AUDA-NEPAD
AFRICAN UNION DEVELOPMENT AGENCY

Policy Framework for Applications of Genome Editing in African Agriculture

AUDA-NEPAD AU High Level Panel on Emerging Technologies (APET) Technology Report

This policy document is the product of the African Union High Level Panel on Emerging Technologies (APET). It is part of a larger effort by the African Union Development Agency (AUDA-NEPAD) to promote knowledge and learning, share ideas, provide open access to its research, and contribute to development of policies within AU Member States. The papers featured in the Policy Framework for Applications of Genome Editing in African Agriculture are considered to have a bearing on the mission of AUDA-NEPAD, and its strategic objectives, as aligned to the AU Agenda 2063, which is a Pan African Vision of an integrated, prosperous, and peaceful Africa, driven by its own citizens, representing a dynamic force in the international arena.

Citation: AUDA-NEPAD APET Genome Editing Policy Framework, September 2022

230 15th Road, Randjespark, Midrand, Johannesburg, 1685,
South Africa

+27 11 256 3600
www.nepad.org

Twitter @Nepad_agency
#TheAfricaWeWant

ISBN: 978-1-7764306-5-9

Jan 2023

TABLE OF CONTENTS

TABLE OF FIGURES

LIST OF TABLES

LIST OF ANNEXURES

ACRONYMS

ABBC	Africa Biennial Biosciences Communication
AI	Artificial Insemination
APET	African Union High-Level Panel for Emerging Technologies
AU	African Union
BSV	Brown Streak Virus
CEN–SAD	Community of Sahel–Saharan States
CFTS	Confined Field Trials
COMESA	Common Market for East and Southern Africa
CPB	Cartagena Protocol on Biosafety
CRISPR/CAS9	Clustered Regularly Interspaced Short Palindromic Repeats/CRISPR-associated protein 9
DNA	Deoxyribonucleic Acid
EAC	East African Community
ECCAS	Economic Community for Central African States
ECOWAS	Economic Community of West African States
GABA	γ-Aminobutyric Acid
GDP	Gross Domestic Product
GED	Genome editing
GMOS	Genetically Modified Organisms
HDR	Homology Directed Repair
HE	Homing Endonucleases
HOSG	Heads of States and Governments
IGAD	Intergovernmental Authority on Development
IITA	International Institute of Tropical Agriculture
IITA	International Institute of Tropical Agriculture
ILRI	International Livestock Research Institute
IPLCS	Indigenous People and Local Communities
KEPHIS	Kenya Plant Health Inspectorate Service
LMO	Living Modified Organism
MSTN	Myostatin
NBMA	Nigeria's National Biosafety Management Agency
NBTS	New Breeding Techniques
NHEJ	Non-Homologous End Joining
ODM	Oligonucleotide-Directed Mutagenesis
PS	Pluripotent Stem
R&D	Research and Development
RECS	Regional Economic Communities
SADC	Southern African Development Community
SDN	Site-Directed Nuclease Technology
SNCT	Somatic Nuclear Cell Transfer Cells
STI	Science, Technology, and Innovation
STISA-2024	Science, Technology, and Innovation Strategy for Africa
TALENS	Transcription Activator Like Effector Nucleases
UMA	Arab Maghreb Union
USDA	United States Department of Agriculture
ZFNS	Zinc-Finger Nucleases

ABOUT THE AU AND AUDA-NEPAD

THE AFRICAN UNION (AU)

The African Union (AU) is a continental body consisting of all 55 African countries. It was established on 26th May 2001 and launched on 9th July 2002 in with the aim of replacing the (OAU). The most important decisions of the AU are made by a semi-annual meeting of the Heads of State and Government of its member states. The AU's secretariat is based in Addis Ababa, Ethiopia.

The AU was established following the 9th of September 1999 Sirte Declaration of the Heads of State and Governments of the Organisation of the African Unity (OAU). The AU is based on a common vision of a united and strong Africa and on the need to build a partnership between governments and all segments of civil society, in particular, women, the youth and the private sector, in order to strengthen solidarity and cohesion amongst the peoples of Africa. As a continental organization, it focuses on the promotion of peace, security, and stability. The development work of the AU is guided by the AU Agenda 2063, which is a 50-year plan to harness Africa's comparative advantage to deliver on the vision of "The Africa We Want".

THE AFRICAN UNION DEVELOPMENT AGENCY - NEPAD (AUDA-NEPAD)

The NEPAD Planning and Coordinating Agency (NEPAD Agency), following a decision at the 31st Ordinary Session of the Assembly of AU Heads of State and Government in July 2018, transformed into the first development agency of the AU, the African Union Development Agency – NEPAD (AUDA-NEPAD), with a strengthened mandate to fast-track the realization of Agenda 2063.

AUDA-NEPAD is a strategic framework for pan-African socio-economic development. AUDA-NEPAD is spearheaded by African leaders to address critical challenges facing the continent, including poverty, development, and Africa's international marginalization. AUDA-NEPAD provides unique opportunities for African countries to take full control of their development agendas, to work more closely together and to cooperate more effectively with international partners.
The AUDA-NEPAD delivers its services through four (4) broad and interrelated functions, namely: a) Foresight and Technical Advisory services: b) Brokering Technical and investment financing Partnerships; c) Executing Flagship Projects; and d) Monitoring, Learning and Reporting.

THE AFRICAN UNION HIGH LEVEL PANEL ON EMERGING TECHNOLOGIES (APET)

The first Specialized Technical Committee on Education, Science, and Technology (STC-EST I) requested the AU Commission and AUDA-NEPAD to advise Member States and RECs on matters of technology prospecting, including regulatory and ethical requirements that need to be put in place in order for the continent to benefit from emerging technologies. The Ministers further directed the NEPAD Agency to establish a system for obtaining expert contribution on the matters of technology development, acquisition, and deployment for socio-economic development.

The Chairperson of the African Union Commission (AUC), HE Dr Nkosazana Dlamini Zuma, thus appointed ten (10) eminent experts from various fields to serve on the African Union High Level African Panel on Emerging Technologies (APET) in December 2016. The High Level-APET has been constituted in recognition of the need to harness both existing and emerging technologies for the economic development of Africa. The appointed ten (10) eminent experts are drawn from diverse professional backgrounds and are critical in terms of providing evidence-based analyses and recommendations that should inform policy direction at the continental, regional and national level on the utilization of existing and emerging technologies.

The High Level-APET consists of eleven leading experts in various fields, representing both gender and geographical demographics and is Chaired by Prof. Yaye Kène-Gassama Dia, Professor in Plant Biotechnology at the University Cheikh Anta Diop de Dakar and Vice-Chair of the board of the National Academy of Science and Technology of Senegal and Chair of the basic, applied sciences and innovation section. She is a former Minister of Science and Technology of Senegal, and acting on behalf of the Senegal Government, as chair of AMCOST II (African Ministerial Council on Science and Technology). Other members of the panel include: Prof. Roseanne Diab, former Executive Officer of the Academy of Science of South Africa (ASSAf) and Emeritus Professor in the School of Environmental Sciences, University of KwaZulu-Natal; Professor Berhanu Abegaz, former Executive Director of the African Academy of Sciences; Professor Francine Ntoumi, Director of the Foundation Congolaise pour la Recherche Médicale and Senior Lecturer at University M. Ngouabi, Congo-Brazzaville; and Professor Abdallah Daar, Emeritus Professor of Clinical Public Health and Global Health at the Dalla Lana School of Public Health, University of Toronto, with a cross-appointment in the Department of Surgery. Dr Rachel Chikwamba, Council for Scientific and Industrial Development (CSIR) Group Executive: Advanced Chemistry and Life Sciences; Prof. Dr Shireen Assem, Vice-President for Research at the Agricultural Research Center (ARC) in Egypt; and Prof. Karim Maredia, Professor and Director of the World Technology Access Programme (WorldTAP) in the College of Agriculture and Natural Resources at Michigan State University, East Lansing, Michigan, USA. Dr William Wasswa, Senior Lecturer at Mbarara University of Science and Technology at the Department of Biomedical Sciences and Engineering and Head of the Department of Biomedical Sciences and Engineering and leads the Advanced Medical Imaging and Artificial Intelligence Laboratory in the department. Dr Wasswa is an AfyaBora Global Health Leader Postdoctoral Fellow, an innovator and recipient of the UK Royal Academy of Engineering Africa Prize for Innovation 2020. Prof Abubakar Sani Sambo, Usmanu Dan Fodiyo University, Sokoto, Nigeria, Professor of Mechanical Engineering who has worked in various capacities spanning several years, including being the Vice-Chancellor of a University of Technology for many years. He was also a Special Adviser to the President of Nigeria on Energy, Vice Chairman of the World Energy Council for Africa and former Director-General for the Energy Commission of Nigeria for 8 years. He is an Officer of the Order of the Niger, OON.

Apart from offering advice to the AU and its Member States on harnessing emerging technologies for economic development, the High Level-APET shall also make recommendations on the nature of regional institutional arrangements that are required to promote and sustain common regulatory approaches to the application and use of such technologies and propose a strategy and policy on emerging technologies in Africa.

AUTHORS (APET)

Chairperson
Yaye Kène Gassama, Université Cheikh Anta Diop de Dakar, Senegal

Panel Members
Abdallah Daar, University of Toronto, Canada
Berhanu M. Abegaz, University of Johannesburg, South Africa
Francine Ntoumi, Fondation Congolaise pour la Recherche Médicale, Congo-Brazzaville
Karim Maredia, Michigan State University (MSU), East Lansing, Michigan, USA
Rachel Chikwamba, Council for Scientific and Industrial Research (CSIR), South Africa
Roseanne Diab, University of KwaZulu Natal, South Africa
Shireen Assem, Agricultural Research Center (ARC), Egypt
Abubakar Sani Sambo, Usmanu Dan Fodiyo University, Sokoto, Nigeria
William Wasswa, Mbarara University of Science and Technology, Uganda

Secretariat
Aggrey Ambali, Technical Advisor, AUDA-NEPAD
Justina Dugbazah (Coordinator) – AUDA-NEPAD
Barbara Glover - AUDA-NEPAD
Chifundo Kungade - AUDA-NEPAD
Nonhlanhla Dhlamini – AUDA-NEPAD

Research and Drafting Team
Samuel Timpo (AUDA-NEPAD), Silas Obukosia (AUDA-NEPAD), Modupe Adeyemo (AUDA-NEPAD), Sunday Akile (AUDA-NEPAD), Aggrey Ambali (AUDA-NEPAD), Jeremy Ouedraogo (AUDA-NEPAD), Justina Dugbazah (AUDA-NEPAD), Barbara Glover (AUDA-NEPAD), Florence Nazare (AUDA-NEPAD), Peter Dotse Junior (University of Ghana), Bhekani Mbuli (University of Johannesburg).

External Reviewers: These individuals provided comments on the draft policy framework.
Hennie Groenewald (Biosafety South Africa, Cape Town, South Africa), Martin Lema (National University of Quilmes, Bernal, Argentina), Muffy Koch (Simplot Plant Sciences Boise, Idaho, USA), and Morven McLean, (Bill & Melinda Gates Agricultural Innovations [Gates Ag One], Seattle, Washington, USA).

Acknowledgements
We thank the various experts identified by the African Union High-Level Panel, who made contributions and comments on earlier versions of this report.

EXECUTIVE SUMMARY

The African Union (AU) Agenda 2063 aspires for a prosperous Africa based on inclusive growth and sustainable development in a way that recognises that this would entail investments in science, research, innovation, and emerging technologies. The African Union further notes that the science of biotechnology is advancing rapidly, and new tools and technologies are emerging worldwide that offer tremendous opportunities for enhancing agricultural productivity within AU Member States. Genome editing is one such recent advancement with various bio-economic applications in sectors such as agriculture, industry, health, and environment. Thus, exploring the potential benefits of genome editing as a tool for socio-economic development can align with the priority areas of STISA-2024. Currently, AU Member States are debating and considering safety and regulations on the utilisation of emerging tools on genome editing for their crops and animal improvement programmes. These considerations are also focused on plants, animals, fisheries, forestry, microorganisms, and pharmaceutical improvement programmes. African countries have realised the need for coherent approaches to the assessment of the genome editing techniques.

