Objection against general release of three 2,4-D GM maize varieties:

1. Corteva's 2,4-D herbicide tolerant maize: DAS-40278-9
2. Corteva's Stacked 2,4-D and glyphosate herbicide tolerant maize: NK603 x DAS-40278-9
3. Corteva's Stacked 2,4-D, glyphosate and glufosinate herbicide tolerant, and Bt insecticidal maize: MON89034 x TC1507 x NK603 x DAS-40278-9

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April 2019

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Acknowledgements

The African Centre for Biodiversity acknowledges Dr Eva Sirinathsinghji for her contribution as researcher and author of this objection and our donors for supporting this work.

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SUMMARY OF KEY CONCERNS

- The three GM varieties were shown in field trials conducted in South Africa to worsen yields when compared with non-GM counterparts. Claims of efficacy and increased yield protection in the applications are thus deliberately misleading and unsubstantiated.
- GM varieties are all tolerant to the toxic chemical, 2,4-D. Their approval will represent the first GM maize in South Africa to be tolerant to 2,4-D. Two of the three varieties are also stacked with glyphosate-tolerance traits. Commercial release of these seed is expected to dramatically increase pesticide application and thus human and environmental exposure.
- Increased pesticide exposure will serve to increase pesticide drift and crop damage, as well as weed resistance, providing at best, a short -term solution for weed management, while increasing damage and thus crop loss in neighbouring fields.
- Pesticide exposure will increase human exposure to 2,4-D, glyphosate and glufosinate, all of which are widely associated with serious adverse health effects including cancers and birth defects. While other countries are removing them from their shelves, they are now being dumped on South Africa.
- The three GM varieties have not been adequately characterised at the molecular level raising concerns for unintended effects that may have environmental or human health impacts.
- Safety assessments have been entirely inadequate to substantiate any claims of safety. No feeding studies on the stacked traits have been performed, and many assumptions on safety have been made that are challenged by independent data showing harm.
- Socio-economic impacts have not been assessed, and claims of benefits associated with improved yields is out of line with their own data showing worse yields than non-GM counterparts. The GM industrial model is acknowledged to be unsustainable and a direct threat to future food production.
- Alternative agro-ecological systems such as push-pull climate adapted technologies are proven, low-cost methods that not only improve farmer yields and livelihoods, but also biodiversity and soil fertility – factors that are all necessary to preserve long-term food security.
- We urge a shift away from GM industrial systems of food production based on precautionary measures to protect human and environmental health, as well as food security for the people of South Africa.

INTRODUCTION

The African Centre for Biodiversity (previously 'Biosafety') (ACB) was established in 2003 and registered in 2004. ACB carries out research, analysis, capacity and movement building, and advocacy, and shares information to widen awareness and catalyse collective action and influence decision-making on issues of biosafety, agricultural biodiversity and farmer-managed seed systems (FMSS) in Africa. The ACB's work both informs and amplifies the voices of social movements fighting for food justice and food sovereignty in Africa.

The ACB has played an essential watchdog role on new GMO permits in South Africa for almost 15 years now, adding substantially to the discourse about the scientific assessment of GMOs as well as issues of socio-economic impacts and democratic decision-making, through lodging substantive comments on at least 30 permit applications.

The ACB has consistently opposed the introduction of Corteva's (formally Dow AgroSciences) 2,4-D tolerant genetically modified (GM) maize DAS-40278-9 into South Africa, along with other GM varieties tolerant to 2,4-D. 2,4-D is one of the two herbicide ingredients in the infamous war chemical 'Agent Orange' used to devastating effect during the Vietnam war, leaving in its

wake generations of children affected by birth defects and cancers. In 2012, the ACB launched a petition against its import intended for the South African food supply. In 2017, the ACB published a briefing on the environmental releases of the three GM varieties for field trials, exposing the failures of the GM varieties in field trial performances, to increase yields and the risks of increased pesticide cocktails to be used on South African food systems and environment.

We are objecting to the general release of three GM maize varieties and their associated pesticides 2,4-D, glyphosate, glufosinate and quizalofop, due to concerns surrounding the biosafety risks they pose to human health and the environment. The applicant fails to provide the required information to show safety of the three products and further, makes misleading claims of benefits that contradict their own field trial data showing a complete lack of yield protection or yield gains. These varieties will instead serve to increase sales of pesticides, further exposing farm workers, consumers and wildlife, consolidate corporate control of the South African already corporatized food systems, and further increase inequalities and inequities.

