Plant genetic engineering, climate change and food security

Working Paper No. 72

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Abstract

This paper explores whether crop genetic engineering can contribute to addressing food security, as well as enhancing human nutrition and farming under a changing climate. The review is based on peer-refereed literature, using results to determine the potential of this gene technology. It also provides a brief summary of issues surrounding this genetic enhancement approach to plant breeding, and the impacts on farming, livelihoods, and the environment achieved so far. The genetic engineering pipeline looks promising, particularly for adapting more nutritious, input-efficient crops in the development of the world's farming systems.

Keywords

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Acknowledgements

Contents

1. Introduction

Climate change impacts agro-ecosystems through changes over the long-term in key variables affecting plant growth (e.g. rising temperatures) and through increasing the variability (frequency and intensity) of weather conditions (rainfall, drought, waterlogging and elevated temperature). These changes affect both crop productivity and quality as well as how pathogens or pests attack plants. Climate change may disturb food availability, access, and the stability of food supply, which will likely differ across regions (Wheeler and von Braun 2013). The changing climate is also likely to alter food quality by decreasing protein and micronutrient contents or modifying lipid composition (DaMatta et al. 2010). Climate change comes with an additional challenge – the mitigation challenge. There is a need to reduce greenhouse gas emissions from agriculture as a contribution to cross-sectoral efforts to reduce overall global emissions.

Climate is just one component in an emerging picture of food security concerns in the medium to long term. The FAO (2009) estimates that by 2050 we must produce 50-70% more food, due to a combination of increasing population and shifts in consumption patterns. Even in the absence of climate change, this would be a significant challenge, but recent studies indicate that yields of grains could be reduced by about 5 percent for each degree Celsius of global warming (Lobell et al., 2011). This presents significant challenges, and whilst much can be done to address these challenges by reducing the yield gap, reducing food waste, changing consumption patterns, or increasing the effectiveness of food systems, the option of increasing yield potentials through crop improvement is a promising avenue to explore (building on the success, and failures, of the green revolution in the 1960s and 1970s). However, Ray et al. (2013) recently analyzed trends in crop yields over the past 50 years, and projected likely increases in yield over the next 40 years. They conclude however that current "business as usual" yield increases are not sufficient to double production by 2050. Annual increases are well below the 2.4% necessary (e.g. just 0.9% for wheat). Hence, new approaches to tackle the problem are required.

As noted by *the Crop Science Society of America*, new cultivars may provide means to adapt farming to the changing climate (Boote et al. 2011). There are two ways to adapt crops to new environments: developing new crops (long-term endeavor starting with domestication) or introducing target traits into existing crops through plant breeding, which includes genetic engineering. However, the job of crop improvement is becoming increasingly difficult. Cultivars are needed that are not only high yielding, but are also efficient in use of inputs, tailored to ever more stringent market demands, able to maintain stability under increasing climate variability, and potentially contribute to climate mitigation. These multi-trait demands for new cultivars provide significant challenges for crop breeders, and standard selection approaches struggle under such complexity.

This paper provides a brief summary of issues affecting the deployment of transgenic crops in farming systems, as well as an update on emerging innovative germplasm with desired traits for adapting crops to the changing climate, with emphasis on the most important staples. It also presents a summary of advances in crop genetic engineering for mitigating climate change and improving photosynthesis efficiency.

2. The promise of transgenic crops

Progress in biological sciences benefit from technology advances because the latter "*opens up new horizons and speeds up discovery by orders of magnitude*" (Ehrhardt and Frommer 2012). Plant biotechnology, through engineering genetically modified, or transgenic, crops has already led to significant impacts on canola (or oil rapeseed), cotton, maize, and soybean farming worldwide. Furthermore, in the last 20 years new breeding technologies have emerged, thereby enlarging the toolbox for plant breeding (Lusser et al. 2012). They include, among others, the modification of gene functions through site-directed mutagenesis, the targeted deletion or insertion of genes into plant genomes, or using transgenes to facilitate the breeding process.

A recent meta-analysis reveals that transgenic cotton and maize with insect resistance (due to the *Bt* gene), and transgenic soybean with herbicide tolerance appear to out-yield their conventional counterparts in agronomic and economic (gross margin) terms (Areal et al. 2012). *Bt*-crops perform well in the developing world, with *Bt*-cotton becoming particularly profitable. The net economic benefits of transgenic crops at the farm level were US$ 19.8 billion in 2011 and have been US$ 98.2 billion (in nominal terms) in the period 1996, when farmers started using them, to 2011 (Brookes and Barfoot 2013). Farmers in the developing world captured a good portion of these gains (51.2%). The cultivation of transgenic crops increased global production of canola, cotton, maize, and soybean since their introduction to farming systems in the mid-1990s, e.g. 195 million and 110 million t of maize and soybean grains, respectively. Farmers adopting transgenic crops can also reduce their production costs by lowering pesticide use, labor, and fuel costs (Kaphengst et al. 2010). Demont and Stein (2013) estimated the impact of forthcoming transgenic rice at US$ 64 billion per year. The transgenic rice in the pipeline includes traits such as insect resistance, herbicide tolerance, enhanced adaptation to drought and salinity, and increased β–carotene content.

Transgenic crops provide the means for adapting crops to climate change, particularly in terms of drought and salinity. Transgenic crops can also contribute to climate change mitigation efforts by reducing input use intensity (Lybbert and Sumner 2011). The integration of genetic engineering with conventional plant breeding, within an interdisciplinary approach, will likely accelerate the development and adoption of crop cultivars with enhanced adaptation to climate change-related stresses (Varshney et al. 2011).

