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REVIEW ARTICLE

Next biotech plants: new traits, crops, developers and technologies for addressing global challenges

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Abstract

Most of the genetically modified (GM) plants currently commercialized encompass a handful of crop species (soybean, corn, cotton and canola) with agronomic characters (traits) directed against some biotic stresses (pest resistance, herbicide tolerance or both) and created by multinational companies. The same crops with agronomic traits already on the market today will continue to be commercialized, but there will be also a wider range of species with combined traits. The timeframe anticipated for market release of the next biotech plants will not only depend on science progress in research and development (R&D) in laboratories and fields, but also primarily on how demanding regulatory requirements are in countries where marketing approvals are pending. Regulatory constraints, including environmental and health impact assessments, have increased significantly in the past decades, delaying approvals and increasing their costs. This has sometimes discouraged public research entities and small and medium size plant breeding companies from using biotechnology and given preference to other technologies, not as stringently regulated. Nevertheless, R&D programs are flourishing in developing countries, boosted by the necessity to meet the global challenges that are food security of a booming world population while mitigating climate change impacts. Biotechnology is an instrument at the service of these imperatives and a wide variety of plants are currently tested for their high yield despite biotic and abiotic stresses. Many plants with higher water or nitrogen use efficiency, tolerant to cold, salinity or water submergence are being developed. Food security is not only a question of quantity but also of quality of agricultural and food products, to be available and accessible for the ones who need it the most. Many biotech plants (especially staple food) are therefore being developed with nutritional traits, such as biofortification in vitamins and metals. The main international seed companies continue to be the largest investors in plant biotechnology R&D, and often collaborate in the developing world with public institutions, private entities and philanthropic organizations. These partnerships are particularly present in Africa. In developed countries, plant biotechnology is also used for non-food purposes, such as the pharmaceutical, biofuel, starch, paper and textile industries. For example, plants are modified to specifically produce molecules with therapeutic uses, or with an improved biomass conversion efficiency, or producing larger volumes of feedstocks for biofuels. Various plant breeding technologies are now used in the entire spectrum of plant biotechnology: transgenesis producing proteins or RNAi. Cisgenesis (transgenes isolated from a crossable donor plant) and intragenesis (transgenes originate from the same species or a crossable species), null segregants are also used. To date, the next generation precision gene editing tools are developed in basic research. They include: clustered regularly interspaced short palindromic repeats (CRISPR), oligonucleotide-directed mutagenesis (ODM), transcription activator-like effects nucleases (TALENs) and zinc-finger nuclease (ZFN).

Keywords

Biofortification, biofuels, climate change, editing, food security, GMO, transgenesis

History

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Introduction

In 2013, GM plants were legally commercialized in 27 countries on 175 million hectares, and produced by 18 million farmers (James, 2013). There were 84 million hectares of soybean, 57 million hectares of corn, 24 million hectares of cotton and 8 million hectares of canola (rape seed). This concentration results from the fact that developing a new GM plant is long, costly and knowledge-intensive. On average, $136 million and 13 years are necessary from the initial discovery to the conformity with regulation (McDougall, 2011). Crops and traits that provide the highest returns on investments have therefore been the first plants developed by institutions that have large investment capacities in research

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Table 1. Transgenic soybean, corn, cotton and canola with new biotic stress resistance.

Species	Pipeline	Trait–resistance (products)	Developer	Method
Canola/Rapeseed	Closest to market release	Herbicide (Optimum® Gly and LibertyLink®)	Dupont/Pioneer, USA	Transgenesis
		Herbicide (LibertyLink®, TruFlex Genuity® Roundup Ready®)	Monsanto, USA	Transgenesis
	R&D	Combined herbicides (TruFlex™ Roundup Ready® LibertyLink®), Dicamba	Monsanto, USA	Transgenesis
	Approved in Canada	Herbicide	Cibus, USA	Rapid Trait Development System (RTDS™): ODM
	Confined field trials, Kenya	Stem borer	International Maize and Wheat Improvement Center (CIMMYT), Kenya Agricultural Research Institute (KARI)	Transgenesis
	Pending regulatory approval for market release	Glyphosate, 2,4-D (Enlist™ corn)	Dow AgroSciences, USA	Transgenesis
	Close to market	Herbicide. Suppressed phytate biosynthesis	Dow AgroScience, USA	EXZACT™ precision technology: Zinc Finger Nuclease (ZFN)
Corn	R&D	Corn borer and corn root worm (Optimum®, AcreMax®, XTreme Insect Protection)	Dupont/Pioneer, USA	Transgenesis
	Closest to market release	Herbicides, several pests (Optimum® Intrasect® Insect Protection + Agrisure Viptera®)	Dupont/Pioneer, USA	Transgenesis
	R&D	Corn rootworm, corn borer (protection DP4114), herbicide	Dupont/Pioneer, USA	Transgenesis
	R&D	Corn borer and corn root worm, stalk rot, grey leaf spot, Goss's wilt	Monsanto, USA	Transgenesis
	Closest to market release	Dicamba, glufosinate and glyphosate herbicides (Roundup Ready® Xtend Crop System)	Monsanto, USA	Transgenesis
	Greenhouse containment, South Africa	Maize streak virus	Pannar Seed, University of Cape Town	Transgenesis
Cotton	Closest to market release	Dicamba, glufosinate and glyphosate herbicides (Genuity Roundup Ready Flex®) and pests budworm cotton boll-wormn (Bollgard® II and Genuity Roundup Ready Flex® Bollgard® II and Bollgard III in Australia)	Monsanto, USA	Transgenesis
		Herbicide (GlyTol®, LibertyLink®, GlyTol and GlyTol LibertyLink stacked technology in high-performing FiberMax®) conventional breeding	BayerCropScience, Germany	Transgenesis
Soybean	Closest to market release	Lepidopteran insects, glyphosate, 2,4-D (Enlist™ 2 CRY proteins)	Dow AgroSciences, USA	Transgenesis
		Dicamba herbicide (Roundup Ready® Xtend Crop System)	Monsanto, USA	Transgenesis