During a consultative meeting in June 2019 at the African Union Headquarters, Addis Ababa, Ethiopia, African Union Member States requested knowledge-based advisory support and technical advice from the African Union High-Level Panel for Emerging Technologies (APET). This support and advice would enable African countries to benchmark their genome editing policies, regulatory, and investment frameworks. To this end, this report provides an overview of genome editing as a developmental tool. The report further highlights the potential applications of genome editing in various socio-economic sectors to address lingering challenges in Africa, especially the limited agricultural productivity, benchmarked on the current state and potential applications of the techniques around the globe. Furthermore, the report discusses possible strategic approaches for regulation of genome editing while presenting the global regulatory landscape and proffering considerations for Africa. Highlighted in this report are current achievements and future opportunities for research, development, and innovation within the genome editing landscape. In addition, this report also outlines risk-benefit analyses and considerations for decision-making. The report concludes with overarching considerations for safely harnessing applications of genome editing in an evolving bioeconomy.

Genome editing is being utilized in healthcare, agriculture, forestry, industrial biotechnology, and other bioeconomy sectors. It is more accessible than genetic engineering (transgenesis), with lower relative costs, ease of use and a shortened timeframe for application. However, the development and application of emerging technologies or tools such as genome editing should consider maximising the benefits while reducing potential adverse impacts. To achieve this, critical capabilities should be built and/or strengthened for research and development, commercialisation, trade, and consumer protection, informed by workable and appropriate policy, ethical, and regulatory frameworks.

In adopting a science-based approach towards safely harnessing the potential of genome editing in alignment with the AU Agenda 2063, APET urges African Union Member States to:

1. Recognise genome editing as a rapidly developing suite of tools that enable cost- and time-effective targeted and precise alterations of the genome and thus opening new applications and possibilities for bioeconomic activities in healthcare, agriculture, industrial biotechnologies, and other bioeconomy-related sectors.

2. Strengthen the capacity of Competent National Authorities, scientific/technical reviewers, and decision-makers in assessing the potential risks and benefits of genome editing and in making informed decisions. This can help governments adopt policies that can allow for genome edited products to be assessed for their similarity to

products that have been developed with conventional breeding methods or that are already occurring in nature.

3. As parties to the Cartagena Protocol on Biosafety, recognise the utility of the precautionary approach during new technological uptake and within the context of scientifically sound, case-by-case risk assessments, appropriate risk management measures, and decision-making processes that weigh potential risks against benefits. This includes the perspectives of indigenous people and local communities (IPLCs), as may be appropriate. Assess products of genome editing on a case-by-case basis to determine whether they fall under the mandate of a country or a regional Biosafety Regulatory Framework or be excluded from such biosafety regulatory framework. This can also enable that these products be subjected to other existing regulatory frameworks for conventionally developed products and technologies.

4. Advance policy decisions about genome edited products that are informed by science.

5. Take steps to address the legal and policy uncertainties regarding the preferred regulatory approach for genome editing and derived products. This can be ascertained by developing national guidelines to indicate whether genome editing products will be regulated as under their Biosafety Acts or otherwise.

6. Provide clear guidelines on which genome edited products should be excluded from the scope of their National Biosafety laws. This can be accomplished by adopting implementing regulations and guidelines that provide a clear mechanism for pre-submission consultation processes to determine the most appropriate, science-based and risk proportionate regulatory approach to use for genome edited products. Furthermore, this review should allow for case-by-case assessments of each product based on the presence of a foreign genetic material in the final product as a criterion for a product-based exclusion.

7. Ensure that any regulatory approach that requires that a genome edited product be subject to existing risk assessment and decision-making process as stipulated in their Biosafety Laws and implementing instruments would only apply to situations where the said genome edited product is proven to contain foreign genetic material in the final product.

8. Further ensure, in accordance with their biosafety laws, that only genome edited organism with genetic changes resulting in the stable and permanent integration of a foreign genetic material from outside the orgasm's gene pool are regulated under the country Biosafety Acts, Regulations and supporting Guidelines during development, testing, release into the environment and releases of commercial products.

9. Exclude from their biosafety laws and implementing instruments, genome edited organisms with genetic changes that are achievable through traditional/ conventional breeding or that occur in nature, and which do not contain foreign genetic material from outside the organisms' gene pool in the final product. Such organisms should be regulated under existing laws, regulations and guidelines governing the evaluation and release of conventionally bred organisms and products derived from them.

10. Facilitate clear communication and stakeholder engagement to improve public understanding of, and garner stakeholder support for safely harnessing applications of genome editing.

As a continent, Africa should note that regulatory requirements commensurate with the level of risk are essential if genome editing is to become a tool for African scientists for the development of African economies, improved quality of life for African citizens, and improved cooperation with other regions of the world.

1 INTRODUCTION

1.1 BACKGROUND

Africa's economy is primarily driven by agriculture, trade (net exports), industry, and human resources. The African economy has exhibited a stable growth rate of about 3.4% for 2018 and 2019. The economy grew by 3.9% and 4.1% in 2020 and 2021, respectively (ADB, 2020). The economic outlook for 2022 and 2023 is envisaged to range between 3.6% and 3.8%. Notably, Africa's economic growth is significantly impacted by agriculture that encompasses crop, livestock, and fisheries production. These agricultural activities have demonstrated massive social and economic footprint with over 60% of the sub-Saharan Africa's population being smallholder farmers. As such, approximately 23% of Africa's Gross Domestic Product (GDP) is generated from agriculture (Goedde et al, 2019). Thus, agriculture remains a significant driver for Africa's socio-economic transformation and development.

Regrettably, Africa's full agricultural potential is not yet realised, and the continent remains the most food insecure region on the globe. The deteriorating food security situation is further worsened by the negative effects of climate change on agricultural productivity. This is accompanied by the degradation of natural resources including the unsustainable utilisation of land, water, and energy, and compounded by rapid population growth (AFSLD, 2019). Consequently, food and nutrition security, healthcare and environmental sustainability are currently among the continent's biggest challenges.

The AU seeks to address these challenges through the implementation of its Agenda 2063. The AU's Agenda 2063 encapsulates a vision of an integrated, prosperous, and peaceful Africa. Thus, science, innovation, and emerging technologies can transform and enhance Africa's agricultural productivity thereby contributing significantly to this vision of the Africa we want. Achieving this will however require deliberate efforts to formulate science-based, environmentally friendly, sustainable, risk proportionate and pragmatic policies.

In addition to such enabling policies, there is also a need for investments in research and development aimed at applying science, technology, and innovation to enhancing productivity (in relevant sectors) and efforts to drive production, industrialisation and value addition towards improved healthcare delivery, and food and nutrition security. This would also entail establishing measures to promote sustainable and natural resource management, biodiversity conservation, appropriate access to genetic resources, transfer of relevant technologies, sustainable consumption and production patterns, climate resilience, and renewable energy, among others.

1.2 THE AFRICAN UNION SCIENCE, TECHNOLOGY, AND INNOVATION STRATEGY FOR AFRICA (STISA-2024)

The AU Science, Technology, and Innovation Strategy for Africa (STISA-2024) is one of the continental strategies developed to respond to the demands of science, technology, and innovation (STI). STISA-2024 is aimed at positively impacting socio-economic development across critical economic sectors such as agriculture, energy, environment, education, manufacturing, food production, and healthcare. Among other things, STISA-2024 is aimed at the eradication of hunger and achieving food security through an innovation-led, knowledge-based economy. As such, this is being pursued by enhancing STI readiness in Africa in terms of infrastructure, professional and technical competence, and entrepreneurial capacity as well as enabling science-guided regulatory frameworks. This is further accomplished by implementing specific policies and programmes in science, technology and innovation that are addressing societal needs in a holistic and sustainable manner.

The African Union's Agenda 2063 is the continent's blueprint for sustainable development and economic growth. The AU's Agenda 2063 notes that sustained investment in new technologies

and continuous innovation in key sectors of the economy remain a requisite for sustainable growth, competitiveness, and economic transformation. Thus, STISA-2024 remains a key strategy in attaining the AU Vision of the "Africa We Want". This is expected to contribute by meeting the knowledge, technology, and innovation demands in various AU economic and social sector development frameworks. This implies that the AU leadership, through this strategy, considers science, technology, and innovation as development tools necessary for socio-economic transformation on the continent.

1.3 RATIONALE FOR A POLICY FRAMEWORK ON GENOME EDITING IN AGRICULTURE

The AU's Agenda 2063 recognises STI as multi-functional tools and an enabler for achieving continental development goals. Further to this, the AU's Agenda 2063 emphasises that Africa's sustainable growth, competitiveness, and economic transformation require continuous investment in emerging technologies and constant innovation in economic areas of agriculture, clean energy, education, and health. In agriculture, innovation in plant and animal breeding have the potential to advance global agricultural sustainability goals and transform African societies. Such innovation comes in the form of new breeding techniques (NBTs), which include Site-Directed Nucleases (e.g., genome editing), Oligonucleotide-Directed Mutagenesis, RNA-Directed DNA-Methylation, Cisgenesis, Intragenesis, Grafting using GM plants, Reverse breeding, Agroinoculation, Synthetic Genomic, and Agroinfiltration. This policy document focuses on genome editing and does not provide a detailed treatment of the various NBTs.

Genome editing (GEd) is a relatively recent development in the field of molecular biology and is increasingly assuming an important space in the technological and regulatory discourse as it offers significant potential for sustainable agricultural development and food security. GEd is also a rapidly developing suite of tools that has enabled targeted and precise alteration of the genome with a high degree of specificity, opening new applications for agricultural advancements. GEd is a more cost-effective precision breeding tool for uptake by AU Member States and can contribute to helping the continent leapfrog in plant and animal breeding efforts. This is due to the relatively low R&D cost needed to upscale this suite of tools, the ease of use, and shortened timeframe for product developments in key sectors especially in agriculture and related fields.

Regulations, whether for conventional or biotechnological products, are intended to ensure that products released into the environment or placed on the market are considered safe for humans, animals, and the environment. As such, even though all countries seek to promulgate regulatory approaches and processes to protect the common good of human, animal and environmental safety, regulatory details can differ across countries. While regulations differ among countries and regions, there is general agreement in each regulatory regime as to what products and processes are covered (Entine et al, 2021).

Enabling policies that ensure the benefits of genome editing techniques are safely harnessed in Africa will therefore contribute to the realisation of the goals of AU Agenda 2063 and ensure sustainable socio-economic development in the continent. Consequently, AU Member States in noting the need for a coherent approach to the assessment of these techniques, requested for knowledge-based advisory support and technical advice from the African Union High Level Panel for Emerging Technologies (APET). APET notes that the great potential of GEd can be hindered by policy and institutional factors if they do not make for an enabling and efficient environment for technology regulation and access. Thus, APET is recommending that AU Member States should establish enabling regulatory systems that would not only support R&D and innovation but would also assure technology access, commercialisation, and distribution to end-users across the continent.

1.4 SCOPE OF THIS POLICY FRAMEWORK

This policy framework is limited to applications of genome editing in agriculture and excludes applications in human health and other fields.

2 GENOME EDITING AS A DEVELOPMENT TOOL

2.1 INTRODUCTION

A key mandate of the African Union Commission and African Union Development Agency (AUDA-NEPAD) is to provide policy and technical advisory services to Member States to enable them to safely harness modern and emerging technologies or tools such as genome editing.