3. Issues to be considered when deploying transgenic crops

The extent and complexity of biosafety, food safety, and other regulations regarding the use of transgenic crops may, however, limit their impacts in farms and livelihoods. For example, EU regulations severely affect the use of transgenic crops, thereby leading to € 443 and € 929 million of annual revenue foregone by farmers because transgenic crops with herbicide tolerance or insect resistance, respectively, cannot be grown (Park et al. 2011) (this covers transgenic canola, cotton, maize, soybean, and sugar beet).

Biosafety

Irrespective of the *ex-post* or *ex-ante* impact assessments indicated above, issues have been raised regarding the safety of foods derived from transgenic crops and their potential threat to biodiversity and the environment.

Transgenic crops and their derived products are evaluated on a case-by-case basis for food safety, allergenicity, and nutritional composition, among other factors. As stated by Lemaux (2008) available transgenic crops were released after extensive checking by regulatory agencies in the countries where they are grown. The literature regarding food safety assessment has significantly increased since 2006 (Domingo and Bordonaba 2011). Long-term (> 90 days and up to 2 years in duration) and multi-generational (2 to 5 generations) research concerning the effects of diets containing items derived from transgenic maize, potato, rice, soybean or triticale on animal health did not suggest any health hazards (Snell et al. 2012). Animal feeding trials also show that transgenic crops are nutritionally equivalent to their non-transgenic counterparts and can be safely used. Furthermore, Herman and Price (2013) indicated that any unintended chemical composition of crops that could be caused by genetic engineering has not materialized after analyzing literature from 20 years of research. They further pointed out that compositional equivalence research[1] required only for transgenic crops does not seem to be defensible if claiming scientific uncertainty. The commercialized transgenic crops and their derived products are at least as safe (as food) as those produced through conventional breeding.

Environment

One of the most important issues raised regarding transgenic crops relates to the impact of their genotype and phenotype on the environment (Lemaux 2009). There are some concerns that transgenic crops in farming systems may affect biodiversity and non-target organisms, or that their use could lead to new strains of pathogens and pests (a claim that could also apply to conventional breeding of host plant resistance). Transgenic crops may, however, contribute to more sustainable farming. For example, a life cycle assessment shows that transgenic sugar beet that tolerates herbicide would be less harmful to the environment than the conventional crop because of the lower emissions from herbicide manufacture, transport, and field operations (Bennet et al. 2004). Moreover, Brookes and Barfoot (2012a) indicated that transgenic crops grown by farmers were able to lower pesticide spraying by 443 million kg (9.1%) of active ingredients, thereby decreasing the environmental impact associated with herbicide and insecticide use by 17.9%. These authors also highlight that transgenic crops significantly reduced the release of greenhouse gas emissions from their cropping area, which was equivalent, in 2010, to removing 8.6 million cars from the roads.

Transgenic crops with enhanced adaptation to abiotic stress-prone environments, resulting from climate change, may require an updated assessment framework for their regulatory approval[2] because they pose new questions regarding safety and impact (Ortiz et al. 2007). For instance, the use of regulatory genes[3] in crop genetic engineering may lead to a cascading effect on various gene pathways. This differs from the first generation of transgenic crops based on a one gene-one product system. A new framework will need to consider the composition of transgenic plants versus their non-transgenic counterparts under the target abiotic stress, and the impact of regulatory genes on human health and the environment. For example, new phenotypes resulting from transgenic technology for abiotic stressful environments may result in increased competitiveness due to transgene flow into wild populations or may result in escapes of target crops to other environments, where the escapees become weeds.

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[1] Extensive research comparing the chemical composition, nutritional quality and any levels of potentially toxic components of conventional crop and its transgenic counterpart
[2] To control or direct as per guidelines or law
[3] A regulatory gene controls the expression of other gene(s) by encoding a protein or acting at the RNA level

Intellectual property

Some activists are accusing the multinational private seed sector of "patent grab" on "climate ready" crops (etc Group 2010). They base their views on the accelerated pace for patenting adaptation-related biotechnology, which increased from less than 10 in 1995 to almost 200 in 2007 (Agrawala et al. 2012). Patents are, however, an indicator for innovation in agro-biotechnology – they can be used as a proxy for assessing inventive capacity.

As noted by Cohen (2005), vibrant knowledge- and resource-intensive public research seems to be leading the development of transgenic crops in the developing world. Likewise, user-led philanthropy-private-public partnerships could assist the development and deployment of transgenic crops in the developing world. This is shown by the Water Efficient Maize for Africa (WEMA) project, now in its second 5-year phase. WEMA seeks solutions for improving maize in the drought-prone environments of the continent (http://wema.aatf-africa.org). This project involves African public and private organizations, the multinational seed sector, a Center of the CGIAR Consortium, and philanthropy. It aims to make available to smallholder farmers through the African-based seed sector new royalty-free crop cultivars with enhanced adaption to drought-prone environments, initially in Kenya, Mozambique, South Africa, Tanzania and Uganda. Each country will assess the benefits and safety of transgenic maize cultivars following their own approved regulatory framework. Maize with enhanced adaptation to drought-prone environments will bring significant economic benefits to African farmers. LaRovere et al. (2010) indicated that the potential impacts of maize-bred cultivars with enhanced adaptation to drought-prone locations in Angola, Benin, Ethiopia, Ghana, Kenya, Malawi, Mali, Mozambique, Nigeria, Tanzania, Uganda, Zambia, and Zimbabwe ranges from US$ 50 to 90 million per annum, assuming most likely adoption rates (based on previous research and expert advice), and either 3–20% or 10–34% grain yield gains, respectively. Under the more optimistic grain yield gains, about 4 million people (particularly in Nigeria, Malawi and Kenya) can be relieved of poverty.