and development (R&D). Currently, very few biotech plants other than field crops are being commercialized globally. These minor crops were mainly developed by public research and play a secondary economic role relative to commodity crops. Several minor crops were developed with RNAi-mediated for resistance to viruses: the Ringspot Virus in papaya, grown in Hawaii, USA, authorized in Japan (Gonsalves et al., 2010) and in Canada, the Plum Pox Virus in plum, deregulated in the USA (Scorza et al., 2013),and the Bean Golden Mosaic Virus in bean, approved in Brazil (Tollefson, 2011), by chronological order of deregulation. The plants listed in the following tables include those studied in R&D as well those recently commercialized (i.e. since 2013).

Next edible plants with biotic stress resistance

The biotech varieties of soybean, corn, cotton and canola currently in the pipeline are being developed by multinational

Table 2. New edible biotech plants resistant to biotic stresses.

Species	Pipeline	Trait	Developer	Method	References
Apple	Confined filed trials, EU	Scab	ETH Zurich, Switzerland, Plant Research International, University of Wageningen, the Netherlands	Cisgenesis	Vanblaere et al. (2011); Joshi et al. (2011)
Banana	Confined field trials, Uganda	Parasitic nematode and weevil	NARO (National Agricultural Research Organization, Uganda), University of California, University of Leeds, university of Pretoria, biodiversity International, Government of Uganda, Rockefeller Foundation, USAID	Transgenesis	http://www.banana.go.ug/
	Confined field trials, Kenya, Uganda	Banana Xanthomonas Wilt (BXW)	National Banana Research Program, International Institute for Tropical Agriculture (IITA), and Academia Sinica, Taiwan	Transgenesis	Tripathi et al. (2014); Namukwaya et al. (2012)
	R&D	*Fusarium oxysporum* f. sp. *cubense* and *Mycosphaerella fijiensis*	National Banana Research Program, Department of Crop Production, Makerere University, Kampala, National Agricultural Research Laboratories, National Research Organization, Kampala, Uganda; Venganza, Inc., USA	RNAi	Mumbanza et al. (2013)
Cassava	Confined field trials, Kenya, Uganda	Cassava brown streak disease (CBSD) and cassava mosaic disease (CMD)	Donald Danforth Plant Science Center (USA), National Crops Resources Research Institute (Uganda), KARI, USAID, the Monsanto Fund, Bill and Melinda Gates Foundation, and Howard Buffet Foundation	RNAi	Virus-Resistant Cassava for Africa (VIRCA). http://www.danforthcenter.org
Chestnut	Confined field trials, USA	Chestnut blight (caused by the fungus *Cryphonectria parasitica*)	Forest health Initiative (FHI), USDA, (US Department of Agriculture), New York States University, and University of Georgia, USA	Transgenesis	Powell et al. (2005); Maynard et al. (2009)
Citrus	Confined field trials, USA	Greening or Huanglongbing (HLB) (caused by the bacteria *Candidatus Liberibacter asiaticus*)	USDA, Southern Gardens Citrus, Texas A&M University, USA	Transgenesis	Polek et al. (2007)
Cowpea	Confined field trials, Burkina Faso, Ghana, Nigeria	Pod borer (*Maruca vitrata*)	African Agricultural Technology Foundation (AATF), Network for the Genetic Improvement of Cowpea for Africa (NGICA), Commonwealth Scientific and Industrial Research Organization (CSIRO), IITA, Monsanto, the Kirkhouse Trust, National Agriculture Research Systems in Target countries of West Africa, Institute for Environment and Research in Agriculture (INERA) in Burkina Faso, Institute of	Transgenesis	http://www.aatf-africa.org/

(continued)