Genome editing refers to techniques in which the deoxyribonucleic acid (DNA) is inserted, modified, replaced, or deleted in the genome of a living organism at predetermined locations. Gene editing is an equivalent term to genome editing and may be used interchangeably. Whichever terminology is used, it is important to note that the effects of these techniques are very focused (i.e., gene-focused). However, they are not limited to effects on only individual genes as applications of genome editing can simultaneously target multiple genes. The technology can also have genome wide impacts that may still be analogous to conventional breeding or natural outcomes.

2.2 THE SCIENCE OF GENOME EDITING

Genome editing was first demonstrated in 1996, when a protein domain called “Zinc fingers” was coupled with a Fok 1 endonuclease domain. The two combined domains could cut the DNA at a specific site. This process was termed “Site-specific nuclease (SSN) and the two coupled domains were called Zinc Finger Nucleases (ZFNs) (Kim et al 1996). Since then, three other site-specific nucleases (SSNs) have been engineered based on three different enzymes. These were referred to as meganucleases, also called homing endonucleases (HE), transcription activator-like effector nucleases (TALENS), and CRISPR/Cas system nucleases. A broader term called the Site-Directed Nucleases (SDNs) is now more often used to define applications of the four enzyme families. Even though each nuclease has a different mode of action, they basically do two things, i.e., to recognise and bind to a specific DNA sequence and then cleave the DNA to create site-specific DNA double strand breaks (DSBs). The polypeptide sequence of the SDN that recognises, and binds to the DNA, is called the DNA binding domain. The sequence that cleaves DNA is called DNA cleaving domain.

After the DNA DSB is created at a specific site, the cell uses its natural endogenous DNA repair mechanisms involving DNA polymerase to repair the DSB. Two natural DNA repair mechanisms can be used. These are the homology directed repair (HDR) and non-homologous end joining (NHEJ). During the repair process, changes in the DNA sequence can occur at the repair site. This can result into deletions, insertions, duplications, or inversions. This change in DNA sequence is referred to as genome editing.

Scientists have programmed these SDNs to induce genetic changes of economic importance in organisms such as plants/crops, animals, fisheries, forestry, and microorganisms. Furthermore, there is ongoing research to use SDNs to treat human diseases caused by genetic defects such as sickle cell anaemia. Unlike early genetic engineering techniques which randomly inserted foreign genetic material into a host genome, genome editing targets the mutations or other genetic changes to site specific DNA locations.

Genome editing can also be achieved through other processes for example, using a process called Oligonucleotide-Directed Mutagenesis (ODM). It has been observed that when a short single strand DNA sequence, called an oligonucleotide is complementary to a specific target DNA sequence in a genome, except for one or two nucleotides, mutation is introduced into a host cell. The host organism will use this short DNA as a template when replicating its own DNA. The sequence of the oligonucleotide will be incorporated into the host DNA together with the mutation. This ODM is another method used for genome editing, especially to make economically important mutation of 1-2 bases to serve several useful purposes including for example, making crops tolerant to herbicides.

CRISPR/Cas is the most widely known tool for genome editing. More information about CRISPR/Cas and other techniques and applications of genome editing are found in Annexures 1, 2, 3, and 4.

The range of methods continues to grow, supported by advancements in science and technology.

2.3 PROSPECTS OF GENOME EDITING APPLICATIONS IN CROPS AND ANIMALS

Genome editing techniques are promising tools in plant and animal breeding. These techniques have applications in diverse plant and animal species to generate various site-specific genome modifications using targeted mutagenesis and editing for varied agricultural applications. Through these techniques, scientists can discover novel traits, expand the range of traits, preserve niche or orphan crops, and accelerate trait development in target crops and animals that are essential for food security. Globally, products under research and development using genome editing tools involve various categories of traits including those related to increased plant yield and growth, increased food and feed quality, biotic and abiotic stress tolerance, industrial utilization, herbicide tolerance, product colour/flavour and storage performance (EU-SAGE).

This sub-section highlights the existing opportunities towards harnessing these techniques to target traits that will boost crop and animal productivity and resilience to climate change and redesign the future of agriculture and food security. Notably, some of these crop improvements have been commercialised and others are at advanced stages of product development.

2.3.1 GENOME EDITING APPLICATIONS IN CROPS

Over the last two decades, genetic editing tools such as ZFN and TALENS have been applied in plant breeding research and development (R&D) programmes. This includes important agricultural crops such as maize, soybean, rice, wheat, potato, tomato, and cassava (Zhang et al, 2018; Martnez-Fortun et al, 2017; Ran et al, 2017). However, the CRISPR/Cas system, first described in 2012, has the most applications to date because it is efficient, easy to use, and cost effective. Notably, the CRISPR/Cas has been used to genetically edit many crops for the purposes of R&D, including rice, maize, wheat, soybean, barley, sorghum, potato, tomato, flax, rapeseed, Camelina, cotton, cucumber, lettuce, grapes, grapefruit, apple, oranges, and watermelon.

There are examples of genome editing research and product developments in Africa (see Table 1). Recently, as a new frontier for agricultural innovation, the Kenya National Biosafety Authority (KNBA) approved three projects on genome editing under the Biosafety (Contained use) Regulations being at the research stage. These are genome editing of yam expressing disease resistance and enhanced Vitamin-A, grass peas with improved morphological, biochemical, and developmental traits, and sorghum lines (LGS1 loci) for resistance against Striga (Striga hermonthica). However, KNBA has received an early consultation application for the sorghum project to enter the open trials stage, for which the regulator's analysis indicates that the genome edited sorghum (would not involve presence of foreign genetic material in the final product), hence it is not GMO and therefore is not subject to regulation under the Biosafety Act, 2009. Banana was also developed for resistance to fungal infections, more especially the Fusarium wilt and Black sigatoka, and bacterial infections (Banana Xanthomonas Wilt, BXW). The cassava was also improved for nutritional quality (KNBA, 2022). It has also found applications for grass pea to enhance nutritional and agronomic traits (ILRI), as well as maize for resistance to Maize Lethal Necrosis (by CIMMYT/KALRO) (KNBA, 2020).

Table 1. Examples of ongoing genome editing research projects in Africa as of the year 2022 (Karembu and Ngure, 2022)

S/N	PROJECT TITLE	CHALLENGE(S) BEING ADDRESSED	PROJECT OBJECTIVE(S)	TARGET GENE(S) / T AND PHENOTYPE(S) / P	COUNTRY
1	Evaluation of Striga resistance in Low Germination Stimulant 1 (LGS1) mutant sorghum	Control of parasitic weed Striga	To evaluate LGS1 gene knock-out in conferring Striga resistance in sorghum	T: LGS 1 P: Reduction of Striga germination stimulant activity	Kenya
2	Genetic improvement of banana for control of bacterial wilt disease	Susceptibility to Xanthomonas wilt disease of banana in East Africa	To develop genome edited banana resistant to bacterial wilt disease	T: Disease susceptibility 'S' genes P: Banana wilt disease resistance	Kenya
3	Application of reproductive biotechnologies to develop a transgenic goat as a model for genetic control of animal diseases	Susceptibility of goat to animal trypanosomiasis disease	To generate African indigenous goat carrying the APOL 1 transgene that confers resistance to trypanosomiasis	T: APOL 1 P: Trypanosomiasis disease resistance	Kenya
4	Gene editing to control maize lethal necrosis in Africa for improved maize productivity and grain harvests	Susceptibility to maize lethal necrosis (MLN) disease	To introduce resistance against MLN disease directly into parent inbred lines of popular commercial maize varieties, which are currently susceptible to the disease	T: Quantitative trait locus (QTL) P: Resistance against MLN disease	Kenya
5	Mutation of energy homeostasis in maize to develop lines tolerant to drought, genotoxic and oxidative stresses	Drought susceptibility of maize	Metabolic engineering of Poly(ADP-ribosyl)ation pathway (a stress response pathway) to broaden stress tolerance in plants by maintaining energy homeostasis during stress conditions To knock-down the maize PARP gene expression using CRISPR/CAS9 gene editing as a strategy for abiotic and genotoxic stress tolerance	T: Poly(ADP-ribose) polymerase (PARP1 and PARP2) P: Maize tolerant to drought, DNA damage and oxidative stresses	Kenya
6	Improving oil qualities of Ethiopian mustard (*Brassica carinata*) through application of CRISPR/CAS 9-based genome editing	High level of erucic acid in *Brassica carinata* varieties released earlier beyond the nutritionally acceptable level	2 of 4: To develop *B. carinata* genotype with low erucic and glucosinolate for food and feed application To develop *B. carinata* genotypes with wax ester for industrial application	T: For food – FAE1 and FAD2 For feed – GTR1 and GTR2 For industry – FAR and WS	Ethiopia
7	Application of targeted gene editing for development of high yielding, stress	No sources of resistance to rice yellow mottle virus	To develop novel sources of resistance to devastating rice yellow mottle virus through gene editing	T: P: Resistance against rice yellow mottle virus	Uganda

	resistant and nutritious crops	No sources of resistance to maize lethal necrosis	To develop novel sources of resistance to maize lethal necrosis through gene editing	T: P: Resistance against maize lethal necrosis	
8	High throughput screening of genes associated with the response of cassava to geminivirus South African cassava mosaic virus (SACMV)	Susceptibility of African cassava varieties to cassava mosaic disease (CMD)	1 of 3: To silence genes putatively associated with the response to SACMV infection in susceptible and tolerant cassava landrace protoplasts using CRISPR gene editing	T: Ubiquitin proteasome system genes (e.g., E3 ligases), transcription factor genes (e.g., WRKYs) and resistance genes (e.g., NLRs) P: Resistance to CMD	South Africa
9	Genome editing of potato	Viral infection in potatoes	To produce potatoes resistant to virus infection	T: Eukaryotic initiation factor 4E (Eif4E)	South Africa
10	Developing sal1 mutant drought tolerant wheat using CRISPR/Cas genome editing	Susceptibility of wheat to drought stress which limits crop productivity	To generate transgenic wheat plants with stress tolerance	T: Sal1 P: Stress (drought) tolerance	Egypt
11	Genome editing by CRISPR/Cas9 in sorghum for improving biofuel production and forage quality	Enhancing biofuel production process and improving animal forage digestibility	To develop sorghum plants with low fibre content	T: COMT P: Lignin content	Egypt

In terms of deployment, the number of genome edited products currently on the market is still limited. However, with expectations of the potential products currently in the R&D stages, these genome edited products will soon move through to commercialisation. For example, Sanatech Seed in Japan has commercialized its Sicilian Rouge tomatoes. These tomatoes are genome edited to contain high amounts of γ-aminobutyric acid (GABA). The company is claiming that this can help support lower blood pressure and promote relaxation (media.springernature.com). Calyx in the United States of America (USA) has commercialised a high oleic, low linolenic soybean that contains no trans fats. This oil also has a longer fry and shelf-life than traditional soybean oil. Further to this, developed using the TALEN technology, the USDA deemed the soybean to be non-regulated (Calyx, 2020). The USA's Biotechnology Regulatory Service has deemed dozens of genome edited plants unregulated under 7 CFR 340, Introduction of Organisms and Products Altered or Produced Through Genetic Engineering Which are Plant Pests or Which There is Reason to Believe are Plant Pests. Its decisions are publicly available in the Regulated Article Letters of Inquiry repository (Calyx, 2020). It is also worth noting that in addition to the US, some other countries have likewise already made several decisions on genome edited products that are not within scope of GMO regulations including Argentina, Brazil, Chile, and Colombia.