BACKGROUND TO APPLICATIONS

Corteva (formally Dow AgroSciences) has applied for general release of three GM maize varieties:

1. DAS-40278-9 expressing the aryloxyalkanoate dioxygenase 1 (AAD-1) protein. This maize is tolerant to 2,4-D and quizalofop herbicides.

2. NK603 x DAS-40278-9 stacked event, expressing the 5-enolpyruvylshikimate-3-phosphate synthase (CP4 EPSPS) and its variant CP4 EPSPS L214P protein, and expressing the aryloxyalkanoate dioxygenase 1 (AAD-1) protein. This maize is tolerant to 2,4-D, quizalofop and glyphosate herbicides.

3. MON89034 x TC1507 x NK603 x DAS-40278-9 stacked event, expressing Cry1A.105 and Cry2Ab2 insecticidal proteins; expressing the 5-enolpyruvylshikimate-3-phosphate synthase (CP4 EPSPS) and its variant CP4 EPSPS L214P protein, expressing the aryloxyalkanoate dioxygenase 1 (AAD-1) protein. This maize is tolerant to 2,4-D, quizalofop and glyphosate herbicides and produces insecticidal Bt toxins.

TRIAL DATA SHOWS GM VARIETIES PERFORM WORSE THAN NON-GM VARIETIES

The entire premise of GM crop cultivation is that they are supposed to improve farmers' yields and thus improve food security and farmer livelihoods while improving environmental impacts of food production systems. The applications for the three GM maize varieties make repeated claims of the benefits they exert over conventional varieties. For example, under Section 10 on socio-economic impacts of the three varieties, claims are made for all three GM varieties that:

"Herbicide Tolerance improves yield and grower efficiency",

and additionally for MON89034 x TC1507 x NK603 x DAS-40278-9, that:

"Insecticide tolerance protects yield and improves grain quality".

The applicant also claims that there are "*no other non-genetically modified maize products available with the same benefits as those provided*" (Section 4.9 in all three applications) by the three GM varieties.

The applicant further states (Section 7.3 in the three applications): "*The elimination of weeds which are usually in competition with maize crops may be effectively controlled, simplified and less time-consuming for this genetically modified maize, which will contribute to the* ***quality and yield improvement*** *of the crop as compared to non-GM crops* [emphasis added]."

However, the data from the field trials (Dow AgroSciences LLC, 2016, (Study ID: 161011)) performed by the applicant in South Africa in 2015-2016 exposes the above claims as scientifically baseless, and deliberately misleading.

As shown in Table 1, yields (blue column) were highest for the isoline non-GM variety at 7873 kilograms per hectare (mean average). Both DAS-40278-9 and NK603 x DAS-40278-9 had statistically significantly worse yields, while MON89034 x TC1507 x NK603 x DAS-40278-9 produced non-statistically significant reductions in yield compared with the non-GM variety. For all three varieties, further reductions were seen when sprayed with their corresponding herbicides, than when left unsprayed.

Other important measurements of agronomic performance, such as both early (non shown in Table 1) and final stand counts (grey column in Table 1) – the number of plants that grow, were also significantly reduced in all GM varieties, and were worse following herbicide application. Early vigour, or the ability to grow relatively fast during early growth stages - identified as an important trait associated with the ability of the crop to better suppress weeds – was also significantly reduced in all GM varieties, and again, even worse following herbicide application.

Despite significant reduction in corn borer damage (as measured by percentage of plants damaged, not extent of damage) as a result of Bt insecticidal toxins being expressed in MON 89034 × 1507 × NK603 × DAS-40278-9, this did not translate into any yield protection. The claim that this variety is thus are "efficacious at providing herbicide tolerance to glyphosate and 2,4-D and protection against several lepidopteran insect pests of maize", is completely meaningless.