Table 2. Continued

Species	Pipeline	Trait	Developer	Method	References
			Agricultural Research (IAR) in Nigeria, USAID, Rockefeller Foundation.		
Egg Plant	Pending authorization, India	Brinjal fruit and stem borer (*Euzophera perticella*)	Maharashtra Hybrid Seed Company (in *joint-venture* with Monsanto)	Transgenesis	http://mahyco.tradeindia.com/
	Approved for commercialization in October 2013, Bangladesh	Brinjal fruit and shoot borer (*Leucinodes orbonalis*)	Bangladesh Agricultural Research Center (BARC), Bangladesh Agricultural Research Institute (BARI), Cornell University – College of Agriculture and Life Sciences (CALS), USA, Department of Biotechnology (DBT), Government of India, Indian Council of Agricultural Research (ICAR), Indian Institute of Vegetable Research (IIVR), International Service for the Acquisition of Agribiotech Applications (ISAAA) – Southeast Asia Center, Philippines, India, Sathguru Management Consultants Pvt. Ltd., India, Tamil Nadu Agricultural University (TNAU), India, University of Agricultural Sciences, Dharwad (UAS-D), India, University of the Philippines, Virginia Tech, USA	Transgenesis	http://absp2.cornell.edu/projects/project.cfm?productid=2
Grapevine	R&D	Fungal disease	University of Florida, Fort Valley State University, Georgia	Cisgenesis	Dhekney et al. (2011)
	R&D	Pierce disease (caused by the bacteria *Xylella fastidiosa*)	Pierce's Disease Control Board of the California Department of Food and Agriculture, the US Department of Energy, and the USDA	Transgenesis	Dandekar et al. (2012)
Plum tree	Field trials and confined test plots, EU; deregulated, USA	Plum pox virus	Public Research Project funded by the EU; USA/ARS	RNAi	Scorza et al. (2013)
Potato	Confined test plots, EU	Blight	Biotechnology and Biological Sciences Research Council (BBSRC), UK	Cisgenesis (Gene transfer from South American potato varieties)	http://www.bbsrc.ac.uk
	Confined test plots, EU	Blight	AMIGA European Union Research program	Transgenesis	Van der Voet et al. (2011)
	Confined test plots, EU	Blight	VIB, Hogeschool Gent, Ghent University, and ILVO (Instituut	Transgenesis	http://www.vib.be

			voor Landbouw – en Visserijonderzoek), Belgium		
	Field trials, EU	Blight	Plant Research International, University of Wageningen, the Netherlands	Cisgenesis	Haverkort et al. (2009)
	R&D	Blight	Leibniz Institute of Plant Biochemistry, Germany	RNAi	Eschen-Lippold et al. (2010)
	R&D	Potato virus Y	Shandong Agricultural University, Tai'an, Shandong, China	RNAi	Zhu et al. (2009)
	Field trials, South Africa, USA	Tuber moth – Spunta G2 variety with Cry11a1 gene	University of Michigan, USA, Agricultural Research Council, South Africa, USAID funded	Transgenesis	Douches (2010), Douches et al. (2009
Rice	R&D	Pest, herbicide	BayerCropScience, Germany	Transgenesis	See company website
	n/a	Lepidopteran pests (pink and yellow borers)	Dupont/Pioneer, USA	Transgenesis	See company website
	n/a	Herbicide	Dupont/Pioneer, USA	Transgenesis	See company website
	Confined field trials since 2012, EU	Aphid (virus vector)	Rothamsted public research institute, UK	Transgenesis	http://www.rothamsted.ac.uk/
	R&D since 2008		Borlaug Global Rust Initiative (BGRI): collaborative efforts of 22 research institutions around the world led by Cornell University, USA	Accelerated multilateral plant breeding, genomics	http://www.globalrust.org
Sugarcane	R&D	Pest	Monsanto, USA and Argentina	Transgenesis	See company website
	Field trials since 2010, Argentina	Pest, Herbicide	Obispo Colombres Experimental Research Station and Santa Rosa Research Institute, Argentina	Transgenesis	http://www.eeaoc.org.ar
Sweet Potato	Greenhouse containment, Kenya, Uganda	Weevil	Auburn University, USA, CIP (International Potato Center), Donald Danforth Plant Science Center, Kenyatta Univ., NaCRRI (National Crops Research Institute, Uganda), NARL (National Agricultural Research Laboratories, Uganda), University of Ghent, University of Puerto Rico, Gates Foundation, Howard G. Buffet Foundation	Transgenesis	sweetpotatoknowledge.org
Tomato	R&D	Bacterial spot disease	Kansas State University, USA	Transgenesis	Horvath et al. (2012)
	Development	Biotic stresses	Morflora, Israel	TraitUp™ technology: plasmid (used in seed treatment, no insertion into chromosomes)	See company website
Walnut	R&D	Bacteria, fungus, virus, nematode	UC Davis, USA	Grafting on GM rootstock on wild-type scion	Haroldsen et al. (2012)
Wheat	R&D (on wheat and other field crops)	Aphid and whitefly	John Innes Center, Genome Analysis Center, Rothamsted Research and University of Greenwich, UK	RNAi	https://www.jic.ac.uk /

Table 3. Biotech plants tolerant to abiotic stresses.

