DEVELOPMENT AND DEPLOYMENT OF INSECT RESISTANT MAIZE

Proceedings of a Workshop Held
August 15-20 1999 at Whitesands Hotel Mombasa, Kenya

M. Siambi, S.N. Mugo and J.A.W. Ochieng, Editors

Sponsored by the

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IRMA PROJECT DOCUMENT 2

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The Kenya Agricultural Research Institute (KARI) was established in 1979 with the express mission of increasing sustainable agricultural production by generating appropriate technologies through research, and disseminating these to the farming community. Inherent to this mission is the protection, conservation, and improvement of the basic resources, both natural and human. Such resources are critical for Kenya's agricultural development and expansion of the nation's scientific and technological capacity. KARI has an extensive history of productive collaborators with national and international institutes and universities, as well as with the private sector.

CIMMYT® (www.cimmyt.cgiar.org) is an internationally funded, nonprofit scientific research and training organization. Headquartered in Mexico, the Center works with agricultural research institutions worldwide to improve the productivity, profitability, and sustainability of maize and wheat systems for poor farmers in developing countries. It is one of 16 similar centers supported by the Consultative Group on International Agricultural Research (CGIAR, www.cgiar.org). The CGIAR comprises about 60 partner countries, international and regional organizations, and private foundations. It is co-sponsored by the Food and Agriculture Organization (FAO) of the United Nations, the International Bank for Reconstruction and Development (World Bank), the United Nations Development Programme (UNDP), and the United Nations Environment Programme (UNEP). Financial support for CIMMYT's research agenda also comes from many other sources, including foundations, development banks, and public and private agencies.

CIMMYT supports Future Harvest,® a public awareness campaign that builds understanding about the importance of agricultural issues and international agricultural research. Future Harvest links respected research institutions, influential public figures, and leading agricultural scientists to underscore the wider social benefits of improved agriculture-peace, prosperity, environmental renewal, health, and the alleviation of human suffering (www.futureharvest.org).

The Novartis Foundation for Sustainable Development provides major funding for the project. The Foundation is dedicated to fostering sustainable development in poor countries of the South through its support of programs and projects in the areas of sustainable agriculture, health, and social development. It is also an active player in development policy debate through its preparation and dissemination of research analysis. Further information about the Foundation may be found at its web site (www.foundation.novartis.com).

The Insect Resistant Maize for Africa (IRMA) Project was launched in 1999 as a collaborative effort between CIMMYT and KARI. Its primary goal is to increase maize production and food security for African farmers through the development and deployment of maize that offers resistance to destructive insect especially stem borers. To achieve this goal, project scientists will identify conventional and novel sources of resistance to stem borers and incorporate them into maize varieties that are both well-adapted to Kenya's various agroecological zones and well-accepted by its farmers and consumers. Varieties and technologies that are appropriate for other African nations may be extended to them for their use.

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1.0 FOREWORD

Insect pests exact untold damage on crops of agricultural worth, especially in sub-Saharan Africa. The damage is manifested as moderate to high yield losses, translating into reduced on-farm productivity. This sad situation feeds into the vicious cycle of food insecurity and poverty in the region.

Many of the options available to redress this distressful situation may not be viable for various reasons, including toxicity of chemicals to people and the environment and the emergence of pesticide resistance among target insects. In the final analysis, Integrated Pest Management (IPM) will have to be adopted to reduce pest damage and yield losses attendant thereto.

It is universally believed that genetic resistance to insect pests forms the first line of defense against damage to crop plants by pests, as this is the most cost effective and environmentally friendly option. Developing resistance to insect pests may be achieved by means of conventional plant breeding and biotechnologically mediated breeding, including marker-assisted selection and genetic engineering, and genome transformation. Conventional breeding for resistance to insects is not only slow, but also not very effective for such an intractable character.

It is in light of the foregoing scenario that a biotechnology oriented project was recently promulgated in Kenya. The goal of this project is to develop and deploy insect resistant maize varieties, with the prime motive of increasing on-farm productivity of maize, which is the mainstay of Kenya's cereal staples. This will serve KARI's vision of putting more food on the table for the vast majority of rural and urban poor.

