--------------|-----------------------------|-----------------------------|-----------------------------------------|
| 1 Argentina  | 2005                        | 2005                        | 2005                                    |
| 2 Australia  | 2003                        |                             |                                         |
| 3 Brazil     | 2008                        | 2008                        | 2008                                    |
| 4 Canada     | 2002                        | 2002                        | 2002                                    |
| 5 China      | 2002<sup>a</sup>            | 2002<sup>a</sup>            |                                         |
| 6 Colombia   | 2006                        | 2006                        | 2007                                    |
| 7 European Union | 2006<sup>b</sup>      | 2006<sup>b</sup>            | 2009                                    |
| 8 Honduras   |                             |                             |                                         |
| 9 Japan      | 2002                        | 2002                        | 2005                                    |
| 10 Malaysia  | 2013                        |                             | 2013                                    |
| 11 Mexico    | 2003                        |                             |                                         |
| 12 New Zealand | 2003                     |                             |                                         |
| 13 Panama    | 2012                        |                             | 2012                                    |
| 14 Paraguay  | 2012                        | 2012                        | 2012                                    |
| 15 Philippines| 2003<sup>c</sup>          | 2003<sup>c</sup>           | 2013                                    |
| 16 Singapore | 2014                        |                             |                                         |
| 17 South Africa | 2002                     |                             | 2012                                    |
| 18 Korea     | 2002                        |                             | 2004                                    |
| 19 Taiwan    | 2003                        |                             |                                         |
| 20 Turkey    |                             |                             | 2011                                    |
| 21 USA       | 2001                        | 2001                        | 2001                                    |
| 22 Uruguay   | 2011                        | 2011                        | 2011                                    |

<sup>a</sup>Renewal 2009, 2012; <sup>b</sup>Expires 2016; <sup>c</sup>Renewal 2008.

Based on ISAAA (2015).
are limited. However, published field studies have reported that TC1507 maize prevented significant yield loss due to *S. frugiperda* infestation compared with non-Bt maize as evidenced by reduced foliar injury, whorl damage, and larval survivorship (Buntin, 2008; Siebert et al., 2008; Hardke et al., 2011).

In the Philippines, two TC1507 maize hybrids, their near-isoline hybrids, and a conventional local hybrid were planted in 12 locations over 2 seasons during 2006–2007, to evaluate their performance against the Asian corn borer (*Ostrinia furnacalis*). Significantly lower insect damage and higher yields were observed with the TC1507 event compared with near-isoline hybrids across the multi-site trials (Thompson et al., 2010).

Global Regulatory Acceptance and Commercial Status of TC1507 Maize

Maize with the Herculex® I trait has been authorized in most of the major grain trading countries in the world. To date, more than 20 countries have authorized use of TC1507 for food and/or feed purposes, of which 10 grow it in commercial scale (Table 3) (ISAAA, 2015). In the US, field trials conducted from 1997 to 2000 in at least 20 States and in Puerto Rico demonstrated that event TC1507 exhibited the desired agronomic characteristics and did not pose a plant pest risk prior to authorization from the USDA and EPA, and prior to review by the US Food and Drug Administration (FDA) in 2001 (Mendelsohn et al., 2003; USDA APHIS, 2001; US FDA, 2001). Authorization from the Canadian Food Inspection Agency followed in 2002 (CFIA, 2002). Thereafter in 2003, TC1507 was launched for commercial cultivation and for food/feed uses in the US and Canada (Rowe et al., 2012). Subsequently, TC1507 has become a common component in breeding stack products in the US (Table 4). For example in 2010, more than 150 hybrids contained event TC1507 either as a single event product (26; number of US maize hybrids) or in breeding stack products: TC1507 x NK603 (10), TC1507 x 59122 (31), TC1507 x 59122 x NK603 (37), and MON88017 x MON89034 x TC1507 x 59122 (52) (McLaren and Copping, 2011).

TC1507 was first authorized for commercial planting in Argentina in 2005 and maize containing events stacked using conventional breeding techniques including TC1507 were authorized soon thereafter (Trigo, 2011). In Honduras, field trials with TC1507 were initiated in 2006, followed in 2009 by initial limited scale commercialization and subsequent full commercialization in 2010 (GAIN Honduras, 2012). TC1507 has been approved for import

| Breeding stack products with TC1507 | Commercial name |
|-------------------------------------|-----------------|
| TC1507 x 59122                      | Herculex® XTRA® |
| TC1507 x NK603                      | Herculex® I Roundup Ready® |
| TC1507 x 59122 x NK603              | Herculex® XTRA® Roundup Ready® |
| TC1507 x MON810 x NK603             | Optimum® Intrasect® Roundup Ready® |
| TC1507 x 59122 x MON810             | Optimum® Intrasect® XTRA® |
| TC1507 x 59122 x MON810 x NK603     | Optimum® Intrasect® XTRA® Roundup Ready® |
| MON89034 x TC1507 x NK603           | Power Core™ XTM |
| MON89034 x TC1507 x NK603 x DAS40278| Power Core™ Enlist™ |
| MON89034 x TC1507 x MON88017 x 59122x DAS40278 | SmartStax® Enlist™ |
| TC1507 x 59122 x MON810 x MIR604 x NK603 | Optimum® Intrasect® XTreme |
| TC1507 x MIR604 x NK603             | Optimum® TRIssect® |
| Bt11 x MIR162 x TC1507 x GA21       | Agrisure Vipera® 3220 |
| Bt11 x 59122 x MIR604 x TC1507 x GA21| Agrisure® 3122 |
| 5307 x MIR604 x Bt11 x TC1507 x GA21| Agrisure Duracade® 5222 |
| 5307 x MIR604 x Bt11 x TC1507 x GA21| Agrisure Duracade® 5122 |
| TC1507 x MON810                      | Optimum® Intrasect® |

