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SPECIAL ISSUE ON GENOME EDITING

# Genome editing for crop improvement: A perspective from India

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## Abstract

Human population is expected to reach to about 10 billion by 2050. Climate change affects crop production, thus posing food security challenges. Conventional breeding alone will not bridge the gap between current level of crop production and expected levels in the decades to come in the food production systems. Rate of genetic gain with time has remained narrow considerably. Biotechnology-enabled crops developed through genome editing will have a part to play in improving crop productivity, meeting food, nutrition security besides catering to regional preferences and fetching valuable foreign exchange. Political, social, economical proposition, scientific will, retailer and consumer acceptance are a must for genome editing (GE) to succeed and add value in the food value chain. This will also help to make agriculture a lucrative profession and attract youth. Therefore, the present review looks into existing regulations governing crops developed using biotechnology in India, institutes involved in genome editing, prospects of new tools developed in this sphere such as DNA-free editing systems, nanotechnology, their applicability in crop improvement efforts, social and future prospects taking cue from recent global developments. This will make GE more appealing to stakeholders and defray any safety concerns.

**Keywords** CRISPR/Cas9 · Nanotechnology · Regulation · Acts · Rules

## Introduction

Crop improvement is an ongoing process for several thousands of years (Voss-Fels *et al.* 2019). Much of the effort in the early years focussed on natural variations, selection from related species and some spontaneous mutations (Huang *et al.* 2016). Later on, artificial hybridisation came into the picture by Fairchild in 1716 (Goulet *et al.* 2017). Then, in 1930, Stadler used X-rays to induce mutation and assisting in a new era of mutagenesis breeding including chemical means (Uaauy *et al.* 2017). Thus, plant breeding has evolved over time accompanying new innovations including precision breeding (Hartung and Schiemann 2014). Molecular breeding includes gene editing and marker-assisted selection (see NAAS 2020). Recently, the term new plant technology (NBTs) is used to include all recent developments in the biotechnology field to improve crops (Lusser *et al.* 2011; Limera *et al.* 2017). Since, ushering the first green revolution was in fact an orchestrated efforts by various stakeholder in the late 1960s (see Swaminathan 2006). Further, our country has achieved self-sufficiency in food and passed legislation for the Right to Food Act, 2013 (Website 2 n.d.), which means greater efforts need to be invested in local food ecosystems and strengthen them in face of biotic and abiotic challenges. To persistently sustain and further increase food production will need additional incorporation of all developed relevant tools using genomics, genome editing (GE), artificial intelligence and deep learning among others (Mahood *et al.* 2020). Parkhi *et al.* (2018) presented early success of GE in India. Genome editing in its elementary form involves allelic variants which are identical to their naturally occurring counterparts (Schmidt *et al.* 2020). For more details on CRISPR/Cas, see excellent reviews recently published by Barman *et al.* (2020) and Wada *et al.* (2020). From economic point of view, genome-edited crops could be far less expensive to develop and more acceptable to the general public than genetically modified GM crops (Lassoued *et al.* 2019a, 2019b). GE results in editing endogenous genes. These results in developing allelic diversity alter endogenous gene activity (Gaj *et al.* 2016). This is similar to random mutagenesis and thus could be a robust alternative to crops developed by mutagenesis

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(Abdallah *et al.* 2015). GE composes of related types of advanced molecular technique which is used for precise modification of target sequence (Gaj *et al.* 2013). The CRISPR/Cas system was initially discovered in 1987 from prokaryotic organism and is fairly common in bacterial and archaeal genomes (Barrangou and Marraffini 2014). Ever since, the practical application of the GE technology to edit any gene was deciphered in 2012 by Charpentier and Doudna in prokaryotic organism (Ding *et al.* 2016) and later by Zhang and Church group in 2013 for eukaryotic cells. There has been tremendous spout of interest in this field (see Lander 2016). Basically, GE takes place by the error prone (non-homologous end joining, NHEJ) or precise (homology directed repair, HDR) both involving DNA break and repair. Later, David Lui found base editor system which is a significant improvement over the NHEJ and HDR repair caused by CRISPR/Cas system, with causing DNA break and no off-target risks. For details in base editing, see Mishra *et al.* (2020). Several other implementations of CRISPR/Cas system are the use of dead Cas9 fusions which can help in activating (VP64, SunTag, SAM associated box; Rees and Liu 2018) or interference (KRAB—Kruppel associated box) and suppression of any genes (Gilbert *et al.* 2013). Mainly two classes of CRISPR/Cas are available for DNA and RNA editing, among which class II-types II and V induces double-stranded breaks (DSB) (see Moon *et al.* 2019). These breaks are repaired by NHEJ (random) or HRD (precise) (Liu *et al.* 2018). For other details of genome editing and their applications, please refer to the Bhattacharya *et al.* (2020).

