Comments on the ARC appeal against the decision not to approve the general release of genetically modified potato, SpuntaG2, in South Africa (17/3/1-ARC-VOPI-08/039)

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1. Introduction

The African Centre for Biosafety welcomes the opportunity afforded to it by the Appeal Board to participate in the appeal process in the Agricultural Research Council's (ARC's) appeal against the decision not to approve the general release of genetically modified potato, SpuntaG2, in South Africa (17/3/1-ARC-VOPI-08/039). We have engaged vigorously and constructively in the various regulatory processes related to SpuntaG2 since 200. In this regard, our activities have included:

- Objection to the application for a permit for additional trials with insect resistant Bt Cry V Genetically Modified Potatoes, June 2004;
- Additional Comments and Objections to Continued Trials of GM Potatoes, March 2006;
- Publication of a comprehensive study titled 'Hot Potato GM potatoes in South Africa-a critical analysis'; (We made copies of the booklet available to every member of the Executive Council: GMO Act, under the direction of the Registrar);
- Objection to the commercial release of the Agricultural Research Council (ARC) genetically modified potato, September 2008;
- Consultations with various stakeholders including consumers, social movements and retailers;
- Set up a consumer petition in the short time afforded us to comment on the commercial release application; and
- Alerted the Biosafety Focal Points of our SADC neighbours to the application under consideration and sent them our publications.

These documents have all been made available to the Registrar: GMO Act 1997 and can also be accessed on our website at http://www.biosafetyafrica.org.za/index.php/Potato/menu-id-100023.html

We trust that the information that we have gathered and the insights we have gained in this process will be of value to the Appeal Board in the course of its deliberations.

2. Summary of ACB's key responses to the ARC's objections

The Agricultural Research Council (ARC) has put forward objections to the socio-economic and technical findings of the Executive Council (EC), which resulted in the rejection of their application for commercial release of SpuntaG2. A summary of the ACB's responses to the ARC's ground for appeal are set out below.

Socio-economic issues

- The African Centre for Biosafety has previously submitted

comprehensive comments on the potential socio-economic impacts of commercially releasing the tubermoth resistant potato. Our key finding was that the adoption rate of this technology was going to be minimal, as small and commercial farmers alike did not feel that the technology would be of benefit to them or assist in increasing production. Ultimately the potential negative impact of consumer rejection far outweighed any potential benefits.

Furthermore, retailers, guided by consumer preferences, were not supportive of the application. Potatoes South Africa objected to the application, being the first time that organised agriculture has lobbied against a new GM crop. Furthermore, the inevitable transboundary movement of GM potatoes to neighbouring countries, where biosafety regimes are incomplete, and in context where no labelling and segregation systems are in place in South Africa, constitutes a serious legal and ethical concern.

- The ARC has contrived a process whereby they insist that a commercial release is necessary in order to carry out participatory trials with small-scale farmers to assess a wide array of factors including productivity, storage, taste, marketability, risk assessment and management and transboundary movement. This is not a valid process as such assessments should form part of the pre-release trial studies. We question why the ARC is intent on foisting this technology on the most vulnerable farmers in the country, rather than engaging in consultation with the sector and designing appropriate research in response.

- Monitoring and risk management measures of insect management, transboundary movement and adventitious mixing have not been adequately addressed.

- The potato tubermoth (PTM) is not a prevalent pest and is a particularly low priority for small-scale farmers, the majority of which are based in KwaZulu Natal, not Ceres where this pest is more prevalent. This will remain the case even if engineered into a stacked trait variety. Public funds should be spent on farmer-need driven research and ecologically sustainable solutions.

Technical issues

- The expression levels of npt11 and Cry1Ia1 in SpuntaG2 tubers has not been analysed and laboratory findings on the efficacy of transgenic Bt in controlling PTM in storage are meaningless.

- Several food safety issues are outstanding, including problems with the acute toxicology studies, nutritional composition and possible allergens.

- Studies carried out on the effects on non-target organisms produced confusing and meaningless results.

3. Background

The SpuntaG2 potato is modified to reduce potato tuber moth (PTM) infestations during storage. SpuntaG2 contains the Cry1Ia1 transgene that is a Bt insecticidal toxin active against all Lepidoptera (moths) and Coleoptera (beetles).