Based on ISAAA (2015).
The Brazilian Regulatory Authority, Comissão Técnica Nacional de Biossegurança (CTNBio), assessed safety of TC1507 and authorized commercial cultivation in 2008 (CTNBIO, 2008). Subsequently, breeding stacks containing TC1507, such as TC1507 × NK603 and MON89034 × TC1507 × NK603, were also authorized in 2009 and 2010, respectively. As of early 2011, 448 total GE maize varieties, derived from 9 different transformation events, were registered in Brazil. Of these, 120 varieties carried TC1507 as a single event product and 44 were breeding stack products containing TC1507 × NK603 (Marinho et al., 2012).

Event TC1507 maize received full European Union (EU) authorization for import, food, and feed in March, 2006, following safety assessments from the European Food Safety Authority (EFSA) including molecular characterization, toxicology, and allergenicity as well as agronomic and compositional equivalence. These EFSA Opinions concluded that 1507 maize is unlikely to have an adverse effect on human health or the environment in the context of its use as or in food, feed, and processing (EFSA, 2004, 2005a, 2005b). The authorization was renewed in 2011 following a further positive EFSA Opinion in May, 2009 (EFSA, 2009a). Moreover, EFSA has published several other safety Opinions on breeding stack products containing the 1507 event, notably 59122 × 1507 × NK603 (EFSA, 2009b), MON89034 × 1507 × MON88017 × 59122, and all sub-combinations of the individual events for food and feed uses, import, and processing (EFSA, 2010). Multiple positive EFSA Opinions regarding safety have been received on a currently pending 1507 maize EU cultivation submission made in 2001 (Rowe et al., 2012).

In Asia, GE maize is largely imported for food, feed, and processing (FFP) use in Japan, Korea, Taiwan, and China. As of 2013, Taiwan has authorized 18 single maize events and 32 breeding stack event combinations for FFP purposes only. The single event TC1507 was registered in 2003 followed by at least 12 breeding stack event combinations containing TC1507 (FDA, 2014). Korea imported 8.2 million metric tons of GE maize for food and feed use in 2012. The use of TC1507 maize for food and for feed in Korea was authorized in 2002 and 2004, respectively, which was followed by authorization of TC1507 × NK603 maize for food and for feed uses in 2004 and 2008, respectively. Later, 12 GE maize breeding stack products containing event TC1507 were authorized (GAIN Korea, 2013). To date, the Philippines is the only Asian country where GE maize is commercially cultivated and consumed, which has been on-going since 2003. In 2013, GE maize was planted in 750,000 hectares of which 90% was stacked maize expressing a Bt trait (James, 2013). Field trials to obtain cultivation authorization for TC1507 as a single event product and in the TC1507 × MON810 × NK603 breeding stack product were completed in 2012. In addition, TC1507 maize and TC1507 × MON810 × NK603 maize were determined to be as safe as conventional counterparts for FFP, and have been allowed for import since 2003 and 2012, respectively (DA-BPI, 2012, 2013a). TC1507 and the following breeding stack products: TC1507 × MON810 × NK603, TC1507 × MON810, and TC1507 × NK603 were recently approved for cultivation in the Philippines (DA-BPI, 2013b). Event TC1507 has also received full approval in South Africa including a general release permit for commercial cultivation in 2012 (DAFF, 2014). Efforts are underway to obtain authorizations to cultivate TC1507 in other countries where farmers will benefit from an insect control perspective.

Conclusions

Maize is an important food crop grown widely in many countries, while primarily cultivated in North and South America, EU, China, Indonesia, and India. Maize grain and its derivatives have been safely consumed by humans for centuries without health concern. GE maize was introduced in 1996 and has been commercially cultivated on millions of hectares in 17 countries without any reported safety incidents. In addition to countries with on-going
commercial cultivation, Japan, Mexico, Korea, Taiwan, and China use significant quantities of GE maize obtained through import for food and feed purposes (Rowe et al., 2012). Other than the presence of the introduced gene(s), GE maize varieties are comparable with their non-GE counterparts with respect to composition, nutrition, and safety.

The robust information generated from over a decade of food, feed, and environmental safety assessments has established that TC1507 is as safe as conventional maize. Laboratory and field experiments conducted with TC1507 demonstrated that the introduced genes are stably integrated and follow the expected Mendelian inheritance pattern for a dominant gene. Additionally, these studies indicated TC1507 maize is substantially equivalent to its non-GE counterpart. A thorough safety assessment has been conducted, and no adverse effects are anticipated on non-target organisms from cultivation of TC1507 maize. It is unlikely that TC1507 maize would become a weed or that the introduced genes would flow to related wild species or other microorganisms resulting in a deleterious environmental impact. Similar conclusions were drawn by OECD and Center for Environmental Risk Assessment-International Life Sciences Institute while reviewing the food, feed and environmental safety of Cry1F protein (OECD, 2007; CERA, 2013).

