Genetically Engineered Crops in Developing Countries
Eds.: DVR Reddy, P Ananda Kumar, P Lava Kumar, G Loebenstein &
C Kameswara Rao. **Studium Press LLC, Houston, USA. 2014;** pp. 377-397

CHAPTER

13

Current Status and Future Direction for Research on Agricultural Biotechnology in Ghana

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ABSTRACT

Biotechnology and related fields of science have over the years been used extensively to revolutionize agriculture and crop production. Although Genetic Engineering (GE) technology has rapidly progressed worldwide and several initiatives on the

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broadening of biotechnology research and outreach exist in Africa, GE is yet to be applied in crop improvement in Ghana and as a result GE crops are not under cultivation with the exception of approval fortransgenic high protein sweetpotato under confined field trials. The worldwide exposure to biotechnology has led to a myriad of institutions in Ghana to develop applied research and training in modern agricultural biotechnology with the aim of improving crop yields and quality. This chapter provides a brief description of current and emerging uses of biotechnology in sweetpotato engineered with high essential amino acids and enhanced protein levels. Attempts to improve protein content of yam (Dioscorea rotundata), an important staple in Ghana by GE are also described. A protocol for yam organogenesis and regeneration, minimum inhibitory concentration of antibiotic level in the selection media following co-cultivation, and Agrobacterium-mediated transformation are discussed. This successful transient transformation and regeneration protocols could be adopted in GE the crop for enhanced protein content of selected root crops. Several programs have been initiated in Ghana to build national capacity in modern biotechnology with respect to human resource and infrastructure development that include establishment of the National Biosafety Committees and its Institutional / technical subcommittees and the National Biosafety framework. A brief survey on the current status and future requirements for biotechnology in Ghana was conducted in 2012 involving farmers, regulators, researchers and lecturers in the field of biotechnology and related areas in Ghana. It is evident that biotechnology can be meaning fully integrated into the existing crop improvement programs. It is apparent that GE crops can have a dramatic impact on the development of Ghana by enhancing its food production and ensuring its nutritional requirements and economic growth, thus attaining food security.

***Key words*:** Dioscorea rotundata, sweetpotato, Ghana, ASP-1, protein, Biotechnology

1. INTRODUCTION

Biotechnology in the context of evolution of agricultural practices is literally defined as the use and manipulation of biological living materials to produce or modify products or processes to address problems in all areas of agricultural, biomedical and industrial production and processing. Modern agricultural biotechnology encompasses current and emerging aspects of molecular markers; genomics and bioinformatics; recombinant DNA (rDNA) technology; expression profiling; systems biology; proteomics; molecular diagnostics; tissue culture; mutation breeding and genetic engineering as applied to improve yields and quality of mostly high value crops, livestock and other farming processes. In this chapter we mainly focus on genetic engineering that involves the use of recombinant DNA technology for the introduction of desirable traits into crop plants. The first genetically engineered

(GE) crop was deregulated in 1996 and since then there has been an upward trend in commercial cultivation of transgenic crops. The major traits of focus have been pest resistance, herbicide tolerance and crop quality introgressed into such crops of interest as soybean, maize, canola and cotton. Currently 160 million hectares of farmlands in some 29 developed and developing countries, that include USA, Brazil, Argentina, India, Canada, China, Paraguay, Pakistan, South Africa and Uruguay, are under GE crops (James, 2011). This represents a 94-fold increase in hectarage making GE crops the fastest adopted crop technology in the history of modern agriculture (ISAAA, 2011). Although GE technology has rapidly progressed worldwide and several initiatives on the broadening of biotechnology research and outreach exist in Africa, GE crops are yet to make an impact in Africa. They are currently grown only in South Africa, Burkina Faso and Egypt. In Ghana GE is yet to be applied in crop improvement and as a result GE crops are not under cultivation with the exception of approval for a confined Field Trial to test transgenic High-protein sweetpotato (Egnin and Quain, personal communication; http://edition.myjoyonline.com/pages/news/20121).

Ghana is a country in West Africa located Latitude $4^0$44'N and $11^0$11'N; Longitude $3^0$11'W and $1^0$11'E with a 550 km long coastline and a land area of 230,940 sq km, comprising of 8,520 sq km water. The population is estimated at 24.22 million (2000 National population and housing census), with a population growth of 2.4% per annum. Agriculture is the backbone of Ghana's economy contributing up to 34% of GDP (2007 Ghana Statistical Services). Within the agriculture subsector, food crop production contributes 66.2% of the GDP. Agriculture (production up to marketing) offers employment to about 60 percent of the population. A 2011 study by the Forum for Agriculture Research in Africa (FARA) revealed that agricultural biotechnology in general is being increasingly adopted as a strategic tool for improving crops and livestock productivity in Africa. Several institutions in Ghana have initiated applied research and training in modern agricultural biotechnology with the aim of improving crop yields and quality (FARA, 2011).

2. CURRENT RESEARCH ON PRODUCTION AND EFFORTS ON DEREGULATION OF GENETICALLY ENGINEERED CROPS IN GHANA

This section briefly presents the current and emerging uses of biotechnology in sweetpotato GE with high essential amino acids and enhanced protein levels (Egnin and Prakash, 1995 and 1997; Prakash *et al*., 1997; Shireen *et al*., 2001 and 2002; Egnin *et al*., 2001 and 2002; http://spaceresearch.nasa.gov/docs/highlight, 2003; Chassy *et al*., 2007 and 2008), for which a Standard Operating Procedure (SOP) has been developed and more importantly an application for contained laboratory experiments and confined field trial in Ghana has been approved (Egnin *et al*., 2012; http://edition.myjoyonline.com/pages/news/20121). Attempts to Improve protein content of yam (*Dioscorea rotundata*), an important staple in Ghana (Quain *et al*., 2011 and 2012b) by GE are also described.