Bioethics

Some activists claim that climate-ready transgenic crops will increase farmers' dependence on such crops, jeopardize biodiversity, and threaten food sovereignty (etc Group 2010). This view contrasts with that of many scientists involved in plant genetic engineering who see their new crops being used in stressful areas, thereby allowing resource-poor farmers to have harvests in most years, ultimately improving food security, and increasing opportunities to enter the market economy. As indicated by Farre et al. (2010), smallholders may not benefit from transgenic crops if the major barriers for their adoption are not overcome. Some of these barriers are political rather than technical.

4. Adapting crops to new climates through transgenic breeding

Genotypic and phenotypic plasticity[4] affect crop adaptation to a given environment (Abberton et al. 2008). Genotypic plasticity depends on the survival of genotypes in a population while phenotypic

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[4] Genotypic plasticity refers to epigenetic and genetic changes transmitted to the offspring, while the ability to change the phenotype as a response to changes in the environment has been named phenotypic plasticity

plasticity results from the interaction between genotypes and the environment. Understanding plant phenotypic plasticity to the changing environment will be key to predicting and managing climate change effects on crops (Nicotra et al. 2010). Genome-wide analysis (GWA)[5] in the model plant species *Arabidopsis thaliana*[6] led to identifying genomic sites related to climatic adaptation (Savolainen 2011). For example, GWA by Hancock et al. (2011) revealed a set of candidate genes for climate change adaptation through genetic engineering.

An interdisciplinary perspective (which includes ecology, evolution, genetics, genomics, physiology and molecular biology, among others) will assist in further deepening the knowledge about plant phenotypic plasticity in a changing climate. Nicotra et al. (2010) list leaf mass per unit area, stomata size and density, height at maturity, flowering time plus size at maturity and phenology, and seed size and number among the key functional traits for investigating plant phenotypic plasticity to climate change.

Bhatnagar-Mathur et al. (2007) suggested that genetic engineering could accelerate plant breeding for adapting crops to stressful environments. They further underline that engineering the regulatory machinery involving transcription factors (TF)[7] provides the means for controlling the expression of many stress-responsive genes. There are various target traits for adapting crops – through genetic engineering – to high CO_2 and high O_3 environments of the changing climate (Ainsworth et al. 2008). Ortiz (2008), Jewell et al. (2010) and Dwivedi et al. (2010, 2013) provide the most recent overviews on research advances in genetic engineering for improved adaptation to drought, salinity or extreme temperatures in crops. The most cited include TF, and genes involved in (a) signal sensing, perception, and transduction (STP), (b) stress-responsive mechanisms for adaptation and (c) abscisic acid (ABA) biosynthesis for enhanced adaptation to drought. Transporter, detoxifying and signal transduction genes as well as TF are cited for tolerance to salinity. Genes related to reactive oxygen species (ROS), membrane and chaperoning modifications, late abundance embryogenesis (LEA) proteins, osmoprotectants/compatible solutes and TF are pursued in crop genetic engineering for temperature extremes.

Improving days to flowering and root systems are not among priority target traits but facilitate crop adaption to climate change; i.e., they are characteristics with co-benefits for enhancing crop performance under a changing climate. More research is needed to identify other plant traits that will enable crops to produce yields under changing climates, thereby increasing their adaptive capacity.

Drought

Duration and intensity of drought has increased in recent years, consistent with expected changes of the hydrologic cycle under global warming. Drought dramatically reduces crop yields. The yield response factor (K_y) can quantify the effects of water deficit on crop growth because it captures the essence of the complex linkages between production and water use by a crop, in which many biological, physical and chemical processes are involved (Steduto et al. 2012). Table 1 summarizes K_y values for several crops. They vary from very sensitive crop responses to water deficits ($K_y>1$), yield

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[5] Research for identifying the association of a genetic variant with trait diversity
[6] Thale or mouse-ear cress has been extensively used in research to understand particular traits and their genetics or physiology, with the aim that findings in this model plant will give insights into crops
[7] A protein binding specific DNA sequences and thereby governing the flow of genetic information from DNA to messenger RNA

reduction directly proportional to reduced water use (K_y=1) and more tolerant to water deficits (K_y<1)

Table 1. Seasonal yield response factor (K_y) clusters for various crops (After Steduto et al. 2012)

K_y	Response to water deficit	Crops
> 1	Very sensitive	Alfalfa, banana, bean, maize, onion, pea, pepper, potato, sugarcane, tomato, watermelon, wheat
= 1	Yield reduction directly proportional to reduced water use	Sugar beet
> 1	More tolerant	Cabbage, cotton, groundnut, safflower, sorghum, soybean, sunflower

Knowledge about molecular responses of plants to drought stress could assist in breeding transgenic crops that withstand water scarcity. Research on *Arabidopsis thaliana* found key physiological traits involved in plant–water relations underlying drought stress responses, but understanding the complex genetic architecture of enhanced adaptation to drought seems to be in its infancy (Juenger 2013). Very recently Bhardwaj and Yadav (2012) did a thorough review of mechanisms adopted by plants to escape, avoid or adapt to drought stress. They stated that enhanced adaptation to drought could be provided directly by metabolites such as trehalose, mannitol, glycinebetaine or indirectly through regulation of gene expression through TF and kinases in signal transduction.