The ARC Bt potato project has a long history linked to a number of international institutions. Funded by the USAID, the project began in the United States and was subsequently transferred to Egypt after market rejection in the USA. After 8 years of research in Egypt it was again rejected by consumers and export markets and abandoned before an attempt at commercial release was made. South Africa subsequently received the project as a 'hand-me-down' as the interests vested in the project pursued their goal to have a GM crop potato commercialised. 8 years of research by the Agricultural Research Council culminated in an application to the South African government for commercial release of SpuntaG2 potatoes in July 2008.

The African Centre for Biosafety submitted a comprehensive objection to this application, in which we submitted independent assessments of the ARC's safety dossier by 3 experts in the fields of molecular biology, food safety and entomology. In addition we accessed the socio-economic studies that were commissioned by the ARC to determine the potential impact of this technology and brought them to the attention of the Executive Council. We also canvassed opinions from various stakeholder groups, including consumer groups, retailers and the potato industry. Our findings pointed overwhelmingly to the rejection of this technology.

The final decision of the Executive Council reflected our findings and the application was rejected on the following grounds:

- The Socio-economic impact study indicates that the commercial farmers do not anticipate this GM crop to present a significant lowering of inputs as the same spraying regime is required to manage other pests which this event does not target
- Small scale farmers identified more pressing challenges relating to production such as lack of water, seed availability, fertilizers, etc
- No evidence is presented that other pest management strategies against PTM have been considered or compared with the release of GM-Spunta

- The applicant presents several arguments of the value of this event for small scale farmers; however, entry of these GM potatoes into the formal trade remains a particular concern. Segregation of GM from non-GM potatoes would require and Identity Preservation System which is currently not in place
- The capacity of small scale farmers to implement risk management measures could potentially be onerous
- Considering the biology of potatoes, vegetative material (tubers) may be used for propagation, which may complicate risk management
- PTM is not a major pest for stored potatoes but rather rodents
- The Western Blot of transformed potatoes was limited to protein extracts from leaves and there is an assumption that one band represents the Cry1 la1 protein. No data is presented of expression levels in tubers
- Concerns on the toxicity testing by use of an animal feeding study was conducted with cooked (boiled) potato although raw freeze dried potato would have been better suited
- No evidence is presented that known allergens of potato, namely Sol t1 (patatin)are not over expressed in the GM potato
- No actual toxicity data of the cry-protein on the target organism PTM is presented.

On learning that the ARC had appealed this decision and submitted an appeal document to the EC, the African Centre for Biosafety formally requested an opportunity to participate in the appeals process. We also applied through the PAIA process to access the ARC's grounds for appeal in order that we may participate in a constructive and informed manner.

The Registrar informed us 26 November 2009 that access to this document was denied on the grounds of possibly prejudicing the outcome of the appeal. This prompted us to enlist the assistance of a lawyer to access this information and a long and costly process ensued before we finally had sight of the ARC appeal document on 24 February 2011. It is therefore distressing to find that the text of this document is freely available on the AgBioForum website in a 2010 article written by Dr. J. Thompson. (http://www.agbioforum.org/v13n4/v13n4a04-thomson.htm) We question why we were made to waste our limited time and resources on this protracted and unnecessary process.

Nonetheless, we are pleased that the appeals board has seen fit to include us in the appeals process and to afford us an opportunity to respond in writing to the grounds of appeal set forth by the ARC.

4. The African Centre for Biosafety's response to the ARC appeal

In response to the grounds upon which ARC are requesting an appeal (17/3/1-ARC-VOPI-08/039)*, the following specific responses need to be considered in relation to the points raised by the ARC

4.1. Socio-economic considerations:

4.1.1. According the ARC appeal document, the Executive Council's decision to reject their permit based on socio-economic reasons is procedurally flawed as no guidelines are established in the GMO Act to evaluate socio-economic impact. They go on to argue that in the absence of such guidelines the only way to determine such information is to allow the permit for commercial release so that participatory evaluations can be run with farmers.