Entry	Corn Borer Tassel Damage (% of plants with feeding damage)	Corn Borer Cob Damage (% of plants with feeding damage)	Bird Damage to Cobs (% of plants with feeding damage)	Final Stand Count (plants per plot)	Yield (kilograms per hectare)
Isoline (non-GM variety)	9.5 ± 1.9 3–25	30.5 ± 7.7 2–83	0.3 ± 0.3 0–2	34.9 ± 1.7 28–39	7873 ± 976 4871–10255
DAS-40278-9 Unsprayed	6.6 ± 1.9 2–10 (0.209)	32.5 ± 7.7 4–82 (0.791)	0.0 ± 0.3 0–1 (0.443)	29.5 ± 1.7 20–34 (0.002*)	6707 ± 976 2187–9662 (0.014*)
DAS-40278-9 Sprayed with 2,4-D	4.6 ± 1.9 0–13 (0.038*)	33.9 ± 7.7 13–70 (0.650)	0.2 ± 0.3 0–1 (0.660)	27.9 ± 1.7 20–35 (<0.001*)	6023 ± 976 1563–8862 (<0.001*)
NK603 × DAS-40278-9 Unsprayed	6.8 ± 1.9 0–12 (0.241)	31.5 ± 7.7 11–75 (0.891)	0.6 ± 0.3 0–5 (0.510)	28.5 ± 1.7 23–36 (<0.001*)	6158 ± 976 3703–9618 (0.001*)
NK603 × DAS-40278-9 Sprayed with Glyphosate + 2,4-D	6.4 ± 1.9 2–15 (0.178)	36.3 ± 7.7 14–74 (0.440)	0.4 ± 0.3 0–2 (0.826)	26.3 ± 1.7 17–32 (<0.001*)	6146 ± 976 2442–10207 (0.001*)
MON 89034 × 1507 × NK603 × DAS-40278-9 Unsprayed	1.0 ± 1.9 0–5 (0.001*)	2.8 ± 7.7 0–23 (0.001*)	0.5 ± 0.3 0–3 (0.583)	28.0 ± 1.7 22–35 (<0.001*)	7748 ± 976 5634–10677 (0.781)
MON 89034 × 1507 × NK603 × DAS-40278-9 Sprayed with 2,4-D + Glyphosate	0.3 ± 1.9 0–3 (<0.001*)	2.5 ± 7.7 0–12 (0.001*)	0.3 ± 0.3 0–1 (1.000)	27.5 ± 1.7 16–38 (<0.001*)	7203 ± 976 5101–9467 (0.157)

Table 1: Yield data from field trials of the three GM maize varieties in South Africa across 2015-2016 season. Adapted from Table 6 of Dow AgroSciences LLC, 2016, (Study ID: 161011).
Notes and Abbreviations: Mean ± SE, range, and P-value (statistical significance test) are provided for each entry and characteristic. P-Value = P- Value for comparing DAS-40278-9, NK603 × DAS-40278-9, MON 89034 × 1507 × NK603, and MON 89034 × 1507 × NK603 × DAS-40278-9 with the isoline. P-values <0.05 were considered significant and are denoted with *.

The inaccurate and misleading claims made to promote commercialisation of these three GM maize varieties makes a mockery of the scientific and regulatory process. The applicant has presented data to show that the three GM varieties are outperformed by non-GM counterparts and thus present NO potential benefits for South African farmers or consumers. Instead, these varieties present biosafety risks to farmers, consumers and the environment, serving to increase exposure to toxic herbicides and inadequately tested GM crops.

We urge the South African authorities to reject the approval of these GMOs based first, on a complete lack of evidence that they will improve yield, and secondly, on the basis that the applicant has attempted to mislead authorities with regards to how these varieties have performed to date in national trials.

GM VARIETIES WILL LEAD TO INCREASED HERBICIDE USE

The commercial release of all three varieties designed to tolerate applications of 2,4-dichlorophenoxyacetic acid, commonly known as 2,4-D, will represent the first GM maize to be cultivated in South Africa that are tolerant to this herbicide. The aryloxyalkanoate dioxygenase-1 (AAD-1) enzyme gene inserted into the GM varieties to confer tolerance to 2,4-D, also confers tolerance to aryloxyphenoxypropionate herbicides such as quizalofop.

All three varieties, by design, will increase 2,4-D use, raising serious concerns over increasing pollution of South African farming systems, environment and staple food supply with a chemical that has been linked to both human and environmental toxicity (see section below). The stacked varieties will also be tolerant to glufosinate and/or glyphosate, resulting in crops that will be sprayed with a cocktail of pesticides, further increasing risks associated with their use.