Deployment of the new technology will, perforce, be accompanied with ex-post impact assessment to verify that this insect resistant maize will have achieved measurable impact on food (read maize) availability to the poor populace. So far, two workshops have been held, the objective of which is to bring the new technology on board. It is my hope that all parties involved (CIMMYT and KARI, financially supported by the Norvatis Foundation for Sustainable Development) are ready to implement this novel technology for posterity. I believe our agricultural research system is ripe for innovative technologies. Nevertheless, we intend to adopt these technologies judiciously, carefully, and in a responsible manner, in order to sustain the agricultural resource base.

Cyrus G. Ndiritu
DIRECTOR KARI

2.0 Background

The workshop brought together scientists from Kenya Agricultural Research Institute (KARI), the International Maize and Wheat Improvement Center (CIMMYT), and the donor, the Novartis Foundation for Sustainable Development, to consult, share information, and plan the way forward for development and deployment of insect resistant maize in Kenya using all available genetic tools. This action followed CIMMYT's initiative to develop and avail insect resistant maize germplasm in developing African countries. The Novartis Foundation for Sustainable Development was approached as a possible donor. Kenya was singled out to serve as an example, after which the technologies and experiences will be extended to other willing African countries. KARI was approached due to its expressed interest in technologies that combat the ravages of stem borer and reduce dependence on chemical pesticides in maize production. The idea of this workshop was to solicit views of KARI scientists and to promote development of the project.

The five-day workshop involved 43 participants and was divided into three sessions. Sessions I and II were devoted to presentations on the background of the workshop, the need for the project, and key scientific approaches and achievements on development of insect resistant maize. Information on biosafety regulations and the stand of participating institutions towards biotechnology in general, and GMOs in particular, was also shared.

These two sessions also provided an opportunity for the scientists to share their individual views towards the project and the utility of the emerging novel biotechnology in maize production. The summaries of these presentations and the ensuing discussions form the first part of this document.

The third session was devoted developing a workplan. Discussions were held in three groups namely:

- Product development
- Product deployment
- Impact assessment.

Each group addressed broad objectives and developed logic frames (logframes) that linked activities to objectives while spelling out the expected outputs and their timelines.

The proceedings from the third session were developed into a separate document, the Strategic Plan of the IRMA project. The next stage will, of necessity, involve promoting this novel technology to the wider Kenyan public.

3.0 Technical Presentations

3.1 Official opening remarks and objectives of the workshop

Joseph A.W. Ochieng, KARI Headquarters

Dr. Ochieng opened the workshop on behalf of the Director of KARI. The objectives of the workshop were two-fold. The basic facts that are important to understanding the expectations and outputs of the project are as follows:

1) Insect pests cause significant losses in maize production;
2) Chemical control has environmental drawbacks, capital input implications, and presents hazards to farmers;
3) Conventional breeding has not achieved as much as expected due to the nature of inheritance of resistance to stem borer; and
4) Biotechnological approaches will reduce the time required to obtain resistance due to the novel approaches they employ.

The objectives of the workshops were outlined as follows:

1) To facilitate discussions by KARI and CIMMYT maize research scientists about problems of insect pest damage and to examine available options to identify strategies for developing pest resistant maize varieties.
2) To address policy issues regarding biosafety in handling GMOs, particularly transgenic maize.
3) To state the case for the need to use transgenic maize as an important option for control of maize stem borers.

The goal of this collaborative effort was to reduce maize yield losses due to stem borer, and make more food available to the small-holder farmer. This would be achieved by developing stem borer resistant maize varieties. The net result would be a positive step toward the alleviation of poverty among rural and urban poor in Kenya and a technology model for developing countries in Africa that rely on maize as a staple food.

The workshop discussed the potential of maize germplasm as a putative source of resistance to stem borer and identified existing maize genotypes as putative recipients of the Bt gene. Different biotechnological approaches and complementary conventional breeding methods were identified. Apart from enhancing the collaboration, the workshop was meant to identify areas of technology development in which KARI scientists in different disciplines could make tangible contributions. Finally, the technologies to be developed and deployed should be socioeconomically acceptable and feasible for small-holder farmers.