Species	Pipeline	Trait–product	Developer	Method	References
Canola	Licensed to Scotts (USA), Syngenta (Switzerland), Bayer CropScience (Germany), Dupont/Pioneer (USA), Mahyco (India) RiceTec (USA) and DBN (China)	Drought	Performance Plants, Canada	RNAi driven by conditional promoters to suppress farnesyl-transferase; shuts down stomata	See company website
Corn	Field trials, Chile, USA	Drought	Dupont/Pioneer, USA	RNAi: gene-silencing approach to modulate the levels of ethylene biosynthesis	Habben et al. (2014)
	On the market in the USA since 2013	Drought (Genuity® DroughtGard™)	Monsanto/BASF, USA	Transgenesis. Expresses a cold shock protein B from *Bacillus subtilis*, which stabilizes RNA	See company website
	Field trials, Argentina for 3 years	Drought	Argentine firm Bioceres signed a joint venture agreement with the US Company Arcadia Biosciences Monsanto/BASF, USA	Transgenesis	See company website
	R&D – Confined tests plots for 6 years and greenhouse containment, Kenya, Uganda, South Africa.	Drought	Water Efficient Maize for Africa (WEMA): Bill and Melinda Gates Foundation, Howard Buffet Foundation, AATF, national agricultural research organizations in Kenya, Mozambique, South Africa, Tanzania and Uganda, farmers groups, seed companies, IRRI, Monsanto	Conventional breeding, marker assisted selection, transgenesis	http://wema.aatf-africa.org
	R&D	Drought, flooding/submergence, salinity	BayerCropScience, Germany and International Research Institute for Rice (IRRI)	Genetic engineering, marker assisted selection	See company website
	R&D	Salinity	Michigan State University, USA	Transgenesis (corn obtained with barley gene transfer)	Nguyen & Sticklen (2013)
	R&D	Flooding/Submergence (anaerobic germination)	Australian Center for Plant Functional Genomics (ACPFG)	Transgenesis	Kretzschmar et al. (2012)
	R&D	NUE	Monsanto	Transgenesis	See company website
	R&D	NUE	Dupont/Pioneer	Conventional breeding, molecular breeding and transgenesis	See company website
	R&D	NUE	Improved Maize for African Soils (IMAS) project: USAID, IRRI, Bill and Melinda Gates Foundation, Dupont/Pioneer, KARI, South African Agricultural Research Council (ARC)	Conventional breeding, molecular breeding and transgenesis	http://www.cimmyt.org/
Rice	R&D	Salinity	ACPFG, Waite Research Institute	Transgenesis	Plett et al. (2010)

	R&D – 2 years of field trials, USA	Drought, NUE, salinity	Arcadia biosciences; ACPFG, Waite Research Institute	Transgenesis: expresses isopentenyltransferase from *Agrobacterium*, which catalyzes the rate-limiting step in cytokinin synthesis; Technology licensed to developers who have put the gene into their own varieties of soybean, wheat, rice, cotton, sugar beets, sugarcane and tree crops (see Bioceres corn, soy, sugarcane and wheat in this table)	See company website
	R&D in Asia (Bangladesh, India, Nepal) and Africa (Benin, Burkina Faso, Ethiopia, Gambia, Ghana, Guinea, Madagascar, Mali, Mozambique, Nigeria, Rwanda Senegal, Tanzania, Uganda)	Drought, submergence (anaerobic germination), salinity, iron toxicity, cold	Stress Tolerant Rice for Africa and South East Asia (STRASA): IRRI, Bill and Melinda Gates Foundation, AfricaRice, AGRA, producer organizations, NGOs, national research entities	Genomics, molecular markers, trait introgression, participatory variety selection	http:://www.strasa.irri.org
	R&D in India and Bangladesh	Flooding/submergence tolerance	IRRI, Philippines	Marker assisted backcrossing, QTLs, transgenesis	http://www.cimmyt.org/
	R&D	NUE	NUE NERICA: Arcadia Biosciences, African Agricultural Technology Foundation (AATF) and International Center for Tropical Agriculture (CIAT)	Transgenesis	See company website
Rice and peanut	Field trials, India	Drought	University of Calcutta (rice), the International Crops Research Institute for the Semi-Arid-Tropics (ICRISAT) (peanuts) – University of Tokyo, Japan, International Research Center for Agricultural Sciences	Transgenesis (expresses DREB1A transcription factor under the control of the rd29A promoter)	http://www.icrisat.org/
Ryegrass	R&D	Drought	ViaLactia Biosciences, New Zealand	Intragenesis	Bajaj et al. (2008)
Sugarcane	Commercial production in Indonesia in 2014 following approval in 2013	Drought	PT Perkebunan Nusantara XI; Sugarcane University of Jember (East Java, Indonesia); Ajinomoto	Transgenesis, expresses choline dehydrogenase (betA) which accumulates membrane protectant glycine betaine	Marshall (2014)
	Field trials, Argentina	Drought	Argentine firm Bioceres signed a joint venture agreement with the US Company Arcadia Biosciences	Transgenesis	See company website
	R&D in Brazil	Drought	Centro de technologia canavieira (CTC), Brazil and BASF, USA	Transgenesis	http://www.ctcanavieira.com.br/
Soybean	R&D – Field trials for 3 years in Argentina	Drought	Argentine firm Bioceres signed a joint venture agreement (Verdeca) with the US Company Arcadia Biosciences	Transgenesis – overexpresses Hahb-4, from sunflower thought to inhibit ethylene-induced senescence	See company website

(*continued*)