However, the GMO Act, NEMA and the Cartagena Protocol on Biosafety all contain provisions for the consideration of socio-economic issues in decision making.

The EC is obliged to apply their minds to socio-economic issues in accordance with NEMA section 5(a), and Regulation 5 of the GMO Act stipulates that such an assessment may include (but is not limited to) information on the impact of the activity on the following:

- continued existence and range of diversity of the biological resources;
- access to genetic and other natural resources previously available;
- cultural traditions, knowledge, and practices;
- income, competitiveness or economic markets; and food security.

Article 26 of the Cartagena Protocol also allows for the consideration of socio-economic issues in decision-making, stating that,

> "The Parties, in reaching a decision on import under this Protocol or under its domestic measures implementing the protocol, may take into account, … socio-economic considerations arising from the impact of living modified organisms …".

Furthermore, the argument that a commercial release is necessary in order to carry out participatory trials with small-scale farmers to gather a wide array of information, including productivity, production constraints, appearance, storage, taste and marketability has no precedent. The aim of general release is market uptake and penetration and these issues need to be determined before market release. These aspects should clearly be part of trial release studies.

It is our contention that this research did not originate from a consultative

process in response to the real needs of small scale farmers, but is rather being imposed upon them. The ARC's own socio-economic study on the potential impacts of this technology on small scale farmers advised that a one size fits all technology approach is not appropriate. It recommended "adapting current technologies to local conditions" rather than "developing new technologies, which, due to their generic nature, are not adapted to local conditions and might not be adopted as a result".[i]

The same study also highlighted the incredible vulnerability of small-scale potato farmers in South Africa and the fact that they have no margin for risk. Diale Mokgojwa, head of Potato South Africa's emerging farmer programme, confirmed with us in a meeting that emerging farmers in their programme were nervous of the technology and preferred that their mentors who are more experienced farmers take the risk. And yet the ARC continues to insist on foisting this technology on these farmers. In the meantime these farmers are calling out for assistance with other more pressing production constraints, particularly water[ii]. Public funds would be better spent on attending to constraints identified by the farmers themselves.

In terms of assessing marketability, surely market research is possible without participatory trials with farmers. As shown in our original objection, the ACB has consulted widely with consumer groups and retailers and there is a strong feeling that this potato represents a threat to the market. It is for this reason that Potatoes South Africa has opposed this application. Over 90% of our potato exports are sold into neighbouring countries. Our investigations showed that export markets will be at risk due to the fact that our SADC neighbours are not prepared to receive genetically modified crops as their regulatory systems are not in place.

4.1.2. The ARC states that the intention behind developing a PTM resistant potato has been misunderstood, which is to create a stacked variety potato in a step-wise fashion that will address "all the major production constraints", beginning with tubermoth resistance.

Stacked varieties are not necessarily inherently desirable. For example, in October 2010 the New York Times reported on the massive farmer rejection of Monsanto's new "Smartstax maize, which incorporates 8 different traits into one seed. Farmers complained that they were expected to pay for traits that they did not want and refused to buy the product[iii]. This brings us back to the recommendation that a one-size-fits-all approach is inappropriate, and more so for farmers who do not have access to all the trappings of capital intensive agriculture, such as chemicals, irrigation and mechanization. If the ARC is in fact at the beginning of a process of developing a stacked variety and is beginning with a trait that clearly no one wants, it is likely that farmers will reject the final product.

In addition, best biosafety practice dictates that stacked varieties must be

evaluated as new events in themselves, as opposed to relying on safety information about their parental lines. In this case, the biosafety information supplied by the applicant was not of a high enough quality to allow the EC to bring this single trait to the market, regardless of whether or not the ARC is intending to develop a stacked variety in the future.

4.1.3. It is particularly important to be sure of genetic stability over several generations when it comes to potatoes and there was an intention to study the transgenic stability in the field trials. Despite the fact that there are claims of a specific PCR detection method for identity preservation, this has not been demonstrated and validated (if new data is available, this needs to be made publically available). The applicant has established the appropriate PCR methods to amplify the transgene (Appendix VII) as well as individual transgene elements such as Cry1Ia1, but did not use these tools to monitor transgenic stability in the field during the numerous field trials that have been carried out. This is particularly pertinent to potato cultivation since there is the established practice of propagating first class seed potato for 8 generations until it loses certification. Therefore, the molecular analysis needs ensure that the SpuntaG2 has genetic stability and integrity that is similar to the non-GM Spunta over several generations. These important experiments have not been carried out nor are they proposed to be part of the post-release monitoring program.