3.2 Perspectives on Bt technology

Klaus Leisinger, Novartis Foundation for Sustainable Development

It is imperative that the discussion on the use of Bt technology be as candid and as open as possible. All available information must be provided because potential detractors will use the information gap to rally support against the project as has been experienced in Europe and elsewhere. The Novartis Foundation (NF) has no hidden agenda in providing funds for the project and does not intend to draw any royalties or other economic benefits from the success of this project. The NF position, as partners in this project, is that the project's major goal is to develop technologies that will be of direct benefit to the small-holder farmer in developing countries. The fact that the scientific soundness of the Bt gene technology is not being questioned is encouraging to the Foundation, and therefore, gives it confidence as a partner in this noble endeavor.

Debates on biotechnologically engineered food resources are currently raging in all continents. The debate in Europe is different because it has political undertones. One must take into account the fact that Europe is a food surplus environment while the developing world is food deficient. That way, the debates can be taken within their proper context rather than made to appear universally relevant. If this project is to succeed, more time must be spent in the establishment of a firm foundation, both in terms of biosafety and information dissemination. It is our suggestion that KARI should go in for the strictest biosafety standards to avoid embarrassment in future. The scientists should not underestimate the power some of NGOs in whipping up public emotions. Those who will be working in this collaborative project must themselves be fully convinced that the technology is sound, offers a novel approach to dealing with an old problem, and that the accrued benefits will eventually make a difference to the lives of small-holder farmers in sub-Saharan Africa. One very important point to bear in mind in this project is the fact that whatever product is developed, it must be acceptable to the farmers. These are the people we are all working for in this collaborative project.

This project is envisaged as a North/South collaborative effort being facilitated by CIMMYT. This is an institution of outstanding repute for research in maize, and whose ability to handle the technology leaves no doubt in our minds as proven by the modest gains made to date. There will be an opportunity for intensive discussions on what has been done so far. In addition, CIMMYT has an outstanding track record on addressing the concerns for food security in sub-Saharan Africa. It has to be remembered that the African continent missed out on being part of the "Green Revolution". This time round, Africa should not be left behind. It must be clearly understood that the issue of GMOs, and for that matter any genetic manipulation of organisms, is not a biological issue but one that transcends ethical, political, and social boundaries.

3.3 Problems of maize stem borer in Kenya and possible areas of project activities

Macharia Gethi, KARI/RRC Embu

Dr. Gethi provided background about maize production and consumption in Kenya. Available data on yield loss due to stem borers were from farmer perceptions from field surveys. The perception of yield loss due to pests is difficult to quantify by interviewing farmers. This is because of compounding effects of other factors that contribute to the yield gap. Yield losses attributed to stem borers are 20-50%. Stem borer is the most important pest in all maize growing regions of Kenya. Stem borer proliferates in the minor and major seasons, however, it is prevalent in lower elevations. Other pests of maize include cutworm, aphids, African bollworm, termites, and chaffer grubs.

The geographical distribution of the major stem borer species is as follows:

1) The pyralids, *Chilo partellus* (spotted stem borer) and *Chilo orichalcociellus* Strand (coastal stem borer) in low altitude, warmer zones below 1,200m, and *Eldana* Saccharina in the Lake Victoria region, where it also attacks sugarcane and maize; and

2) The noctuids *Busseola fusca* Fuller in higher altitude, cooler areas above 1,500m and *Sesamia calamistis* hamp in all zones.

Stem borer has a wide host range especially in the families of wild grasses that have stems large enough to accommodate larvae feeding e.g., *Eldana sacharina* is known to attack sedges. In the native grasses, larvae densities do not reach high levels found in cultivated crops. The introduction of maize about 100 years ago in East Africa has undoubtedly had an impact on the abundance of lepidoptrean stem borer by providing a highly nutritious food source with little or no resistance to the pest. This is evident by the fact that larvae survival and pupal weight is higher when reared on maize than on selected wild grasses.

The possible areas of project impact will be development of tolerant germplasm in target areas depending on the prevalence of the stem borer species, i.e., *Chilo* in the coastal and midaltitudes, and *Busseola* in midaltitude zones.