In the United States for example, where 2,4-D tolerant crops were recently approved, the US Department of Agriculture estimated a 200-600 % increase in use over a 10-year period (USDA EIS, p4-34), while independent scientists calculated a 30-fold increase after 9 years of cultivation (Benbrook, 2012). Such increases would be expected to be replicated in South Africa.

Increased pesticide use will increase pesticide drift and crop damage

Another concern for increased 2,4-D use, is the issue of pesticide drift that can lead to damage of neighbouring farmer's fields and wild plants. 2,4-D is a volatile herbicide and can thus travel over long distances and re-deposit itself on plants far from the site of application. It is responsible for the most episodes of crop damage in the US according to two recent surveys by US state pesticide regulators (AAPCO, 1999 & 2005). It is also active against most fruit and vegetables, making any drift a threat to the production of food crops and thus food security and farmer livelihoods.

Increased pesticide use will hasten the development of herbicide resistant weeds

The cultivation of 2,4-D tolerant crops has been developed to combat the epidemic rise of weeds that have developed resistance to glyphosate. However, it is only a matter of time before the same situation will arise under increased application of 2.4-D herbicides. The use of 2,4-D thus represents at best, a short-term techno-fix for weed management. Indeed, 2,4-D resistant weeds are now becoming a problem in the US, where major weed pests that are already resistant to glyphosate, are now showing resistance to 2,4-D (Kumar et al., 2019).

Increased pesticide use will increase exposure to toxic pesticides

There is a growing body of evidence demonstrating a link between exposure to pesticides and a wide spectrum of pathologies, ranging from neurological symptoms, developmental problems and cancer. However, the safety of the three pesticides that will be applied to the GM maize varieties was completely dismissed in the safety assessments (as single pesticides, or in combination), despite the expectation that their cultivation will increase human and wildlife exposure to the herbicides via food residues and/or environmental pollution.

2,4-D, one of the two ingredients in 'Agent Orange' war chemical used to devastating effect in the Vietnam war, is a synthetic hormone that mimics a type of plant hormone, called auxins. Hormone mimics are widely associated with adverse health effects including developmental and reproductive toxicity as a result of hormone disruption. 2,4-D has been linked to increases in birth abnormalities in high use areas in the US (Garry et al., 1996). Consistently, 2,4-D has been suggested to interfere with male reproduction, with effects including disrupted testosterone levels and spermatogenesis; reduced mortality of human sperm; and increased sperm abnormalities in farm workers (Swan, 2003; Lerda & Rizzi 1991), reduced testosterone and increased leutinising and follicle-stimulating hormones in male rats (both involved in male and female reproduction).

Farm workers have also been shown to suffer higher rates of the non-Hodgkin lymphoma (NHL), a cancer of the lymph node, as a result of 2,4-D exposure in numerous studies across various countries (Hardell et al., 1999; McDuffie et al., 2001; Zahm et al., 1990). The herbicide was recently categorised as a Group 2B possible human carcinogen by the World Health Organisation's Cancer arm, the International Agency for Research on Cancer (IARC). Studies also show increases in oxidative stress in exposed maize farmers, which may be behind 2,4-D's carcinogenic effects (Lerro et al., 2017).

Other modes of toxicity have recently been uncovered including inducing perturbations of the gut microbiome in laboratory mice at occupationally relevant doses (Tu et al., 2019). The gut microbiome confers various health benefits, and its disruption is increasingly associated with a large number of disease conditions ranging from cancers to neurological problems such as anxiety and autism. The authors suggest that the relationship between the gut microbiota and environmental contaminants is largely underestimated and should be comprehensively considered with regards to assessing toxicity of environmental chemicals.