Table 3. Continued

Species	Pipeline	Trait–product	Developer	Method	References
Tomato	R&D – Greenhouse studies, India	Drought	Indian Agriculture Research Institute	Transgenesis: over expressing osmotin-encoding genes under the control of the 35S CMV promoter	http://www.iari.res.in/
Wheat	R&D – Field trials, Egypt	Drought	Agricultural Genetic Engineering Research Institute, Egypt	Transgenesis: expresses HVA1 gene from barley, which confers osmotolerance	http://www.arc.sci.eg
	R&D, Argentina, Brazil, Paraguay, Uruguay	Drought, salinity	Trigall genetics: joint venture created between Florimond Desprez, France and Bioceres, Argentina	Transgenesis (HB4® technology)	See company website
	R&D	Drought, NUE	Limagrain	Transgenesis	See company website
	Field trials for 3 years, Argentina	Drought	Argentine firm Bioceres signed a joint venture agreement with the US Company Arcadia Biosciences (Verdeca)	Transgenesis	See company website

corporations. During the first years, transgenesis was used to transfer one trait in the varieties commercialized (mainly, resistance to one pest or tolerance to one herbicide). The new biotech plants being developed through transgenesis and directed against biotic stresses contain traits and combine resistance to several pests, tolerance to several herbicides or both (Stein & Rodriguez-Cerezo, 2009; Table 1).

Although wheat and rice represent significant market shares of global agricultural production, consumption and trade, biotech varieties of wheat and rice are currently not on the market, despite the fact that Iran commercializes insect resistant rice [transgenes from *Bacillus thuringiensis* (Bt)] in 2006. Also, regulation has become increasingly stringent over the years, and has discouraged most developers (especially in the public sector) from working on plants other than commodity crops (Miller & Bradford, 2010). In fact, market approvals of biotech plants with comparably lower economic value on world markets than commodity crops increased steadily in the 1990s but then declined, mainly due to the regulatory burden imposed prior to market release. Ironically, as markets have become more restrictive, the prospects of developing long lasting disease resistance systems have improved significantly, as a result of a better understanding of the molecular components of the plant immune system, and the development of faster and less expensive technologies to sequence the genome (Dangl et al., 2013).

In some countries, wheat is grown from farm-saved seeds at a higher rate than in other species, and investing in wheat genetics has therefore lower returns than investing in corn or soybean genetics for seed companies. Nevertheless, R&D investment in wheat genetics is growing at full speed to address the global stagnation of wheat yields (while yields of corn and soybean continue to grow; Fischer et al., 2014) and as the use of combined and diverse biotechnologies (including marker assisted selection, genomics, association genetics, high-throughput phenotyping and genotyping) is expanding. For example, the Wheat Initiative is an international consortium coordinating research on wheat launched under the French Presidency of the G20 in 2011. Several major seed groups are currently engaged in wheat breeding to develop transgenic or hybrid varieties. TALENs and the CRISPR-Cas9 system can be used to generate wheat resistance to powdery mildew (Wang et al., 2014).

New biotech crops currently being developed with agronomic traits include tuber plants, cowpeas, fruit trees, grapevine, sugarcane and vegetables (Table 2).

Edible plants with abiotic stress tolerance

Several crops can be genetically modified to improve their water use efficiency (Marshall, 2014; Waltz, 2014) or tolerate drought. This trait is more difficult to transfer to plants than herbicide tolerance or pest resistance as a wider range of genes are usually involved, and several pathways have been explored to insert this traits into plants. The first biotech drought tolerant corn varieties have been cultivated in the USA since 2013.

Resistance to abiotic stresses is being introduced in other species than the soybean, corn, cotton and canola currently on the market. It includes not only drought tolerance, but also

Table 4. Nutritional traits in biotech edible crops.

Species	Pipeline	Trait–Product	Developer	Method	References
Alfalfa	Deregulated by USDA in November, 2014	Low lignin (to increase alfalfa digestibility by livestock)	Monsanto and Forage Genetics International	RNAi	http://www.aphis.usda.gov/brs/aphisdocs/12_32101p_dpra.pdf
	R&D	Low lignin (to increase alfalfa digestibility by livestock)	J.R. Simplot company, USA	Intragenesis	Week et al. (2008)
Apple	Close to market, Canada and USA	Arctic® Apples, non-browning	Okanagan Specialty Fruits	RNAi	See company website
Banana	R&D, Australia, Uganda	Banana21, enriched in pro vitamin A and iron, disease resistance, drought tolerance	Center for Tropical Crops and Biocommodities in Queensland University, national research program on banana in Uganda, funded by the Bill and Melinda Gates Foundation	Transgenesis	http://www.banana21.org/
Barley	R&D, field trials, EU	Improved phytase activity	Aarhus University, Denmark	Cisgenesis	Holme et al. (2012, 2013)
Camelina	R&D	Long chain omega 3 polyunsaturated fatty acid	Rothamsted Research Center, UK	RNAi	Ruiz-Lopez et al. (2013)
Cassava	R&D, confined field trials, Kenya and Nigeria	Increased nutrient (zinc, iron, protein and vitamin A) levels	Biocassava Plus program – Donald Danforth Plant Science Center, ETH Zurich, NARS, Bill and Melinda Gates Foundation	Transgenesis	Sayre et al. (2011)
Corn	R&D	HarvestPlus Program: Vitamin A biofortification	Center for International Tropical Agriculture (CIAT), International Food Policy Research Institute (IFPRI), Consultative Group for International Agricultural research (CGIAR), Bill and Melinda Gates Foundation, Asian development Bank, Austrian Ministry of Finance, Canadian International Development Agency, International fertilizer Group, International Life Science Institute, Royal Danish Ministry of Foreign Affairs, Swedish International Development Agency, Syngenta Foundation for Sustainable Agriculture, United Kingdom Department for International Development, USAID, USDA, World Bank, World Food Program	Conventional breeding, Transgenesis	www.harvestplus.org
		Vitamins A, B9 and C enriched corn	Lleida University, Spain	Transgenesis	http://www.udl.es/