Past experiences and information from the maize database indicate that the project should focus on these three zones. However, once resistant varieties have been developed, the regional breeding programs that are not within the initial three zones can utilise them. For the past 30-40 years of maize research in Kenya, efforts have been mainly on breeding for higher yields while ignoring losses caused by other stresses. Thus far, the maize varieties available commercially are all susceptible to stem borer and yield losses will still occur despite use of chemical insecticides.

3.4 Conventional breeding for resistance to stem borer in Kenya

Charles J.M. Mutinda, KARI/NARC Muguga

The most damaging stage of stem borer is the larval stage. Leaf lesions, dead heart, and stem tunneling, characterise the damage. Conventional techniques for identifying sources of resistance involve rearing of good quality insects for artificial infestation, and evaluation of the damage caused. Artificial infestation may involve use of eggs or first instar larvae placed in the leaf whorls. Damage evaluation can be done using a foliar damage rating, dead hearts four weeks after infestation, and stem tunneling at harvest.

Conventional breeding methods show that inheritance of resistance to stem borer is polygenic, with additive genetic variance as the major mode of gene action. Therefore, a recurrent selection breeding program that enables accumulation of alleles for resistance to stem borer should be effective in increasing resistance. A definitive breeding protocol may be recurrent selection with full-sib or half-sib with S1 or S2 selection to develop improved populations from which pedigree breeding can be initiated to develop inbred lines for developing hybrids and/or synthetics.

Successful host plant resistance projects are dependent on

1) Availability of genes for resistance in the original population,
2) An efficient insect rearing technique,
3) An efficient artificial infestation method,
4) Efficient evaluation of genotypes, and
5) Suitable breeding procedures to incorporate resistance into suitable genetic backgrounds.

These will be required in the proposed project.

Studies at ICIPE have led to identification and development of resistant lines such as MP701, MP702, MP704, and populations ICZ1-CM, and ICZ2-CM. These studies also show that Katumani Composite B has moderate resistance to stem borer, while Kitale inbred line A is highly susceptible.

3.5. Biotechnology approaches to breeding for resistance to insects

Duncan Kirubi and Zachary Muthamia.

Insect resistant maize varieties possess heritable qualities that allow infested cultivars to give higher yields than similar cultivars that do not have these characteristics. The mechanisms of plant host resistance include antibiosis, antixenosis (non-preference) and tolerance. In antibiosis, the resistant genotype, when fed upon, will affect the life cycle of the insect, causing death or abnormal behavior. Antixenosis refers to the lack of attractiveness of the host as food or shelter for the insect due to plant vigor. Tolerance is the ability of the host plant to support a certain population level of insects due to plant vigor or the ability to repair the damaged tissue without loss of quality or yield.

Conventional breeding has advantages and disadvantages. It is an economically and socially acceptable and an environmentally safe method of crop protection. However, it is slow and difficult to incorporate the genes that confer resistance against the pests because of the cycles of recurrent selection required. On the other hand, marker-assisted selection (MAS) is much quicker if chromosomal locations of the genes that control resistance to stem borer and suitable genetic markers for this region are identified. Therefore, transfer of important quantitative genes takes less time and is more effective. The effects of genotype x environment interaction are also avoided when using MAS.

Some important aspects to consider in the development of transgenic maize are

1) Availability of a suitable vector to carry the required gene;
2) Whether the target gene (Bt) is to be inserted in the nucleus or in the chloroplasts;
3) Modification of foreign DNA to increase expression in plant cells;
4) A methodology to deliver plasmid DNA into plant cells;
5) Selection methodology to identify the transformed cells;
6) Tissue culture technique to identify the transferred cells; and
7) Tissue culture methodology to recover viable plants from transformed cells.

Once these are decided on, the available techniques to deliver the foreign gene to plants are

1) Microinjection,
2) Electroporation,
3) Silicon carbide fibers,
4) Particle bombardments using the biolistics or particle gun (gene gun), and
5) Agrobacterium tumefasciens mediated transfers.

