Potential of Biotechnology and Application of Genomics to Indigenous and Traditional Leafy Vegetables in West Africa

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Keywords: biotechnology/genomics, Sub-Saharan Africa, indigenous leafy vegetables

Abstract

The application of biotechnology/genomics has been very useful in many fields. In agriculture, they have been used for the development of new genetic lines, rapid multiplication of improved crop planting materials and the detection and elimination of diseases in useful plants. However, the techniques developed from these important disciplines have not been applied to many indigenous and traditional leafy vegetables (ITLVs) that are sustaining the lives of poor rural people in Sub-Saharan Africa (SSA). This paper examines the importance of ITLVs in West Africa as well as the limitations of popularising them. An attempt is made to throw light on some of the tools of biotechnology/genomics that have the potential to solve many problems militating against the commercialisation of ITLVs in West Africa.

INTRODUCTION

Only about 30 crop species provide 95% of the world's food energy whereas over 7000 species have been known to be used for food and are either partly or fully domesticated (Williams and Haq, 2002). In Africa, many of the underutilised species are indigenous leafy vegetables (ILVs) and traditional leafy vegetables (TLVs). The ILVs originated from Africa while the TLVs were introduced over a century ago and, due to long use, have become part of the food culture of Africa (Smith and Eyzaguirre, 2007). It has been reported that no fewer than one thousand African plants could be consumed as leafy vegetables (Spore 116). They also contribute to a more balanced diet for many communities, especially disadvantaged people (Shippers, 2000). Their nutritive values are comparable or often higher than the exotic vegetables (Table 1). The ITLVs have many uses in the diet of Africans. They provide high amounts of vitamins, especially vitamin A, and the young leaves also provide some protein. The green vegetables are very rich in minerals such as Ca, K, and Fe as well as some essential micro-nutrients as compared to cabbage and lettuce. They are also a source of crude fibre which is very important for the prevention of constipation. In addition, ILVs have medicinal uses in many African communities. Several ILVs are still used for prophylactic and therapeutic purposes in rural communities (Ayodele, 2005) and it has been reported that they also contain non-nutrient bioactive phytochemicals that have been linked to prevention of some heart and degenerative diseases (Smith and Eyzaguirre, 2007).

ITLVs play an important role in income generation and subsistence in sub-Saharan Africa (SSA). They are unique in that they directly address poverty and food security in rural areas of SSA. In addition, they have a positive impact in all the UN millennium Challenge goals (Table 2).

THE LIMITATION OF POPULARISING ILVs

The development of ILVs is hindered by many limitations. It is very regrettable that most influential Africans look down on the ILVs. They prefer the exotic vegetables and have patronised them. People therefore erroneously regard the ILVs as the food of poor people despite the high nutritive value. Consequently, the production of exotic vegetables is very lucrative as compared to the ILVs. It is therefore not uncommon to spot exotic vegetables in market gardens in the cities while the ILVs are cultivated by peasant women farmers in the villages. There is a need to consciously and seriously promote and publicise the use of ILVs in SSA.

It is also very clear that the ILVs have received less attention in research. They are not normally among the research priorities of SSA. Lack of research has led to the following:

1. Cultivars have not been identified for multiplication and distribution to farmers. Pure lines of the ILVs have not been identified to make good use of specific characteristics such as high yield and disease or drought resistance. There is a need to develop pure lines of the ILVs and to identify and document their characteristics.
2. Disease and pest problems have not been seriously investigated and documented. Growers who face these problems often do not know what to do. The lack of literature and pest information is a big problem for extension officers who should be teaching farmers improved methods of protecting their crops from pests and diseases.
3. There is a lack of certified seeds. Good quality seed is very important in any agricultural production venture. Modern certification requires experts who will produce breeder seeds for multiplication. However, the few plant breeders in SSA have concentrated their efforts on the breeding of arable crops and exotic vegetables. A strong Africa-based commercial vegetable seed sector devoted to serving smallholder farmers has long been a missing link in creating a sustainable seed system for ILVs. Hence, almost all the ILV seeds in SSA have not entered the certification scheme and farmers save their own seeds for planting. The seeds are often of low viability and vigour, infested with seed-borne diseases and are not true-to-type.
4. The production practices of most of the ILVs are based on what the peasant farmers have developed over many years and passed on from generation to generation. There is a need to develop good husbandry practices based on scientific principles from knowledge of plant morphology, anatomy and physiology. Due to the lack of development of agronomic technology, handbooks for good husbandry practices to obtain maximum yield in terms of seeds and leaves are absent in most cases.
5. Post-harvest losses and post-harvest processing techniques for most ILVs have not been investigated. Farmers and traders have not been educated in the correct handling of the leaves and appropriate processing techniques to reduce post-harvest losses are lacking. The losses are so great that farmers and traders are forced to sell the produce at very cheap prices which renders the production and sale of ILVs as a non-lucrative business.
6. Recipes have not been developed for most of the local vegetables, especially ILVs. The younger generation of Africans in the cities do not normally patronise the consumption of ILVs because they are not conversant with their preparation. It will be very useful if recipes are developed and documented and if food shows popularised the consumption of ILVs.
7. Identification of ILVs is inadequate. In Africa, ILVs are known by their local names; however, a local name may refer to more than one cultivar. It is therefore imperative that taxonomists should clearly identify cultivars by both their scientific and common names so that scientists can compare their characteristics and select the superior ones.

BIOTECHNOLOGY TECHNIQUES AVAILABLE FOR IMPROVEMENTS OF ITLVs

Biotechnology has been identified as one of the leading technologies of the 21st century with the potential to address economic, social and environmental issues affecting the poor in developing countries. Most of the techniques and applications in the field of biotechnology seem to be generally accepted and this can be applied to the characterisation, development and improvement of ILVs in SSA (http/www.africa-union.org/root/au/AUC/Departments/HRST/biosafety/AU_Biosafety.htm).

Biotechnology is an interdisciplinary subject whose practice requires cooperation between diverse groups of specialists ranging from scientists, engineers, social scientists and farmers (Schmauder, 1997). Biotechnology can be viewed as any technique that uses living organisms or substances from those organisms to make or modify a product, to improve plants or animals or to develop microorganisms for specific uses (Persley, 1992), The approaches taken by each or a combination of the specialists addressing problems may involve both simple and complicated techniques in an unfamiliar field. Some of the aspects of biotechnology that are currently being used in crop production are genetic engineering, cell and tissue culture, somatic embryogenesis, protoplast fusion, molecular markers, cryopreservation and somaclonal variation (Monti, 1992; Swiader et al., 1992; Quainoo et al., 2008a; Quainoo, 2009).

Genetic Engineering

Although the terms biotechnology and genetic engineering are sometimes used interchangeably, biotechnology has a considerably broader meaning. Genetic engineering is more specific in application and involves isolation, characterisation and recording of genetic material and its transfer to foreign organisms (Swiader et al., 1992). The transfer of genetic materials into new or suitable hosts may lead to a change in a hereditary trait of an organism. Genetic engineering seems to complement rather than replace other traditional breeding techniques and should be considered for the improvement of leafy vegetable crops in SSA.

Cell and Tissue Culture

Plant tissue culture, which includes the culture of embryos, seeds, meristems, leaves, shoot and root tips, stems and pollen grains, should be developed for the cloning and genetic improvement of superior plants of ILVs with desirable traits. Somatic embryogenesis, which is the formation of embryos from somatic cells, has been routinely used both as a means of propagation for some plants, as well as a valuable model for investigating the structural, physiological and molecular events occurring during embryo development (Stasolla et al., 2003). Somatic embryogenesis has proven useful for clonal propagation, micropropagation, production of mutants, artificial seeds and materials for use in genetic engineering (Redenbaugh et al., 1988; Li et al., 1998; Hu et al., 2002). The health quality, marketability, storage and distribution of leafy vegetables could be enhanced by generating and propagating uniform materials through somatic embryogenesis.