2.3.2 GENOME EDITING APPLICATIONS IN ANIMALS

Genome editing techniques and methodologies such as the CRISPR/Cas9 and complimentary tools are efficiently and precisely manipulating animal genomes to produce genome edited animals. However, several limitations and challenges for the genome editing implementation in animals still need to be overcome and optimised. This includes the minimisation of off-target effects, indel noise in the target site, delivery of CRISPR/Cas9, and production of chimeric offspring. However, it is encouraging to note that the prime editing technique has recently been reported to be effective in genetically engineering the animal genome with few bi-products. In addition, these techniques have exhibited enhanced target accuracy, improved target range, and delivering small size nucleases. Thus, research and development from African scientists that improve the methodologies and techniques to positively advance the field of genome editing in animals and promoting personalised disease models to enhance desirable economic traits should be encouraged.

Successful genome editing has been reported in cattle, sheep, goats, pigs, horses, fish among others (Menchaca, 2020). See Annexure 5. Animal welfare should be pursued when dealing with animals to promote ethical research when carefully choosing useful traits for economic advancement. For example, the replication of natural mutation "double muscle" trait using genome editing may be controversial in mammals. Furthermore, it remains important to choose techniques that can efficiently minimise off-targets to avoid any unintended health impact to the first animals obtained.

Genome editing in animals has aimed to increase productivity and diseases resistance. Below are some examples, summarised as follows where genome editing was used:

a) The enhancement of muscle growth and development as target myostatin gene (MSTN). This involves the deletion in the MSTN gene which has resulted into enhanced muscle development in cattle, sheep, goats, and channel catfish.

b) The enhancement of animal welfare. For example, the TALENs was utilised to produce dehorned cattle. This precluded the conventional dehorning of cattle, which can be painful and time consuming.

c) The development of sterility in Atlantic salmon is desired to avoid genome edited escapees from rearing ponds from interbreeding with wild types. Genome editing approaches have been used to produce sterile Atlantic Salmon.

d) Genome editing has potential applications in complementing artificial insemination (AI) in areas where AI is hampered by limited resources for research infrastructure. One approach has been to use sterile host animals that are used to distribute high value transplanted germplasm. Success has been reported in producing sterile pigs and chicken through genome editing.

e) Developing pigs that are resistant to viral infections such as Porcine Reproductive and Respiratory Syndrome Virus (PRRSV). PRRSV is the most economically important disease of pigs globally causing a loss of approximately $2.5 billion per year in USA and Europe. Genome editing has been applied to knock out of the CD163 gene or the SRCR5 (scavenger receptor-cysteine rich domain). Approaches using genome editing in either one of these gene resulted in resistance to PRRSV.

f) Resistance to African swine fever caused by African Swine Fever Virus. The disease in endemic to regions of Sub-Saharan and has recently spread into Europe and Asia. Warthogs are resistant to ASFV because of the RELA gene in their genome. Using ZFN based genome editing, scientists have edited a domestic pig gene to produce the warthog RELA protein and acquire resistance to African swine fever.

3 REGULATORY LANDSCAPE FOR MODERN BIOTECHNOLOGY AND GENOME EDITING: A GLOBAL AND AFRICAN PERSPECTIVE

3.1 INTERNATIONAL BASIS FOR REGULATING MODERN BIOTECHNOLOGY

The primary international forum deliberating the regulation of genome editing is the Convention on Biological Diversity (CBD, 2007), along with its subsidiary agreements concerned with the biosafety of living modified organisms (LMOs); Cartagena Protocol on Biosafety to the CBD (CPB, 2000), and Nagoya Protocol on access to genetic resources and the fair and equitable sharing of benefit arising from their utilization (NP, 2011).

Article 8 (g) of the CBD obliges Parties to "establish or maintain means to regulate, manage or control the risks associated with the use and release of LMOs" (CBD, 2007). The CPB was negotiated and adopted in 2003 with the primary aim of maximising the benefits of biotechnology for biodiversity conservation while minimising adverse effects to the same. Premised on that, Article 1 of the Cartagena Protocol on Biosafety (CPB) aims "to contribute to ensuring an adequate level of protection in the field of the safe transfer, handling and use of LMOs resulting from modern biotechnology that may have adverse effects on the conservation and sustainable use of biological diversity, taking also into account risks to human health, and specifically focusing on transboundary movements" (CPB, 2000).

The definition of a genetically modified organism (GMO) may vary between different jurisdictions. However, most countries have based their definition on the Cartagena Protocol on Biosafety (CPB) and its definition of a Living Modified Organism (LMO). The CPB defines a LMO as ''any living organism that possesses a novel combination of genetic material obtained using modern biotechnology'' (CPB, 2000). The CPB also defines "Living organism" as any biological entity capable of transferring or replicating genetic material, including sterile organisms, viruses, and viroids; and "Modern biotechnology" as the application of:

a. In vitro nucleic acid techniques, including recombinant deoxyribonucleic acid (DNA) and direct injection of nucleic acid into cells or organelles, or

b. Fusion of cells beyond the taxonomic family,

that overcome natural physiological reproductive or recombination barriers and that are not techniques used in traditional breeding and selection.

From these definitions, a "novel combination of genetic material" means a stable insertion in the genome of one or more genes or DNA sequences that encode proteins, RNA, double-stranded RNA

or regulatory sequences, that could not occur through conventional breeding or is not found in nature or is not the result of spontaneous or induced mutagenesis. Thus, a novel combination may simply be defined as an organism whose DNA has been altered with the addition or substitution of foreign genetic material. A novel combination of genetic material and foreign genetic material are terminologies that are used interchangeably in literature, as the latter could be defined as novel combination of genetic material from sexually non-compatible species through the use of modern biotechnology techniques. Likewise, this policy considers both terminologies to have the same meaning.

Article 2 of the Protocol enjoins each Party to take necessary and appropriate legal, administrative, and other measures to implement its obligations under this Protocol. Indicative of this provision is for Parties to the Protocol to have the foresight for future technological developments. It is important to note that while this is deliberated upon under the CBP/CBD in the context of synthetic biology discussions, there is currently no consensus on either the inclusion or scope of applicability of the Protocol to genome edited products.

In Africa, governments have generally started the development of their national biosafety systems within the last 20 years under guidance of the CPB. The CPB emphasizes the application of the "precautionary approach" in biosafety decision-making. As rightly observed by some authors, this presents difficulty as it fails to provide guidance on effective risk assessment and reaching clear risk-based decisions that allow access to beneficial biotechnology products that have low risk.

It is generally the responsibility of national governments to formulate and implement science-based and efficient biosafety policies. It is important to note that regulatory considerations for products of genome editing can be effectively managed under existing legal instruments with or without minor revisions. This can inform how policies are implemented with specific guidance on how to identify and exclude products that do not contain foreign genetic material. It is also important to note that in some jurisdictions, a government may choose to provide a register of genome edited organisms or products that come under regulations applied to conventionally bred plants and for which categorical exemptions are granted for exclusion from GM regulations.

3.2 CURRENT REGULATORY STATUS OF GENOME-EDITED ORGANISMS WORLDWIDE

Policies are useful to guide the safe implementation of new products such as those derived from technology and innovation (Whelan et al, 2020). In a productive sector based on biological processes, such as the agro-industry sector, regulation is a tool that should be used to preserve the "welfare" of society as it adopts innovations. In other words, the enactment and application of regulations is part of policymaking, where the aim is to establish frameworks for safe and adequate development within the innovation system (Whelan et al, 2020). The regulatory status of genome-edited organisms has been discussed in various countries, and the regulatory approaches differ across countries. The discussions have been gaining traction globally as genome edited products progress in the pipeline. Consequently, several agencies in countries such as Argentina, Australia, Brazil, Canada (Health Canada), Chile, Japan, and the USA (USDA) have adapted their regulatory approach to apply the appropriate level of regulation, including regulations applicable to conventional varieties, on genome edited varieties with no foreign gene integration (Lema, 2019).

For Nigeria, the biosafety legislation was amended to include regulation of genome editing. Guidelines were subsequently adopted in December 2020 to clarify regulatory oversight. Subsequently, Kenya, Malawi, and Ethiopia have adopted national guidelines. The aforementioned four African national guidelines all provided for certain genome edited products not to be regulated under biosafety laws. As positive momentum for risk-based regulation builds, many African countries

have expressed interest in developing regulations or guidelines for genome editing including Burkina Faso, Eswatini, Ghana, Mozambique, Rwanda, Senegal, and Togo. However, in contrast to recent science-based risk analysis, the government of South Africa in 2021, classified all genome edited plants as genetically modified crops based on its interpretation of the definition of the country's legislation, but this decision is being appealed.

Notably, several countries including Argentina, Australia, Brazil, Canada, Chile, Japan, and the USA do not regulate genome edited varieties with no foreign gene integration in the final product (Figure 1).

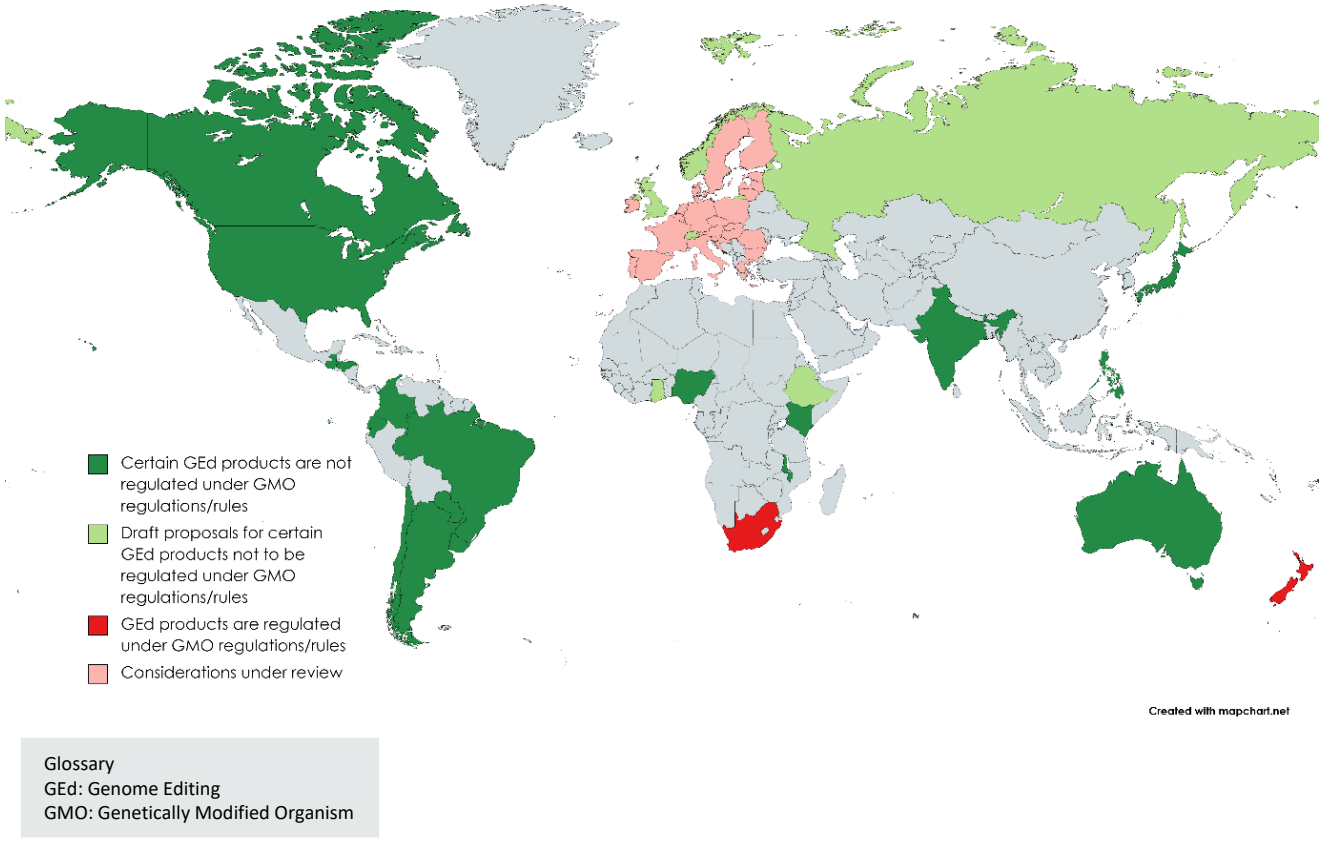


Figure 1. Regulatory approaches for genome edited products in various countries (as at 2022)

Concerns were raised as far back as a decade ago as to whether products of NBTs that do not contain foreign genetic material should be subject to GMO legislation. For instance, the European Academies Science Advisory Council (EASAC), a body of national science academies of the EU Member States and a voice of independent science advice, mobilising Europe's leading scientists to guide EU policy for the benefit of society, held the scientific opinion that products of NBTs that do not contain foreign genetic material should not fall under GMO legislation. EASAC noted that in instances where the commercialised crop developed through NBTs is free of foreign genetic material it cannot be distinguished from those developed by conventional techniques thereby posing detection and regulatory challenges. advisors noted that in some cases the product cannot be distinguished from one generated by conventional techniques. The argument was that the new techniques can allow much more precise and targeted changes compared with mutagenesis used in conventional breeding. In such cases, the changes in the genome are induced by chemicals or radiation, and thereby, creating multiple, unknown, and unintended mutations.