Bt transgenic maize technology

Bacillus thuringiensis is a soil borne bacteria, which produces crystal, or

Cry, proteins exhibiting highly specific insecticidal activity to insect larvae. The endotoxins are nontoxic to mammals, and it is widely used as a biological control agent because of its specificity for a certain group of insects. Bt toxins can be classified as follows:

1) *Cry I* against Lepidopteran larvae
2) *Cry II* against Lepidopteran and Dipteran larvae
3) *Cry III* against Coleopteran larvae
4) *Cry IV* against Dipteran larvae
5) *Cry V* against Lepidopteran and Coleopteran larvae

Bt transgenic maize with resistance against the European corn *borer (Ostrinia nubilalis)* has been developed. However, some Lepidopteran larvae have been shown to develop resistance under laboratory and field conditions, thus development of resistance in field populations is possible and appropriate resistance management technology needs to be developed.

The following aspects have to be considered in development of this project:

- Bt genes have not been extensively tested for toxicity to tropical stem borer, as there are only a few reports on *Chilo* and *Busseola* from South Africa. Deployment of transgenic maize with Bt was recently initiated in South Africa.
- Due to binding specificity of Bt toxins, each species should be screened against a standard library of Bt toxins to determine the best constructs to be used within a region.
- Stem borers in the tropics are more aggressive in feeding and have several life cycles per cropping season.
- Identification of the *cry* genes to be used and an evaluation of how effective are they in controlling target pests?
- How will the transgenic plants be deployed in the field to minimize development of resistance?

Suggested strategy for development of transgenic stem borer resistant varieties for Kenya:

1) Incorporate different constitutively expressed *Cry* toxins in various adapted lines;

2) Develop synthetics from 6-10 lines expressing different toxins and other valuable characteristics;
3) Number of *cry* genes will depend on the expense of transformation and/or backcrossing to incorporate *cry* genes, and the number of available constructs; and
4) Hybrids with Bt could follow later.

3.6 Biosafety regulations-status and issues

Benjamin Odhiambo

The Kenya Agricultural Research Institute has made a deliberate effort to tackle issues of biosafety associated with production and handling of GMOs. Realising the need to safeguard both people and the environment, KARI has developed institutional guidelines for its crop and livestock research.

KARI has had previous experience with tissue culture techniques (pyrethrum, ornamentals, potatoes, strawberry, and bananas). The application of genetic manipulation and recombinant DNA technology is currently limited to livestock. KARI has already formed an institutional biosafety committee drawing membership from the Ministry of Health, the International Livestock Research Institute (ILRI), the Department of Veterinary Services, the University of Nairobi, and Jomo Kenyatta University of Agriculture and Technology.

The major role of KIBC is to advise the KARI management on formulation and implementation of institutional guidelines for research and development of GMOs. In addition, the committee certifies compliance of facilities for DNA work depending on the category of risk, and approval of proposals from the institute for the category of risk for onward transmission to the natural biosafety committee.

The Director-KARI appoints a biosafety officer (BSO), who is a member of KIBC. The biosafety officer does periodic inspections of biocontainment facilities and ensures adherence to the set standards. The officer also provides feedback to KIBC about the state of facilities and any violations of the established guidelines. The guidelines define four levels of physical biocontainment and associated laboratory practices, containment equipment, and special laboratory design required at each level.

Proposals for release of live organisms outside containment facilities must be directed to the National Biosafety Committee indicating objectives of the project, nature of organisms and of the genetic material, and control of the released organisms.

3.7 Efficacy and deployment of transgenic plants for stem borer management

David Bergvinson

Transgenic plants expressing *Bacillus thuringiensis* δ-endotoxins are now being used commercially in several crop species. These toxins have demonstrated good control of temperate *(Ostrinia nubilalis)* and tropical (*Diatraea grandiosella* and D. *saccharalis)* stem borer in maize. Resistance to B. thuringiensis toxins has been reported in over 11 species in laboratory and field studies, demonstrating the need for resistance management strategies to prolong the efficacy of this valuable pest management tool within an integrated control program. Resistance involves reduced binding of toxins to midgut epithelial cells and is generally considered to be a recessive trait. Resistance management will require the use of spatial and temporal refugia, which may require unique schemes for each pest complex. Information was presented on the mode of action of *Cry* toxins, resistance mechanisms, interaction of transgenic plants and biocontrols, and management/deployment strategies for transgenic maize in tropical ecologies.

