PART II: Risk Assessment (Annex III of the Cartegena Protocol on Biosafety)

COMMON FORMAT FOR Risk Assessment

(In accordance with Annex III of the Cartagena Protocol on Biosafety)

	Risk assessment details	
1.	Country Taking Decision:	South Africa
2.	Title:	Application for Commodity Clearance of Genetically Modified Organisms (GMO) - MON 89034 × MIR162
3.	Contact details:	Monsanto Company, represented by Monsanto S.A.(Pty) Ltd Monsanto Company 800 N. Lindbergh Boulevard St. Louis, Missouri 63167 USA Monsanto House, Building No. 4 Fourways Office Park Corner Fourways Boulevard and Roos Streets Fourways South Africa
	LMO information	
4.	Name and identity of the living modified organism:	Multi-event stack MON 89034 × MIR162
5.	Unique identification of the living modified organism:	MON-89Ø34-3 × SYN-IR162-4
6.	Transformation event:	MON 89034 × MIR162
7.	Introduced or Modified Traits:	B. Altered growth, development and product quality **Pest resistance** - Insect resistance
8.	Techniques used for modification:	MON 89034 × MIR162 maize was obtained by conventional breeding of two single maize products: MON 89034 and MIR162. MON 89034 has been developed by Monsanto Company while MIR162 was developed by Syngenta.

9. Description of gene modification:	Conventional breeding techniques were used to develop MON 89034 × MIR162, a combined-trait maize product that confers insect resistance. Each biotechnology-derived trait contributes specific benefits to the final combined product as follows: MON 89034 produces two insecticidal proteins that protect against feeding damage caused by European corn borer (*Ostrinia nubilalis*) and other lepidopteran insect pests. Cry1A.105 is a modified *Bacillus thuringiensis* (*Bt*) Cry1A protein and Cry2Ab2 is a *Bt* (subsp. *kurstaki*) protein. The combination of the two insecticidal proteins provides enhanced insect control and offers an additional insect-resistance management tool. MIR162 produces the *Bacillus thuringiensis* (*Bt*) Vip3Aa20 protein, which protects against feeding damage caused by fall armyworm (*Spodoptera frugiperda*), corn earworm (*Helicoverpa zea*) and other lepidopteran insect pests. MIR162 also expresses the phosphomannose isomerase (PMI) enzyme from *Escherichia coli*, as a plant-selectable marker.
	Characteristics of modification
10. Vector characteristics (Annex III.9(c)):	Not applicable, MON 89034 × MIR162 was obtained through conventional breeding methods.
11. Insert or inserts (Annex III.9(d)):	Like MON 89034, MON 89034 × MIR162 expresses the Cry1A.105 and Cry2Ab2 insecticidal proteins derived from *B. thuringiensis*, providing protection against certain lepidopteran insect pests such as maize stalk borers. Like MIR162, MON 89034 × MIR162 expresses the Vip3Aa20 protein variant of the native insecticidal Vip3Aa protein from *Bacillus thuringiensis* strain AB88. The Vip3Aa20 protein is insecticidally active against a number of lepidopteran pests of maize. MIR162 also contains the the *pmi* gene, obtained from *Escherichia coli* strain K-12. It encodes for the enzyme phosphomannose isomerase (PMI) which was utilized as a plant selectable marker during the development of MIR162.
Recipient organism or parental organisms (Annex III.9(a)):	
12. Taxonomic name/status of recipient organism or parental organisms:	Common name: Maize Family name: Gramineae Genus: *Zea* Species: *mays* (2n+20)
13. Common name of recipient organism or parental organisms:	Maize or Corn
14. Point of collection or acquisition of recipient or parental organisms:	MON 89034 × MIR162 was produced using elite parent lines into which the MON 89034 and MIR162 events have been introgressed independently, using conventional breeding techniques. The original transformations that produced the individual events used privately owned germplasm acquired for this purpose.