Genetic engineering may be one of the biotechnology tools for developing crop cultivars with enhanced adaptation to drought (Ruane et al. 2008). It should be seen as complementary to conventional plant breeding rather than as an alternative to it. The function of a TF such as the *Dehydration-Responsive Element Binding* (*DREB*) gene in water stress-responsive gene expression has been extensively investigated (Sakuma et al. 2006). The main research goal was to gain a deep understanding of this TF for developing transgenic crops targeting drought-prone environments (Ortiz et al. 2007). For example, the *DREB1A* gene was placed under the control of a stress-inducible promoter from the *rd29A* gene and inserted via biolistic transformation[8] into bread wheat (Pellegrineschi et al. 2004). Plants expressing this transgene demonstrated significant adaptation to water stress when compared to controls under experimental greenhouse conditions as manifested by a 10-day delay in wilting when water was withheld. Saint Pierre et al. (2012) indicated, however, that these transgenic lines did not generally out-yield the controls under water deficit in confined field trials. Nonetheless, they were able to identify wheat lines combining acceptable or high yield under enough irrigation that also showed stable performance across the water deficit treatments used in their experiments; i.e., severe stress, stress starting at anthesis, and terminal stress.

Farooq et al. (2009) discuss the advances in transgenic breeding for drought-prone environments. In their review, they noted the testing of 10 transgenic rice events[9] under water scarcity. It seems that the transgenic expression of some stress-regulated genes leads to increased water use efficiency (WUE)[10].

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[8] Biolistic transformation (also known as particle bombardment) refers to DNA delivery into plant cells by high velocity gold or tungsten particles

[9] Unique DNA recombination taking place in one plant cell and thereafter to be used for generating entire transgenic plant(s)

[10] WUE is the ratio of biomass produced to the water used

For example, the *Arabidopsis' HARDY* (*HRD*) gene improved WUE by enhancing photosynthetic assimilation and reducing transpiration (Karaba et al. 2007). The enhanced photosynthesis assimilation and efficiency observed in transgenic rice bearing this gene seems to result from an increase of both leaf biomass and bundle sheath cells. This finding shows the benefits of using a plant model system as source of variation for crop breeding. Likewise, this research suggests that exploring genetic engineering to improve photosynthesis in a C_3 crop such as rice using C_4 traits could be beneficial (further details are given below in Section 6).

Drought affects maize throughout its life cycle but mostly when stress occurs before and after flowering. Nelson et al. (2007) used functional genomics to identify the transcription factor *AtNF-YB1*, which confers enhanced adaptation to *Arabidopsis* under drought. Thereafter, their bioinformatics[11] analysis led to identifying maize homologs to this TF. They selected *ZmNF-YB2* for genetic engineering it into an elite maize inbred. Transgenic maize lines showed enhanced adaptation to drought as measured by their chlorophyll content, stomatal conductance, leaf temperature, reduced wilting, and maintenance of photosynthesis. Enhanced adaptation to the stress translated into grain yield advantage under drought. The best-performing transgenic maize had about 50% increase in grain yield vis-à-vis the controls, whose grain yields were 50% below that expected under full watering.

Further research by a multinational seed company (Castiglioni et al. 2008) showed that bacterial RNA chaperones[12] also provide enhanced adaption to drought stress and improved grain yield in maize under water scarcity. The gene encoding cold shock protein B (*CspB*) from *Bacillus subtilis* – a soil bacterium – was used for genetic engineering MON 87460 maize. *CspB* allows the transgenic maize plant to react more quickly to drought, slowing its growth and conserving water, thereby making water available for key plant functions after the onset of drought stress. Based on this transgenic event, the DroughtGard™ hybrid maize was bred and released for farming in the USA in 2013.[13] Prior to the release, the company facilitated 250 large-scale on-farm trials (on about 4000 ha) of DroughtGard™ in the western half of the US Great Plains. Under stress, a DroughtGard™ hybrid used 261 mm of water from the soil while the control used 338 mm of water from the soil; i.e., their WUE rates (or the amount of water for producing 1 bushel or 25.4 kg of maize grains) were 0.59 and 0.44, respectively. At harvest, DroughtGard™ had more grain yield than non-transgenic maize hybrids with enhanced WUE (up to 0.4 t ha^{-1} greater in some locations of the western Great Plains). In theory, DroughtGard™ can save about 2.5 mm of water inputs per hectare; i.e., 5 trillion liters of water, which translates to providing water to the US city of Denver (Colorado) for one month.