Protoplast Fusion

Fusion of protoplasts facilitates mixing of two or more whole genomes and could be exploited in crosses at interspecific, intergeneric, or even inter-kingdom levels, which is not possible by conventional techniques due to incompatibility. The fact that isolated protoplasts are devoid of walls makes them easy tools for undergoing fusion in vitro. Mixing two genomes opens the door to gene transfer and a study of gene expression, stability of several traits, and cell genetic changes (Sink, 1984; Widholm, 1983). Protoplast fusion of ILVs may regenerate into new whole plants which may be reproducible and more efficient.

The Use of Molecular Markers in Detecting Pathogens

Traditionally, indexing of plant propagating materials for pathogens has been carried out through visual inspection of seeds (Slack and Singh, 1998) and laboratory tests, including enzyme linked immunosolvent assay (ELISA). The interpretation of ELISA test results may be difficult because sometimes it is unclear whether low ELISA extinction values are due to the presence of low quantities of virus or to non-specific binding of the conjugate (Van de Vlugt et al., 1997). This phenomenon often leads to false-negatives (virus concentrations too low) and false positive (background too high) test results.

Additional healthy plants or plant parts may become infected with diseases but may not show any symptoms for a considerable period of time as a result of latency problems. Removal of infected materials can prove difficult because plants in the latent period of infection are not always evident.

Rapid advances in molecular biology based on nucleic acids have given rise to sensitive diagnostic and detection tools for pathogen detection in plant materials. Recently PCR-based capillary electrophoresis was used to demonstrate the presence of the cocoa swollen shoot virus in all component parts of the cocoa seed and newly emerging seedlings from infected seeds (Quainoo et al., 2008b). This technology also has an important role to play in germplasm conservation and distribution and can be applied to ILVs production in SSA.

Cryopreservation

Cryopreservation is the preservation of viable cells, tissues and organs in liquid nitrogen at -196°C (Benson, 1999) and their storage for indefinite periods without genetic erosion. Cryopreservation is increasingly used to conserve crop plant germplasm, thus providing long term storage methods for plant genetic resources which cannot be maintained using conventional preservation methods, such as seed banking (Sakai, 2004). Quainoo (2009) has demonstrated that somatic embryos of individual cocoa genotypes subjected to low temperature storage did not show any significant difference in survival post cryopreservation. This implies that the trueness of cocoa somatic embryos can be maintained no matter the length of storage time in liquid nitrogen. This technique can be applied to ILVs to preserve pollen and seed propagated species (Towill, 2005).

Somaclonal Variation

Regeneration protocols based on repeated passage of materials through culture have been reported to increase the production of mutant regenerants (Plader et al., 1998). This variation was termed by Larkin and Scowcroft (1981) as somaclonal variation and is defined as genetic and phenotypic variation among clonally propagated plants of a single donor clone (Kaeppler et al., 2000). Somaclonal variation has been attributed to the plasticity of the plant genome to adapt to environmental conditions and includes genomic and epigenetic changes (Joyce et al., 2003). Epigenetic changes are often heritable only through somatic division and changes normally revert to parental phenotype following meiosis (Kaeppler et al., 2000).

Somaclonal variation is undesirable for clonal propagation of woody and ornamental plants and transgenic plants or in large scale mass propagation (Heinze and Schmidt, 1995). Although it is not studied much today, culture derived variation has the potential to improve crops (Jayasankar, 2005) and can be applied to ILVs.

BIOTECHNOLOGY IN FUTURE VEGETABLE CROP IMPROVEMENT

Today, it is possible to deliberately manipulate the genome of plants at the molecular level, transferring desirable genetic materials from one organism to another, creating a new desirable organism in the process (Swiader et al., 1992). The potential of biotechnology in agriculture is enormous and promises to open a whole new era of opportunities for crop production. This can eventually be applied or used as the standard for ILV crop improvement programmes in SSA. This will give growers new options in pest and disease control, selection of cultivars, production of superior crops and packaging of their products for specialised markets (De Wet, 1983).