(continued)

Table 4. Continued

Species	Pipeline	Trait–Product	Developer	Method	References
Canola	R&D	High content in omega 3 fatty acids	BASF	Transgenesis	See company website
Grape vine	Non-regulated status by USDA	Increased anthocyanin production (red berries)	University of Florida, USA	Intragenesis	Espinoza et al. (2013)
Pineapple	Deregulated by USDA in January 2013	Enriched in lycopene, ‘‘Rosé’’ variety	Del Monte, USA	Null segregants	See company website
Potato	R&D	Enriched in protein	National Institute of Plant Genome Research, India	Transgenesis	Chakraborty et al. (2010)
	R&D	Reduced production of acrylamide after cooking	University of Wisconsin-Madison, USA and USDA/Agricultural Research Service, USA	RNAi	http://www.wisc.edu/
	Deregulated status from USDA in November 2014, pending authorization by Canada and Japan	Innate™, non-browning biotechnology, reduced production of acrylamide	J. R.Simplot company	Intragenesis	See company website Holme et al. (2013).
Potato, tomato, strawberry	R&D	Vitamin C biofortified	New Zealand Institute for Plant and Food Research Limited, Simplot company, and School of Biological Sciences, University of Auckland, New Zealand	Transgenesis	Bulley et al. (2012)
Rice	Close to market, Bangladesh, China, India, Indonesia, Philippines, Vietnam	Golden Rice, biofortified in beta-carotene	ETH Zurich University and Fribourg University, Syngenta, funding by the Rockefeller Foundation and the Bill and Melinda Gates Foundation, USAID, Department of Agriculture of the Philippines, Swiss federal fund. Field trials conducted by IRRI	Transgenesis	Ye et al. (2000), Paine et al. (2005), Alberts et al. (2013)
	Close to market, Bangladesh	Rice enriched in zinc and iron	Harvest Plus program and Australian Research Council	Transgenesis	Johnson et al. (2011)

Sorghum	Field trials, Burkina Faso, Kenya, Nigeria, South Africa (greenhouse containment)	Biofortification with iron and zinc	The Africa Biofortified Sorghum (ABS) Project: AATF, Africa Harvest, CSIRO, ICRISAT, NARS, Dupont/Pioneer, Donald Danforth Plant Science Center, University of California, University of Pretoria, Gates Foundation, Howard G. Buffett Foundation	Transgenesis	http://biosorghum.org/
Soybean	On the market, USA	Plenish® high oleic	Dupont/Pioneer	RNAi	See company website
		Vistive® Gold low saturated high oleic	Monsanto	RNAi	See company website
	R&D	Oil content and feed efficiency varieties	Dupont/Pioneer	Transgenesis	See company website
		Stearidonic acid omega-3 varieties, high threonine	Monsanto	Transgenesis	See company website
Tomato	R&D	Enhanced shelf life	National Institute of Plant Genome Research, India	Transgenesis	Meli et al. (2010)
	R&D	Enriched in anthocyanins	John Innes Center, UK	Transgenesis	Butelli et al. (2008)
	R&D	Enhanced carotenoid and flavonoid content	Cell Signaling Laboratory, Stazione Zoologica, Naples, Italy; Royal Holloway, University of London, Biological Sciences, Egham, Surrey; DNA Plant Technology, Oakland, California, USA; Seminis Vegetable Seeds, Inc. Woodland, California, USA; CNRS/ENS Paris, France; Laboratory for Plant Physiology, Wageningen University, the Netherlands	RNAi	Davuluri et al. (2005).
Wheat	R&D	Gluten-free (coeliac disease)	Instituto de Agricultura Sostenible, Spain	RNAi	Gil-Humanes et al. (2008), León et al. (2009)
	R&D	High amylose	CSIRO, Australia, Biogemma UK Limited	RNAi	Regina et al. (2006)

Table 5. Examples of therapeutic molecules produced from biotech plants and a fungus by transgenesis.