Glyphosate toxicity has been widely documented in both independent and industry data. Most recently Bayer, formally Monsanto, lost two high profile court cases in the US where Monsanto was found guilty of not warning of the cancer risks of glyphosate herbicides that had resulted in two people suffering from NHL. 11,200 further cases are pending and it is now estimated that Bayer could be liable to pay US$ 30 billion in compensations (Sustainable Pulse, 2019). These court cases have also revealed aggressive PR strategies by Monsanto to cover up evidence of cancer links, including ghost-writing of scientific papers and pushing favourable studies (McHenry, 2018), in efforts to delegitimise the IARC's 2015 decision to classify glyphosate as Group 2A probable human carcinogen, spending an estimated US$ 17 million in a single year. In recent days a draft US federal report from the Agency for Toxic Substances and Disease Registry, tied to the Centres for Disease Control and Prevention (CDC), echoed the IARC decision, acknowledging a link to NHL (ATSDR, 2019). These recent developments have rightly prompted numerous national and regional bans or restrictions on the herbicide, including a complete ban on production and import in Vietnam (Reuters, 2019) and Malawi (Sustainable Pulse, 2019b), ban on sales in France, and numerous regional bans across European, Asian and Latin American nations, national bans on home use. Glyphosate has also been linked to reproductive and developmental toxicity, disruption of the microbiome, and liver disease at legally permitted levels for human exposure.

Glufosinate has also been shown in many studies to have adverse toxic effects on humans, such that its use was restricted in 2013 by the European Union. Toxic effects of glufosinate have been linked to its glutamate neurotransmitter-mimicking effects. This has been shown to disrupt brain signalling, resulting in learning and memory deficits, structural changes in the brain and impaired brain development in laboratory animals (Herzine et al., 2002; Calas et al., 2008; Meme et al., 2009; Lantz et al., 2014; Laugeray et al., 2014). In humans, paternal exposure has been linked to developmental defects in their children (García et al., 1998).

We urge the South African authorities to reject the approval of these GM maize varieties, and not fall behind other nations that are taking precautionary steps to protect the health of their citizens from unsustainable, destructive agrochemical practices.

MOLECULAR CONSIDERATIONS

Characterising the genetic modification is necessary at the level of the genome to identify the location of the integration site of the transgene, stability of the transgenes as well as the number of copies of the transgene integrated into the maize genome. Any disturbances at the genomic level could have consequences for the transcriptomic, genomic or metabolomic activity of the plant, which may have adverse health, environmental and/or agronomic implications such as alterations in nutrient or toxin levels.

Description of the recombinant DNA before and after modification

The transgenic material in the three GM varieties have been generated synthetically and therefore has no history of safe use in nature. A detailed description of the sequence of the transgenes should therefore be provided. Independent analysis of data provided to Indian authorities for parental lines NK603 and MON 89034 found unintended modifications in the inserted transgenic DNA (Then, 2013).

No such information is provided to justify claims that the inserted into the three GM varieties is stable. Southern blotting analyses performed to assess the integrity and copy number of transgenes insertions are unable to detect subtle DNA rearrangements, requiring additional techniques such as polymerase chain reaction to allow for more complex analyses.

The data provided does not confirm the integrity of the transgene sequences, nor does it substantiate claims made by the applicant that the integrated DNA is stable, and that only a single copy of the transgene is present in each GM variety.

Characterisation of the indel

The applicant does not provide any details on the specific location of the transgenes in any of the GM varieties. There is no sequence information or description of the flanking genomic DNA provided. The applicant therefore does not provide information to confirm a lack of disruption to endogenous maize genes or regulatory sequences. The applicant should provide details showing a lack of disruption to the endogenous maize genome.

Lack of molecular characterisation to rule out unintended effects

Genetic modification is associated with unintended effects at the level of the genome, transcriptome, proteome and metabolome. For example, studies have shown that expression of Bt toxins Cry1A.105, Cry2Ab2 an EPSPS proteins in GM maize can cause changes in global proteome levels in GM maize varieties, resulting in impacts on metabolic pathways (Agapito-Tenfen et al., 2014). The NK603 parental line used for two of the GM maize varieties in the applications, has also been shown to have altered proteome and metabolome profiles. Using unbiased 'omics' global profiling techniques to assess such changes, Mesnage et al., (2016) documented altered levels of proteins and metabolites indicative of oxidative stress, alterations in levels of enzymes involved in glycolysis metabolism, as well as TCA cycle involved in energy production in NK603 maize. Metabolome alterations also included a 28-fold rise in polyamines, which play multiple roles in cell growth, survival and proliferation; they can be either toxic in certain contexts.