Results from safety studies on Cry1F and PAT proteins and grains derived from TC1507 suggest TC1507 is unlikely to impact mammalian health through dietary consumption. The safety information provided here supports the conclusion that TC1507 presents negligible risk to human health and low risk to the environment.

Safety data on TC1507 have been submitted to regulatory agencies in over 20 countries that have authorized its use for cultivation and/or food and feed uses; 10 of which have been consuming TC1507 grains for at least a decade. Testimony to TC1507 product safety includes the extensive cultivation of TC1507 maize hybrids in Argentina, Brazil, Canada, and the US as well as its consumption in over 20 countries without any safety issues. Increasing authorization of event TC1507 as a component of breeding stack products further demonstrates the safety and global acceptance of TC1507. As TC1507 maize has been widely cultivated and consumed by humans and animals without incident, and in combination with the extensive safety data available, it is therefore concluded that TC1507 maize has a history of safe use for cultivation and food/feed purposes.

**DISCLOSURE OF POTENTIAL CONFLICTS OF INTEREST**

GBB, BD, TLF, GSL, RJL, MEHL, JS, and JAA are employed with DuPont Pioneer; RAH and SLE are employed with Dow AgroSciences LLC, both of which market transgenic seed, including TC1507 maize.

**REFERENCES**

Bajaj S, Mohanty A. Recent advances in rice biotechnology – towards genetically superior transgenic rice. Plant Biotechnol J 2005; 3:275–307; PMID:17129312; http://dx.doi.org/10.1111/j.1467-7652.2005.00130.x
Barfoot P, Brookes G. Key global environmental impacts of genetically modified (GM) crop use 1996–2012. GM Crops and Food 2014; 5(2); PMID:24637726; http://dx.doi.org/10.4161/gmcr.28449
Brooks KJ. (Toxicology & Environmental Research and Consulting, The Dow Chemical Company, Midland, MI). PAT Microbial Protein (FL): acute oral toxicity study in CD-1 mice. 2000 Jan. 45p. Indianapolis, IN: Dow AgroSciences LLC; Unpublished Technical Report No.: 991249
Buntin GD. Corn expressing Cry1Ab or Cry1F endotoxin for fall armyworm and corn earworm (Lepidoptera: Noctuidae) management in field corn for grain production. Fla Entomol 2008; 91(4):523–30; http://dx.doi.org/10.1653/0015-4040-91.4.523
Bystrak P. Toxicity of the Cry1F protein to neonate larvae of the monarch butterfly. Huxley, IA: Mycogen Seeds; 2000 May; 23p. Unpublished Technical Report No.: GH-C 5073
CAC. Guideline for the conduct of food safety assessment of foods derived from recombinant-DNA plants. CAC/ GL 45-2003; 2nd edition. Geneva: Codex Alimentarius Commission; 2009.
Castle LA, Wu G, McElroy D. Agricultural input traits: past, present and future. Curr Opin Biotechnol 2006; 17(2):105–12; PMID:16483761; http://dx.doi.org/10.1016/j.copbio.2006.01.011
A review of the environmental safety of the Cry1F protein. Washington, DC: Center for Environmental Risk Assessment, ILSI Research Foundation [Internet]. 2013 [cited 2014 Jan 29]. Available from: http://www.cera-gmc.org/files/cera/uploads/Cry1f-monograph-rev1.pdf

Craig W, Tepfer M, Degrassi G, Ripandelli D. An overview of general features of risk assessments of genetically modified crops. Euphytica 2008; 164:853–80; http://dx.doi.org/10.1007/s10681-007-9643-8

CTNBIO. Technical Opinion no. 1679/2008 - Commercial release of genetically modified corn, Herculex corn (TC1507) [Internet]. 2008 [cited 2013 Dec 12]. Available from: http://cera-gmc.org/docs/decdocs/09-060-001.pdf

Delaney B. Safety assessment of foods obtained from crops developed using biotechnology. Gen Appl Sys Toxicol 2009; 1–14; http://dx.doi.org/10.1002/9780470744307.gat139

Delaney B, Astwood JD, Cunhy H, Conn RE, Herouet GC, Macintosh S, Meyer LS, Privalle L, Gao Y, Mattsson J, Levine M. ILSI International Food Biotechnology Committee Task Force on Protein Safety. Evaluation of protein safety in the context of agricultural biotechnology. Food Chem Toxicol 2008; 46(2): S71–97; PMID:18348900; http://dx.doi.org/10.1016/j.fct.2008.01.045

Drottar KR, Krueger HO. (Wildlife International Ltd., Easton, MD). Bt Cry1F delta-endotoxin: A 48-hour static-renewal acute toxicity test with the cladoceran (Daphnia magna) using bacterially expressed Bt Cry1F delta-endotoxin, and pollen from maize expressing Bt Cry1F delta-endotoxin. 1999 Sept. 21p. San Diego, CA: Dow AgroSciences/Mycogen Corporation; Unpublished Technical Report No.: 354A-111

EFSA. Opinion of the scientific panel on genetically modified organisms on a request from the commission related to the notification (Reference C/ES/01/01) for the placing on the market of insect-tolerant genetically modified maize 1507, for import and processing, under Part C of Directive 2001/18/EC from Pioneer Hi-Bred International/Mycogen Seeds (Question No EFSA-Q-2004-011). EFSA J 2004a; 124: 1–18; http://dx.doi.org/10.2903/j.efsa.2004.124