4.1.4. The risk management measures of insect management, transboundary movement and adventitious mixing have not been addressed in field trials, but the applicant wishes to carry out general release in order to carry out these assessments. Clearly, most of these risk assessments should have been part of field trials and there is a legal obligation to monitor transboundary movements (Biosafety protocol) of GMOs so that this method should also be used in a monitoring programme (the monitoring programme needs to be active and not merely rely on feedback of agronomic performance from farmers and consumers as the applicant has proposed).

4.1.5. The risk management measures do not address the different routes of gene escape; namely, human error, adventitious mixing through pollination and animal-dispersal of tubers in the field as well as through horizontal gene flow. The applicant only assumes that vegetative propagation is important and state that SpuntaG2 carries no additional risks of escape, but has failed to provide any evidence to support this claim.

4.1.6 The PTM does not seem to be a prevalent pest in South Africa. Of all the field trials documented to date, many failed simply because there was poor infestation of PTM at that location. Since 2001, only 4 (out of more than 20 field trials) delivered results that demonstrated the benefits of SpuntaG2 in controlling PTM. Many of the others had no data on efficacy, because the natural infestation of PTM was too low. In one case the applicant irresponsibly released PTM at a location in an attempt to get infestation (Report 2002/3,

page 12), but still failed "Despite release of 30000 moths, the level of infestation was low".

4.2. Technical issues:

4.2.1. Experiments were carried out to quantify the levels of npt11 as well as Cry1Ia1 the in the SpuntaG2. Unfortunately, in both cases this analysis was only carried out on the leaf tissue and not the tubers (Appendix V). Obviously, the levels in both leaves and tubers need to be analysed, since the claimed benefit of SpuntaG2 is the protection of potato tubers during storage. The levels of this Bt toxin need to be assayed in the tubers during the storage period (in addition to levels in other parts of the plant). The assumptions and extrapolations used to estimate the levels in tubers are not valid and there is no reason why the applicant cannot/did not carry out these tests- there has been several field trials and the opportunity to measure Bt levels in various parts of the plants, including tubers. Furthermore, the results presented for the Bt levels in leaves (Figures V.1 and V.2) are inconclusive since a only 1, 1.5 and 2 ug Cry1Ia1 was used as the standard and this produced a (saturating) signal that cannot be accurately quantified (Figure V.2) against the amounts in SpuntaG2. Also noteworthy is the poor specificity of the antibody used and the cross reactivity of a band immediately below the Cry1Ia1 band of 82.1 kDa and the fact that the standard also shows a band immediately below the 82.1 kDa Cry1Ia1, but this is barely discernible due to the saturating signal of the standards (V.2). The use this antibody for quantification is therefore unreliable. The data is also of poor scientific quality since the results show no replication and standard errors for these determinations that need to be accurately quantified.

4.2.2. The acute toxicology testing of Cry1Ia1 protein was also carried out, but there are several problems with this study. Cry1Ia1 expressed from the bacteria, *E.coli*, was used and the biochemical characteristics of this Cry1Ia1 compared to that expressed *in planta* (ie in SpuntaG2) have not been firmly established. The estimates of exposure are presumptions based on the highly questionable levels that may be in the tubers (estimated to be 10 fold less that the leaves). Furthermore, the potato was tested in the cooked form and this is not the form that non-target organisms in the field will be exposed to. This means that the tests with cooked potato are not invalid, but merely inappropriate and therefore uninformative for determining safety and lack of toxicology. The food safety testing of SpuntaG2 was limited to acute toxicological testing in a rat feeding study (Appendix XXI) and details of the cooking procedures have been omitted. In this feeding study, only means of starting weights of the rats are given. Furthermore, the high variation (standard deviation values, SD) at the beginning of the experiment can hide the growth and developmental changes. Despite this fact, the male rats grew to a greater size when fed SpuntaG2 compared to the controls (Appendix XXI, table 2), the applicant dismissed this merely because males and females were not similarly affected. Additionally, the clinical chemistry

parameters that were measured have unacceptably high variability; some had SD values +/- 30% or more (e.g. Ab lymphocytes). Such a high intra-sample variability will mask any differences between groups and this can be seen if a two- way analysis of variance of the results in Table 4 is carried out.