[3]EASAC is the voice of independent science advice, mobilising Europe's leading scientists to guide EU policy for the benefit of society. It brings together the National Academies of Science of the EU Member States, Norway, Switzerland, and United Kingdom. https://easac.eu/ EASAC (2013). Planting the future: opportunities and challenges for using crop genetic improvement technologies for sustainable agriculture. EASAC policy report 21 (Halle, Germany: European Academies Science Advisory Council).

plants in England, is limited to field trials and R&D activities. The second step intended to enable marketing of genome edited organisms has been also drafted and is currently under discussion.
Regulatory authorities in a host of countries follow this same EASAC conclusion that genome editing, in cases where no novel combinations of genetic material have been created, should be no more regulated than a product of conventional mutagenesis. For example, Argentina has been a case of reference, where in 2015 the country enacted regulatory criteria to assess if organisms resulting from new breeding techniques (NBTs) are to be regarded as genetically modified organisms (GMOs) or not (Whelan & Lema, 2015).

Following Argentina's regulatory approach to genome edited products, some countries in Latin American such as Brazil, Chile, Colombia, Ecuador, Guatemala, Honduras, and Paraguay have embraced the same approach to regulate genome edited products on a case-by-case basis and allow exclusions from GM regulation when there is no novel combination of genetic material (Lema, 2019). A synopsis of existing regulations on genome editing is provided by Entine et al (2021).

In order to advance regulatory approaches that are proportionate to risk and avoid arbitrary distinctions across similar products, the United States Department of Agriculture recently revised its biotechnology regulations when it introduced the SECURE Rule in 2020 with it coming into effect in 2021. Amongst other key provisions, the SECURE Rule establishes specific instances of exemptions for plants modified by genetic engineering where the modification could otherwise have been made through conventional breeding or are already occurring in nature, as is the case for some specific categories of genome edited plants defined by the USDA. By applying a risk-proportionate approach to regulatory oversight, it is expected that the SECURE Rule will be supportive of innovation by expanding the number and diversity of developers to include small and medium sized enterprises and public research institutions, and to increase the number and variety of traits being developed through biotechnology (Hoffman 2022).

In Europe, the current regulatory approach treats genome-edited products as GMOs stemming from the 2018 judgment from the European Court of Justice which declared "organisms obtained by means of techniques and methodologies of mutagenesis constitute GMOs within the meaning of that provision" and "only organisms obtained by means of techniques/methods of mutagenesis which have conventionally been used in a number of applications and have a long safety record are excluded from the scope of that directive". In 2021, the European Commission published a study on New Genomic Techniques that indicated "the current GMO legislation is not fit for plants produced by targeted mutagenesis and cisgenesis, and that it needs to be adapted to scientific and technological progress in order to be resilient, future-proof and uniformly applied" and that the Commission "intends to initiate a policy action tailored to plants derived from targeted mutagenesis and cisgenesis" (EC, 2021).

Two countries in Oceania have different regulations. For example, Australia gave notice of "Gene Technology Amendment (2019 Measures No. 1) Regulations 2019," which is modified law of "The Gene Technology Act 2000" on April 4, 2019. The Australian office of gene technology regulator (OGTR) will not regulate the use of gene editing techniques in plants, or animals, that do not introduce new genetic material or allow breaks to repair using natural machinery, whereas foreign genetic material template guided repair of DSBs will be regulated. According to Fritsche et al (2018), "in 2014, New Zealand's Environmental Protection Authority ruled that plants produced via genome-editing methods, where no foreign genetic material remained in the edited plant, would not be regulated as LMOs". However, this decision was overturned by the High Court. Currently, New Zealand considers all gene edited organisms as LMOs.

3.3 EMERGING REGULATORY APPROACHES IN GENOME EDITING IN AFRICA

In Africa there have been several important steps made regarding regulatory policy formulation on genome editing. It is encouraging to note that sub-Sahara African governments have started defining their own regulatory approaches for genome edited plant and animals.

The AU has been steadfast on proposing more enabling and science-based approaches towards emerging technologies such as modern biotechnology, gene drives, and genome editing. While the AU's work on genome editing started only recently, its reports on gene drives have clearly embraced the technology as a realistic option for effective disease control: Freedom to Innovate 2007 (Juma & Serageldin, 2007) and APET Report 2018. Preceding these reports was the decision of the topmost organ, Summit of Heads of States and Governments (HoSG), 3-4 July 2017, on the technology of Gene Drive for Malaria Control (AU, 2017).

AUDA-NEPAD held a workshop that was attended by 35 AU Member States on the Science and Regulation of Genome Editing and their products. The key outcome of that meeting was that AU Member States recommended a science-based approach to genome editing. Prompted by these developments, some governments on the continent are considering the inclusion of genome editing and other emerging technologies within their regulatory frameworks.

A case in point is Nigeria, where, in August 2019, the National Biosafety Management Agency (NBMA) amended its Biosafety Act to include genome editing. In 2020, Nigeria developed and adopted genome editing guidelines. The guidelines recognise that some genome edited products are equivalent to the conventional products and should be regulated under the Seed Act. Some genome edited products have GMO equivalent intermediary steps, which can be eliminated later. The GMO intermediary step is regulated under the Biosafety Act while the null-segregant is regulated under the Seed Act.

From the positive momentum noted in Nigeria, Kenya, and Malawi, it is expected that other nations and governments with such programmes can enhance their agricultural productivity by following suit. This is because the agenda on food security is a matter of concern to most countries in the region. Scientists have advocated for the development of science-based regulatory policies for genome editing. This can take into consideration the low risk associated with many of the products and their similarity to products developed by conventional breeding or that occur in nature, which are considered safe and remain regulated under conventional seed laws.

3.4 CONSIDERATIONS FOR A HARMONISED APPROACH FOR AFRICA

Products developed using genome editing techniques can be effectively assessed using existing instruments with minor revisions to how they are implemented or with guidance on how to identify low-risk products. However, it is important to note that as per the definitions of the CPB and the national legislation for LMOs and modern biotechnology, not all genome edited products would fall under the scope of biosafety regulation. The use of genome editing would thus require biosafety policies that would decide on the regulatory status of a proposed product as to whether it should come under biosafety or conventional regulation.

Divergent regulatory approaches to new breeding techniques (NBTs) may be a result of different economic, social, and political pre-requisites (Entine et al, 2021). Such divergence may not pose problems when applied to locally produced and consumed products. This is even though time and cost of getting local products through the regulatory process could prove prohibitive. However, non-compatible regulatory processes are

[4]The African Union (AU) High Level Panel on Emerging Technologies (APET) three reports on emerging technologies that are believed to help drive Africa's sustainable socio-economic and structural transformation launched in June 2018.

problematic when applied to commodities entering international trade. Global trade in agricultural goods allows harvesting of economic benefits across regions. However, to facilitate such trade, globally harmonised and compatible regulations, and policies, can be an asset for such processes. it should be feasible to align policies, technical regulations, standards, and specifications for a harmonised regulatory system. A well-structured harmonised regulatory system should confer benefits such as cost efficiency, adequate shared technical capacity to ensure a high level of safety, creation of more competitive markets, facilitation of cross-border trade, and creation of standardized and transparent processes that will assure predictability in international trade.

STISA-2024 remains a key strategy in accelerating Africa's transition to an innovation-led, knowledge-based economy. The implementation of STISA-2024 is not only at the national level, but also at regional and continental levels. Regional collaboration is an efficient regulatory approach for Africa and beyond. Visible on the continent today are regional economic communities (RECs) with structures and platforms that coordinate and guide the conversation surrounding NBTs. Groups such as the Economic Community for Central African States (ECCAS), Common Market for East and Southern Africa (COMESA), the Economic Community of West African States (ECOWAS), the Southern African Development Community (SADC), and East African Community (EAC), Arab Maghreb Union (UMA), Community of Sahel–Saharan States (CEN–SAD), and Intergovernmental Authority on Development (IGAD) are among those recognized by the AU for the delivery of services to the people of the continent. In the recent past, each of these regional bodies have initiated processes, policies, and guidelines for new breeding techniques (NBTs) that could leapfrog access to safe genome edited products.

4 OVERARCHING CONSIDERATIONS IN REGULATING GENOME EDITING

4.1 APPLICATION OF THE CARTAGENA PROTOCOL ON BIOSAFETY DEFINITION OF LMOS/GMO

Presently, there are still some uncertainties and debate as to whether genome edited products should be regulated as GMOs or treated as conventional (Araki & Ishii, 2015; Rinaldo & Ayliffe, 2015). There is also a need for a decision on this considering the benefits to both producers and consumers (Curtin et al, 2012; Belhaj et al, 2013; Ribarits et al, 2014; Araki & Ishii, 2015; Rinaldo & Ayliffe, 2015). It may be helpful to first address the legal uncertainty about which genome edited products will be regulated as GMOs or as conventional.

The Cartagena Protocol on Biosafety may contribute to resolving these uncertainties regarding the status of genome edited products as GMOs. This will be helpful considering that most AU Member States are Parties to this Protocol. Noting certain applications of genome editing may result in varieties that cannot be uniquely identified from conventionally developed ones, the definition of a living modified organism (LMO) in the Cartagena Protocol on Biosafety is instructive. The Cartagena Protocol on Biosafety defines an LMO as any living organism that possesses a novel combination of genetic material obtained using modern biotechnology. It is important to note that the "novel combination" is not arbitrarily used but linked to the term "recombinant DNA". Thus a DNA molecule that "combines" sequences from different origins is referred to as a "gene construct". Therefore, the criterion to consider the genome edited organisms as an LMO would be the presence of a foreign genetic material or novel combination of genetic material in the target genome (Hartung & Schiemann, 2014; Araki & Ishii, 2015).

Box1: Some examples of Genome Edited Crops produced through SDN1, SDN2, ODM, and Null-segregants

Site directed Nuclease 1 (SDN1). Most countries globally consider SDN1 as equivalent to conventional bred crops because they are developed though generation of indels such as those achieved through conventional breeding or that are already occurring in nature. They are to be released under laws regulating conventionally bred crops.

Site directed Nuclease 2 (SDN2). Crop genome edited using SDN2 could be regulated as conventional depending on the country's sovereign decision in which case they will be subjected to the national regulation for releasing conventionally bred crops.

Oligonucleotide Directed Mutagenesis (ODM). ODMs give small genetic changes including indels that are identical to conventionally bred crops and could be exempted from the GM Biosafety Regulations and subject to country specific laws such as Seed Act Regulations for release of conventionally bred crops.