Integrated pest management has historically placed a great emphasis on the development of host plant resistance. The development of conventional host plant resistance often involved quantitative traits at several loci, making the resistance durable but difficult to achieve. With the advent of transformation techniques, genes, which confer resistance to pest organisms, have been inserted into crop plants including maize, rice, soybean, potato, tobacco, and cotton.

Among the biological pesticides, bacteria have been the most successful group of organisms identified for insect control on commercial crops. The best examples of bacterial insecticides come from two soil bacterium, *Bacillus thuringiensis* and B. *sphaericus.* Insecticidal crystal proteins, called δ-endotoxins, produced by these bacteria are highly toxic to certain pests, yet cause little or no harm to humans, to most beneficial insects, and to other nontarget organisms. After being activated by midgut proteases, B. thuringiensis toxins bind to epithelial brush border membrane vesicles (BBMV), creating pores that result in cell lyses. Incorporation of genes encoding d-endotoxins into maize has provided astonishing levels of resistance and tremendous excitement in crop protection. However, concerns over environmental hazards and widespread resistance in pest populations have restricted the deployment of toxin producing plants.

Transgenic plants containing insecticidal proteins will feature prominently in agricultural systems in both developed and developing countries. Entomologists, breeders, molecular biologists, and population ecologists need to determine how best to deliver this technology to provide good pest control and reduce environmental hazards (including gene flow and retarding the development of resistance in pest populations). To achieve these objects, we need to better understand the pest biology, behavior, and response to insecticidal proteins; the temporal and spatial expression of toxins in transgenic plants; the dynamic of different refugia strategies in resistance management; the impact of toxin producing plants on biological control; and how to deliver this package to resource poor farmers.

4.0 Discussion from technical presentations

Question **Who are the clients for these products?**

Answer Resource-poor farmers in midaltitude areas. We may wish to focus on small-scale farmers, but it is alright if benefits spill over to the large-scale farmers.

Question **Has the farmer been consulted in the formulation of objectives?**

Answer Farmer problems were identified from farm surveys. Farmers will be invited to the Stakeholders Meeting in March 2000.

Question **How will the product be labeled for the market?**

Answer We could label transgenic maize using aleurone or endosperm color or any other easy identification method.

Question **What are the relationships among Ciba-Geigy, Norvatis Company, and the Norvatis Foundation for Sustainable Development?**

Answer The merger of Ciba Geigy with Sandoz created Norvatis. The Novartis Foundation for Sustainable Development is devoted to humanitarian purposes, and it has projects in health, agriculture, and social development. The Foundation is independent of Novartis Company

Question **What of Intellectual Property Rights (IPR) issues in this project?**

Answer On IPR, members of the CGIAR review recommended that CIMMYT should try to establish IPR. Intellectual property rights do not mean you have to sell the product. However, a company using these materials should not protect the germplasm in order to restrict its access by other CIMMYT partners. This project will not work with Norvatis Foundation materials, so the question of IPR does not arise among the CIMMYT, KARI, and NF partners, but may arise with other partners if their products are involved.

Question **What are the effects of transgenic maize on parasitoidsused for biological control of stem borer? A biosafety issue?**

Answer These will be addressed within the project activities through laboratory and field studies. We will need to evaluate the efficacy of Bt against other control agents and other beneficial and nontarget organisms.

Question **What of Bt resistance management?**

Answer Interactions between farmer practices and insect resistance management will need to be studied to establish compatibility of farmers' practices and use of Bt genes in maize. Training of farmers on use of refugia for maintaining heterozygous plants, in order to avoid homozygous double recessive genetic situations.

Question **Will coast Kenya be included since the region is a hot spot for stem borer of various species?**

Answer All agroecological zones will be addressed, depending on severity of the problem and value of products.

Question **What is the LD50 of Bt gene/spray?**

Answer Not of concern since Bt has no known mammalian toxicity. Expression of Bt genes is often specific to targeted plant parts. Of concern is the possibility of stem borers developing resistance to Bt toxins, and gene flow into other plant species.