Eventually, production of specialised ILV crops with particular traits such as high protein, oil and starch would be developed. The composition of vegetables will be modified to improve the amino acid balance and vitamin content to meet the nutritional requirements of both humans and domestic animals. The shelf life of leafy vegetable products can be improved and this may have positive implications for postharvest storage.

The technology has the potential for generating ILVs with resistance to certain herbicides and to control plant pathogens such as viruses, as has been used to make tomatoes resistant to *Tobacco mosaic virus*. There is also the potential of reducing the time taken to produce new leafy vegetables through hybridisation. Tissue culture techniques can be used to multiply single vegetable plants with desirable traits into several uniform copies within a short time and space through micropropagation (Widholm, 1983). Anther culture has been successfully used by seed companies in the development of inbred broccoli and pepper lines.

PROBLEMS AND CONCERNS OF BIOTECHNOLOGY IN SSA

The importance of biotechnology to agricultural development in SSA depends on the availability of skilled human resources, laboratory facilities and chemical reagents which are often lacking. Usually skilled personnel who have received their training at the expense of government seek employment outside the country. Working opportunities in SSA are often inadequate or unattractive, contributing to reduced availability of personnel. Where there are trained personnel to carry out research, lack of funds or the availability of basic chemicals and reagents have been the cause of the slow pace of biotechnology application to leafy vegetables in SSA. Even where chemicals and reagents are present lack of appropriate storage facilities may become a problem. As a result of these challenges improvement of ILVs has mainly been carried out using the conventional breeding techniques.

THE CURRENT CROP RESEARCH FOR ILVs IN WEST AFRICA

Unfortunately, almost all countries in West Africa do not have national research programmes on ITLVs. It was reported that Nigeria was planning to have national programmes on underutilised crops (William and Haq, 2002). Despite the fact that some interested scientists are working on ILVs, the work is not co-ordinated and their impacts are not felt. William and Haq (2002) pointed out correctly that the first step towards ITLVs research needs to be the identification of regional common species that could constitute the starting material for planned and multi-national research and developmental activities. Therefore the following ITLVs (Table 3) which are in almost all countries in SSA should be considered seriously for future biotechnological development.

CONCLUSION

One of the key drivers of present day agricultural innovation is biotechnology/ genomics and, since its rapid evolution in the last century, it has transformed the science underpinning the agricultural biotechnology sector. This has resulted in unprecedented social impact. As with all tools, full potential is realised through human initiative and few countries in SSA, like Ghana and Nigeria, are blessed with sufficient human resources in the field of biotechnology. However, these human resources are scattered in academia, research institutions and other public organisations across the country. What is needed is to identify and gather this human potential to address various challenges facing ITLVs. Concerted development of ITLVs can create a platform for increased health and economic improvement of the people leaving a legacy for future generations.

ACKNOWLEDGEMENTS

The presentation of this paper at Kuala Lumpur, Malaysia has been supported by Crops for the Future (CFF) through a grant provided by the UK Department for International Development (DFID) for the benefit of developing countries. The views expressed are not necessarily those of CFF or DFID. Also, thanks to Sean Mayes of School of Biosciences, University of Nottingham for his technical advice on this paper.

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Tables

Table 1. Nutritional content of fresh of ITVS and exotic vegetables commonly grown in West Africa.

Vegetable	Water (g)	Protein (g)	Fat (g)	Carbohydrate (g)	Fibre (g)	Ca (mg)	P (mg)	Fe (mg)	β-carotene (µg)	Niacin (mg)	Ascorbic acid (mg)
**Corcolus oliterius*	80.4	4.5	0.3	12.4	2.0	360	132	7.2	6419	1.2	80
**Amaranthus crientus*	84.0	4.6	0.2	8.3	1.8	410	103	8.9	5716	1.2	64
**Hibiscus subdariffa*	85.6	3.3	0.3	9.2	1.6	213	93	4.8	4135	1.3	54
Cabbage (exotic)	90.1	1.7	0.4	4.1	2.9	52	41	0.7	385	0.54	49
Lettuce (exotic)	95.6	0.7	0.3	1.9	0.6	19	18	0.4	5.0	0.3	3

Source: PROTOA, 2004.