Many other applications of gene editing technology are being discovered nowadays (Website 1 n.d.).

**Current Status and Regulation of Genome-Edited Crops** A recent report by NAAS (National Academy of Agricultural Sciences, India, July 2020) estimated that genome editing market is a billion dollar industry now. Therefore, there needs to be a clear policy in this field to gain from such advances. DNA edits are classified into three types. SDN-1 is one or few base pair (bp) changes (similar incurred by using base editors; see Mishra *et al.* 2019), SDN-2 is few bp changes (the definition of a few varies greatly, for example, Gao 2018, restricts changes to 20 bp) and SDN-3 is typically long indels or gene replacements (Gao 2018, Draft document on GE, India). Table 1 shows list of institutes in India working on genome editing. The list is not exhaustive but representative. The NAAS 2020 report further noted that mutation falling under SDN-1 and SDN-2 is indistinguishable from those obtained from mutation breeding and should be made available to the farming community in the shortest possible time. In January of 2020, the Department of Biotechnology under Ministry of Science and Technology, Government of India, came up with draft guidelines for public consultation. The "Draft document on Genome Edited Organisms: Regulatory Framework and Guidelines for Risk Assessment" was released (available at ibkp.dbtindia.gov.in>ShowPublicConsulationImg>20200109173448583_Draft_Regulatory_Framework_Genome_Editing9jan2020.pdf). The consultation period ended on 8 Feb 2020, and various stakeholders were invited to submit their comments by then. The basic risk assessment associated with genome-edited crops is in line with "Risk Assessment Framework and Guidelines for the Environmental Risk Assessment of Genetically Engineered Plants 2016" which is available online at geacindia.gov.in. There is a fundamentally distinct difference between genome-edited crops and genetically modified crops in that GE deals with precise editing of endogenous genes similar to what could have been achieved by random mutation causing gene edits by chemical or physical mutagenised crops. GM crops on the other hand contain foreign DNA sequences from related or other organisms which gives distinct phenotype. The GE draft document advocated that risk assessment would be according to the complexity of edits with single base edits and a few base edits falling under group I, deletions in group II and DNA replacement (foreign or synthetic) in group III. The regulatory approval road map differs from group to group. Exception can occur, like singly base pair change conferring herbicide tolerance or weediness related traits will require additional biosafety studies. Data on comprehensive biosafety assessment needs to be submitted to the approval committee including biology, delivery method, molecular basis of edits, group to which edits belong, molecular characterisation, integration pattern of donor DNA or cassette, off-target study, phenotype and biosafety among others (for more information, please refer to the draft guidelines document, 2020). The final guidelines pertaining to genome-edited crops are awaited at the time of writing of this review. Earlier, the definition of biotechnology and genetically modified organisms (GMO) is considered wide and is covered under Rules, 1989, under the provision of EPA, Environment Protection Act of 1986. Broadly, there are six agencies which oversee the development of genetically modified crops. Initially, regulation of genome engineering technologies in India covering the entire spectrum of genetically modified organisms is in accordance with EPA rules 1989 (Rules for manufacture, use of genetically engineered organisms or cells, 1989 (refer to geacindia.gov.in)). The Ministry of Environment, Forest and Climate change is the final authority in close collaboration with State Governments and DBT (Department of Biotechnology). The competent agency composes of (1) RDAC: rDNA Advisory Committee; (2) IBSC: Institutional Biosafety Committee; (3) RCGM: Review Committee on Genetic Manipulation; (4) GEAC: Genetic Engineering Appraisal Committee; (5) SBCC: State Biotechnology Coordination Committee; and (6) DLC: District Level Committee. India is also a signatory to convection on biological diversity including Cartagena and Nagoya