Null segregants. These are genome edited crops that are developed using similar techniques as used in developing genetically modified crops but which the GM intermediate are segregated out through breeding. The plant should be subject to biosafety regulations of GM up to the stage of removal of the GM intermediate after which they should be subjected to the country's conventional regulation for releasing and commercializing conventionally bred crops such as seed laws.

4.2 REGULATORY PATHWAYS AND INFORMATION REQUIREMENTS FOR GENOME EDITING

The first step would be for African countries to evaluate if their biosafety laws and/or regulations are applicable to genome edited products and, if they are, are there categories of genome edited plants or animals that may be excluded or exempted by definition. In the case of the latter, regulatory authorities may wish to consider the evidence-based approach i.e., diagnostic criteria or protocols that will be required to demonstrate the absence of foreign genetic material if that is a prerequisite to an exemption or exclusion from GM regulation. Harmonization of such protocols across countries for determination of the absence of foreign genetic material in a candidate product will offer advantages for technology review, dissemination, and trade. To this end, AUDA-NEPAD is to convene a meeting of experts from multiple African countries to define specific model protocols for national consideration, which will be shared with AU member states and/or will actively participate in any multilateral efforts established for this same purpose.

In general, African countries that have reviewed these options have identified genome edited products that could have been produced by conventional breeding or that are already occurring in nature as having the same risks as conventional breeding and so are not regulated as GMO, but in a manner like conventional varieties. These would include SDN1 and SDN2 products with gene knock outs, deletions of any size, and insertions of short sequences because of the inherent DNA repair mechanisms. They also exclude from regulations the introduction of cis-genes that are part of the breeding gene pool of the host species and rearrangements in the genome that do not introduce open reading frames with high sequence similarity to known allergens or toxins. On the other hand, genome edited products that introduce foreign genetic material into the genome to introduce a new trait are considered GMOs and are regulated like GMOs.

This approach then provides the basis for exemption/exclusion or otherwise to biosafety regulation and includes:

a) A pre-submission consultation process to determine regulatory status. This is noting that there are a few examples of some jurisdictions in which developers can self-determine. i.e., in the US and Australia while in India, the Institutional Biosafety Committee is tasked with making the determination that a genome edited plant is free from exogenous DNA and therefore not subject to any further regulation under the Rules, 1989. which govern GM.

b) A case-by-case assessment of the applications.

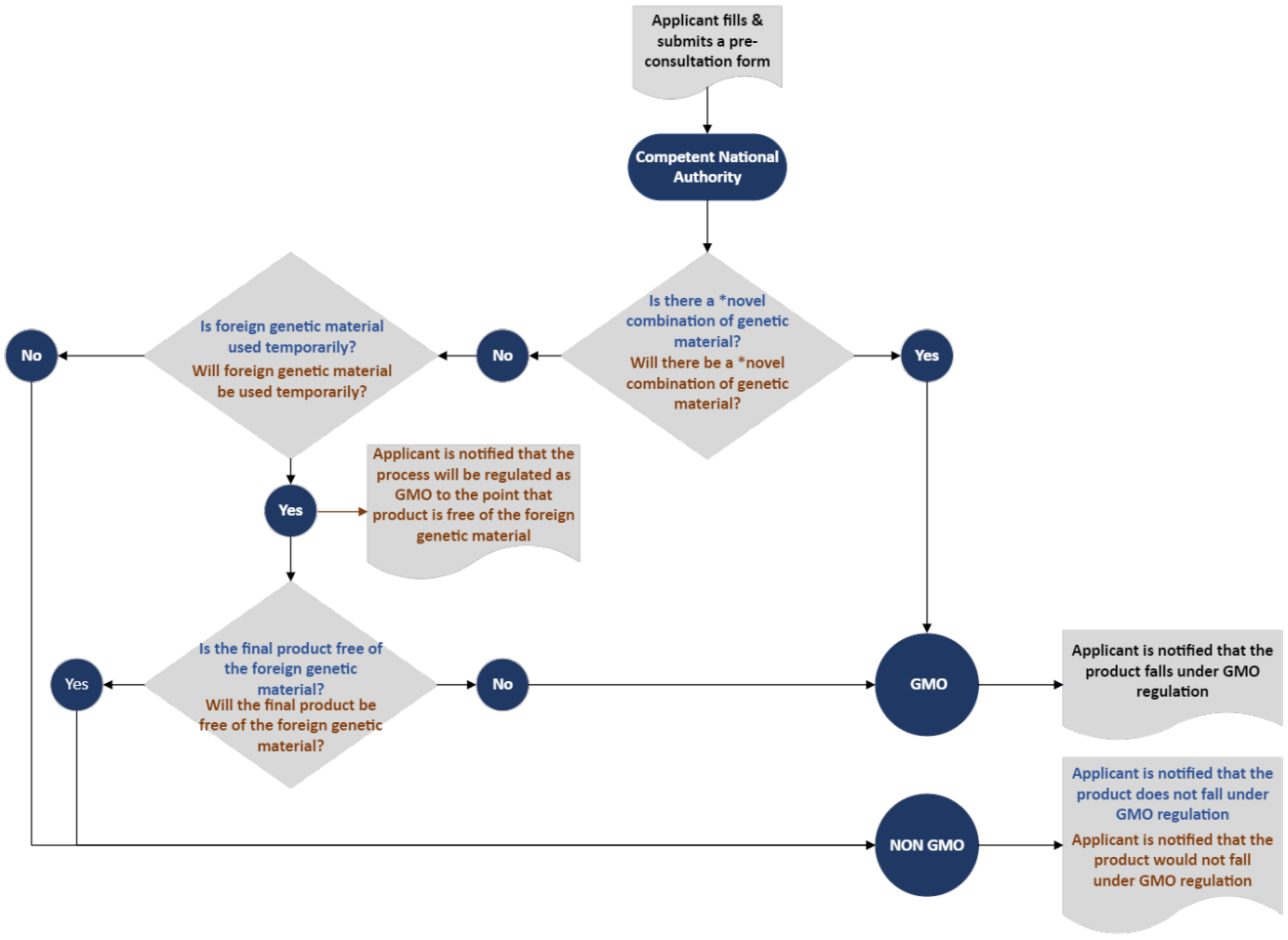
c) A product-based regulatory approach to determine presence of a foreign genetic material or novel combination of genetic material in final products.

The first step in the case-by-case assessment of a genome edited crop application is to provide an early consultation pathway (pre-submission consultation) to differentiate between which genome edited crops are equivalent to conventionally bred crops and which are GMOs. This early consultation pathway aims to help National Competent Authorities for Biosafety to determine when and how to exclude a genome edited product from the scope of the national Biosafety Act in a stepwise and systematic way to ensure predictability in decision making. During early consultation process, regulators will examine submissions made by applicants to determine whether the final genome edited product developed or anticipated does or will have a stable and permanent insertion of a foreign genetic material. Regulators also examine if the genome editing process used by the applicant involves the temporary use of foreign genetic materials.

Below are possible outcomes from the early consultation process and their regulatory implications:

Outcome of early consultation review	Regulatory implication
1. The final genome edited product developed or anticipated does contain a stable and permanent insertion of foreign genetic material	Product falls within the scope of the biosafety act and will be regulated as a GMO
2. The final genome edited product developed or anticipated **DOES NOT** contain a stable and permanent insertion of foreign genetic material	Product **DOES NOT** fall within the scope of the biosafety act and will be regulated as would any conventional crop (non-GMO)
3. The process of developing and testing a genome edited product involves intermediate steps that may contain foreign genetic material temporarily	Product / process will be considered to be under the scope of the biosafety act until such a stage in the development and testing process where the applicant can demonstrate the absence of the foreign genetic material

An example of such a decision tree is presented below (Figure 2).



*An organism whose DNA has been altered with the addition or substitution of foreign genetic material

Figure 2. Early or pre-submission consultation procedure for determining if a genome edited organism will be regulated, or not, as genetically modified

The decision tree includes two possibilities, (a) an already developed product and (b) a product to be obtained. Regarding (b) an early regulatory determination at the project design stage is very important to foster new projects, especially in developing countries, and therefore countries should allow for both (a) and (b). Such criteria also include cooperative links between the regulatory frameworks for GMOs and for conventional products, to avoid any safety or legal gap.

There may be instances of a temporary use of foreign genetic material during the early stages of R & D. The introduced foreign genetic material may subsequently be selected out (also called null-segregants) and is thus absent in the final products. Such research may be considered to fall within the scope of the Biosafety Act until such a time when the foreign genetic material is removed. At this point, the product should no longer be regulated by the Biosafety Act but rather be regulated under other existing laws, regulations, and guidelines governing the evaluation and release of conventionally bred organisms. There should be scientific evidence that the foreign genetic material has been removed.

A genome edited product that is deemed to be a GMO would be subjected to the decision-making criteria as defined in national policies, laws, and regulations for GMOs. These criteria include submission of application form and dossier, check for completeness, public participation (where required), risk assessment review, recommendations, and decision-making. The decision-making process for biosafety activities and GMOs seeks to ensure accountability and transparency. Accountability is achieved by compliance to the regulatory regime while transparency is provided through decision documents.

Genome edited products that are deemed not to be GMOs will not require any additional regulation under the biosafety laws but follow regulations applicable to products of conventional breeding. Genome edited products that are deemed to be GMOs will be regulated under the national biosafety law.

4.3 CLASSIFICATION OF GENOME EDITED ORGANISMS

The genome edited organisms can be classified as follows:

4.3.1 GENOME EDITED ORGANISMS/ PRODUCTS DETERMINED TO BE GMOS

The general guidance is that genome edited organisms with genetic changes resulting in the stable and permanent integration of a foreign genetic material from outside the organism's gene pool are determined to be GMOs. This category should be regulated under national biosafety laws. The regulations and associated guidelines should be implemented during the development, testing, release into the environment, and the subsequent releases of commercial products. Each AU Member State should exercise its sovereign rights in developing guidelines for oversight on such genome edited organisms. The conduct of risk analysis will be as outlined in the national Biosafety Regulatory Framework.

4.3.2 GENOME EDITED ORGANISMS/ PRODUCTS THAT ARE DETERMINED TO BE NON-GMOS

The general guidance is that genome edited organisms with genetic changes that are achievable through traditional and conventional breeding or that are already occurring in nature are determined to be non-GMOs. These products do not contain foreign genetic material from outside the organisms' gene pool in the final product counterparts. This category should be regulated outside the scope of National Biosafety Acts and implementing instruments. Such organisms should be regulated under existing laws, regulations, and guidelines governing the evaluation and release of conventionally bred organisms and products derived from them. Each AU Member State should exercise a science-based approach and within their sovereign rights in developing the guidelines for oversight on genome edited organisms.

4.4 OFF-TARGETS IN GENOME EDITED ORGANISMS

Off-target edits vary depending on the methods under consideration. Both the ZFNs and TALENs function as dimers with one monomer as the DNA-binding domain responsible for sequence specificity and other monomer being the Fok I nuclease catalytic domain that is responsible for the cleavage (Zhao & Wolt, 2017; Malzahn et al, 2017; Bortesi & Fischer, 2015). ZFN deploy low affinity and specificity resulting in higher off-target activity than TALENs (Zhao & Wolt, 2017). TALENs on the other hand is more of the cut efficiency that varies depending on the target sequence. TALENs cannot target methylated DNA which is most common in plants. CRISPR/Cas9 is widely utilised in the genome editing activities in plants. However, they have limited potential off target than TALENs (Zhao & Wolt, 2017).

Currently, research activities on economically important organisms for both plants and animals using genome editing are progressing rapidly with many published studies demonstrating promising results. As for any technology, safety and non-safety concerns have been raised to include weed and pest adaptation challenges and other unexpected side effects. However, it is important to note that these concerns are not peculiar to genome editing techniques. For example, the potato lines from conventional breeding were found to produce hazardous levels of steroidal alkaloid demissidine, a toxin not found in both parents, Solanum tuberosum and Solanum brevidens (Laurila et al., 1996).