Species	Pipeline	Trait: Molecule produced – Disease treated	Developer	References
Arabidopsis thaliana	Marketed in the EU	Intrinsic human factor – Vitamin B12 deficiency	Cobento Biotech A/S, Denmark	See company website
Barley	Confined field trials, USA	Lyzozyme, lactoferrin	Washington State University, USA	http://www.aphis.usda.gov/brs/ph_permits.html
Carrot	Approved in the USA, Brazil, Israel, Mexico, Chile, Uruguay	ELELYSO™ (taliglucerase alfa) – Gaucher disease type I	Protalix, Israel and Pfizer, USA	See company website
	Clinical tests (I)	Acetylcholesterase – Biodefense, nerve agent attacks	Protalix, Israel	See company website
Corn	Confined field trials, USA	22 pharmaceutical and/or industrial molecules expressed in corn seeds, including Hepatitis B virus surface antigens, Brazzein sweet proteins, plus proteases, hydrolases, endo- and exo-cellulases and protease inhibitors	Applied Biotechnology Institute, USA	http://www.aphis.usda.gov/brs/ph_permits.html
	Clinical tests (I)	Lactoferrin (for the prevention of antibiotic-associated diarrhea)	Ventria Bioscience, USA (acquires Meristem Therapeutics' Recombinant Lactoferrin Intellectual Property, France)	See company website
Plasmodium (fungus)	R&D	Inhibiting substances in mosquitoes	University of Maryland, USA; John Hopkins School of Public Health, UK; University of Westminster, UK	http://www.umd.edu/ Fang et al. (2011)
Potato	R&D	One pharmaceutical protein	Planton, LLC, USA	http://www.aphis.usda.gov/brs/ph_permits.html
	Clinical tests (I)	Capsid virus Norwalk protein	Arizona State University, USA	Tacket et al. (2000)
Potato and Lettuce	Clinical tests (I)	Hepatitis B virus surface antigen	Thomas Jefferson University, USA (transgenic potato) Polish NAS, Poland (transgenic lettuce)	Potato: Kapusta et al. (1999) Lettuce: Thanavala et al. (2005)
Rice	R&D	Rotavirus – Diarrhea	University of Tokyo, Japan	Tokuhara et al. (2013)
	R&D	Lactoferrin, lysozyme, serum albumin, transferrin and 13 other pharmaceutical protein combinations	Ventria Bioscience, USA	http://www.aphis.usda.gov/brs/ph_permits.html
	R&D	Human Serum Albumin	Wuhan University, China	He et al. (2011)
Safflower	Phase (I/II) clinical tests finished	Insulin (SBS-1000) – Diabetes	SemBioSys, Inc., Canada	See company website
	R&D, USA	Human proinsulin	SemBioSys, Inc., Canada	See company website
Spinach	Clinical tests (I)	Rabies virus glycoprotein (vaccine)	Fraunhofer Center for Molecular Biotechnology, USA	Yusibov et al. (2002)
Tobacco mosaic virus	Confined field trials, USA	Aprotinin	Kentucky Bioprocessing, LLC	http://www.aphis.usda.gov/brs/ph_permits.html
Tobacco	Confined field trials, USA	Immunoadhesin against the protective antigen of *Bacillus anthracis*, antibody against botulinum neurotoxin (author wording changed ok?), or an immunoadhesin against human major group rhinoviruses in tobacco	Planet Biotechnology Inc., USA	http://www.aphis.usda.gov/brs/ph_permits.html
	Approved in the EU	Mouse Guy's 13 IgG (CaroRx™) (for the prevention of infection	Planet Biotechnology Inc., USA	See company website

(*continued*)

Table 5. Continued

Species	Pipeline	Trait: Molecule produced – Disease treated	Developer	References
		by *Streptococcus mutans* involved in tooth decay)		
	R&D	Antibody Rhinox – Rhinovirus prophylaxy (RhinoRx™)	Planet Biotechnology Inc., USA	See company website
	R&D	Human adenosine deaminase – Severe immunodeficiency	University of East London, UK	Singhabahu et al. (2013)
	R&D	Monoclonal antibodies – Rabies	University of London, UK	Both et al. (2013)
	Clinical tests (I)	Collagen	CollPlant Ltd., Israel	See company website
	Clinical tests (I)	ZMapp™ (against Ebola virus)	Mapp Biopharmaceutical, USA	See company website

resistance to high salinity and flooding, as well as nitrogen use efficiency (NUE) of the plant, to minimize nitrogen fertilizer treatments while maintaining high yields. Rice appears as the species where most of this type of research is conducted, followed by other cereals like wheat, barley and sugarcane (Table 3).

Edible plants with nutritional traits

There is a variety of nutritional traits currently being explored in soybean, corn and canola with direct benefits for consumers. The most advanced products are varieties of soybean with a modified fatty acid profile (high oleic), while varieties of corn and canola are still at the R&D stage. Targeted markets for these products are either population in developed countries for a healthier diet, or with vitamin deficiency developing countries. Nutritional traits are also being introduced into new species, the most famous example and closest to market release being Golden Rice. Interestingly, health benefits to consumers can be found into new species (such as the recently deregulated Innate™ variety of potato; Table 4).

Biotech plants with non-food uses

Molecular farming – therapeutic applications

Biotech plants with therapeutic uses are developing (Williams, 2012), and the European Academies of Science Advisory Council (EASAC) listed biotech plant protein applications in human health (European Academies of Science Advisory Council, 2013), including enzyme replacement therapy, hormone therapy, cytokine therapy, transferrin therapy, monoclonal antibody and a vaccine (Table 5). For further details, see Mangan (2014).