**Table 1.** Representative list of Indian institutions working on crop genome editing

| Sr. no. | Institutions | Specific areas of interest | Source (website) | Publications (if any) |
|---|---|---|---|---|
| 1 | NIPGR (National Institute of Plant Genome Research) | Nutritional improvement of Indian oilseed mustard using CRISPR-Cas9-mediated genome editing | http://www.nipgr.ac.in/home/home.php | Not available |
| | | RICE/Maize-CRISPR/Cas9/Cpf1 based genome editing system to engineer and improve root architecture and stress/nutrient response/abiotic stress | NIPGR website http://www.nipgr.ac.in/home/home.php http://www.nipgr.ac.in/research/dr_asarkar.php http://www.nipgr.ac.in/research/dr_amarpal.php | Farhat *et al.* (2019) Das A *et al.* (2019) |
| | | Biofortification of banana (Collaboration with National Agri-Food Biotechnology Institute, Mohali) | http://www.nipgr.ac.in/research/dr_ashutosh.php | Kaur *et al.* (2020) |
| 2 | Bose Institute, Kolkata | "Developing an optimized toolkit for inducible genome editing and regulation of gene expression in tomato plant: implications in adjusting complex traits via synthetic biology approach" | DBT sponsored project / http://www.jcbose.ac.in/uploads/ADVT/19/p_4.pdf | Not available |
| 3 | Junagadh Agricultural University | (1) Groundnut GE—high oleic acid and low linoleic acid (2) Gene editing in major field crops of Saurashtra—use of CRISPR-CAS9 technology in breeding programme | (1) https://geneticliteracyproject.org/2018/08/03/low-cholesterol-oil-indian-scientists-developing-crispr-gene-edited-groundnut/ (2) http://www.jau.in/attachments/vision2050.pdf | Rajyaguru and Tomar (2020) |
| 4 | ICAR-National Institute for Plant Biotechnology | Targeted editing of potato genome to develop variety-specific true potato seed | https://www.icar.gov.in/nasf/documents/Ongoingproject_VII.pdf | Not available |
| | | CRISPR-Cas9-based genome editing of multiple negative regulators for blast resistance in rice | http://www.nrcpb.res.in/content/%EF%83%BC-crispr-cas9-based-genome-editing-multiple-negative-regulators-blast-resistance-rice | Not available |
| | | Wheat (review article) | | Kumar *et al.* (2019) |
| | | GE-oil seed crops-mustard | http://www.nrcpb.res.in/content/genome-editing-oilseed-brassica-crop-improvement | Not available |
| 5 | IARI—New Delhi (cooperating centres ICGEB, New Delhi NRCPB, New Delhi NRRI, Cuttack IIRR, Hyderabad TNAU, Coimbatore) | Genetic improvement of rice for yield, NUE, WUE, abiotic and biotic stress tolerance through RNA guided Genome editing (CRISPR/Cpf1) Genome edited rice lines of elite mega rice varieties with known/novel alleles of DEP1, CKX2, TB1, SPL14, PP2Cs, DST, miR169a, Os8N3 and eF4g genes which will be useful for direct commercial cultivation or donors in breeding programmes. | https://www.icar.gov.in/nasf/documents/Ongoingproject_VII.pdf | Kumar *et al.* (2020), Gouda *et al.* (2020) |
| | | GE: rice, wheat, soybean | | Thakare *et al.* (2020) |
| 6 | ICGEB—New Delhi (International Centre for Genetic Engineering and Biotechnology) | Redesigning rice crop for improvised grain micronutrient quality using CRISPR-cas9/Cpf1 genome editing Objectives: ¬ To knockout the Fe-sensing genes and iron-binding haemerythrin RING ubiquitin ligases (OsHRZ1, OsHRZ2) for higher accumulation of iron and simultaneously knock out the cadmium transporter gene (OsLCT1) to reduce the translocation of Cd to rice grains (1) Low phytate rice (2) Nutrient use efficiency Herbicide tolerance: (1) AHAS—rice (2) EPSPS/ALS—rice, wheat, maize | (1) https://www.icar.gov.in/nasf/documents/Ongoingproject_VII.pdf (2) https://www.icgeb.org/nutritional-improvement-of-crops/ | Chib *et al.* (2020) Fartyal *et al.* (2018) |
| 7 | NRCPB (National Research Centre on Plant Biotechnology) | Development of haploid inducer line and enhancement of seed meal quality in *Brassica juncea* through CRISPR/Cas mediated genome editing | http://www.indiascienceandtechnology.gov.in/research/development-haploid-inducer-line-and-enhancement-seedmeal-quality-brassica-juncea-through-crisprcas?field_area_id=2356 | Bisht *et al.* (2019) |
| 8 | Rama Devi Women's University | Anthracnose resistance in chilli pepper: CRISPR/Cas9-fused cytidine base editing (CBE) targeting NAC72 | | Mishra *et al.* (2019) |