2.1. Genetically Engineered Sweetpotato

Sweetpotato (*Ipomoea batatas* (L.) Lam) ranks first among crops in the production of edible energy per hectare, and third in the production of edible protein per hectare (Horton, 1987). The crop contributes both to household food production and in many areas a source of income primarily through local sale of freshly harvested roots (Onwueme, 1978; Horton, 1987; Prakash *et al*., 1997; Chassy *et al*., 2007 and 2008). While sweetpotato is primarily used for human food and animal feed it is also an industrial crop serving as a source of starch and ethanol in many countries (Horton, 1987; Prakash *et al*., 1997; Chassy *et al*., 2007 and 2008).

Like many other major food crops, sweetpotato has a nutritional value limited by the low level of sulfur containing and other essential amino acids (Walter *et al*., 1981 and 1985; Prakash *et al*., 1997; Chassy *et al*., 2007 and 2008), required in human diets. It is extremely poor in tryptophan, methionine and lysine, and to a lesser extent in threonine, isoleucine and phenylalanine. Fortunately the storage protein content and the nutritional quality can be improved through genetic engineering (Egnin and Prakash, 1995 and 1997; Gama *et al*., 1996, Prakash *et al*., 1997). A GE sweetpotato, ASP-1, was developed from the genotype PI 318846-3, a cultivar from the Timor islands near Indonesia, with a dry white flesh type root having total storage root protein ranging from 1.3 to 2.84% of the dry weight (Egnin and Prakash, 1995 and 1997; Prakash *et al*., 1997; Shireen *et al.,* 2001 and 2002; Egnin *et al*., 2001 and 2002).

The 292 bp artificial storage protein (*asp-1*) gene used in this transformation was the result of *denovo* synthesis of a KOZAC translational enhancer (8bp) at the C-terminal of its sequence comprised of 80% essential amino acid codons. This *denovo* coding sequence was driven by a 600 bp CaMV35S promoter and a 300 bp 3'-end *Nos* terminator region. The resultant protein was a tetramere with B-turn and a very stable protein. Nutritional value was 6.5 times higher than the Zein protein (Maize) and 3 times higher than the phaseolin (bean) protein. Its structural folding and nature was based on the native Zein and phaseolin seed storage proteins (Jaynes *et al.,* 1985). The *asp-1* was designed to have a stable protein-like structure in plants, comprising 4 helical repeating units each with 20 amino acids. Its helical region was amphipatic, stabilized by several GLu-Lys salt bridges. The helix breaker Gly-pro-Gly-Arg was used as a turn sequence. This design resulted in an antiparallel tetramere (Fig. 1), with an extraordinary stable secondary and tertiary structure even at low concentration (Jaynes*et al*., 1985; Espinoza *et al*., 1989; Destéfano-Beltrán *et al*., 1991; Egnin and Prakash, 1997; Chassy *et al*., 2008).

A binary vector based on the *Agrobacterium tumefaciens* Ti- plasmid vector system was used to transfer *asp-1* and marker genes into sweetpotato. Binary vectors were disarmed but contained origins of replication from *E. coli* and *Agrobacterium* with both right and left borders region of homology with the Ti plasmid. These binary vectors did not require forming a cointegration and were

considerably easier to mobilize into *Agrobacterium* through a freeze and thaw protocols (An, 1998; Egnin *et al*., 1998). They can be introduced into any *Agrobacterium* host containing any Ti or Ri Helper-plasmid, as long as the vir helper function was provided. In this case the Ti or Ri plasmids were devoid of all the oncogenic genes and border sequences. Using these systems, the transgenes of the T-DNA transferred into plant cells by *A. tumefaciens* were stable and irreversible (Zambryski *et al*., 1982). Such a binary vector pBI121-C_2H containing a chimeric *asp-1* gene flanked by a selectable marker gene at its right border and a reporter gene (*uid-A* or *gus-A*) at its left border (Fig. 1) was a gift from Dr. Jaynes (Jaynes *et al*., 1985; Yang *et al*., 1989).

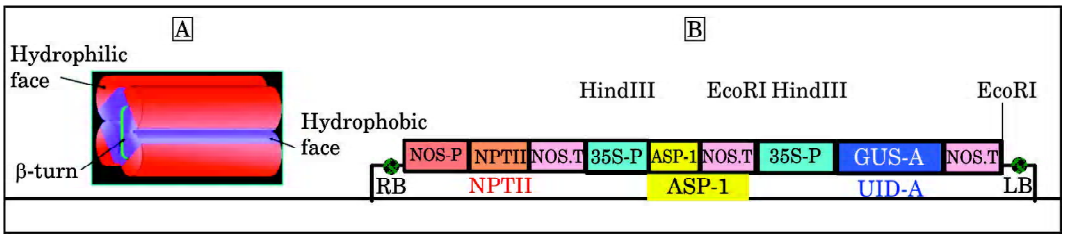

Fig. 1: A: ASP-1 protein tetramere and B: Binary plasmid pBI121C_2H containing *asp*-1 (encoding artificial storage protein rich in essential amino acids), *nptII*, and *uid*-A (GUS). LB = left border and RB = right border; *Hin*d III and *Eco*RI, restriction enzymes; *nptII*= neomycin phosphotransferase gene conferring kanamycin resistanc; Nos-T = Polyadenylation site of the nopaline synthase gene; 35S-P = promoter from the 35S gene of Cauliflower Mosaic Virus (CaMV); Nos-P = Nopaline syntase gene promoter. (Jaynes *et al.,* 1985; Yang *et al*., 1989; Egnin and Prakash, 1995 and 1997).