EFSA. Opinion of the Scientific Panel on Genetically Modified Organisms on an application (reference EFSA-GMO-NL-2004-02) for the placing on the market of insect-tolerant genetically modified maize 1507, for food use, under Regulation (EC) No 1829/2003 from Pioneer Hi-Bred International/Mycogen Seeds (Question No EFSA-Q-2004-087). EFSA J 2005a; 182:1–22; http://dx.doi.org/10.2903/j.efsa.2005.182

EFSA. Opinion of the Scientific Panel on Genetically Modified Organisms on a request from the Commission related to the notification (Reference C/ES/01/01) for the placing on the market of insect-tolerant genetically modified maize 1507, for import, feed and industrial processing and cultivation, under Part C of Directive 2001/18/EC from Pioneer Hi-Bred International/Mycogen Seeds (Question No EFSA-Q-2004-072). EFSA J 2005b; 181:1–33; http://dx.doi.org/10.2903/j.efsa.2005.181

EFSA. Safety and nutritional assessment of GM plants and derived food and feed: The role of animal feeding trials. Food Chem Toxicol 2008; 46(Suppl. 1): S2–70; PMID:18328408; http://dx.doi.org/10.1016/j.fct.2008.02.008

EFSA. Scientific Opinion of the Panel on Genetically Modified Organisms on an application (EFSA-GMO-RX-1507) for renewal of authorisation for the continued marketing of existing products produced from...
maize 1507 for feed use, under Regulation (EC) No 1829/2003 from Pioneer Hi-Bred International, Inc. / Mycogen Seeds. EFSA J 2009a; 1138:1–11; http://dx.doi.org/10.2903/j.efsa.2009.1138

EFSA. Scientific Opinion of the Panel on Genetically Modified Organisms on an application (Reference EFSA-GMO-UK-2005-21) for the placing on the market of the insect-resistant and herbicide-tolerant genetically modified maize 59122 × 1507 × NK603 for food and feed uses, import and processing under Regulation (EC) No 1829/2003 from Pioneer Hi-Bred International, Inc. EFSA J 2009b; 1050:1–32; http://dx.doi.org/10.2903/j.efsa.2009.1050

EFSA. Scientific Opinion of the Panel on Genetically Modified Organisms on an application (EFSA-GMO-CZ-2008-62) for the placing on the market of insect resistant and herbicide tolerant genetically modified maize MON 89034 × 1507 × MON88017 × 59122 and all sub-combinations of the individual events as present in its segregating progeny, for food and feed uses, import and processing under Regulation (EC) No 1829/2003 from Dow AgroSciences and Monsanto. EFSA J 2010; 8:1–37; http://www.efsa.europa.eu/en/search/doc/1781.pdf

Estruch JJ, Warren GW, Mullins MA, Nye GI, Craig JA, Koziel MG. Vip3A, a novel Bacillus thuringiensis vegetative insecticidal protein with a wide spectrum of activities against lepidopteran insects. Proc Natl Acad Sci U S A 1996; 93(11):5389–94; PMID:8643585; http://dx.doi.org/10.1073/pnas.93.11.5389

Evans SL. Equivalency of microbial and maize expressed Cry1F protein; characterization of test substances for biochemical and toxicological studies. San Diego, CA: Mycogen Corporation c/o Dow AgroSciences LLC; Unpublished Technical Report No.: MYCO98-001

Evans SL. Producing proteins derived from genetically modified organisms for toxicity and environmental fate assessment of biopesticides. In: The GMO handbook: genetically modified animals, microbes, and plants in biotechnology, Parekh SR, editor. Totowa, NJ: Humana Press Inc; 2004; 53–83

Faust M, Smith B, Rice D, Owens F, Hinds M, Dana G, Hunst P. Performance of lactating dairy cows fed silage and grain from a maize hybrid with the Cry1F trait versus its nonbiotech counterpart. J Dairy Sci 2007; 90(12):5706–13; PMID:18024763; http://dx.doi.org/10.3168/jds.2007-0480

FDA. Current approvals of genetically modified foods in Taiwan [Internet]. Published by the Food and Drug Administration, Ministry of Health and Welfare. 2014 [cited 2014 Sep 30]. Available from: https://consumer.fda.gov.tw/Food/GmoInfoEn.aspx?nodeID=300#

FSANZ. Final assessment report – Insect-protected and glufosinate tolerant corn line 1507. Canberra, Australia: Food Standards Australia New Zealand [Internet]. 2003 [cited 2013 July 01]. Available from: http://cera-gmc.org/docs/dedocs/05-246-003.pdf

GAIN Colombia – Agricultural Biotechnology Annual. GAIN Report. USDA Foreign Agricultural Service [Internet]. 2013 [cited 2013 Dec 12]. Available from: http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Agricultural%20Biotechnology%20Annual_Bogota_Colombia_6-12-2013.pdf

GAIN Honduras – Agricultural Biotechnology Annual. GAIN Report #HOBT-2012. USDA Foreign Agricultural Service [Internet]. 2012 [cited 2013 Dec 12]. Available from: http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Agricultural%20Biotechnology%20Annual_Tegucigalpa_Honduras_7-16-2012.pdf

GAIN Korea – Agricultural Biotechnology Annual. GAIN Report # KS1336. USDA Foreign Agricultural Service [Internet]. 2013 [cited 2013 Dec 12]. Available from: http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Agricultural%20Biotechnology%20Annual_Seoul_Korea%20-%20Republic%20of%20Sep_17-2013.pdf