**Table 1.** (continued)

| Sr. no. | Institutions | Specific areas of interest | Source (website) | Publications (if any) |
|---|---|---|---|---|
| | | | https://www.imedpub.com/conference-abstracts-files/genome-editing-in-chili-pepper-using-a-crisprcas9.pdf | |
| 9 | TNAU | Genome editing for enhancing disease resistance and nutritional properties in rice Coordination with ICAR-NASF | https://tnau.ac.in/cpmb/research-projects/ | Not available |
| | | CRISPR-mediated genome engineering for developing "Thermo-sensitive genic male sterile lines (TGMS)" in rice (Oryza sativa): Tms5 locus | https://tnau.ac.in/cpmb/plant-biotechnology-externally-funded-projects/ | Nagaraj *et al.* (2019) |
| 10 | National Agri-Food Biotechnology Institute, Mohali | GE-β-carotene rich banana—LCYε | https://vigyanprasar.gov.in/isw/crisper_banana_genome_story.html | Kaur *et al.* (2020) |
| | | The research group is focussed on metabolic engineering banana and wheat for nutritional enrichment. Research focusses on the pro-vitamin A (beta-carotene) biofortification of banana. | | |
| 11 | IGIB (CSIR-Institute of Genomics and Integrative Biology—New Delhi) | Mammalian cell research. | https://www.igib.res.in | Achrya *et al.* (2019) |
| | | Generation of inheritable, transgene-free abiotic stress (salinity and drought) tolerant and semi-dwarf indica rice cultivars using new plant breeding approach \| | https://www.igib.res.in/?q=projects | Not available |
| 12 | IIT Delhi | Cancer research-Ayurvedic herb/Covid 19/Bioinformatics-GE | http://beb.iitd.ac.in/ | Not available |
| 13 | ILS Bhubaneswar (Institute of Life Sciences) | Development of seedless Bhimkol (Musa balbisiana, BB gemone) through CRISPR/Cas9 and mutational approaches. (Bimkol- Banana) DBT fuded project-2018 | https://www.ils.res.in/wp-content/uploads/2018/07/advt-11-2018.pdf | Shrestha *et al.* (2019) |
| 14 | NBRI (National Botanical Research Institute) (CSIR-NBRI-NCP) | GE—tomato, cotton, chickpea, rice and *Brassica*<br>Enhanced post-harvest life and nutrition quality in tomato<br>Tomato root architecture modification for enhanced yield<br>Development of determinate/semi-determinate sympodial cotton varieties for synchronized fibre yield and quality<br>Development of rice varieties with low arsenic accumulation in grain<br>Development of high yielding and short duration mustard/rapa variety<br>Genome-editing of miRNAs and associated miRNA peptides for improving drought stress tolerance in chickpea | https://nbri.res.in/genome-editing-of-plants/ https://nbri.res.in/molecular-scientists/dr-praveen-c-verma/ https://nbri.res.in/media/4.5.4-underway.pdf https://www.biotecnika.org/2019/05/csir-nbri-msc-bsc-phd-life-sciences-research-jobs/ | Not available |
| 15 | CSIR-North East Institute of Science & Technology, Assam | Establishing multiplex CRISPR-Cas9 and CRISPR-Cpf1 genome editing systems for abiotic and biotic stress tolerance in tomato (*S. lycopersicum* L.) and rice (*Oryza sativa*) | http://www.rrljorhat.res.in/1261.php | Saikia *et al.* 2020 and Debbarma *et al.* (2019). |
| 16 | Amity Science Technology and Innovation Foundation | Details not available | https://www.amity.edu/astif/ | Jyoti *et al.* (2019) |