This pBI121C_2H containing the *asp-1* gene was mobilized into the disarmed *A. tumefaciens* strain, C58 containing a helper plasmid for transformation of plant cell (Egnin and Prakash, 1995) and subsequent development of the "High-Protein transgenic sweetpotato" (Egnin and Prakash, 1995 and 1997; Prakash *et al*., 1997; Egnin*et al*., 2001 and 2002). The transgenic plants expressed the ASP-1 protein, and the increased protein content in the roots (6.5to 12% protein as compared to 2.84% protein on a dry weight) was primarily due to enhanced levels of several native proteins, in particular sporamin and ß-amylase. Levels of many essential amino acids, such as methionine, threonine, isoleucine, and lysine were also increased by two to five fold, while tryptophan levels increased by several orders of magnitude. A serving of 300 g of the high-protein sweetpotato could provide all the daily nutritional requirements-EAA- for a child. With a conventional sweetpotato, the child will have to eat a serving of 4 kg sweetpotato to get the daily nutrient requirements of EAA (Egnin and Prakash, 1995 and 1997; Prakash *et al*., 1997; Shireen *et al*., 2001 and 2002; Egnin *et al*., 2001 and 2002, http:// spaceresearch.nasa.gov/docs/highlight, 2003; Chassy *et al*., 2007 and 2008). For

the first time the GE Sweetpotato was approved by the Ghana National Biosafety Committee for confined field trials in Kumasi, and indeed a positive step towards introduction of GE crops (http://edition.myjoyonline.com, 2012).

2.2. Genetically Engineered *Dioscorea rotundata*

Dioscorea rotundata (white yam) is a tuber crop of the family Dioscoreaceae and genus *Dioscorea* with the centre of origin being West Africa and an important staple food crop in Africa (Onwueme, 1978). Yam has 7–100 g dry weight of protein, less than 1–100 g dry wt fat, 80–100 g dry wt carbohydrate, and 3–100 g dry wt dietary fibre (IITA, 1980). It is apparent that carbohydrate is the major food component in yam and this constitutes the main dry matter (Orkwor and Asada, 1998). Yam is a monocotyledonous crop cultivated clonally by using the edible tuber and in a few instances the vine. Sparse flowering coupled with poor synchronisation of male and female flowering phases has become a stumbling block for successful sexual hybridisation. The production of yams in Africa is confronted by several constraints that include post harvest diseases and limited availability of planting material compounded by dormancy. The viruses, fungi, nematodes and insects affect crop yield and consequently returns from the crop. These constraints emphasize the need to carry out breeding programs that will improve crop yield, nutritional value and biotic stress tolerance (Quain *et al*., 2011; 2012b).

Tissue culture can complement the production constraint through the provision of disease free planting materials. Organogenesis, as an *in vitro* methodology for generating whole plants from tissues, and organs, is a vital tool for crop propagation and improvement. Yam breeding programmes have mainly focused on clonal selection from land races and hybridisation of elite genotypes within and between species (Sadik and Okereke, 1975; Akoroda, 1985). Hence the options of organogenesis and somatic embryogenesis must be explored. Several regeneration attempts have been reported in the development of callus and plantlets fromstem segments of *D. opposite* (Nagasawa and Finer, 1989), tubers of *D. rotundata* (IITA, 1974; Ng, 1984), as well as *D. alata* (Mantell *et al*., 1978). Zygotic embryos have also been used in the development of somatic embryos in *D. rotundata* (Osifo, 1988). Initiatives in crop breeding programs for the improvement of quality including protein content, as well as longer shelf-life, and disease resistance, using various important West African yam varieties need to be undertaken (Quain *et al*., 2011).

Initially efforts were made to develop an efficient regeneration protocol coupled with *Agrobacterium*-mediated transformation for *D. rotundata*, using an intron-containing a glucuronidase (*gus A* or *uidA*) gene, under the transcriptional control of CaMV 35S promoter serving as a reporter. As a result an optimized protocol was developed in crop breeding programs for the improvement of protein content, using nutritional protein gene such as *asp-1* gene, longer shelf-life and disease resistance, using various important West African yam varieties (Quain *et al*., 2011).

2.2.1. *Major findings*

Yam organogenesis

The observations made at the embryo induction stage of the Quain *et al*. (2011) protocol indicated that 3 days exposure of petiole explants to 2,4-D supplemented medium was ideal for the *Dioscorea* species, whereas as a longer duration of 1 week or more as described by Egnin *et al*., (1995, 1998) was not desirable. Following three days of incubation on the step one medium of the two-step organogenesis protocol, as adapted and modified from Gosukonda *et al*. (1995*a*), petioles were slightly swollen at the basal end (Fig. 2) confirming the reports by Gosukonda *et al*., 1995a; Dessai *et al*., 1995. Shoot production was obtained 21 days on the stage two treatments with higher shoot regeneration frequencies in explants incubated in an inverted vertically manner followed by a vertically upright position. As shown in Fig. 2 and Table 1, medium supplemented with TDZ (0.2 mg L-*1*) alone had 7+1.35 shoots regenerating per petiole explant; however, significantly higher number of shoots per explant (9±1) were obtained on medium supplemented with a lower concentration of TDZ (0.05 mg L-1) in combination with 2iP (0.02–0.2 mg L-1) (Quain *et al*., 2011).