Question **Might present maize imports into Kenya be transgenic?**

Answer We do not know, as at the moment there is no quick and useful way to tell whether particular maize is transgenic or not. Imports are usually declared as a control measure.

Question **What is the time factor in issuance of a permit to introduce Bt maize?**

Answer Due to biosafety concerns, there are established procedures to apply and be granted permission to import GMOs into Kenya. There are forms that go through scrutiny by institutional and national biosafety committees. However biosafety research on Bt maize has shown the product to be safe. Anticipation of problems that are realistically non-existent will stall the progress of the project.

Question **What type of germplasm will be imported?**

Answer The Bt genes at CIMMYT are currently in a hybrid background, CML216 x CML72, used for its ease of transformation. This will then be backcrossed to target and adapted germplasm.

Question **Who will negotiate for gene constructs?**

Answer CIMMYT.

Question **How will impact be assessed?**

Answer One project staff member based in Kenya is a socioeconomist, who will assess impact throughout the project cycle.

Question **Are the Bt genes stable across stress environments?**

Answer We need to consider G x E interaction. Experiments under drought and irrigation have shown that the gene expression is not different under these two environments.

Question **Is there evidence of significant losses due to stem borer in Kenya?**

Answer Data is spotty-over extrapolation from a few locations to a wider region. This is an opportunity to go out and systematically collect data on yield losses and farmers' perceptions of these losses. Farmers' perceptions of yield loss estimation are higher than actual losses. These data will arm us in case questions arise later

Question **Where are we in the project? Has it already started and only now linked up with KARI?**

Answer Certain aspects of the project have been on-going in the time leading up to this workshop. The work of Dr. Natasha Bohorova has laid the basis for transformation of CIMMYT materials for use in tropical germplasm for tropical environments. Most of the work in other places has been on temperate materials. It is fair to say that the current CIMMYT materials are not the most appropriate materials for this environment, therefore, we are beginning all over.

Question **Is gene flow an issue?**

Answer Gene flow is an issue in Mexico because of its being a center of origin for maize. Gene flow is less likely to be an issue here in Kenya.

Question **Does the Bt gene come as one gene or multiple genes? And, how do we develop the defined product?**

Answer Bt comes as single dominant gene. We could develop one or multiple genes and transfer it or them to target germplasm through a series of backcrosses. Multiple genes are preferred in order to slow down development of resistance by targeted borers.

Question **Why has work not been done on Kenyan borers?**

Answer Due to regulations governing insect and host importations.

Question **Is Bt going to be introduced into hybrids or synthetics? And, what about the question of the cost of seed?**

Answer The project is clearly targeting the small-scale farmers in sub-Saharan Africa-mainly through OPVs, but hybrids will also be developed where they are required. All Bt genes act as single dominant loci , and can be used in inbred or hybrids.. There will be at least one copy of the gene in each plant.

Question **How much background work is required to transform materials before getting to the level of transgenics?**

Answer These processes will be the subject of training throughout the project cycle. We may not wish to transform Kenyan germplasm at this particular time, but will select important ones and backcross them with already transformed genotypes to introduce the Bt gene into elite and adapted Kenyan germplasm. Materials developed as such can then be grown everywhere and at all locations without a problem. Insertion of a gene is what is considered in the regulatory process for US and Europe. Backcrossing is not considered in the regulatory process because it is assumed that the gene is already safe.

Question **Conceptual reservations appear to exist. Scientists should feel convinced of the efficacy of the technology deep down in their hearts. Orelse they may not withstand public criticism.**

Answer The project is acceptable in Kenya, subject to some provisions. This meeting was called to iron out any differences that may exist or doubts in the minds of the scientists before opening it up to other stakeholders.