All three GM varieties also carry genetic elements such as the t-nos terminator and the cauliflower mosaic virus 35S promoter that are associated with the production of novel RNA variants and genetic rearrangements respectively (Ho et al, 1999; EFSA 2009). Finally, the use of the vector Agrobacterium tumefaciens has been shown to induce genetic deletions, insertions, chromosomal rearrangements, translocations, scrambling of sequences and epigenetic (chemical modifications of DNA) perturbations (Jupe et al., 2019). This vector was used to develop NK603, and MON89034 parental lines.

The three varieties have not been adequately characterised at the molecular level. The applicant should be asked to provide further data, including global 'omics' profiling data for all three GM maize varieties to ensure a lack of unintended molecular effects that may have health or environmental implications.

SAFETY ASSESSMENT

Establishing the food and feed safety of the three GM maize varieties is essential considering that maize is not only consumed by humans and animals in South Africa, and is **an important staple crop consumed on a daily basis**.

A number of claims of safety are made in the safety assessment that are questionable. For example, the applicant concludes that the three GM maize varieties are compositionally equivalent to conventional varieties of maize; 2) they proteins produced by the transgenes are not acutely toxic to mammals; 3) have a history of safe use; 4) the proteins have no structural similarities to known toxins or other biologically active proteins that could cause adverse effects, and 5) the transproteins are rapidly digested in mammalian gastrointestinal systems.

However, without thorough safety assessments such as chronic toxicological feeding studies, such claims remain unsubstantiated, as detailed below.

Substantial Equivalence

The principle of 'substantial equivalence' for risk assessment is not a risk assessment but an analytical exercise that compares arbitrary comparators of GM crops to any variety or composite of varieties of conventional crops, such as levels of total fibre, limited number of minerals, fat and sugar.

The applicant claims that all three varieties are compositionally and nutritionally equivalent to their non-GM counterparts. However, no details of the results are presented in the applications, making it impossible to substantiate their claims. Indeed, details were only attainable through regulatory publications from the European Food Safety Authority (EFSA) (EFSA, 2019), which was not presented in the applications. This data shows that there were significant differences for one of the GM varieties MON 89034 x TC1507 x NK603 x DAS-40278-9. Samples taken from a single season field trial in the US in 2010 were analysed, finding that levels of 23 of the 82 constituents measured were significantly different in the GM variety, including iron, folic acid, isoleucine, manganese and b-carotene. Further, taking samples from a single season in the US, not South Africa is of limited relevance for a South African risk assessment. Different environmental conditions, as well as different levels of herbicide applications are likely to influence the composition of the plants, making the data provided inadequate for drawing conclusions on equivalence and safety. Further, the limited data that does exist shows substantial non-equivalence, making claims of equivalence deliberately misleading. Similarly, EFSA (EFSA, 2016) data for DAS-40278-9 shows that samples taken from a single season in 2009 from a US field trial found levels of 20 of the 59 constituents measured, to be significantly altered in both 2,4-D sprayed and unsprayed crops, while 26 were significantly different when sprayed with

quizalofop. No EFSA data was found for NK603 x DAS-40278-9, and none was provided by the applicant to be able to confirm their claims of substantial equivalence for that variety.

Such narrow definitions of equivalence are also challenged by more detailed compositional analyses done by independent researchers. As highlighted above recent 'omics' profiling techniques that look at thousands of genes, proteins and metabolite expression levels, found significant changes in the NK603 parental line, including levels of potentially toxic polyamines (Mesnage et al., 2016).

No claims of safety can be made on the three GM varieties on the basis of the compositional data provided by the applicants. **We urge that the applicant is required to provide more detailed analyses including 'omics' profiling to confirm a lack of altered compositional profiles for all three varieties. Further, samples should be analysed from field trials conducted in South Africa, under various environmental conditions including more extreme weather conditions, such as drought, and with differing levels of herbicide applications. This is need to more accurately reflect the South African context within which such crops would be cultivated, in order to provide relevant, biologically meaningful data.**

Toxicology

No data was provided by the applicant on any feeding studies performed in mammals for the three GM maize varieties. No data appears to exist for feeding studies using the whole plant.

The only data available on acute toxicity are mouse feeding studies for DAS-40278-9 documented in an EFSA assessment (EFSA, 2016). This data shows that mice fed on the AAD-1 protein expressed in the GM variety, caused altered biochemical blood parameters such as lower haemoglobin levels in males, high serum alkaline phosphatase in males. Males also had higher prostate weights. This goes against the claims made by the applicant that the AAD-1 protein "is not acutely toxic and does not cause any adverse effects".