Fig. 2: Explant Regeneration Patterns of Yam Basal Petioles. A: Initial Petiole explant; B: Swollen petiole explant base with emerging shoot primordial; C: Fully developed plantlet 21 days post-initial culture *Source*: Quain *et al.,* 2011.

Table 1: Mean number of shoots obtained in the various treatments of explants following a three-day incubation on MS 2,4-D; *Source*: Quain *et al*., 2011.

Treatment	***Mean number of regenerants/explant***
0.2 mg l^{-1} TDZ	7±1.35
0.05 mg l^{-1} TDZ + 0.2 mg l^{-1} 2iP	9±0.1
0.05 mg l^{-1} TDZ + 0.02 mg l^{-1} 2iP	7.5±0.23
0.05 mg l^{-1} TDZ + 0.05 mg l^{-1} 2iP	8±1.03

Note: No regeneration obtained in cultures of *D. rotundata* (yam) accessions BA 97 001 (local name – Labrekor) and *D. alata* accession SO 89/97A (local name – afase).

Substantial improvement in yam regeneration was observed as compared to previous protocol where only a maximum of four shoots were regenerated per nodal cutting explants (Ashun, 1996). In the present study high potency for shoots regeneration in yam from the basal portion of yam petioles was demonstrated. Therefore coupled with successful transformation, prospects for generation of GE yam using *A. tumefaciens* (Quain *et al*., 2011) have improved. Although petioles were reported to be recalcitrant in regeneration studies (Dodds *et al.*, 1992) experiments by Gosukonda *et al*. (1995 a and b) have demonstrated that the petioles could be used for regeneration and hence suitable for subsequent genetic manipulation.

Minimum inhibitory concentration of antibiotic used in selection media

The following experiments were aimed at determining the minimum inhibitory concentration (MIC) of antibiotic that served as the selective agent. The MIC ensures that the concentration used allowed transformed cells to regenerate into shoots without exerting lethal effect on the explant tissues (Egnin *et al*., 1998; Quain *et al.,* 2011). Prior to yam transformation, petiole explants were subjected to selection pressure in the two stage regeneration experiment to determine the minimum tolerance level of kanamycin. A kill curve of 150 mg/l was obtained as indicated in Table 2. This concentration did not result in callus production and shoot production; however the explants were viable. Kanamycin concentrations up to 80 mg/l in media with control cultures resulted in shoot regeneration with excess phenolic exudes, however, this was not observed in 100–150 mg/l treatments. When antibiotic levels were increased to above 150 mg, explants were brown and necrotic without any viable development. Excess of phenolic exudates in yam culture were problematic as they decreased explant viability and regeneration as well as the effectiveness of Agro-infection. However, pretreatment of washing in half strength MS prior to Agro-infection and plating on medium supplemented with 2,4-D, reduced polyphenolic exudate.

Table 2: Kill curve of yam minimum inhibitory concentration of Kanamycin.

Kanamycin mg/L	***Explant development***
0	Multiple shoot regeneration
25 to 80	Shoot regeneration
100	Explant swelling and callus production but no shoot production
140	Very small callus production but no shoot p
150 possible kill curve	No callus production and no shoot production
180	Explant browning, no callus production, arrested development

Agrobacterium-mediated transformation

We have developed an efficient *Agrobacterium*-mediated genetic transformation and regeneration system, applicable across a range of yam cultivars, to efficiently produce GE plants (Quain *et al*., 2011). An intron-containing *gusA* as a reporter gene was used to develop the protocol along with several investigative factors (polarity of the petiole during different cocultivation days). Transformation frequency was evaluated by scoring the number of sectors expressing GUS activity on the petiole explants, petioles vertically inverted followed by a vertically upright position, resulted in higher transformation frequencies regardless of the strain used. The optimized protocol yielded transient transformation frequencies ranging from 25% (C58) to 65% (EHA101) for petioles. The disarmed *A. tumefaciens* strain EHA101 was superior in facilitating the transfer of *uidA* gene to yam cells compared to the disarmed strain C58.

This successful transient transformation and regeneration protocols of putative transgenics could be adopted in engineering the crop for enhanced protein content (Fig. 3). The high level of transformation efficiency was encouraging since few monocots were natural hosts of *Agrobacterium.* The breakthrough study herein opens up substantial opportunities for biotechnology work in the area of genetic engineering of yam with desired traits (Quain et *al*., 2011).

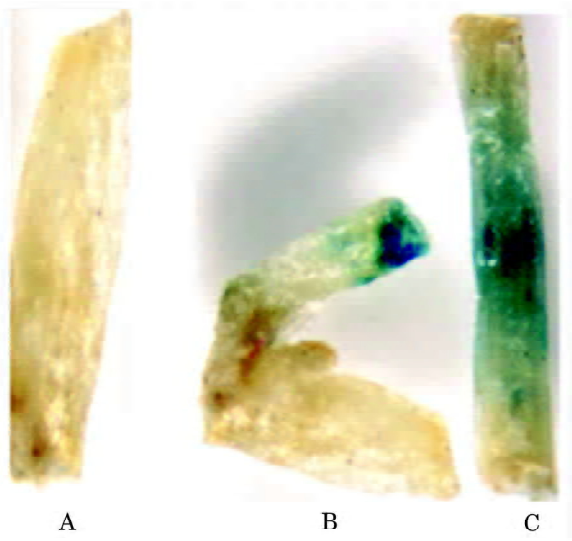

Fig. 3: Transient expression Pattern of Yam Petiole following cocultivation with *Agrobacterium*/pIG-121-Hm. Transformation efficiency was assessed with GUS hystochemical assay and recorded as the area of explant surface expressing blue. A: Control untransformed explant; B: Explant cocultivated with Strain C58; C: Explant transformed with Strain EHA101; Quain *et al*., 2011.