5.0 List of participants

	Name	Discipline	Institution
1.	Mr. W.K. Malinga	Entomologist, Center Director, Kibos	KARI/NSRC-KIBOS
2.	Mr. Wilson Muasya	Breeder	KARI/NDFRC-KATUMANI
3.	Dr. Moses Siambi	Agronomist	KARI/NDFRC-KATUMANI
4.	Mr. Samuel N. Njihia	Entomologist	KARI/NARC-MUGUGA
5.	Dr. Charles W. Kariuki	Entomologist	KARI/NARC-MUGUGA
6.	Mr. Z.K. Muthamia	Breeder	KARI/NDFRC-KATUMANI
7.	Dr. George A. Ombakho	Breeder	KARI/NARC-KITALE
8.	Dr. Jane M. Ininda	Breeder	KARI/NARC- MUGUGA
9.	Mr. Patrick Kalama	Entomologist	KARI/NARC-KITALE
10.	Dr. Charles Mutinda[1]	Breeder	KARI/NARC-MUGUGA
11.	Dr. Margaret Mulaa	Entomologist	KARI/NARC-KITALE
12.	Dr. J.A.W. Ochieng	Breeder, Assist. Director, Food Crops	KARI-HQTS
13.	Dr. Dennis Friesen	Agronomist	CIMMYT-NAIROBI
14.	Dr. Klaus M. Leisinger	Administrator	NOVARTIS FOUNDATION
15.	Dr. Schimit	Administrator	NOVARTIS FOUNDATION
16.	Dr. Jost Frei	Administrator	NOVARTIS FOUNDATION
17.	Mrs. B.D.S Salasya	Socioeconomist	KARI/RRC-KAKAMEGA
18.	Dr. S.M Wangia	Socioeconomist	KARI-HQTS
19.	Dr. F.M Muriithi	Socioeconomist	KARI/RRC-EMBU
20.	Mr. W.S. Chivatsi	Breeder	KARI/RRC-MTWAPA
21.	Dr. Duncan T. Kirubi[2]	Breeder	KARI/RRC-EMBU
22.	Mr. Geoffrey M. Kamau	Agronomist	KARI/RRC-MTWAPA

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23 Dr. Rahab W. Muinga	Animal Production, Center Director, RRC Mtwapa	KARI/RRC-MTWAPA
24 Dr. Eliud C.K. Ngugi	Breeder	KARI-KATUMANI
25 Dr. Stephen N. Mugo[3]	Breeder/ Physiologist	CIMMYT- MEXICO
26 Dr. Dave Hoisington[4]	Breeder/ Director ABC	CIMMYT- MEXICO
27 Mr. P.J. Ninnes	Administrator	CIMMYT MEXICO
28 Mrs. Josephine M. Songa[5]	Entomologist	KARI/NDFRC-KATUMANI
29 Mr. David Poland	Information	CIMMYT-MEXICO
30 Dr. David Bergvinson	Entomologist	CIMMYT-MEXICO
31 Dr. Alpha O. Diallo	Breeder, Liaison officer, Kenya	CIMMYT NAIROBI
32 Dr. Munene Macharia	Entomologist	KARI/NJORO
33 Dr. Ben. Odhiambo	Biotechnologist, Head KARI Centre	KARI/NARL-NAIROBI
34 Dr. Shivaji Pandey	Breeder, Maize Program Director	CIMMYT-MEXICO
35 Dr. Natasha Bohorova	Molecular Biologist	CIMMYT-MEXICO
36 Dr. Wilson Songa	Pathologist, Deputy Director, KEPHIS	KEPHIS-NAIROBI
37 Dr. Prabhu Pingali	Socioeconomist, Director Economics Program	CIMMYT-MEXICO
38 Dr. Macharia Gethi	Entomologist	KARI/RRC-EMBU
39 Ms. Elizabeth Wekesa	Socioeconomist	KARI/RRC-MTWAPA
40 Dr. Kiarie Njoroge	Breeder, Coodrinator Maize Research	KARI/NDFRC-KATUMANI
41 Ms. Grace Kimani	Information	KARI-HQTS
42 Dr. Wilfred Mwangi	Socioeconomist, Director of Agriculture, Kenya	MOA/CIMMYT
43 Dr. Hugo De Groote	Socioeconomist, IRMA project	CIMMYT-NAIROBI
44 Mr. J. Kasango	Research Assistant, AMS project	CIMMYT-NAIROBI
45 Dr. Luta Muhammed	Socioeconomist	KARI/NDFRC-KATUMANI

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