The public Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV, Mexico) announced in 2010 the development of cisgenic[14] maize with enhanced adaptation to severe drought and extreme temperatures[15]. Under drought stress a maize plant begins

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[11] Collection, classification, storage, and analysis of complex biochemical and biological information related to molecular genetics and omics using computers and based on mathematics and information theory
[12] Proteins aiding in the process of RNA folding by preventing misfolding or by resolving misfolded structures
[13]http://www.genuity.com/corn/Pages/Genuity-DroughtGard-Hybrids.aspx or
http://www.monsanto.com/products/Pages/droughtgard-hybrids.aspx
[14] Cisgenic plants ensue from genetic engineering using a natural gene from a crossable (i.e., sexually compatible) species in the recipient plant
[15]http://www.2000agro.com.mx/biotecnologia/el-cinvestav-crea-maiz-resistente-a-sequia-y-temperaturas-extremas/

producing sugars such as trehalose, which can be broken by the glycoside hydrolase enzyme trehalase. When this sugar is not destroyed, the plant shows enhanced adaptation to drought stress. Hence, antisense RNA expression was used for silencing trehalase in the popular maize inbred line B73 (derived from Iowa Stiff Stalk Synthetic), which was used in the public maize sequencing project and has been one of the most successful parents of North American and other hybrids worldwide. The CINVESTAV announcement indicated that their cisgenic maize (CIEA-9) required 20% less water, endured high temperatures (up to 50°C), and had better flowering plus ears than local cultivars. CIEA-9 seeds also germinated at 8°C, demonstrating their ability to withstand cold at early development stages. Due to these promising results, CINVESTAV filed a petition for field-testing of CIEA-9 (Cabrera Ponce et al. 2011), and trials are ongoing in Sinaloa (northwest Mexico).

Salinity

Soils affected by salinity are found in more than 100 countries, and about 1/5 of irrigated agriculture is adversely affected by soil salinity. Therefore, breeding salt-tolerant crops should be a priority because salinity will likely increase under climate change. Mumms (2005) lists some candidate genes for salinity tolerance, indicating the putative functions of these genes in the specific tissues in which they may operate. Genes involved in tolerance to salinity in plants limit the rate of salt uptake from the soil and the transport of salt throughout the plant, adjust the ionic and osmotic balance of cells in roots and shoots, and regulate leaf development and the onset of plant senescence. The most promising genes for the genetic engineering of salinity tolerance in crops, as noted by Chinnusamy et al. (2005), are related to ion transporters and their regulators, as well as the C-repeat-binding factor. The recent genome sequencing of *Thellungiella salsuginea*, a close relative of *Arabidopsis* thriving in salty soils, will provide more resources and evidence about the nature of defense mechanisms constituting the genetic basis underlying salt tolerance in plants (Wu et al. 2012).

Advances have occurred in the quest for breeding transgenic rice and tomato showing salt tolerance. Plett et al. (2010) were able to show an improved salinity tolerance in rice by targeting changes in mineral transport. They initially observed that cell type-specific expression of *AtHKT1;1* – a sodium transporter – improved sodium (Na^+) exclusion and salinity tolerance in *Arabidopsis*. Further research explored the GAL4-GFP enhancer trap[16] to drive expression of *AtHKT1;1* specifically in the root cortex in transgenic rice plants. The transgenic plants had a higher fresh weight under salinity stress due to a lower concentration of Na^+ in the shoots. They also noted that root-to-shoot transport of $^{22}Na^+$ decreased and was correlated with an upregulation of *OsHKT1;5* – the native transporter responsible for Na^+ retrieval from the transpiration stream. Moghaieb et al. (2011) bred transgenic tomato plants producing ectoine – a common compatible solute in bacteria living in high salt concentrations. Ectoine synthesis was promoted in the roots of transgenic tomato plants under saline conditions, which led to increased concentration of photosynthates in improving water uptake. Likewise, the photosynthetic rate of ectoine-transgenic tomato plants increased through enhancing cell membrane stability in oxidative conditions under salt stress.

Heat

Global warming will reduce yields in many crops: about 6% and 5% average yield loss per 1°C in C_3 and C_4 crops, respectively, whose optimum temperature ranges are 15–20°C and 25–30°C (Yamori et

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[16] Transgenic construction inserted in a chromosome and used for identifying tissue-specific enhancers in the genome

al. 2013). The extent of yield loss depends on crop, cultivar, planting date, agronomy and growing area. For example, an increase of 1°C in the night time maximum temperature translates into a 10% decrease in grain yield of rice, whereas a rise of 1°C above 25°C shortens the reproductive phase and the grain-filling duration in wheat by at least 5%, thereby reducing grain yield proportionally. Heat stress will exacerbate climate change impacts in the tropics, while it may put agriculture at risk in high latitudes where heat-sensitive cultivars are grown today. Hence, new cultivars must be bred to address heat stress.

Ainsworth and Ort (2010) suggested giving priority to traits improving photosynthesis for adapting to heat stress. However, plants have various mechanisms to cope with high temperatures, e.g. by maintaining membrane stability, or by ion transporters, proteins, osmoprotectants, antioxidants, and other factors involved in signaling cascades and transcriptional control (Wahid et al. 2007; Hasanuzzaman et al. 2013). Furthermore, Gao et al. (2008) noted that *bZIP28* – a gene encoding a membrane-tethered TF – upregulated[17] in response to heat in *Arabidposis*. Some of these genes can be used in crop genetic engineering for enhancing plant adaptation to heat stress. For example, some stress-associated genes such as *ROB5* – a stress inducible gene isolated from bromegrass – enhanced performance of transgenic canola and potato at high temperatures (Gusta 2012). Likewise, Katiyar-Agarwal et al. (2003) introduced *hsp101*–a heat shock protein gene from *Arabidopsis* – in basmati rice. This transgenic rice had a better growth in the recovery phase after suffering heat stress.