In the specific instance of genome edited organisms, the increased precision of the SDN techniques was expected to result in the less likely occurrence of unwanted mutations as compared to transgenesis or induced mutagenesis (Carlson et al, 2013; Ribarits et al, 2014). However, the targeted gene also remains in its normal chromosomal context, reducing the possible effects on gene expression (Ribarits et al, 2014). However, in the case of plants and as has been done in conventional breeding, (a) random genetic changes are occurring in each generation just from natural and spontaneous mutations and (b) any possible negative effects of these mutations (however they are caused) are eliminated through subsequent plant breeding and screening programs. Safety and non-safety concerns are best addressed through appropriate regulatory systems that can quickly identify the absence of any new hazards associated with genome edited products compared to conventionally bred products. This would ensure that products are safe and would allow safe and new products to reach users, and subsequently benefit the consumers (Bruce et al, 2013; Graham et al., 2020; Hartung & Schiemann, 2014; Smyth et al, 2014). With the rapid evolution of the molecular tools for genetic characterization, prescribed methods for determining off-target edits could quickly become obsolete or not fit for purpose. Hence it is recommended that Regulators review whether the methods used were appropriate.

4.5 SCIENCE-BASED REGULATION: AN APPROPRIATE APPROACH TO RESOLVING GOVERNANCE ISSUES IN GENOME EDITING RESEARCH AND APPLICATION

Genome editing can result in products that are either GMOs or are indistinguishable from conventionally bred ones. Whatever the case, the products will either fall within the scope of biosafety or other existing laws. Therefore, the focus is not to promote legal amendments to the scope of existing laws to include genome editing. Rather, it is to clarify the regulatory approach through development of guidelines.

Guidelines for genome edited products may involve multiple regulatory authorities. This is because genome edited products will either be a GMO or a conventional organism and in which case, existing multi-sectoral regulatory systems would apply. These are established in many African countries, but not necessarily effective.

The robustness of national biosafety regulations

varies from one country to another. Robustness can be determined by available human and financial resources, available infrastructure, as well as pragmatic and science-based decision making that is not impeded by politics. Some countries have rationalised their regulatory processes to deal with these constraints. These were aimed towards making the regulatory processes fit for purpose (Wendy, 2016).

5 RECOMMENDATIONS

Through decisions of the AU Assembly, the continent commits to safely harness modern biotechnology and emerging technologies or tools including genome editing as socio-economic developmental tools. This entails creating an enabling environment that would ensure that these technology benefits are harnessed while minimising potential risks. In adopting a science-based approach towards safely harnessing the potential of genome editing and its contribution to the AU Agenda 2063, the African Union High Level Panel for Emerging Technologies (APET) has the following recommendations:

5.1 ESTABLISHING AND STRENGTHENING AFRICA'S NATIONAL BIOSAFETY REGULATORY AUTHORITIES AND SCIENTIFIC REVIEWERS

a) Genome editing is a rapidly developing suite of tools that enable cost- and time-effective targeted and precise alterations of the genome and thus opening new applications and possibilities for bioeconomic activities in healthcare, agriculture, industrial biotechnologies, and other bioeconomy-related sectors.

b) African Union should undertake high level advocacy and strengthen the capacity of Member States on Genome Editing, using the Calestous Juma Executive Dialogues (CJED), which is an established APET Platform.

c) African countries can strengthen the capacity of Competent National Authorities, scientific/technical reviewers, and decision-makers through the CJED Platforms, in assessing the potential risks and benefits of genome editing and in making informed decisions. This can help governments adopt policies that can allow for genome edited products to be assessed for their similarity to products that have been developed with conventional breeding methods or that occur in nature.

d) African countries, as parties to the Cartagena Protocol, should recognise the utility of the precautionary approach during new technological uptake and within the context of scientifically sound, case-by-case risk assessments, appropriate risk management measures, and decision-making processes that weigh potential risks against benefits. This includes the perspectives of indigenous people and local communities (IPLCs), as may be appropriate.

5.2 RISK ASSESSMENTS AND DECISION MAKING FOR GENOME EDITED PRODUCTS

a) Genome edited products should be assessed on a case-by-case basis to determine whether they fall under the mandate of a country or a regional Biosafety Regulatory Framework or be excluded from such biosafety regulatory framework. This can also enable that these products be subjected to other existing regulatory frameworks for conventionally developed products and technologies.

b) Policy decisions on genome edited products should be informed by science.

c) AU Member States should take steps to address the legal and policy uncertainties regarding the preferred regulatory

approach for genome editing and derived products. This can be ascertained by developing national guidelines to indicate whether genome editing products will be regulated as under their Biosafety Acts or otherwise.

d) African countries should formulate and provide clear guidelines through which certain genome edited products should be excluded from the scope of their National Biosafety laws. This can be accomplished by adopting implementing regulations and guidelines that provide a clear mechanism for pre-submission consultation processes to determine the most appropriate, science-based and risk proportionate regulatory approach to use for genome edited products. Furthermore, this review should allow for case-by-case assessments of each product based on the presence of a foreign genetic material in the final product as a criterion for a product-based exclusion.

e) African countries should also ensure that any regulatory approach that requires that a genome edited product be subject to existing risk assessment and decision-making process as stipulated in their Biosafety Laws and implementing instruments would only apply to situations where the said genome edited product is proven to contain foreign genetic material in the final product.

f) Further ensure, in accordance with their biosafety laws, that only genome edited organism with genetic changes resulting in the stable and permanent integration of a foreign genetic material from outside the orgasm's gene pool are regulated under the country Biosafety Acts, Regulations and supporting Guidelines during development, testing, release into the environment and releases of commercial products.

G) Exclude from their biosafety laws and implementing instruments, genome edited organisms with genetic changes that are achievable through traditional/ conventional breeding or that are already occurring in nature, and which do not contain foreign genetic material from outside the organisms' gene pool in the final product. Such organisms should be regulated under existing laws, regulations and guidelines governing the evaluation and release of conventionally bred organisms and products derived from them.

5.3 STRENGTHENING COOPERATION, COLLABORATION, AND INVESTMENTS ON BIOSAFETY REGULATION, POLICY IMPLEMENTATION, AND INFRASTRUCTURE

a) As the genome editing techniques are rapidly advancing and evolving, AU Member States should consider strengthening their South-South and/or North-South cooperation and collaborations in accessing, evaluating, and applying genome edited products before production.

b) African countries should also develop mechanisms that can strengthen their cooperation towards developing harmonised systems at regional and continental levels on regulating genome editing.

c) African countries should also enhance their investments in capacity building and strengthening of human resources and research infrastructure suitable for genome editing advancements.

d) African countries should strengthen their

genome editing research, development, and innovation, education, product development, and product stewardship programmes.

5.4 IMPROVING AND STRENGTHENING BIOSAFETY REGULATION AND POLICY AWARENESS, AND OUTREACH PROGRAMMES

a) African countries should emphatically establish and implement outreach and communication programmes to create greater awareness and sensitisation with various stakeholders through the various forms of traditional and social media platforms. Such efforts will help address miscommunication, misinformation, and misunderstanding of emerging technologies or tools such as genome editing.

b) AU Member States should enhance their curriculum for science education and communication programmes, at basic, secondary, and tertiary levels. This should include cutting edge areas of genome editing to benefit Africa's technology and product development. This can help strengthen capacity within AU Member States in raising the next generation of Africa leaders on emerging technologies.

c) Given the experiences of GMOs, the regulatory approvals alone will not be sufficient to bring the products to farmers and end-users. Therefore, in addition to an enabling policy environment and product development platforms, African countries would have to strengthen technology delivery and seed systems in utilizing genome editing techniques. This can help the products efficiently reach smallholder and commercial farmers to create the desired impacts.

To take advantage of the lessons learned from the controversy related to the use of GMOs and to avoid the miscommunication considered as the main factor of the slow-paced adoption of GMOs in Africa, a well-resourced science-based communication strategy is required for better adoption of genome edited products in Africa. Ultimately, products derived from genome editing are intended for commercialization and placement on the market. Hence the importance of a communication strategy that promotes a balanced, science-based, and transparent public education and awareness creation for a supportive and enabling environment for adoption.

Saner (2007) enumerates reasons for public participation, among which include: potentially improve public policy, a more informed and engaged public, more solid support for regulatory decisions, and greater public confidence in government. Communication therefore includes these activities: inform or educate to help understand a policy or program; gather information to anticipate communication challenges; facilitate discussion among stakeholders; engage citizens for shared agenda setting and generate options; as well as partner or reach an agreement with stakeholders.

The question of production and use of seeds along with derived products from genome editing is highly complex as it affects many sectors including agriculture, environment, transport and trade, health innovation, food, industry, consumers, natural habitats and biodiversity, intellectual property, affecting international economic relations and even development assistance.

Efficient and effective multi stakeholder communication is the main lever for product acceptance, consensus building to facilitate decision-making, and stakeholder engagement. While a policy provides a set of rules to guide the production, the use, and the dissemination of a new product in the market, its application within national systems, will depend mostly on public understanding, acceptance, and adoption by

actors and end users. Therefore, a roll-out of a communication strategy is needed to facilitate the implementation of the policy on genome editing. Through the process of communication with key actors, the socio-cultural and ethical aspects, gender, and youth inclusivity, will be sufficiently highlighted and provided with sufficient space for expression to feed into policy making and implementation.

Multi stakeholder participation must operate upstream, involving farmers, scientists, regulators, etc. in the process of co-creation of the product, enabling knowledge generation at local level through platforms such as national level CJEDs. The importance of social media should be emphasized as an effective public relations and advocacy tool.

6 CONCLUSIONS

Delivering on Africa's agricultural potential to ensure food security while safeguarding biodiversity will require, among other things, investments in and adoption of affordable technologies. However, these emerging technologies should enhance agricultural productivity, promote animal and fishery welfare, improve climate resilience and carbon storage while reducing emissions, and ensure sustainable utilisation of land and other natural resources. The CBD recognises access to and transfer of technologies, including modern biotechnology, as relevant to the conservation and sustainable utilisation of biodiversity.

The application of modern biotechnology has substantially contributed to more sustainable agriculture, enabling better healthcare, enhanced food security, improved supplies of potable water, more efficient industrial processes, sustainable afforestation and reforestation, and detoxification of hazardous waste. Therefore, modern biotechnology can be increasingly deployed across the various biological kingdoms. Further to this, the genome improvements can expand the opportunities for agricultural breeding, industrial biotechnology, and human gene therapies and vaccines. Consequently, exploring the potential benefits from genome editing as a tool for socio-economic development aligns with the priority areas of STISA-2024.

Notwithstanding, genome editing has policy implications, acceptance issues, and safety considerations. Thus, Africa's ability to use existing and emerging biotechnologies effectively will depend largely on developing risk commensurate regulatory requirements and investment in physical, human, institutional, and societal capacity. As a continent, Africa should note that regulatory requirements commensurate with the level of risk are essential if genome editing is to become a tool for African scientists for the development of African economies, improved quality of life for African citizens, and improved cooperation with other regions of the world.

7 REFERENCES

Africa Food Security Leadership Dialogue (AFSLD) (2019). Communique. Africa Research Bulletin: Economic, Financial and Technical Series. Retrieved from https://doi.org/10.1111/j.1467-6346.2019.09069.x

African Development Bank (ADB) (2020). Africa's economic performance and outlook amid COVID–19. ISBN 978-9973-9848-8-3 (electronic). Retrieved from https://www.afdb.org/sites/default/files/documents/publications/afdb20-04_aeo_supplement_full_report_for_web_0705.pdf.