Table 2. The role of ITLVs in achieving the Millennium Development Goal of the United Nations in Sub-Saharan Africa.

Millennium development goals		Impact of ILVs development
Goal 1	Eradicate extreme poverty	ITLVs are intensively cultivated and generate higher profit per unit area than arable crops, especially in SSA where land is scarce and labour is cheap. Vegetables are likely to generate more jobs per hectare than other agricultural enterprises which benefits landless labourers and peasant farmers. ITLVs are affordable and are sometimes the main sources of micronutrients to most rural communities in SSA. Overcoming malnutrition improves health, productivity and raises income of the poor.
Goal 2	Achieve universal primary education	Incorporating ITLVs in the ongoing school feeding programme in SSA like Ghana is likely to increase enrolment of pupils in schools. Micronutrient-rich food is known to nourish the brain and thereby improve the intelligence of children. Parents in Africa are likely to encourage and sponsor intelligent and brilliant children to be in school.
Goal 3	Promote gender equality and empower women	ITLVs production is normally referred to as a woman's job in almost all the communities in SSA. Thus any intervention to promote them increases the income of the rural women. Increased consumption of ITLVs will improve the health of women which will subsequently increase their productivity and income.
Goal 4	Reduce child mortality	Increased access to ILVs in the diet of children will help them to withstand better most of the diseases that cause child mortality.
Goal 5	Improve maternal health	Majority of rural farmers in Africa suffer from micronutrient deficiency, especially iron and vitamin A. ILVs are the cheapest and most appropriate sources of these essential micronutrients for expectant mothers.
Goal 6	Combat HIV/AIDS, malaria and other diseases	Incorporating ITLVs in the diet of children and expectant mothers enhances the proper function of their immune systems, helping consumers to resist many infectious diseases (HIV/AIDS, malaria and tuberculosis) that are prevalent in the SSA.
Goal 7	Ensure environmental sustainability	Most of the ITLVs are more adapted to the environment and more resistant to disease and pests, therefore pesticide use is reduced drastically. The ITLVs produce more profit per unit of water use as compared to most known agronomic and industrial crops.
Goal 8	Develop a global partnership for development	The existence of regional common ITLVs in Africa demands a regional approach where scientists and developmental officers in Africa with proper support from donors come together and develop strategies for crop improvement, utilisation and popularisation. Such collaboration will be a good example of a global partnership aimed at improving the quality of life in developing countries.

Table 3. Major biological problems of four very important indigenous vegetables in sub-Saharan Africa.

African indigenous vegetable	Major biological problems	Biotechnology tool to apply	References
Hibiscus sabdarifa	Different morphotypes, mixture of kenef, rosselle and false rosselle in the market, padagrica beetles, leaf hoppers, short shelf-life.	Generation of desirable seeds and micropropagation.	Schippers (2000) PROTA 2 (2004)
Corchoruso litorius	Numerous morphotypes, seed dormancy caused by impermeability of seed coat, irregular germination, seed shattering, grasshoppers, nematodes and short shelf-life, no improved seed.	Somatic embryogenesis to obtain clonal materials and micropropagation.	Opabode and Adebaayo (2005), Schippers (2000)
Amaranthus species	Confused taxonomy, damping off, leaf and stem rot (resistant cultivar should be breeding target), short shelf life.	Apply genetic markers to identify genetic make up, select for resistance and somatic embryogenesis to generate clonal materials.	PROTA 2 (2004) Schippers (2000)
Abelmoschus esculentus	Polymorphic species, flowers sensitive to emasculation, seed dormancy for at least 2 months, local cultivars undeveloped, flea beetle, crickets, cotton stainer, leaf roller, nematodes, *Fusarium* attack.	Apply genetic markers to identify genetic make up, select for resistance and somatic embryogenesis to generate clonal materials.	PROTA 2 (2004) Schippers (2000)