Gallagher SP, Grimes J, Beavers JB. Wildlife International Ltd., Easton, MD. Transgenic corn expressing Bacillus thuringiensis var. aizawai (Bt) Cry1F delta-endotoxin: A dietary toxicity study with the Northern Bobwhite. 1999 Jul. 38p. San Diego, CA: Mycogen c/o Dow AgroSciences LLC Corporation; Unpublished Technical Report No.: 354-116

Halliday WR. Department of Analytical and Biological Services, Ricerca, Inc., Painesville, OH. Chronic exposure of Folsomia candida to bacterially expressed Cry1F protein. 1998 Dec. 122p. San Diego, CA: Mycogen Corporation; Unpublished Technical Report No.: 7535-98-0078-AC-001

Hammond BG, Koch, MS. A review of the food safety of Bt crops. In Bacillus thuringiensis Biotechnology, Sansinena E. (ed.). New York, NY: Springer Science; 2012; 305–25; http://dx.doi.org/10.1007/978-94-007-3021-2_16

Hanley AV, Huang ZY, Pett WL. Effects of dietary transgenic Bt corn pollen on larvae of Apis mellifera and Galleria mellonella. J Apic Res 2003; 42(4):77–81

Hardke JT, Leonard BR, Huang F, Jackson RE. Damage and survivorship of fall armyworm (Lepidoptera: Noc-tuidae) on transgenic field corn expressing Bacillus thuringiensis Cry proteins. Crop Prot 2011; 30(2):168–72; http://dx.doi.org/10.1016/j.cropro.2010.10.005

Health Canada. Novel food information - food biotechnology: Glufosinate ammonium tolerant corn (T14 & T25) [Internet]. 1997 [cited 2013 December 11]. Available from: http://www.hc-sc.gc.ca/fn-an/gmf-agm/appro/32bg_agrevo-ct_agrevo-eng.php

Heide BR, Heir E, Holck A. Detection of eight GMO maize events by qualitative, multiplex PCR and fluorescence capillary gel electrophoresis. Eur Food Res Technol 2008; 227(2):527–35; http://dx.doi.org/10.1007/s00217-007-0751-4
Herman RA. Thermolability of Cry1F (truncated) delta-endotoxin. Indianapolis, IN: Dow AgroSciences LLC; 2000 Nov. 13p. Unpublished Technical Report No.: GH-C 5144

Herman RA, Ekmay R. Do whole-food animal feeding studies have any value in the safety assessment of GM crops? Reg Toxicol Pharma 2014; 68(1):171–4; PMID: 23851038; http://dx.doi.org/10.1016/j.yrtph.2013.07.003

Herman RA, Wolt JD, Halliday WR. Rapid degradation of the Cry1F insecticidal crystal protein in soil. J Agric Food Chem 2002; 50(24):7076–8; PMID:12428962; http://dx.doi.org/10.1021/jf025630u

Herouet C, Esdaile DJ, Mallyon BA, Debruyne E, Schulz A, Currier T, Hendricks K, van der Klis RJ, Rouan D. Safety evaluation of the phosphinothricin acetyltransferase proteins encoded by the pat and bar sequences that confer tolerance to glufosinate-ammonium herbicide in transgenic plants. Regul Toxicol Pharmaco 2005; 41(2):134–49; PMID:15698537; http://dx.doi.org/10.1016/j.yrtph.2004.11.002

Holck AL, Dromtorp SM, Heir E. Quantitative, multiplex ligation-dependent probe amplification for the determination of eight genetically modified maize events. Eur Food Res Technol 2009; 230(2):185–94; http://dx.doi.org/10.1007/s00217-009-1155-4

Hoxter K, Krueger H, Porch J. (Wildlife International Ltd., Easton, MD). Cry1F Bacillus thuringiensis var. aizawai delta-endotoxin: A dietary toxicity study with parasitic Hymenoptera. 1999a Dec. 39p. San Diego, CA: Dow AgroSciences LLC/Mycogen Corporation; Unpublished Technical Report No.: 354-114D

Hoxter K, Krueger H, Porch J. (Wildlife International Ltd., Easton, MD). Cry1F Bacillus thuringiensis var. aizawai delta-endotoxin: A dietary toxicity study with the ladybird beetle. 1999b Dec. 38p. San Diego, CA: Dow AgroSciences LLC/Mycogen Corporation; Unpublished Technical Report No.: 354-113B

Hoxter KA, Porch JR, Krueger HO. (Wildlife International Ltd., Easton, MD). Cry1F Bacillus thuringiensis var. aizawai delta-endotoxin: An acute toxicity study with the earthworm in an artificial soil substrate. 1999a Dec. 40p. San Diego, CA: Dow AgroSciences LLC/Mycogen Corporation; Unpublished Technical Report No.: 354-112

Hoxter KA, Porch J, Krueger HO. (Wildlife International Ltd., Easton, MD). Cry1F Bacillus thuringiensis var. aizawai delta endotoxin: A dietary toxicity study with green lacewing larvae. 1999b Dec. 28p. San Diego, CA: Mycogen c/o Dow AgroSciences LLC; Unpublished Technical Report No.: 354-115A