4.3.3. The applicant provides the sequence for the site of integration of the transgene, and demonstrates that no unexpected additional recombination events had occurred at the border sequences (Appendix IX). However, it is unclear if the insertion event has resulted in the interruption of a host gene. The applicant's results indicate that the transgene inserted into a host patatin gene (Appendix X1), but this is not referred to in the supporting documents. The biosafety risks associated with such an insertion event are uncertain and include loss of protein function and the generation of new allergens (neo-epitopes). The applicant argues that this insertion does not affect the antigenicity, function or amount of patatin in SpuntaG2 (compared to Spunta), but ARC provides little evidence to support this. The argument that the changes in amount of patatin will be revealed in gross compositional analysis is unfounded; particularly since the ptatin makes up a large proportion of the protein content in potatoes- (the patatin genes in potato encode proteins that compose up to 40% of the soluble protein in the tubers and are a critical nutritional component;(Prat et al. 1990). Therefore, small changes in the amount of patatin will be difficult to detect in these compositional analyses that determine total protein levels. Perhaps more important than the levels of patatin are the risks that the insertion into the patatin gene has resulted in the production of a new patatin gene product with new function and/or antigenicity. This has not been considered and Western blots (for patatin detection) or proteomics studies should be carried out to determine the protein expression profiles for SpuntaG2 compared to Spunta.

4.2.4. The efficacy of transgenic Bt on the protection of potatoes against PTM during storage has been adequately demonstrated from field studies. However, the results of the laboratory analysis are meaningless since:
(i) *Manduca sexta* (hookworm) was used as the target not the problem pest, *Phtorimea operculata* (potato tuber moth);
(ii) The experimental numbers are too low to be confident in any differences (starting with two hookworms) and looking for dose-dependent killing (ie 0 dead, 1 dead, 2 dead). The published assay for PTM uses 10 larvae with five replications, which is required in order to obtain reliable data, but this published and established method was not followed.; and
(iii) There is no reference for comparison. The extracts from Spunta G2 and Spunta potato should be used as controls since the aim of the experiment is too show that the bacterially produced Cry1Ia1 is the same in character and effect as that produce the transgenic potato, but this was not carried out.

There are several other key concerns that have not been adequately addressed to warrant the extension of SpuntaG2 beyond field trials:

- The molecular characteristics of SpuntaG2 have not been determined. The data presented in Appendix VII-X provide evidence of one copy of the cassette that had been integrated into the genome. Appendix IX.7 states that the "intensity of bands in the lanes with G3 genomic DNA is 2-3 times the intensity of the bands in the lanes with SpuntaG2 genomic DNA, consistent with the results of experiment 1 that indicate that G2 has three T-DNA copies". The use of Spunta G3 as the standard is inappropriate since there is no evidence presented (or in the published literature) that Spunta G3 does indeed contain three copies. The basic experiment using the plasmid used for transformation (pSPUD5) at different copies (1-5) alongside the SpuntaG2 transgenic digested with 3-4 different restriction enzymes needs to be carried out so that subsequent analysis of the images can be carried out in a quantitative way to demonstrate the copy number. Furthermore, there should be more than one probe used in the experiment since the transgens may have fragmented and integrated elsewhere in the genome (ie not only Cry1Ia1 but also npt11 and 35SCamV).

- SpuntaG2 has significant compositional differences compared to the non-GM Spunta potato. The Appendix XX detailed the compositional analysis of SpuntaG2 compared to the non-GM Spunta and incorrectly concluded that there were no significant differences observed. The data (Table XX.4) reveals that energy content (Petrus Steyn location) and potassium (all locations) were statistically significant. The applicant argues that since they energy content was not different at all locations this is not important. Similarly, the inconsistent variation in potassium is used to dismiss the significant potassium differences observed. This interpretation is highly questionable and scientifically flawed.