No feeding studies have been conducted or referenced for the stacked event MON 89034 x TC1507 x NK603 x DAS-40278-9, **making it impossible to make any conclusions with regards to the acute toxicity of this variety.**

Independent research challenges claims by the applicant that newly expressed proteins present in MON 89034 x TC1507 x NK603 x DAS-40278-9 have a 'history of safe use', are not allergenic, or do not survive mammalian digestion, based on predictive bioinformatics analyses and simulated in vitro experiments, instead of empirical data based on in vivo animal studies. For example, studies have now linked Cry toxins to immunogenic reactions in mammals. Cry1Ac (which shows some similarity to Cry1A.105) is known to enhance immune reactions Vásquez-Padrón et al., 2000), and thus be potentially allergenic. Several Bt toxins have been shown to be haematoxic, and also to have potential impacts on human health (Shimada et al. 2003; Huffmann et al. 2004; Ito et al. 2004; Mesnage et al. 2012; and Bondzio et al. 2013). Cry toxins have also been shown to survive mammalian digestion, being detected in mice intestines, going against claims that exposure and thus toxicity, is limited (Vázquez-Padrón et al.,1999).

Finally, combinatorial effects may occur due to interactions between the novel transproteins and metabolites produced in the stacked variety. For example, having multiple Bt toxins may have cumulative or synergistic effects on non-target organisms. This is the basis for the EU regulation that requires risk assessment of stacked traits which defines a stacked event derived from conventional breeding of existing single event GM varieties as a "new entity" (Regulation (EC) No 1829/2003). It takes into account the possibility of stacked varieties showing disturbances in transgene and host genome stability, expression of novel proteins, and potential synergistic/ combinatorial interactions between the individual modifications. Such interactions in stacked events have been documented in stacked maize that carries both Bt toxins and glyphosate

tolerance, showing alterations of transgene expression in the stacked versus single event lines (Vilberte et al., 2016).

The applicant however, fails to provide any feeding studies for compositional or allergenicity tests on the stacked varieties, despite the application stating under Section 8: "*For stacked events (e.g. A x B x C), nutritional and compositional data should be provided for the stacked event under consideration, i.e., data for the single events (A, B and C) in the stack or data for a higher-level stack (e.g. A x B x C x D) do not replace the need for specific data relating to the stacked event (A x B x C, in this example) that is the subject of the application*".

Without chronic long-term feeding studies and more detailed toxicological tests that encompass parameters such as immune responses, reproductive toxicity, effects of pesticides, combinatorial effects of multiple transgenes, such conclusions of safety cannot be drawn for any of the three GM maize varieties.

ENVIRONMENTAL RISK ASSESSMENT

The environmental risk assessment fails to assess the full extent of risks posed by the three GM maize varieties. First, it does not take into account the ecotoxicological effects of their corresponding herbicides (either as single herbicides are in combination with Bt toxins), despite growing recognition that agricultural chemicals and industrial farming systems are one key factor that has promoted biodiversity loss (FAO, 2019). Second, it fails to present any data on potential effects various parameters such as on non-target organisms, potential for gene transfer, or effects on biogeochemical processes. Nonetheless, the applicant misleading claims to have conducted a "thorough environmental risk assessment.

Impacts on non-target organisms

The applications for DAS-40278-9 and NK603 x DAS-40278-9 fail to address the issue of toxicity to non-target organisms, thus any conclusions regarding safety to non-target organisms cannot be drawn. With regards to MON 89034 x TC1507 x NK603 x DAS-40278-9, generalised claims that the Bt toxins are toxic only to lepidopteran pests is made. They also make mention of tests on non-target organisms in South Africa, but no data, description of the study or even which non-target organisms were assessed is presented. It is thus impossible to draw conclusions regarding the safety of MON 89034 x TC1507 x NK603 x DAS-40278-9 to non-target organisms.

Independent research challenges the specificity of Cry Bt toxins, raising concerns of their potential impacts on South African biodiversity. For example, existing data shows that many Cry toxins are not as specific as previously thought and have detrimental effects on a variety of beneficial organisms such as pollinators (Ramirez- Romero et al., 2008), pest predators (Hilbeck et al., 2008; 2012) and soil fungi (Castadini et al., 2016).