Icoz I, Stotzky G. Fate and effects of insect-resistant Bt crops in soil ecosystems. Soil Biol Biochem 2008; 40:559–86; http://dx.doi.org/10.1016/j.soilbio.2007.11.002

ISAAA Briefs Nos. 1, 5, 8, 12, 17, 23, 26, 29, 30, 32, 34, 35, 37, 39, 41-44 & 46 [Internet]. 2014 [cited 2014 September 10]. Available from: http://www.isaaa.org/resources/publications/briefs/default.asp

ISAAA. GM approval database [Internet], 2015 [cited 2015 March 06]. Available from: http://www.isaaa.org/gmapprovaldatabase/

James C. Global status of commercialized Biotech/GM Crops: 2013. ISAAA Brief No. 46. Ithaca, NY: ISAAA. ISBN: 978-1-892456-55-9; 2013

Keepe P. Risks from GMOs due to horizontal gene transfer. Environ Biosafety Res 2008; 7(3):123–49; PMID: 18801324; http://dx.doi.org/10.1016/j.ebr.2008014

Kim JH, Kim SY, Lee H, Kim YR, Kim HY. An event-specific DNA microarray to identify genetically modified organisms in processed foods. J Agric Food Chem 2010; 58(10):6018–26; PMID:20438128; http://dx.doi.org/10.1021/jf100351x

Kleter GA, Peijnenburg AA, Aarts HJ. Health considerations regarding horizontal transfer of microbial transgenes present in genetically modified crops. J Biomed Biotechnol 2005; 2005(4):326–52; PMID:16489267; http://dx.doi.org/10.1155/JBB.2005.326

Kuhn JO. (StillMeadow Inc., Sugar Land, TX). Acute oral toxicity study in mice. 1998 Sept. 11p. San Diego, CA: Mycogen; Unpublished Technical Report No.: 4281-98

Kuiper HA, Kok EJ, Davies HV. New EU legislation for risk assessment of GM food: no scientific justification for mandatory animal feeding trials. Plant Biotechnol J 2013; 11(7):781–84; PMID:23786622; http://dx.doi.org/10.1111/pbi.12091

La Paz JL, García-Muniz N, Nadal A, Esteve T, Puigdomènech P, Pla M. Inter-laboratory transfer of a real-time polymerase chain reaction assay for quantitative detection of genetically modified maize event TC1507. J AOAC Int 2006; 89(5):1347–52; PMID:17042186

Ladies GS. Current codex guidelines for assessment of potential protein allergenicity. Food Chem Toxicol 2008; 46(10):S20–3; PMID:18708115; http://dx.doi.org/10.1016/j.fct.2008.07.021

Ladies GS, Bardina L, Cressman RF, Mattsson JL, Sampson HA. Lack of cross-reactivity between the Bacillus thuringiensis derived protein Cry1F in maize grain and dust mite Der p7 protein with human sera positive for Der p7-IgE. Regul Toxicol Pharmacol 2006; 44(2):136–43; PMID:16406630; http://dx.doi.org/10.1016/j.yrtph.2005.11.005

Ladies GS, Cressman RF, Herouet GC, Herman RA, Privalle L, Song P, Ward JM, McClain S. Bioinformatics and the allergy assessment of agricultural biotechnology products: industry practices and recommendations. Regul Toxicol Pharmacol 2011; 60(1):46–53; PMID:21320564; http://dx.doi.org/10.1016/j.yrtph.2011.02.004

MacKenzie SA, Lamb I, Schmidt J, DeGe L, Morrisey MJ, Harper M, Layton RJ, Prochaska LM, Sanders C,
Locke M, Mattsson JL, Fuentes A, Delaney B. Thirteen-teen feeding study with transgenic maize grain containing event DAS-Ø1507-1 in Sprague-Dawley rats. Food Chem Toxicol 2007; 45(4):551–62; PMID: 17097206; http://dx.doi.org/10.1016/j.fct.2006.09.016

Maggi VL. (California Agricultural Research, Inc., Ker-
man, CA). Evaluation of the dietary effect(s) on hon-
eybee development using bacterially expressed Bt
Cry1F delta-endotoxin and pollen from maize expres-
sing Bt Cry1F delta endotoxin. 1999 Dec. 53p. San
Diego, CA: Mycogen c/o Dow AgroSciences LLC;
Unpublished Technical Report No.: CAR 172-99

Makarevitch I, Svitashev SK, Somers DA. Complete
sequence analysis of transgenic loci from plants trans-
formed via microprojectile bombardment. Plant
Molecular Biology 2003; 52(2):421–32; PMID: 12856947; http://dx.doi.org/10.1023/A:1023968920830

Marinho CD, Martins FJ, Amaral J, Gonçalves
LS, Amaral SC, de Mello MP. Use of transgenic seeds in Brazilian agriculture and concentration of agricultural production to large agribusinesses. Genet Mol Res 2012; 11(3):1861–80; PMID:22869542; http://dx.
doi.org/10.4238/2012.July.19.6

McDougall P. The cost and time involved in the discovery,
development and authorisation of a new plant biotech-
ology derived trait. A Consultancy Study for CropLife International [Internet]. 2011 [cited 2015 June 01]. Available from: http://crolife.org/wp-content/uploads/2014/04/Getting-a-Biotech-Crop-to-
Market-Phillips-McDougall-Study.pdf

McLaren J, Copping L. Transgenic maize - the registration
status of lines that have been commercialised: the first in a series that examines the GM crop market. Outlooks Pest Manage 2011; 22(2):66–73; http://dx.
doi.org/10.1564/22apr07