15. Characteristics of recipient organism or parental organisms related to biosafety:	Maize is the world's third leading cereal, following rice and wheat, in terms of production and area harvested. It has a long history of safe use as a raw material for processed products, and direct uses as a human food or animal feed. Today, maize is produced on every continent except Antarctica, and is exported and imported as viable grain for use as foods or feeds, or directly in processing, without risk to the environment. According to the OECD [Consensus Document on the Biology of *Zea mays* subsp. *mays* (Maize), 2003], "*Maize has lost the ability to survive in the wild due to its long process of domestication, and needs human intervention to disseminate its seed*". In addition, "*maize is incapable of sustained reproduction outside of domestic cultivation*", and "*maize plants are non-invasive in natural habitats.*" Despite the fact that maize frequently appears as a volunteer plant in a subsequent rotation, it has no inherent ability to persist or propagate. In all regions of the world, volunteer plants are managed with herbicides, tillage, or manual removal of plants. As such, maize is not considered to be a pest anywhere in the world. When it occurs outside of cultivation, it has no impact on the conservation and sustainable use of biological diversity. Gene flow from maize occurs through dispersal of seed and pollen-mediated exchange of genes to sexually compatible plants. Since maize has no biological mechanism to scatter seed, low-level, incidental dispersal of viable grain occurs as a result of human-based activities such as transport and harvesting operations. As was noted by the OECD, the few plants that might result from incidental release will not persist or meaningfully reproduce without human intervention. Gene flow via pollen is only possible to other maize plants throughout the world except in Mexico and Guatemala where wild relatives occur. Maize reproduces sexually, is a wind-pollinated, monoecious species with separate staminate (tassels) and pistillate (silk) flowers, which encourages natural cross-pollination between maize plants. The distance that viable pollen can travel depends on prevailing wind patterns, humidity, and temperature. Generally, the pollen dissemination period lasts three to seven days. Because incidental release of maize during importation occurs at very low levels, and because maize is not competitive, pollen-mediated gene flow between local maize and rare volunteers has had no effect on the conservation and sustainable use of biological diversity.

16. Centre(s) of origin of recipient organism or parental organisms:	Maize is thought to have its origin in Mexico, from where it spread northward to Canada and southward to Argentina. Although secondary centres of origin in South America are possible, the oldest archaeological evidence of domesticated maize (5000 B.C.) was discovered in the valley of Tehuacan in Mexico (Benson and Pearce 1987). Several theories on the origin of maize have been proposed; the two theories most adhered to being that either teosinte (a wild relative of maize that is endemic to parts of Mexico and Guatemala) or a wild pod maize that is now extinct was the wild ancestor of maize (Benson and Pearce 1987; Brown, et al. 1984). Maize is a member of the genus *Zea,* which is broken into 2 sections: ZEA and LUXURIENTES. The section ZEA includes one species (*mays*), which includes three subspecies: ssp. *mays,* ssp. *mexicana* (formerly *Euchlaena mexicana),* and ssp. *parviglumis*. The former subspecies is known as maize while the latter comprise a portion of the complex known as teosinte. Furthermore, ssp. *mexicana* and ssp. *parviglumis* are further separated into several races (OECD 2003). Section LUXURIENTES encompasses 3 species: an annual *Z. luxurians,* and perennials *Z. diploperennis* and *Z. perennis*. While the classification of *Zea* continues to be modified, teosintes are the only know wild relatives of maize capable of forming hybrids in nature. Outcrossing and gene exchange between teosinte and maize has been reported with annual teosinte (*Zea mays* ssp. *mexicana*) (2n = 20) and maize (*Zea mays L.*) (2n = 20). A frequency of one F1 hybrid (maize × teosinte) for every 500 maize plants or 20 to 50 teosinte plants in the Chalco region of the Valley of Mexico was reported. However, newer information shows that annual teosintes may be a separate species because of the level of genetic isolation and that hybrids that do form are highly unsuccessful in introgressing genetic material (OECD 2003). Regardless, Mexico and parts of Central America are regarded as the centre of genetic diversity for maize. The natural distribution of teosinte is limited to the seasonally dry, suropical zone with summer rain along the western escarpment of Mexico and Guatemala and the Central Plateau of Mexico. The belief that Central America and southern Mexico are both the centre of origin and a centre of diversity for maize was supported by Vavilov (1992).
17. Centres of genetic diversity, if known, of recipient organism or parental organisms:	Refer to point 16 above