- The nutritional profiling is limited and does not assess known anti-nutrients. In terms of anti-nutrients, only the glycoalkaloids have been measured. A major omission to these studies is that the levels of antinutrients, trypsin inhibitors and potato lectin have not been measured. The data presented of the total glycoalkaloid levels in the replicates were also too variable to be certain that the differences between Spunta and SpuntaG2 were not significant (freeze dried samples from 2007- the standard deviations, SD, must be shown for these values so that the validity of the data can be assessed. Furthermore, in the general chemical analysis did not explain why the analyses had to be repeated and amended and how the analysed samples in the three sets of data were related to each other.

- Effects on non-targets. Evidence from the literature has shown that lacewings fed on aphid pests that had eaten Bt-maize took longer to develop and were two to three times more likely to die. Earthworms

have been shown to be affected and significant reductions in populations of the beneficial parasites *Microplitis* sp. (88.9% reduction) and *Campoletis chloridae* (79.2% reduction) were detected in Bt cotton field (Birch, *et al.* 1997, Marvier, M. 2001). The effects of Bt may be considerable and long-lasting since transgenic plants release Bt into the soil where it can remain for up to 234 days (Koskella, J. and G. Stotzky. 1997, Tapp, H. and G. Stotzky. 1998). The applicant carried out field trials to measure the effect on biodiversity and non-target insects (Apppendix XXII), but these studies were carried out at only 3 of the locations (Table XXII.1)- what became of the data from Patensie and Kokstad? The details of the traps (pitfalls and sweep nets) are not given and it appears that the frequency, number or layout of the of traps were insufficient- the monitoring non-target effects amounts to 4 to 6 days in a period of 3 years which will only provide a snapshot in time and the considerable intrasampling variability observed may obscure real differences between samples so that a two way ANOVA is required. Despite these limitations, the comparison of the SpuntaG2 with the non-GM Spunta for effects on non-target anthropods, did reveal differences: in Hemipptera and Diptera at Roodeplaaat (Table XXII.20 and Table XXII.21); Hymenoptera at Ceres (Table XXII.23) and thrips and Aphids at Perys Steyn (Table XXII.25 and Table XXII.26). These differences were all significant, however the applicant chooses to uses non-applicable controls (Spunta and SpuntaG2 grown at different localities) to erroneously conclude that there are no differences: "For any treatment to have a significant and stable effect on any organism the effect has to be present over time and repeated under different environmental conditions. We could not prove any of the aforementioned in trials..." (pg 29, Appendix XXII). This approach of using inappropriate controls is either a deliberate attempt to obfuscate the data or the aims have clearly been lost- what would be the point of choosing different locations if one would suffice?

- **Conclusion**

The ARC's response still does not address the fact that tubermoth resistant potatoes are not seen as a useful technology by small-scale and large scale farmers alike. The fact that the potato industry is not supportive of this project is testament to the fact that this research is not a response to real farmer needs on the ground and is not a prudent way to use limited public funds. The ARC is intent on foisting this controversial product onto the most vulnerable farmers in South Africa, even to the extent of using participatory trials with small-scale farmers as spurious reason for commercial release of Spunta G2 potatoes. This commercial release has the potential to damage the potato market and puts small-scale farmers in the frontline of this risk.

There are also numerous flaws in the design and interpretation of the applicant experiments as well as gross omissions in the biosafety tests carried

out to date. In the light of current scientific evidence that SpuntaG2 poses unacceptable risks for the human health and the environment, it should not be approved for general release and the decision of the Executive Council not to approve should be upheld.

[i] Smallholder Potato Production Activities in South Africa: A Socio-Economic and Technical Assessment of Five Cases in Three Provinces. TGB Hart (HSRC) and HJ Vorster (ARC). December 2006

[ii] ibid

[iii] The New York Times, 4 October 2010. Monsanto's Fortunes Turn Sour http://www.nytimes.com/2010/10/05/business/05monsanto.html?_r=1 accessed 13 January 2011