5. Mitigating climate change through transgenic crops

Agriculture contributes significantly to greenhouse gas (GHG) emissions. As indicated by Philippot and Hallin (2011), plant breeding should therefore give priority to developing cultivars that can be used in farming systems with reduced GHG emissions. In this regard, transgenic crops have been contributing to lower GHG emissions through reducing fuel use, due both to less pesticide applications and increasing the area grown under conservation agriculture, which involves practices such as "no-till" or "reduced-till". Brookes and Barfoot (2012b) estimated that farming with transgenic crops since 1996 has led to additional soil carbon sequestered, equivalent to 133,639 million t of CO_2. Likewise, transgenic poplar trees overexpressing cytochrome P4502E1 – a key enzyme in the metabolism of a variety of halogenated compounds – increased the rates of metabolism and removal of volatile environmental pollutants such as hydrocarbons, including trichloroethylene, vinyl chloride, carbon tetrachloride, benzene, and chloroform (Doty et al. 2007).

Nitrous oxide and dioxide are potent GHGs released by manure or nitrogen (N) fertilizer, particularly in intensive cropping systems. Crops are bred for N-use efficiency (NUE) because this trait is a key factor for reducing N fertilizer pollution, improving yields in N-limited environments, and reducing fertilizer costs. There are various genetic engineering activities for improving NUE in crops (Shrawat and Good 2008). The gene *Alanine aminotransferase* from barley, which catalyzes a reversible transamination reaction in the N-assimilation pathway, seems to be a promising candidate for accomplishing this plant breeding target. Transgenic plants overexpressing this enzyme can increase N-uptake at early stages of growth. This gene technology was licensed to a private biotech company, which was founded with the aim of promoting sustainable agriculture (Daemrich et al. 2008). A patent issued a few years ago gave this company the rights to use this gene technology in major cereals –

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[17] Process of increasing the response to a stimulus

wheat, sorghum, rice, maize and barley – as well as in sugarcane. They have been testing the technology with rice in China, and researching further with rice and wheat in India, and assessing its value for maize and rice in sub-Saharan Africa through private-public partnerships.

Keeping N in ammonium form will affect how N remains available for crop uptake and will improve N-recovery, thus reducing losses of N to streams, groundwater and the atmosphere. There are genes in tropical grasses such as *Brachiaria humidicola* and in the wheat wild relative *Leymus racemosus* that inhibit or reduce soil nitrification[18] by releasing inhibitory compounds from roots and suppressing *Nitrosomonas* bacteria (Subbarao et al. 2007). Their value for genetic engineering crops for reducing nitrification needs to be further investigated. Almost one-fifth of global methane emissions are from enteric fermentation in ruminant animals. Apart from various rumen manipulation and emission control strategies, genetic engineering is a promising tool to reduce these emissions. The amount of methane produced varies substantially across individual animals of the same ruminant species. Efforts are ongoing to develop low methane-emitting ruminants without impacting reproductive capacity and wool and meat quality. A recent study to understand why some sheep produce less methane than others, by Rubin et al (2014), deployed high-throughput DNA sequencing and specialized analysis techniques to explore the contents of the rumens of sheep. The study showed that the microbiota present in sheep rumen was solely responsible for the differences among low and high methane-emitting sheep. It was further observed that the expression levels of genes involved in methane production varied more substantially across sheep, suggesting differential gene regulation. There is an exciting prospect that low-methane traits can be slowly introduced into sheep.

6. Re-engineering cereal photosynthesis

Photosynthesis involves the use of the sun's energy to obtain sugar and oxygen after binding CO_2 and water. There are two types of photosynthesis in the major crop species: C_3 and C_4. In C_3 photosynthesis CO_2 is first incorporated into a 3–carbon compound and the photosynthesis enzyme Ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco) is involved in CO_2 uptake. In C_4 photosynthesis CO_2 is first incorporated into a 4–carbon compound and phosphoenolpyruvate carboxylase (PEP) is the enzyme involved in the uptake of CO_2. C_4 photosynthesis occurs in inner cells and requires the special Kranz anatomy, while C_3 photosynthesis takes place throughout the leaf.

Photosynthetic efficiency in C_4 species (maize, sorghum, sugar cane) can exceed that of C_3 species (rice, wheat) by up to 50% at temperatures above 21 to 23°C. This is due to photorespiration suppression in the former. C_3 plants are well adapted to environments with cool temperatures. Genetic engineering C_4 photosynthesis into C_3 plants has been advocated to improve photosynthetic efficiency (Hibberd et al. 2008), which should translate into increasing biomass and grain yields per unit of water transpired. Ghannoum (2009) warns, however, that C_4 photosynthesis is highly sensitive to water stress (more so than C_3 photosynthesis). Another genetic engineering approach considers introducing components of a high efficient CO_2-concentrating mechanism from blue-green algae (cyanobacteria) into chloroplasts of C_3 plants. Mathematical modeling suggests that photosynthesis may improve up to 28% by introducing single-gene cyanobacterial bicarbonate transporters BicA and SbtA into C_3 chloroplasts (Price et al. 2013).

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[18] The process of oxidation of ammonium (NH4+) ions into nitrite (NO2-) ions and then nitrate (NO3-) ions by microorganisms during the nitrogen cycle in the soil

There have been many attempts at introducing transgenes into nuclear and plastid genomes to increase photosynthetic efficiency (Maurino and Weber 2013). This very ambitious genetic engineering undertaking may prove to be very difficult – introducing C_4 photosynthetic enzymes and changes in leaf anatomy and biochemistry into C_3 species will depend on several hundred genes. Denton et al. (2013) indicate that the genetic basis of C_4 photosynthesis remains mostly unknown, but there are some advances in understanding it through comparative genetic analysis of C_3 and C_4 species facilitated by the completion of their genome sequencing.