This is the first report of schematic development towards organogenesis that can be utilized for GE of local Ghanaian *Dioscorea rotundata* ('Pona') (Quain *et al*., 2011) and we expect that the protocol can be successfully adapted for other *D. rotundata* accessions.

3. FUTURE REQUIREMENTS FOR AGRICULTURAL BIOTECHNOLOGY IN GHANA

An earlier report of a survey conducted by Olemba *et al*. (2010) indicated that Ghana is leveraging adequate human resources in biotechnology and related disciplines in molecular biologists, virologists, plant breeders, geneticists, pathologists, microbiologists, physiologists, entomologists and tissue culture specialists. A total of 50 scientists were interviewed, nearly 2/3rds of them had PhD degrees with specialization in crop breeding, biochemistry, physiology, molecular biology, and tissue culture, with most of the graduate training in biotechnology acquired overseas. However in recent times, a number of students are now undergoing training in modern biotechnology related disciplines in Ghanaian universities such as University of Ghana's Crop Science and Botany Departments; Biochemistry, Crop Science Biotechnology Departments of Kwame Nkrumah University of Science and Technology, and University of Cape Coast are offering undergraduate and post-graduate course in plant biotechnology (Quain *et al*., 2012a). Additionally, various institutions have a wide range of facilities to address their respective functions mainly in the areas of molecular biology, tissue culture and analytical sciences to meet institutional research mandates. All the eight institutions surveyed have molecular and tissue culture laboratories with supporting analytical and general laboratory capability in Molecular markers, PCR, ELISA, nucleic acid extraction and cloning (Olembo *et al*., 2010). Thus, the potential exists to improve on biotechnology in the areas of GE, genetic characterization, port of entry diagnostics of promising materials.

We report here, in support of Olemba *et al*. (2010) conclusion, that in Ghana, tremendous efforts have been made to build national capacity in modern biotechnology with respect to human and resource development. National Biosafety Committees and its Institutional/technical subcomittees, were formed to enhance biotech education and outreach, organizational structure and infrastructural capacity (Quain *et al*., 2012a; Tuskegee University & Ghana-CRI USDA-FAS Project, 2009). The Council for Scientific and Industrial Research (CSIR) of Ghana since 2006 has been involved in several projects and programs that contributed to the application of biotechnology. These include the West Africa Agriculture Productivity Program (WAAPP) where the CSIR-Crops Research Institute (CRI) is the national center of specialization in Biotechnology. The Open Forum for Agriculture Biotechnology (OFAB) is a platform that network biotechnology stake holders can utilize to share knowledge, experiences, and, explore new avenues of

bringing the benefits of biotechnology to the African farmer and investor. It is a collaborative initiative between the African Agricultural Technology Foundation (AATF) and International Service for the Acquisition of Agri-biotech Applications (ISAAA) AfriCenter. The Ghana chapter of this program is housed in and run by scientists of the CSIR. The project on Strengthening Capacity for Safe Biotechnology Management in sub-Saharan Africa (SABIMA), an initiative by Forum for Agricultural Research in Africa (FARA), is designed to enhance capacity building in biotechnology and biosafety in Africa, with CSIR as the focal point (FARA, 2009). The CSIR in collaboration with Biotechnology and Nuclear Agriculture Research Institute (BNARI), through this project, are in the process of formulating policies for the safe application of biotechnology tools in various research programs. The CSIR/CRI-Kumasi and Tuskegee University secured a USDA-FAS 3-year funded project on sweetpotato, aimed at bringing changes in Ghana for creating an environment for introducing GE Crops through stakeholder and scientist training and Confined Field Trial (CFT) demonstration.

3.1. Ghana Biosafety Framework

Cartagena Protocol is a binding international agreement under the Convention on Biological Diversity (CBD) for implementing biosafety regulations. The Cartagena Protocol on Biosafety (CPB) obligates countries to establish biosafety procedures for trans-boundary movement and handling of all living genetically modified organisms that could have effects on the conservation and sustainable use of biological diversity, in addition to effects on human and livestock health. According to the Secretariat of the CBD 2012 Cartagena Protocol on Biosafety Ratification list (Secretariat of the CBD. 2012), 163 countries have instruments of ratification or accession deposited with the UN Secretary General and Ghana is one of the countries.

The President of the Republic of Ghana signed the Ghana Biosafety Act, 2011 (Act 831) in December 2011 following the passage of the Biosafety Law by the Ghanaian Parliament, on June 21, 2011. The Ghana Biosafety Act, 2011 (Act 831) establishes the National Biosafety Authority as the administrative body responsible for all issues related to Biotechnology in Ghana. The Act establishes biosafety regulations that will govern procedures for contained research work and field trials on biotechnology products; release into the environment, commercialization, importation, exportation and transit of agricultural biotechnology products. The Act does not apply to pharmaceuticals for human use (Ghana Biosafety Act 831, 31stDecember, 2011).