The herbicides associated with the GM crop varieties are also associated with adverse ecotoxicological effects. Glyphosate is well established to exert toxic effects on wild-life including with many studies showing toxicity to a variety of species including aquatic organisms, amphibians, soil organisms including worms, insects including endangered species (Sirinathsinghji & Ho, 2015). A recent paper also showed it to be toxic to the microbiome of honeybees, threatening their ability to defend against pathogens (Motta et al., 2018). 2,4-D has also been shown to exert ecotoxicity, and is known to degrade poorly in the environment. Toxic effects have been reported in soil organisms including worms (Singh & Singh, 2015) as well as fish species (Gallagher and Di Giulio 1991; Coady et al., 2013).

SOCIO-ECONOMIC CONCERNS

Following over 20 years of GM crop cultivation in South Africa, there is no evidence that GM crops have improved on the levels of hunger and food insecurity which remain high, with an estimated 46 % of households still going hungry every day.

The generalised claims that approval of these GM maize varieties will "help in improving yields", and "enhance" cultural traditions, are also challenged by their own field data, as well as independent studies highlighting the outperformance of GM varieties in South Africa, by local open-pollinated varieties that are better adapted to local agro-ecologies and fluctuations in weather conditions (Fischer, Van den Berg and Mutenga, 2012). GM regions such as the US are also lagging behind the EU (almost a completely GM-free region) in cereal grain production (Heinemann et al., 2013), highlighting the lack of benefits associated with the costly technology of expensive seed and their need to be used alongside additional chemical inputs.

Food production and yields across Southern Africa are already negatively affected by degraded soils, pests and climate change. Industrial farming practices, including the high use of external inputs, chemical pesticides and monoculture practices have been shown to contribute to these problems as highlighted in this report, and to result in drastic biodiversity decline - a threat to future food production (FAO, 2019); and to also be one of the largest contributors to climate change. The UN FAO (2017) also recently reported that it is a "myth" that pesticides are necessary to feed the world, are a global burden on human health, with catastrophic impacts on the environment and human health as a whole." This system is unsustainable and a direct threat to food production and socio-economic circumstances of the South African people in an era of unpredictable climate change.

Pesticide toxicity that accompanies these varieties also needs to be considered. The EU has estimated that the economic burden of illnesses induced by pesticide exposure could reach billions of euros. In Europe, unlike South Africa, diets are largely free of GM herbicide tolerant crops, while South African staple foods are set to be further polluted with such chemicals.

The socio-economic impacts of these three varieties of maize need to be further analysed, and the misleading claims of benefits made by the applicant. We urge the South African authorities to ignore such claims.

ALTERNATIVES

While the GM varieties serve to offer an unsustainable path of further industrialisation of the food system, further consolidate the corporate domination of the South African food system and pollute our foods and lands with war chemicals, solutions exist that can not only increase yields, but also offer holistic environmental solutions that increase biodiversity, protect against climate change and thus improve long-term resilience and environmental and human health. For example, push-pull systems developed specifically for Southern and Eastern African agricultural systems, by African and international agricultural experts have shown huge successes in preventing pest destruction by major pests including corn stemborers and the fall armyworm, with adoption of the technology increasing significantly in the last 15 years as a result. It is a low cost technology, proven to increase biodiversity and presence of natural pest predators, soil fertility, as well as provide additional crops for animal fodder, and production of drought-tolerant intercrops. This technology is thus highly suited to small-holder farmers, and has most crucially been shown to increase incomes and livelihoods (see Khan et al., 2014 and Khan et al., 2018 for reviews).

It is crucial that we move towards agro-ecological solutions such as push-pull technologies before we undermine our future food production systems any further by GM industrial monoculture systems that merely serve the purpose of producing profits for foreign multinationals.

CONCLUSIONS

The ACB firmly rejects the approval of the three varieties of GM maize to enter South Africa for general release, based on inadequate evidence of safety to both human health and the environment, as well as concerns over socio-economic impacts they will have over the people of South Africa.

We urge that the authorities take a precautionary approach in rejecting these varieties to protect people and the environment.

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