18. Habitats where the recipient organism or parental organisms may persist or proliferate:	As noted by OECD (2003), maize is not invasive of natural habitats, does not persist or disperse anywhere in the world without the human intervention. Early domestication and diversification through selection occurred in Meso-America. Maize is grown across a wide range of ecological conditions including soil types, altitude and rainfall. Currently, maize is grown over a wide range of conditions because of its many divergent types that have been bred for this purpose. The bulk of the maize is produced between latitudes 30°and 55°, with relatively little grown at latitudes higher than 47°latitude anywhere in the world. The greatest maize production occurs where the warmest month isotherms range between 21°C and 27°C and the frost-free season lasts 120 to 180 days. A summer rainfall of 15 cm is approximately the lower limit for maize production without irrigation. Experience with maize imported for use as foods or feeds, or directly in processing, has demonstrated that stable populations do not establish, persist or proliferate as a result of this practice.
Donor organism or organisms (Annex III.9(b)):	
19. Taxonomic name/status of donor organism(s)	MON 89034 × MIR162 was obtained from conventional breeding of the single events MON 89034 and MIR162. The donor organisms used in development of the single events MON 89034 and MIR162 were *Bacillus thuringiensis* and *E. coli* strain K-12.
20. Common name of donor organism(s):	Bacteria
21. Point of collection or acquisition of donor organism(s):	The organisms are ubiquitous in nature.
22. Characteristics of donor organism(s) related to biosafety:	Not applicable, since the donor organisms are ubiquitous in nature and therefore do not pose a threat to biodiversity.
Intended use and receiving environment	
23. Intended use of the LMO (Annex III 9(g)):	The intent of this application is for the commodity import of maize containing MON 89034 × MIR162
24. Receiving environment (Annex III.9(h)):	This is an application for Commodity Clearance, therefore the product will not be cultivated in South Africa.
Risk assessment summary	
25. Detection/Identification method of the LMO (Annex III.9(f)):	Detection methods for detection of MON 89034 DNA has been provided to the Registrar. Monsanto has intellectual property rights that govern the use of each event specific detection method in the Republic of South Africa. DAFF shall use the Information solely for the purpose of facilitating or permitting regulatory approval under the Genetically Modified Organism Act, Act 15 of 1997 and to carry out or enforce the requirements of those other laws of the Republic of South Africa which regulate MON 89034 × MIR162 and its use, and for no other purpose. The event specific detection method for detection of MIR162 has been validated for use by the EURL GMFF (European Union Reference Laboratory for Genetically Modified Food and Feed) and can be found on the EURL GMFF website. http://gmo-crl.jrc.ec.europa.eu/summaries/MIR162_validated_Method.pdf

26. Evaluation of the likelihood of adverse effects (Annex III.8(b)):	Based on the nature of the recipient species (unable to proliferate) and the lack of related and wild species with which MON 89034 × MIR162 can outcross, the likelihood of adverse effects from out-crossing to other related species is negligible. Transgenic maize hybrids with similar genes have been grown around the world and in South Africa for several years without any recorded impact on the environment other than those created by conventional maize production. Any volunteers could, like conventional maize, be removed by current agricultural practices such as ploughing and the use of herbicides.
27. Evaluation of the consequences (Annex III.8(c)):	Studies conducted with MON 89034 × MIR162 confirmed that this stack is agronomically and compositionally equivalent to conventional maize and has no increased tendency towards weediness or an increased susceptibility of tolerance to insects normally associated with maize. Thus, should any of the potential risks materialize, the consequences would be negligible. No potentially adverse effects were detected based on extensive characterization of MON 89034 × MIR162, which included molecular analysis, expression analysis, compositional analyses and phenotypic evaluation. Testing of MON 89034 × MIR162 demonstrated no changes in its ability to persist in the environment without human intervention or to become invasive compared to conventional maize. As such, the potential consequences to biodiversity resulting from MON 89034 × MIR162 for commercial use, including food, feed or processing, are the same as with conventional maize. Any volunteer seed germinating in subsequent growing seasons would be detected in the fields that were planted and destroyed using chemical or mechanical means.
28. Overall risk (Annex III.8(d)):	Considering the potential risks and the consequences should the potential risks materialize the overall risk of importing, cultivating or field testing MON 89034 × MIR162 is extremely low.
29. Recommendation (Annex III.8(e)):	No risks have been identified and therefore other than the containment parameters that might apply through the permit conditions, no additional actions need to be taken.
30. Actions to address uncertainty regarding the level of risk (Annex III.8(f)):	The potential risks for the specific product is negligible; hence no additional actions are required except compliance with the conditions contained in the permit.
	Additional information
31. Availability of detailed risk assessment information:	More information regarding the safety of MON 89034 × MIR162 is contained in the application preceding this section.
32. Any other relevant information:	None
33. Attach document:	*Not applicable to applicant*
34. Notes:	None