The national biosafety framework is a combination of policy, legal, administrative and technical instruments. It was developed to ensure an adequate level of protection for the safe transfer, handling and use of GE crops. Introduction of these crops may affect adversely the conservation and sustainable use of biological

diversity. The Ghana Biosafety Framework, evolved through the contribution of the scientists, several ministries and stakeholders, was to ensure that the framework was tailored to suit the conditions in Ghana. It has the following elements:

(a) A Government policy on biosafety, which in part is a broader policy on biotechnology. The current policy duration to the framework is in the National Science and Technology Policy, which states that innovative and modern technologies including biotechnology shall be harnessed to address problems in agriculture, health and industry. The policy duration is further strengthened by the constitutional obligation to promote agriculture and industry and at the same time ensure protection of the environment and our national resources.

(b) A regulating system set in place to address safety in the field of modern biotechnology. This includes a biosafety Act (a proposed bill under consideration), a set of National Biosafety Guidelines and regulation/guidelines to be made periodically to guide practices in modern biotechnology.

(c) An administrative system to handle requests for permits for certain activities such as release of Living Modified Organisms (LMOs), which is the focus of the administrative guidance document.

(d) A decision making system that includes risk assessment and management for the release of LMOs, a guidance document on "Risk Assessment of Genetically Modified Organisms in Ghana" has been developed to assist in the decision making process.

(e) Mechanism for public participation information sharing. Guidance document on public participation. Information sharing and access to Justice with respect to Genetically Modified Organisms has also been developed.

The scope of the National Biosafety bill (2004) draft law regulates all activities in biotechnology including contained use, releases into the environment and placement in the market, export and transit of GE crops. The National Biosafety Committee through training had built the capacity to review confined field trial application through its Institutional and technical biosafety subcommittees. Three applications were received for consideration and approved for Confined Field Trials: 1) Introduction of BT-Cowpea in Ghana, 2) Protein quality improvement in sweetpotato and 3) Nitrogen use efficiency and salt tolerance in rice (http://edition.myjoyonline.com/pages/news/2012).

3.2. Survey Results on the Current Status and Future Requirements to Generate Genetically Engineered Crops in Ghana

A brief survey on the status and opinion of biotechnology in Ghana was conducted in 2012 for the purpose of this publication. Due to previous awareness creation and stewardship trainings by the authors (Tuskegee University and Ghana CSIR-CRI USDA-FAS Project), farmers, regulators, researchers and lecturers in the field

of biotechnology and related areas in Ghana, were requested to participate in the survey.

3.2.1. *Results of survey*

Results from the first 40 survey respondents are summarized herein. In terms of type of organization only 3 respondents out of the 40 (7.5%) were from the private sector, the remaining 92.5% were from the government sector, indicating that biotechnology is mainly utilized by governmental organizations. Therefore the involvement of the government in biotechnology initiatives in Ghana is crucial. However, the private sector needs to be motivated to be involved in biotechnology since they are the major beneficiaries. The OFAB initiative is an ideal forum to encourage the private sector to utilize Biotechnology.

The distribution in Fig. 4 highlights the importance of research in organization focus (60%). Clearly some of the respondents are both into research and teaching fields. Teaching is the second highest category (25%) and administrative the least (1.7%). This response is a clear indication that currently in Ghana, potential organizations to be involved in GE are in the research institutions, while the regulatory and administrative aspects calls for improvement over a period of time.

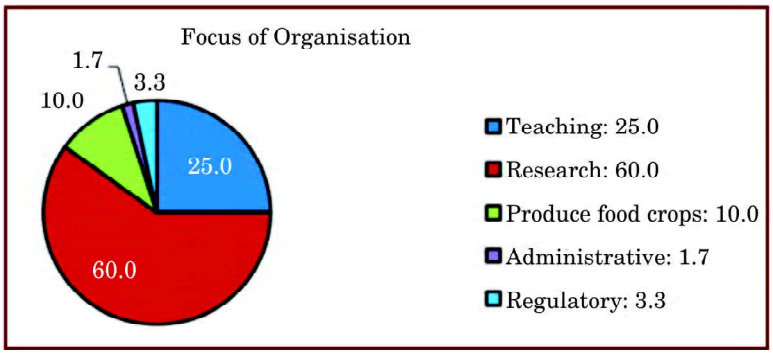

Fig. 4: Pie Chart Depicting, from results of a survey, the distribution of biotech organisationfocus in Ghana.

Respondents were asked to describe biotechnology in terms of their areas of interest and from their response,biotechnology is potentially used in micropropagation for multiplication of vegetatively propagated plant material, molecular breeding for rapid selection of desired progeny and germplasm conservation. The use of polyclonal and monoclonal antibodies as well as DNA probes in diagnostics, was also apparent. Since relatively less emphasis was laid on utilizing genetic engineering for crop improvement, our presumption is that, GE crops if introduced in Ghana, should be mainly to complement existing conventional crop breeding and food production programs. Researchers and lectures are using

biotechnology tools to improve conventional research efforts, however, a limited number of respondents are engaged in GMOs regulatory systems, and to a lesser extent in the generation of GE crops.

Although no specifications (since they could have encountered GE outside Ghana) were given, only three out of the 40 respondents indicated that they had encountered GE crops in their line of work or professional development in GE crop producing countries. The response to the question "Do you have the capacity to screen for genetically modified crops and their products" (Fig. 5), 62.5% indicated lack of facility; 37.5% had the facility and 5% did not have the infrastructural facility but both the groups possessed necessary skills. Considering that, 38% of respondents have facilities to screen for GEs, hence we can conclude that, adequate facilities are available to handle GE crops in Ghana.