Manipulating Rubisco[19] has been regarded as a primary target for enhancing photosynthesis, thus improving both crop yield and input efficiency (Parry et al. 2013). Mathematical modeling suggests that genetic engineering plants expressing different types of Rubisco in sunlit and shade leaves may maximize C gains at current and elevated CO_2 levels (Zhu et al. 2004). Hanson et al. (2013) give an overview on manipulating Rubisco properties through plastid genetic engineering and how plastid operons could be changed for expressing various genes involved in pathways or controlling enzymes enhancing photosynthetic rates or reducing photorespiration.

7. Beyond climate change adaptation and mitigation: the transgenic pipeline

In spite of the positive impacts of farming transgenic crops, there are few transgenic traits and cultivars used commercially (Lemaux 2006). Herbicide tolerance, host plant resistance to insects and viruses, crop nutrient composition, and extended shelf life were the main traits of the first generation of approved transgenic cultivars. These included transgenic cultivars of alfalfa (lucerne), canola, cotton, eggplant, maize (including sweet corn), papaya, potato, rice, squash, soybean, sugar beet, and tomato. The newest release includes enhanced maize adaptation to drought-prone environments. A very recent e-conference convened by the United Nations Food and Agriculture Organization (FAO) highlighted some of the traits and crops in the genetic engineering pipeline ensuing from both private and public endeavors targeting the developing world (Ruane 2012). Traits are related to host plant resistance to pathogens (bacteria, fungi, nematodes, viruses) and insects, tolerance to herbicides, enhanced food and feed quality (β-carotene, fatty acid profiles, high lysine, low phytate content), enhanced adaptation to stressful environments (due to drought, heat and salinity), and improved input efficiency (nitrogen, water). Crops covered include banana, bean, cabbage, canola, cassava, chickpea, cotton, cowpea, eggplant, groundnut (peanut), maize, potato, rice, pigeonpea, sorghum, soybean, sugarcane, and wheat. Tammisola (2010) indicated that the global biofuel demand may pave the way for further use of genetic engineering for improving bio-energy crops and tapping the crop wild relatives gene reservoir.

Governments will play, through their regulation guidelines, a major role in determining whether or not some of these transgenic crops become new cultivars in farming systems. Stein and Rodríguez Cerezo (2009) argued that since each country regulates transgenic crops at its own pace and using its own rules, the global trade of such crops will be likely affected by this asynchronous approval system.

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[19] Enzyme involved in the first major step of carbon fixation

Functional foods for enhancing human health and better feed sources

Crop genetic engineering can deliver food with human health benefits beyond basic nutrition (Pew Initiative on Food and Biotechnology 2007). The targets include micronutrient-dense crops, increased protein content in main staples, oil crops with healthy fatty acid profiles, foods with enhanced antioxidant levels, and better dietary fibers and prebiotic/probiotics (Newell-McGloughlin 2008). As indicated by Goldman (2011), the interest in functional foods[20] coincides with an increasing consumer demand for healthy and nutritious food. The use of genetic engineering in producing functional foods does not challenge current regulatory frameworks *per se*, but it may expose some regulatory classification boundaries.

There have been significant advances in developing rice with enhanced β-carotene content (Datta et al 2007). This Golden Rice (as popularly known) ensues from incorporating transgenes for carotenogenesis in grains without changing other agronomic traits. Current versions of Golden Rice are based on a transgene from maize and thus represent a transgenic event between two cereals that are relatively closely related in an evolutionary sense. Stein et al. (2006) did an ex-ante impact analysis and noted that in India alone 1.4 million healthy life years could be saved annually if Golden Rice were consumed widely. They further indicated that Golden Rice compares favorably to alternative vitamin A interventions such as supplementation: the cost for saving one disability adjusted life year (DALY) is < US$ 20 for Golden Rice vis-à-vis US$ 134 to 599 for supplementation. Recent research shows that β-carotene in Golden Rice is as good as pure β-carotene in oil at providing vitamin A to children (Tang et al. 2012). Between 100 to 150 g of cooked Golden Rice (about 50 g dry weight) will likely provide approximately 60% of China's recommended nutrient intake of vitamin A for 6- to 8-year old children. Likewise, a stable transgenic maize plant with elevated amounts of β-carotene, ascorbate and folate in its endosperm was bred by genetically altering simultaneously their respective metabolic pathways (Navqi et al. 2009). This genetic engineering achievement could pave the way for breeding highly nutritious maize, which is the main staple of disadvantaged people in many parts of Africa and Latin America.

Holme et al. (2012) using cisgenesis with an endogenous phytase gene were able to improve the phosphate bioavailability (from 30 to 60%) of barley grains that are widely used for feeding monogastric animals such as chickens and pigs. This proof-of-concept of cisgenic plant engineering demonstrates the feasibility of using a crop's own genetic resources as well as the variation available in related wild species.

Engineering fatty acids and oils for nutritious diets

Each human consumes on average 25 kg per year of fats and oils, of which 80% are from plant sources. Genetic engineering can change fatty acid content of oilseed plants with the aim of improving human health or producing nutritional fatty acids absent in crops (Damude and Kinney 2008). These include fatty acids associated with lowering risks of coronary heart disease (Haslam et al. 2013).