McNaughton JL, Zeph L. Broiler study nutritional evalua-
tion of Bt Cry1F maize corn from Bacillus thuringiensis
subsp. aizawai and phosphinothricin-n-acetyltransferase. Poult Sci 2004; 83(Suppl. 1):s399–400; http://www.poul
tryscience.org/meeting-abstracts/jam04/398.PDF

Mendelsohn M, Kough J, Vaituzis Z, Matthews K. Are Bt
coats safe? Nat Biotechnol 2003; 21(9):1003–9; PMID:12949561; http://dx.doi.org/10.1038/nbt0903-
1003

Meyer T. Comparison of amino acid sequence similarity of Cry1F and PAT proteins to known allergen proteins. Johnston, IA: Pioneer Hi-Bred International, Inc.; 1999 Aug. 24p. Unpublished Technical Report No.: PH99-013.

NCGA. National corn yield contest: 2011 Winners corn
yield guide. Washington DC: National Corn Growers
Association [Internet]. 2011 [cited 2013 July 01].
Available from: www.ncga.com

NCGA. National corn yield contest: 2012 Winners corn
yield guide. Washington DC: National Corn Growers
Association [Internet]. 2012 [cited 2013 July 01].
Available from: www.ncga.com

OECD. Consensus document on general information concerning the genes and their enzymes that confer
tolerance to phosphinothricin herbicide. Series on har-
onisation of regulatory oversight in biotechnology, number 11, ENV/JM/MONO(99)13. Paris: Organisation for Economic Co-operation and Development; 1999.

OECD. Module II: herbicide biochemistry, herbicide metabolism and the residues in glufosinate-ammonium (phosphinothricin)-tolerant transgenic plants. Series on Harmonisation of Regulatory Oversight in Biotechn-
ology, Number 25, ENV/JM/MONO(2002)14. Paris: Organisation for Economic Co-operation and Development; 2002.

OECD. Consensus document on the biology of Zea mays
subsp. mays (Maize). Series on Harmonisation of Regu-
larly Oversight in Biotechnology, Number 27, ENV/JM/MONO(2003)11. Paris: Organisation for Economic Co-operation and Development; 2003.

OECD. Consensus document on safety information on transgenic plants expressing Bacillus thuringiensis-derived insect control proteins. Series on Harmonisa-
tion of Regulatory Oversight in Biotechnology, Number 42, ENV/JM/MONO(2007)14. Paris: Organisation for Economic Co-operation and Development; 2007.

Park KW, Lee B, Kim CG, Kim DY, Park JY, Ko EM, Jeong SC, Choi KH, Yoon WK, Kim HM. Monitoring the occurrence of genetically modified maize at a grain receiving port and along transportation routes in the Republic of Korea. Food Control 2010; 21(4):456–61; http://dx.doi.org/10.1016/j.foodcont.2009.07.006

Pawlowski WP, Somers DA. Transgene inheritance in plants genetically engineered by microprojectile bombardment. Mol Biotechnol 1996; 6(1):17–30; PMID: 8887358; http://dx.doi.org/10.1007/BF02762320

Que C, Chilton MM, de Fontes CM, He C, Nuccio M, Zhu T, Wu Y, Chen JS, Shi L. Trait stacking in transgenic crops-challenges and opportunities. GM Crops Food 2010; 1(4):220–9; http://dx.doi.org/10.4161/gmcr.1.4.13439

Raybould A, Higgins LS, Horak MJ, Layton RJ, Storer
NP, Fuente JMDL, Herman RA. Assessing the eco-
logical risks from the persistence and spread of feral populations of insect-resistant transgenic maize. Transgenic Res 2012; 21(3):655–64; PMID:22002083; http://dx.doi.org/10.1007/s11248-011-9560-4

Raybould A, Kilby P, Graser G. Characterising microbial protein test substances and establishing their equiva-
ience with plant-produced proteins for use in risk assessments of transgenic crops. Transgenic Res 2013; 22(2):445–60; PMID:23065372; http://dx.doi.org/10.1007/s11248-012-9658-3

Rimachi LFG, Alcantara JD, Aquino YV, Ortiz R. Detect-
ing adventitious transgenic events in a maize center of
Sindt J, Drouillard J, Eoe L, Kessen T, Sulpizio M, Montgomery S, Rice D, Hinds M, Smith B, Owens F, Dana G, Hunst P. Effect of corn containing the Cry1F protein on performance of beef heifers fed a finishing diet based on steam-flaked corn. Prof Anim Sci 2007; 23(6):632-6

Snell C, Bernheim A, Bergé JB, Kuntz M, Pascal G, Paris A, Ricroch AE. Assessment of the health impact of GM plant diets in long-term and multigenerational animal feeding trials: a literature review. Food Chem Toxicol 2012; 50(3-4):1134-48; PMID:22155268; http://dx.doi.org/10.1016/j.fct.2011.11.048

Stauffer C, Zepf L. Compositional analysis of maize MPS hybrid line 1507. Johnston, IA: Pioneer Hi-Bred International, Inc. and Des Moines, IA: Woodson-Tenet Laboratories, Inc; 2000 Jul. 34p. Unpublished Technical Report No.: 98-09-RA-NGLP-012