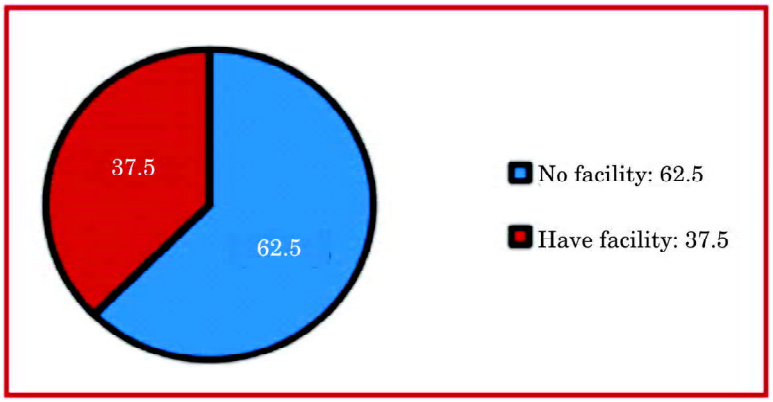

Fig. 5: Pie Chart Results of the survey on the "capacity to screen for genetically modified crops and their products?"

Respondents were also asked to list the priority areas in biotechnology, taking in to account that the research and development in biotechnology in Ghana is in its nascent stage. Over 80% of the respondents are currently involved in biotechnology related research areas and their observations are summarized in Table 3.

Utilizing the existing facilities the respondents would like to use tissue culture and molecular biology techniques for the improvement of crops. Marker assisted breeding is preferred to enhance the chances of identifying the progeny with desirable characteristics in conventional breeding programs. These biotechnology related activities are vital, thus will ultimately contribute towards germplasm conservation, cataloging, and gene discovery with its subsequent utilization in generating GE crops

Table 3: Preferred areas of research by Ghanaian scientists.

• Molecular marker assisted breeding towards development of crop lines with tolerance to biotic and abiotic stresses, using QTLs
• Molecular Marker assisted selection
• Disease diagnostics
• Genotyping and germplasm characterization
• Genetic engineering theory to aid in teaching
• Tissue culture for production and rapid multiplication of sanitized planting materials.
• Tissue culture for germplasm conservation
• Genotyping to remove duplicates in conserved germplasm collections
• Molecular tools for validation of traits
• Molecular tools for identification of somaclonal variants
• Trait pyramiding, mutagenesis and double haploid production

The section of the survey dealing with "genetic engineering" questioned the interest of researchers to introduce GE crops into Ghana. Out of the 40 respondents, 26 (65%) were interested in introducing GE crops as part of their research. The majority of them proposed a time line for a two-year period and a few for periods extending up to 10 years. The primary aim in the introduction of GE crops,as shown in Fig.6, is for research (29%). Priority areas are in food (24%) followed by teaching (17%), and animal feed (17%). The application in pharmaceuticals received less attention with 13%.

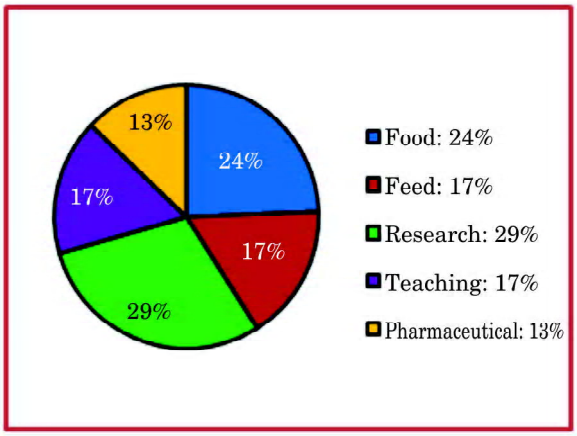

Fig. 6: Pie chart results of the survey related to "Purpose of Transgenic Crops you wish to Introduce"

The biosafety law in Ghana was given Presidential approval on 31st December 2011. As reported in the Ghanaian Time (a national daily), on the 8th of May 2012 (http://newtimes.com.gh/story/hey-wait-what-s-this-biosafety-act-about), science and research received minimum attention in Ghana. When asked about the threats that GE poses, many of the respondents admits that there are some perceived threats to the utilization of GE crops, and they are summarized below:

- Preservation of local germplasm/land races may be compromised, since outcrossing from pollen derived from GE crops is a real threat.
- Ghanaians may not accept the introduction of GE crops.
- GE crops may not be the panacea for all the shortcomings in introducing high yielding crops with enhanced nutrition. It is perceived to complement the traditional crop breeding
- The majority of people are unaware of the benefits that GE technology offers
- It may be difficult to comply with national laws and regulations
- The technology is expensive and largely in the hands of multinationals. It can only be accessed after fulfilling largely the business interests. As a result the seed may not be available at affordable prices

The listed threats are genuine concerns for the stakeholder, whether science-based or not, attention must be paid to them in order to design sound academic training modules to help researchers in Ghana to introduce GE crops. Although, threats are rated as being moderate (Fig. 7), the benefits of agricultural biotechnology remain unknown to Ghanaian producers, the general public and decision makers, who are relatively risk-averse or misinformed based on our informal surveys during the workshop opinion surveys of 2008 and 2009 (unpublished document). The respondent concerns are a challenge for The National Biosafety Committee of Ghana, whose main responsibility is to educate scientists, administrators and general public the risks as well the benefits of introducing GE crops.