So far, there are 10 transgenes that have led to the accumulation of high value fatty acids in plants.

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[20] Food with additional functions (most often related to promoting human health, preventing a disease, or enhancing performance) because of adding new ingredient(s) or increasing existing ingredient(s)

The genetic engineering of such oils in crops will require addressing regulatory issues because modifying plant composition to enhance its nutrient profile needs safety tests as well as oversight measures – especially to demonstrate that the changes will benefit most consumers and do not harm people.

Host plant resistance to tackle old and emerging pathogen epidemics

Weather influences how pathogens and pests affect and interact with crops and their host plant resistance, and thus climate change could have wide-ranging impacts on pests and diseases (Dwivedi et al. 2013). Late blight, which is caused by *Phytophthora infestans*, ranks as the most damaging potato pest. Late blight accounts for 20% of potato harvest failures worldwide, translating into 14 million t valued at € 2.3 billion. Global warming will increase late blight spread, e.g. expanding its range above 3,000 m in the Andes (Ortiz 2012 and references therein). Chemical control may lead to more aggressive strains of the pathogen and chemical control is often regarded as being environmentally damaging. Cisgenic potato cultivars with late blight resistance are becoming available and will impact growers, consumers and the environment favorably (Haverkort et al. 2008). Related wild *Solanum* species can be sources of alleles for enhancing host plant late blight resistance in potato. For example, *Solanum bulbocastum* – a wild relative with high resistance to late blight from Mexico – was used to breed the cultivar Fortuna using genetic engineering. Cisgenesis allows inserting several host plant resistance genes from wild species in one step without linkage drag[21].

A recent outbreak of *Xanthomonas campestris* pv. *Musacearum* led to the devastating *Xanthomonas* wilt of banana in the Great Lakes Region of Africa, thereby threatening the food security and income of millions of East and Central African people who depend on this crop. Transgenic banana plants with the *hypersensitivity response-assisting protein* (*Hrap*) gene from sweet pepper did not show any infection symptoms after artificial inoculation of potted plants with *Xanthomonas* wilt in the screenhouse (Tripathi et al. 2010). Selected transgenic banana plants with putative host plant resistance to *Xanthomonas* wilt are ongoing confined field-testing in East Africa, where elevated temperatures, due to the changing climate, will likely favor banana production.

8. Outlook

Crop breeding through genetic engineering begins with the discovery of genes and includes several stages (and years) of development and testing (including field trials). Transgenic crops are released for farming after assessing any potential risks to human health, food safety, the environment, and biodiversity. Crop physiology, plant genetics, and genomics are providing a better understanding of the physiological and molecular controls for key adapting traits in different agro-ecosystems.

Genetic engineering provides innovative methods for modern plant breeding to adapt crops to agricultural systems facing new challenges brought by the changing climate. New breeding methods, relying on genetic engineering, can accelerate the pace of improving crops, or be more precise for transferring desired genes into plant germplasm. Some limited target traits already available in

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[21] Reduction in cultivar fitness because of deleterious genes (often from a crop wild relative or exotic germplasm) introduced along with beneficial alleles

transgenic cultivars include those adapting agriculture to climate change and reducing their emissions of greenhouse gases.

However, only a handful of success stories exist, many of them focused on traits that reduce inputs or enhance the nutritional quality of crops. Rommens (2011) includes trait efficacy in the field, critical products concepts, freedom-to-operate, industry support, identity preservation and stewardship, regulatory approval, and last but not least, retail and consumer acceptance among the main barriers for farming more transgenic crops. Although the technologies demonstrate potential to reduce crop losses, food waste, and enhance nutritional quality, there remains only limited evidence that transgenic cultivars can increase yields and contribute to greater yield stability under a changing climate through exploitation of abiotic resistances. Nonetheless, farmers' surveys reveal that increased yields are among the benefits for growing transgenic crops (Carpenter 2010). Such a finding results from yield increases because of reduced losses from insect pests and weeds. In most developing countries, crop yields are low and yield gaps are large because of low input use, poor soil health, and pests and diseases. Genetic engineering has a lot promise in increasing overall adaptive capacity of agriculture but emphasis on good agricultural practices, including maintenance of soil, water, and genetic resources and increasing irrigation and fertilizers remains critical to increasing production.

Globally, there is increasing emphasis on climate-smart agriculture. Climate-smart agriculture ensures a sustainable increase in agricultural productivity and income along with strengthening resilience to climate change and reducing or removing greenhouse gases emissions, wherever possible. The examples listed in earlier sections demonstrate how genetic engineering can facilitate yield increases in stress-prone environments. There are, however, no examples yet in the literature to demonstrate that molecular or conventional breeding can result in simultaneous improvements in productivity, resilience, and the adaptation and mitigation capacity of agriculture.

Given that current trends in yield increase are insufficient to double food production by 2050 (Ray et al. 2013) and keep up with population and shifting consumption patterns, new approaches to the problem are needed. Transgenic cultivars could be a piece of that puzzle, building on success stories such as that of WEMA, but it is crucial that safety concerns are adequately addressed. Further research should benchmark conventional approaches to crop improvement, compare them with likely yield increases available from transgenic approaches, and explicitly address climate impacts to ascertain the true potential of transgenic technologies in adapting to and mitigating climate change in the long term.

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