Industrial applications: starch, biofuels, paper and textile industries

Several pathways are being explored in the plant production of molecules for the chemical industry. The production of the metabolite of interest can be induced in plants (e.g. a high content of starch in potatoes or sugar in sugarcane), or biomass production can be altered to more easily produce bioethanol or increased quantities of biodiesel in first-generation biofuels (Table 6). In addition to the plants listed in the table below, RNAi technology is studied at the research stage in corn, cotton and jothropha, respectively for cellulosic bioethanol at Anhui Agricultural University, China (Li et al., 2013), for fiber length at the Centre of Genomics and Bioinformatics, Academy of Sciences of Uzbekistan and at the Uzbekistan Ministry of Agriculture and Water Resources (Abdurakhmonov et al., 2013), and for oil content for biodiesel at the Laboratory of Plant Molecular Biology, The Rockefeller University, New York, USA.

Conclusion

Biotechnology uses are clearly expanding to a wider range of plants to address diverse pressing issues in agriculture for both food and non-food purposes. New crops (outside of commodity crops) and traits (outside of herbicide-tolerance and pest-resistance) are being increasingly developed in developing and emerging countries, often thanks to research consortia with joint public/private research efforts. There are now various breeding techniques that are very effective when used in combination. These are not alternatives to transgenesis, but serve to complement each other. In addition to marker assisted selection, several modern breeding technologies include cisgenesis/intragenesis, RNA interference, novel DNA-editing technologies producing genetically edited organisms (GEOs) which do not have inserted genes from other organisms (site-directed mutagenesis through various techniques, oligonucleotide directed changes and others). To date, new editing tools have not been applied in fruit and vegetable breeding (Kanchiswamy et al., 2014).

Interestingly, the cost and time required to develop new plants considered as genetically modified organisms by regulatory authorities seem to discourage a number of developers, especially in the public sector and small and medium enterprises (Ammann, 2014; Chassy, 2010; Kuntz & Ricroch, 2012), from using classic recombinant DNA technologies, and aforementioned breeding techniques are sometimes preferred (Camacho et al., 2014). There is a continuum in science progress and discoveries, and biotechnologies are not static. Whether these editing technologies will develop further depends on several factors: it will not only depend on how efficient and appropriate they will prove to be in the various plants of interest for mankind, but also on how stringently they will be regulated. If they are regulated

Table 6. Biotech plants with industrial applications.

Species	Pipeline	Trait – Product, industry	Developer	Method	References
Aspen trees	Confined field trials, USA	Phytoremediation in soil and water of volatile organic compounds, e.g. benzene, chloroform, trichloroethylene, toluene	University of Washington, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
Camelina	R&D	Enhanced oil yield and improved growth – Aviation fuels	University of Hong Kong, China, Donald Danforth Plant Science Center, USA	Transgenesis	Zhang et al. (2012), Collins-Silva et al. (2011), Choudhury et al. (2013)
	Confined field trials, USA	Production of wax esters (to be used as biolubricants) and higher oleic acid levels	University of Lincoln, Nebraska, USA	Transgenesis	Nguyen et al. (2013)
Eucalyptus and poplar	R&D, Brazil, China, Israel. Commercialization expected in 2015 in Brazil	Fast growth, bigger trees than average, pulp production for paper	FuturaGene, ArborGen and others	Transgenesis	Sedjo (2011); Mansfield et al.(2012); Wang et al. (2013).
Eucalyptus	R&D	Freeze	Arborgen Inc., USA	Transgenesis	See company website
Miscanthus	R&D, USA	Nitrogen Utilization Efficiency Increase	Ceres, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
Poplar	R&D	Bioethanol production – Industrial and agricultural biotechnology	Bio Base Europe Initiative, EU	Transgenesis	Biobase website
	R&D	Different growth types	Oregon State University, Michigan Technological University, USDA/ARS	Cisgenesis	Han et al. (2011).
Potato	Field trials, UE	High amylopectin	BASF, EU	Intragenesis	Holme et al. (2013)
Switchgrass	R&D, non-regulated by USDA	Drought Tolerance Increased/ AP-Nitrogen Utilization Efficiency Increase	Ceres, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
	R&D, deregulated by USDA	Delayed wilting	Ceres, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
	R&D, deregulated by USDA	Biomass conversion efficiency	Ceres, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
	R&D	PQ-Lignin Levels Decreased	University of Tennessee, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
	R&D	PQ lignin modification	University of North Texas, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
	R&D	Altered Lignin Biosynthesis	Noble Foundation, USA	Transgenesis	http://www.aphis.usda.gov/brs/ph_permits.html
Sugarcane	R&D	Increased sucrose yield neutralizing the fructose 6-phosphate 1-phosphotransferase (PFP)	Universities of Newcastle and Queensland, Australia	Transgenesis	Groenewald & Botha (2008); Patrick et al. (2013)
	R&D	Modification of the chemical structure of lignin	University of Campinas (UNICAMP), Brazil	Transgenesis	Bottcher et al. (2013)
	R&D	Introduction of microbial genes in transgenic sugarcane, producing enzymes degrading cellulose	Syngenta, Switzerland	Transgenesis	Harrison et al. (2011)

like transgenesis (GMOs), whereby the process is regulated rather than the final product, these technologies may not live up to their full potential.

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Declaration of interest

The authors report no declarations of interest.

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