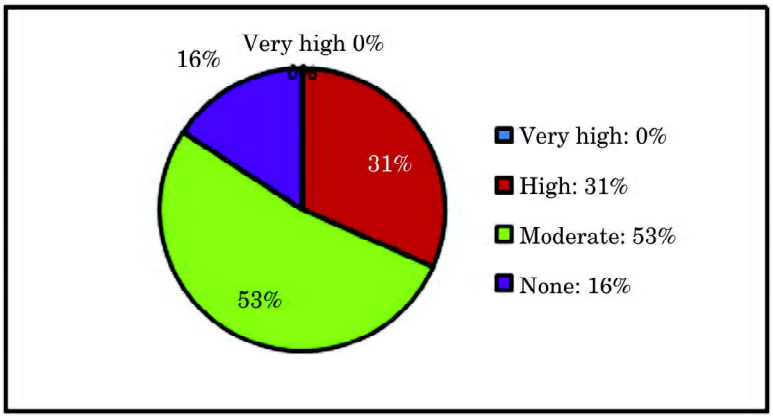

Fig. 7: Survey results from respondents on "How severe are the threats of GE"

We conclude, on the basis of our survey, organization of training activities tailored to generate interest and awareness in GE technology, and additionally efforts need to be made for an effective implementation of a biosafety framework and harness infrastructural capacity, in order to facilitate the introduction of GE crops. The respondents proposed the following requirements for the safe introduction and utilization of GE crops in Ghana:

- Continual education of both the formal and informal sectors on GE related.
- Establish a forum for open discussions on GE related issues
- Well equipped and maintained laboratories, containment and confinement facilities
- Well trained personnel
- Well equipped regulatory bodies
- Encourage use of GE crops and their products.

Potential of biotechnology to increase food production was realized and the respondents were fully convinced of the high impact it can generate to agriculture in Ghana and consequently gave a pivotal role to Ghana in the global market (Figs. 8 and 9).

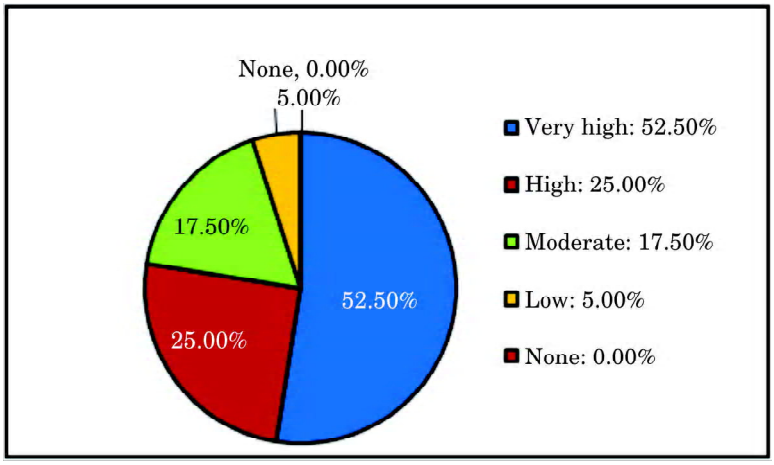

Fig. 8: Opinion to the survey question "What do you presume will be the economic impact of biotechnology on Ghanaian agriculture?"

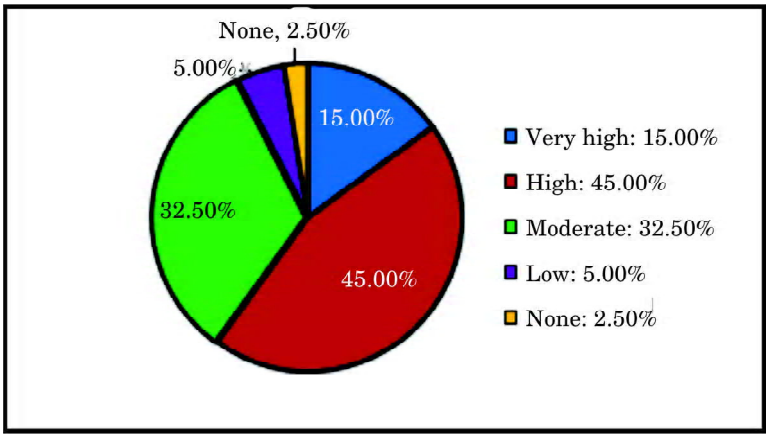

Fig. 9: Ghanaian views on "Where do you presume adapting agriculture Biotechnology will place Ghana in the global market?"

4. CONCLUSIONS

Our attempts to genetically engineer two important staple food crops in Ghana, yam and sweetpotato, with enhanced nutrition, shelf life and stress tolerance were described. It will be necessary to optimize the results utilizing locally cultivated genotypes. GE sweetpotato with enhanced protein is under confined field trials and is yet to be deregulated. Several programs have been introduced in Ghana to build national capacity in biotechnology and to set the stage for deregulation of GE crops. These include human resource and infrastructure development, establishment of the National Biosafety Committee and its Institutional/Technical subcommittees and the National Biosafety Frame Work.

We have conducted a survey to gauge the current status and assess future requirements of biotechnology in Ghana. At the outset we should admit that the sample is abysmally small. Nonetheless it represents a cross section of developers, users and administrators from all the major universities and research organizations in Ghana. We have summarized the results of this survey. It is apparent that the majority is in favor of introducing GE crops in Ghana and expressed faith in the technology towards enhanced food production, nutritional and economic growth. Ghana needs to make substantial investment in cutting edge technologies and in the human resource development. Policies need to be harnessed to ensure biosafety and sustainability of the deregulated GE crops. Ultimate aim of our efforts is to alleviate poverty, hunger and malnutrition in the sub-Saharan sub region

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