protocols and is committed towards biosafety of gene edited crops as well (see, cbt.int, Randhawa *et al.* 2007). This resulted in the introduction of Biotechnology Regulatory Authority of India bill in 2013 but lapsed later on (Ahuja, 2018), which was supposed to provide a single window approval instead of duplication of efforts by various Government Agencies, Department and Ministries.

**Prospects of Genome Editing Technology: the Evolving Landscape** The common form of genome editing involves DNA vectors expressing both Cas9 and Sg RNAs. Crop transformation is another challenge. Here, we present newly developed tools being used in genome editing. Lowe *et al.* (2016) showed that by manipulating morphogenetic regulators like BABYBOOM, WUSCHEL helps in overcoming recalcitrance. Kelliher *et al.* (2019) discovered a one-step haploid editing technology for editing inbreed lines. The authors validated CENH3-based HI system, also known as CRISPR pollen method, by editing VRS-1 LIKE HOMEOBOX PROTEIN and GRAIN WEIGHT 2 gene. Usually, CENH3 works in dicots, whereas MATL and MATRILINEAL (also known as NOT LIKE DAD, PHOPHOLIPASE 1) work in monocots. However, clean technologies, *i.e*. non-vector systems are available today to introduce two components in the plant cell for edits. The newer technologies need more complex technical skill sets and screening is a challenge given the absence of markers and still is in infancy. Other improvements like the use of t-RNA machinery to process sequence and increase pol III transcription. This same strategy is used by Xie *et al.* (2015) to form t-gRNA strings with 20 bp spacer sequence and targeting sequence of interest to be edited. Further, the advantage of construct having PTGS (polycistronic glycine t-RNA) resulted in non-biased editing against standard U3: sssgRNA favouring specific nucleotide at the 5′ end. The advantages of using DNA free or RNP (ribonucleoprotein) are low to no off-target effects and addressing consumer concerns. Other improvements include TREE (transient reporter for editing enrichment) to purify single edited cells in real time (Standage-Beier *et al.* 2019). A simple next generation of hybrid seed technology is developed using CRISPR-edited GMS maintainer by knocking out or inactivating ZMMS26 (*Zea mays* Male Sterile 26 gene) and a DS RED marker. The hemizygous mutated MS 26 gene is sterile and the maintainer line is already labelled by DsRED (red fluorescent protein) and easy to sort (Qi *et al.* 2020). Maher *et al.* (2019) showed that a combination of developmental regulators (like WUS, ipt, STM- SHOOT MERISTEMLESS) is sufficient to induce shoot formation thus circumventing tissue culture methods. Then, a combination of DRs and gene editing reagents created edited shoots. The strategy was used in dicot crops like *Arabidopsis*, tobacco, potato, grape and tomato. Consumer resentment in some areas of the world resulted in low to no penetration of GM technology. Therefore, emphasis on non-vector, non-intregration of foreign DNA in the primary or initial stage of GE development would be beneficial. Here, we look into available technique to undertake DNA-free editing.