Stein HH, Sauber TE, Rice DW, Hinds MA, Smith BL, Dana G, Peters DN, Hunst P. Growth performance and carcass composition of pigs fed corn grain from DAS-OÎ1507-1 (Herculex I) hybrids. Prof Anim Sci 2009; 25(6):689-94

Takabatake R, Futo S, Minegishi Y, Watai M, Sawada C, Nakamura K, Akiyama H, Teshima R, Furui S, Hino A, et al. Evaluation of quantitative PCR methods for genetically modified maize (MON863, NK603, TC1507 and T25). Food Sci Technol Res 2010; 16(5):421-30; http://dx.doi.org/10.3136/fstr.16.421

Thompson GD, Dalmacio SC, Criador IV AR, Alvarez ER, Hechanova RF. Field performance of TC1507 transgenic corn hybrids against Asian corn borer in the Philippines. Philipp Agric Scientist 2010; 93(4):375-83

Tian JC, Wang XP, Long LP, Romeis J, Naranjo SE, Hellmich RL, Shelton AM. Eliminating host-mediated effects demonstrates Bt maize producing Cry1F has no adverse effects on the parasitoid Cotesia marginiventris. Transgenic Res 2014; 23(2):257-64; PMID:24026808; http://dx.doi.org/10.1007/s11248-013-9748-x

Tian JC, Wang XP, Long LP, Romeis J, Naranjo SE, Hellmich RL, Wang P, Earle ED, Shelton AM. Bt crops producing Cry1Ac, Cry2Ab and Cry1F do not harm the green lacewing, Chrysoperla rufilabris. PLoS One 2013; 8(3):e60125; PMID:23544126; http://dx.doi.org/10.1371/journal.pone.0060125

Trigo EJ. Fifteen years of genetically modified crops in Argentine agriculture. Argenbio, Buenos Aires, Argentina [Internet]. 2011 [cited 2015 May 05]. Available from: http://www.argenbio.org/adc/uploads/15_years_Executive_summary_of_GM_crops_in_Argentina.pdf

US EPA. Phosphinothricin acetyl transferase (PAT) and the genetic material necessary for its production in all plants; exemption from the requirement of a tolerance on all raw agricultural commodities. Federal Reg 1997; 62(70):17717-20

US EPA. Bacillus thuringiensis subspecies Cry1F protein and the genetic material necessary for its production (plasmid insert PHI 8999) in corn. Pesticide Fact Sheet, US Environmental Protection Agency
US EPA. Biopesticides Registration Action Document – Bacillus thuringiensis Cry1F Corn [Internet]. 2005 [cited 2015 April 09]. Available from: http://bch.cbd.int/database/attachment?id=10711

US FDA. Memorandum to file concerning insect resistant and herbicide tolerant maize line 1507 [Internet]. 2001 [cited 2013 July 01]. Available from: http://cera-gmc.org/docs/decdocs/bnfM073.pdf

USDA APHIS. Petition for the determination of non-regulated status B.t. Cry1F insect resistant, glufosinate tolerant maize line. Washington DC: United States Department of Agriculture, Animal and Plant Health Inspection Service [Internet]. 2000 [cited 2013 July 01]. Available from: http://www.aphis.usda.gov/brs/aphisdocs/00_13601p.pdf

USDA APHIS. Decision on Mycogen Seeds c/o Dow AgroSciences LLC and Pioneer Hi-Bred International, Inc. petition 00-136-01P seeking a determination of non-regulated status for Bt Cry1F insect resistant, glufosinate tolerant corn line 1507 [Internet]. 2001 [cited 2013 July 01]. Available from: http://cera-gmc.org/docs/decdocs/02122001.pdf

Vachon V, Laprade R, Schwartz JL. Current models of the mode of action of Bacillus thuringiensis insecticidal crystal proteins: a critical review. J Invertebr Pathol 2012; 111:1–12; PMID:22617276; http://dx.doi.org/10.1016/j.jip.2012.05.001

Van Frankenhuysen K. Insecticidal activity of Bacillus thuringiensis crystal proteins. J Invertebr Pathol 2009; 101:1–16; PMID:19269294; http://dx.doi.org/10.1016/j.jip.2009.02.009

Wehrmann A, Van Vliet A, Opsomer C, Botterman J, Schulz A. The similarities of bar and pat gene products make them equally applicable for plant engineers. Nat Biotech 1996; 14(10): 1274–8; PMID:9631092; Available from http://www.nature.com/nbt/journal/v14/n10/pdf/nbt1096-1274.pdf

Whalon ME, Wingerd BA. Bt: Mode of action and use. Arch Insect Biochem Physiol 2003; 54:200–11; PMID:14635181; http://dx.doi.org/10.1002/arch.10117

Wolt JD. A mixture toxicity approach for environmental risk assessment of multiple insect resistance genes. Environ Toxicol Chem 2011; 30(3):763–72; PMID:21298718; http://dx.doi.org/10.1002/etc.427

Wolt JD, Conlan CA, Majima K. An ecological risk assessment of Cry1F maize pollen impact to pale grass blue butterfly. Environ Biosafety Res 2005; 4(4):243–51; PMID:16827552; http://dx.doi.org/10.1051/eb:2006005

Xu Z, Hennessy DA, Sardana K, Moschini G. The realized yield effect of genetically engineered crops: US maize and soybean. Crop Sci 2013; 53(3):735–45; http://dx.doi.org/10.2135/cropsci2012.06.0399