**DNA-Free Editing and Nanotechnology** For quite some time, nanotechnology-based material is being used in agrochemical formulations which are aimed at plant nutrition, crop protection, abiotic and biotic stress resistance (Sanzari *et al.* 2019). These nano-materials are particles ranging from 1 to 100 nm and are chemically or physically linked. Thus, nanoparticles could be used in the delivery of Cas9 protein-gRNA or mRNA (Cas9)-gRNA load inside the cell for genome editing (Jeevanandam *et al.* 2018). The basic requirement of nanoparticle in genome editing is that the material must be permeable, can accumulate the cargo and should be able to retain the payload for extended period of time (Blanco *et al.* 2016). However, the uptake of nanoparticle (NP) is very unpredictable (Sanzari *et al.* 2019). Zhao *et al.* (2017) reported magnetofection technology in cotton involving pollen, magnetic particle and electromagnetic field. This technique has the advantage of being independent of the tissue culture protocol and genotype. Commonly used gene-coating material and cargo include polymers (polyethylenimine, phenyl boronic acid or cell-specific aptamer, a combination thereof including functionalised grapheme oxidase), nanoclay, liposome (lipid like nanoparticles), gold nanoparticles, nanoscrew and coreshell. Cargo delivery involves *Agrobacterium*-mediated transformation including agrolistics, adenovirus and lentivirus (see Wei *et al.* 2020 and Nguyen *et al.* 2020 for details). The emergence of delivery methods of nanotechnology assisted delivery is considered a breakthrough technology. While commonly used Cas9 causes double-stranded breaks in the 4th base pair of PAM sequence, which is then repaired by NHEJ or HDR (Wu *et al.* 2014). However, HDR is active in dividing cells and therefore rare (Devakota, 2018). The Cas9 protein is about 160 kDa (Mout *et al.* 2017) which when co-delivered with RNPs for DNA-free genome editing poses a challenge due to its shear size, and in such situation, Cas9 mRNA may help as well as limit the off-target editing due to its limited stability in the plant cell. Further, the Cas9 protein is positively charged and poses challenge for encapsulation in nanoparticles. The encapsulation of lipid in the outershell of the nanoparticle improves its stability and better protects the DNA and protein from degradation by cellular nucleases and protease. Also, see Chakraborty and Vora (2020). The supremacy of NHEJ over HDR has limited application in plants owing to concerns about possible off-targets (Anshari *et al.* 2020). With the discovery of base editing, this can transform one base to the other without breaks and foregoing the need for a repair template. Base editing can be categorised into broadly four generation. Initially, BE1 was invented with fusing cytidine deaminase, BE2 was an improvement over BE1 in that it

incorporates uracil DNA glycosylase inhibitor (UGI), BE3 is dCas9 replaced by nCas9 (nickase) while BE4 has rat/lamprey/human APOBEC1 with nickase (for detailed review, Bharat *et al.* 2019). NHEJ is preferred approach where loss of function of a gene is desired (Monsur *et al.* 2020). Nowadays, both adenine and cytidine base editors are available which facilitates base conversion (by deamination) in a narrow window (see Nishida *et al.* 2016). Like DNA, RNA editing is done *via* REPAIR (RNA editing for programmable A to I then to G) and RESCUE (RNA editing for specific C to U exchange). However, to reduce the possible off-targets, guanine mismatches is incorporated in gRNA design (Bharat *et al.* 2020; Monsur *et al.* 2020). Another strategy is to introduce uracil DNA glycosylase inhibitor (UGI) into base editor (Komor *et al.* 2016). In another recent development, Liu *et al.* (2020) invented a very fast CRISPR (vfCRISPR) on demand system targeted on living cells with the goal of inducing DSBs with a high resolution. The vfCRISPR system helps in targeting single allele and eliminating off-target activity. Primarily, the vf system works on a caged RNA strategy, which prevents Cas9 from cleaving the DNA strand until activated by light, thus facilitating precise breaks. Therefore, SNP variation in the crop genome is linked to variation in traits and this knowledge can be successfully exploited to create designer crops through base editing (Li *et al.* 2018). Traditionally, exploiting such SNPs variation through conventional breeding would take several years and CRISPR/Cas can achieve the same in less time. Wang *et al.* (2020) developed a series of APOBEC-Cas9 fusion induced deletion system (AFIDS) which when combined with human APOBEC3A (A3A) with uracil glucosidase and a AP lyase results in a robust editing system. This results in predictable, multinucleotide targeted deletions within the protospacer window in rice and wheat. The SWEET14 (Sugar Will Eventually Be Exported Transporters 14) deletion mutants obtained in this study had significantly smaller blight lesions than 1 or 2 bp INDELS, while in wheat, the same group found three predictable mutants of miR396 that averted formation of mature RNA.

**Social Impact** No formal study depicting the perception of genome-edited crops on consumer sentiments was conducted in India till date. However, there are studies available across globe on the social and consumer perception. Earlier, Wehlan and Lema (2017) noted that GE caused dilemma in the minds of policyholders in issues relating to regulatory and safety compliance. Furthermore, due to the absence of exogenous DNA in the final product despite using biotechnological intervention is an additional challenge for enforcing regulatory requirements. Thus, such decision-making will lead to the use of many social science tools to arrive at logical conclusion. Lassoud *et al.* (2019) published the results of a comprehensive study to estimate the cost and time involved for commercialising gene edited crops when they are regulated just like GMO vs. when GE crops are treated at par with conventional crops. The survey results shows that in the first scenario (like GM), this will cost US$24.5 M and take 14 yr. In the second scenario (non-regulated), it will fetch US$10M and can be completed in just 5 yr. There was no significant difference between products developed in North America and Europe based on existing regulations. In another survey conducted in Costa Rica, it was found that though consumers have low knowledge about genome editing but are ready to accept the product owing to high perceived produce, quality and disease resistance (Gatica-Arias *et al.* (2019). While in a study by Kato-Nitta *et al.* (2019) in Japan, it was found that consumers were more positively inclined toward genome-edited crops than genetically modified (GM) crops. Also, the perception widened when choice was between conventionally breed crops and GM crops. Thus, GE crops seem to have a positive edge. A study in Germany conducted on the same topic of GE found that stakeholders in value chain of wheat perceived celiac-safe and fungal tolerant trait positively. Whelan *et al.* (2020) observed that GE resulted in diverse range of products options and wide participation of many start-ups and SMEs. This will result in faster adoption of technology, *i.e.* faster bench to market. Website 5 n.d.gives detailed up-to-date information on GE in human and agriculture space.

**Future of GE Crops** As Nucciu *et al.* (2018) discussed, the efforts undertaken over the past 20 yr in developing drought tolerant crops emphasising that regulations and the fact that food security advantages can accrue from modern biotechnological interventions including gene editing. GE may be seen positively by general public than in the past. Some notable products in the past developed worldwide through GM approaches include Monsanto's DROUGHTGUARD (Website 2 n.d.), Corteva's AQUAMAX (Website 3 n.d.) and NX1-4T sugarcane (Website 4 n.d.). A comprehensive list of global pipeline of biotech crops is reviewed by Parisi *et al.* (2016), from arable to speciality crops. Xu *et al.* (2019) noted that there must be continuous discussion between developers, breeders and consumers in the GE product development space. Further, the type of delivery agents used to regenerating mutants should be emphasised. Therefore, it is expected that the new plant breeding technologies could fill the unfulfilled promise in accelerating crop improvement efforts. Further, the cost of developing gene editing crops is less than that of GM crops, owing largely due to no to low regulatory cost depending on designated marketplace.

## Conclusion

Farmers need new improved varieties to increase crop yield and feed the Nation and the Globe. Plant breeders are

equipped with various tools including marker assisted molecular breeding and genomics to name a few for the purpose. Competition between breeding community is intense given the globalisation of the world economy and warrants adoption of new technique to achieve the desirable end results (please see Zimny *et al.* 2019). Genome editing has been here for the last decade or so, particularly targeted oligonucleotide mutagenesis, meganucleases, zinc fingers and TALENS. However, due to their complexity, it did not result in any new crop product in India though many examples exist outside the country. Particular interest in this field is amplified by the recent discovery of CRISPR/Cas9 GE system. CRISPR/Cas9 is simple, versatile, low cost and stood to democratise the field of GE. In the absence of a clear road map, fresh investments by various parties are unlikely, especially from the past GM experience. India has come up with a draft regulation for genome editing. Further, other interested parties like ICAR (Indian Council of Agriculture Research) and NAAS (National Academy of Agricultural Science) have shown positive intention to make the road map simple (NAAS 2020). SDN-1 and 2 edits are likely to be less regulated, barring few traits like herbicide tolerance and can show up in farmer's field hopefully soon. SDN-3 gene replacements are likely to be treated at par with GM. Food security is paramount for the world's second most populous country. It is hoped that GE tool kit will be included in the plant breeding strategies sooner than later.

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