African Union (AU) (2017). Decision of the African Union: Assembly/AU/Dec.649 (XXIX) at the 29th Ordinary Sitting in Addis Ababa, 3 – 4 July, 2017. Retrieved from: https://au.int/sites/default/files/decisions/37294-assembly_au_dec_642_-_664_xxix_e_1.pdf

Anzalone, A.V., Koblan, L.W. & Liu, D.R. (2020). Genome editing with CRISPR/Cas nucleases, base editors, transposases, and prime editors. Nature Biotechnology, 38, 824 – 844. https://doi.org/10.1038/s41587-020-0561-9

Anzalone, A.V., Randolph, P.B., Davis, J.R. et al. (2019). Search-and-replace genome editing without double-strand breaks or donor DNA. Nature, 576, 149 – 157. https://doi.org/10.1038/s41586-019-1711-4

Araki, M., & Ishii, T. (2015). Towards social acceptance of plant breeding by genome editing. Trends in plant science, 20(3), 145–149. https://doi.org/10.1016/j.tplants.2015.01.010

Belfort, M., & Roberts, R. J. (1997). Homing endonucleases: keeping the house in order. Nucleic acids research, 25(17), 3379–3388. https://doi.org/10.1093/nar/25.17.3379

Belhaj, K., Chaparro-Garcia, A., Kamoun, S., & Nekrasov, V. (2013). Plant genome editing made easy: targeted mutagenesis in model and crop plants using the CRISPR/Cas system. Plant methods, 9(1), 39. https://doi.org/10.1186/1746-4811-9-39

Bortesi, L., & Fischer, R. (2015). The CRISPR/Cas9 system for plant genome editing and beyond. Biotechnology advances, 33(1), 41–52. https://doi.org/10.1016/j.biotechadv.2014.12.006

Bruce, A., Castle, D., Gibbs, C., Tait, J., & Whitelaw, C. B. (2013). Novel GM animal technologies and their governance. Transgenic research, 22(4), 681–695. https://doi.org/10.1007/s11248-013-9724-5

Calyx Letter of Am I regulated Inquiry. March 6, 2020. Accessed June 11, 2020. https://www.aphis.usda.gov/biotechnology/downloads/reg_loi/20-066-01-air-cbidel.pdf

Carlson, D. F., Tan, W., Hackett, P. B., & Fahrenkrug, S. C. (2013). Editing livestock genomes with site-specific nucleases. Reproduction, fertility, and development, 26(1), 74–82. https://doi.org/10.1071/RD13260
Cartagena Protocol on Biosafety (CPB) (2000). Retrieved from https://www.cbd.int/doc/legal/cartagena-protocol-en.pdf

Convention on Biological Diversity (CBD) (2007). Article 8, in situ Conservation. Retrieved from: https://www.cbd.int/convention/articles/default.shtml?a=cbd-08

Crispo, M., Mulet, A. P., Tesson, L., Barrera, N., Cuadro, F., dos Santos-Neto, P. C., Nguyen, T. H., Crénéguy, A., Brusselle, L., Anegón, I., & Menchaca, A. (2015). Efficient Generation of Myostatin Knock-Out Sheep Using CRISPR/Cas9 Technology and Microinjection into Zygotes. PloS one, 10(8), e0136690. https://doi.org/10.1371/journal.pone.0136690

Curtin, S. J., Voytas, D. F., & Stupar, R. M. (2012). Genome engineering of crops with designer nucleases. Plant Genome, 5(2), 42-50. https://doi.org/10.3835/plantgenome2012.06.0008

European Sustainable Agriculture through Genome Editing (EU-SAGE) https://www.eu-sage.eu/genome-search [Accessed on 2 December 2022]

Entine, J., Felipe, M., Groenewald, J. H., Kershen, D. L., Lema, M., McHughen, A., Nepomuceno, A. L., Ohsawa, R., Ordonio, R. L., Parrott, W. A., Quemada, H., Ramage, C., Slamet-Loedin, I., Smyth, S. J., & Wray-Cahen, D. (2021). Regulatory approaches for genome edited agricultural plants in select countries and jurisdictions around the world. Transgenic research, 30(4), 551–584. https://doi.org/10.1007/s11248-021-00257-8

Fonfara, I., Richter, H., Bratovič, M., Le Rhun, A., & Charpentier, E. (2016). The CRISPR-associated DNA-cleaving enzyme Cpf1 also processes precursor CRISPR RNA. Nature, 532(7600), 517–521. https://doi.org/10.1038/nature17945

Fritsche, S., Poovaiah, C., MacRae, E., & Thorlby, G. (2018). A New Zealand Perspective on the Application and Regulation of Gene Editing. Frontiers in plant science, 9, 1323. https://doi.org/10.3389/fpls.2018.01323
Gao, C., Wang, Z., Liang, Q. et al. (2010). Global transcription engineering of brewer's yeast enhances the fermentation performance under high-gravity conditions. Applied Microbiology and Biotechnology, 87, 1821–1827. https://doi.org/10.1007/s00253-010-2648-6

Gao, P., Yang, H., Rajashankar, K. R., Huang, Z., & Patel, D. J. (2016). Type V CRISPR/Cas Cpf1 endonuclease employs a unique mechanism for crRNA-mediated target DNA recognition. Cell research, 26(8), 901–913. https://doi.org/10.1038/cr.2016.88

Gaudelli, N. M., Komor, A. C., Rees, H. A., Packer, M. S., Badran, A. H., Bryson, D. I., & Liu, D. R. (2017). Programmable base editing of A•T to G•C in genomic DNA without DNA cleavage. Nature, 551(7681), 464–471. https://doi.org/10.1038/nature24644

Goedde, L., Ooko-Ombaka, A., & Pais G. (2019). Winning in Africa's agricultural market. McKinsey & Company. Retrieved from https://www.mckinsey.com/industries/agriculture/our-insights/winning-in-africas-agricultural-market

Graham N, Patil GB, Bubeck DM, Dobert RC, Glenn KC, Gutsche AT, Kumar S, Lindbo JA, Maas L, May GD, Vega-Sanchez ME, Stupar RM, Morrell PL. Plant Genome Editing and the Relevance of Off-Target Changes. Plant Physiol. 2020 Aug;183(4):1453-1471. doi: 10.1104/pp.19.01194. Epub 2020 May 26. PMID: 32457089; PMCID: PMC7401131.

Gratacap, R. L., Wargelius, A., Edvardsen, R. B., & Houston, R. D. (2019). Potential of Genome Editing to Improve Aquaculture Breeding and Production. Trends in genetics : TIG, 35(9), 672–684. https://doi.org/10.1016/j.tig.2019.06.006. Epub 2019 Jul 19. PMID: 31331664.

Hammer, R. E., Pursel, V. G., Rexroad, C. E., Jr, Wall, R. J., Bolt, D. J., Ebert, K. M., Palmiter, R. D., & Brinster, R. L. (1985). Production of transgenic rabbits, sheep and pigs by microinjection. Nature, 315(6021), 680–683. https://doi.org/10.1038/315680a0

Han, J. Y., & Park, Y. H. (2018). Primordial germ cell-mediated transgenesis and genome editing in birds. Journal of animal science and biotechnology, 9, 19. https://doi.org/10.1186/s40104-018-0234-4

Han H, Ma Y, Wang T, Lian L, Tian X, Hu R et al. (2014). One-step generation of myostatin gene knockout sheep via the CRISPR/Cas9 system. Front Agr Sci Eng., (1): 2–5. 10.15302/j-fase-2014007.

Hartung, F., & Schiemann, J. (2014). Precise plant breeding using new genome editing techniques: opportunities, safety and regulation in the EU. The Plant journal : for cell and molecular biology, 78(5), 742–752. https://doi.org/10.1111/tpj.12413

He, Z., Zhang, T., Jiang, L., Zhou, M., Wu, D., Mei, J., & Cheng, Y. (2018). Use of CRISPR/Cas9 technology efficiently targetted goat myostatin through zygotes microinjection resulting in double-muscled phenotype in goats. Bioscience reports, 38(6), BSR20180742. https://doi.org/10.1042/BSR20180742

Hoffman N.E. (2022) USDA's revised biotechnology regulation's contribution to increasing agricultural sustainability and responding to climate change. Front. Plant Sci. 13:1055529. doi: 10.3389/fpls.2022.1055529
Jo, Y. I., Kim, H., & Ramakrishna, S. (2015). Recent developments and clinical studies utilizing engineered zinc finger nuclease technology. Cellular and molecular life sciences : CMLS, 72(20), 3819–3830. https://doi.org/10.1007/s00018-015-1956-5

Juma, C., & Serageldin, I. (2007). Freedom to Innovate: Biotechnology in Africa's Development: Report of the High-Level African Panel on Modern Biotechnology

Kalds, P., Zhou, S., Cai, B., Liu, J., Wang, Y., Petersen, B., Sonstegard, T., Wang, X., & Chen, Y. (2019). Sheep and Goat Genome Engineering: From Random Transgenesis to the CRISPR Era. Frontiers in genetics, 10, 750. https://doi.org/10.3389/fgene.2019.00750

Karembu M. (MBS), Ngure G. (2022). Genome Editing in Africa's Agriculture 2022: An Early Take-off. 2nd Edition. International Service for the Acquisition of Agri-biotech Applications (ISAAA AfriCenter), Nairobi Kenya.

Khan S. H. (2019). Genome-Editing Technologies: Concept, Pros, and Cons of Various Genome-Editing Techniques and Bioethical Concerns for Clinical Application. Molecular therapy. Nucleic acids, 16, 326–334. https://doi.org/10.1016/j.omtn.2019.02.027

Kim, Y.G., Cha, J., & Chandrasegaran, S. (1996). Hybrid restriction enzymes: zinc finger fusions to Fok I cleavage domain. Proceedings of the National Academy of Sciences of the United States of America, 93(3), 1156–1160.

Lambowitz, A. M., & Belfort, M (1993). INTRONS AS MOBILE GENETIC ELEMENTS. Annual Review of Biochemistry, 62 (1) 587-622. doi: 10.1146/annurev.bi.62.070193.003103. https://www.annualreviews.org/doi/full/10.1146/annurev.bi.62.070193.003103

Laurila J, Laakso I, Valkonen JPT, Hiltunen R, Pehu E (1996). Formation of parental type and novel alkaloids in somatic hybrids between Solanum brevidens and S. tuberosum. Plant Sci, 118:145-155; http://dx.doi.org/10.1016/0168-9452(96)04435-4.

Lee, M. (2008). EU regulation of GMOs: Law and decision making for a new technology. EU Regulation of GMOs: Law and Decision Making for a New Technology. 1-274. 10.4337/9781848443969.00001

Lee J, Kim DH, Lee K (2020). Current Approaches and Applications in Avian Genome Editing. Int J Mol Sci., 21(11):3937. Published 2020 May 30. doi:10.3390/ijms21113937.
Lema M. A. (2019). Regulatory aspects of gene editing in Argentina. Transgenic research, 28(Suppl 2), 147–150. https://doi.org/10.1007/s11248-019-00145-2

Li, E., Bestor, T. H., & Jaenisch, R. (1992). Targeted mutation of the DNA methyltransferase gene results in embryonic lethality. Cell, 69(6), 915–926. https://doi.org/10.1016/0092-8674(92)90611-f

Li, T., Liu, B., Spalding, M. H., Weeks, D. P., & Yang, B. (2012). High-efficiency TALEN-based gene editing produces disease-resistant rice. Nature biotechnology, 30(5), 390–392. https://doi.org/10.1038/nbt.2199

Liu, Z., Chen, M., Chen, S., Deng, J., Song, Y., Lai, L., & Li, Z. (2018). Highly efficient RNA-guided base editing in rabbit. Nature communications, 9(1), 2717. https://doi.org/10.1038/s41467-018-05232-2

Malzahn, A., Lowder, L., & Qi, Y. (2017). Plant genome editing with TALEN and CRISPR. Cell & bioscience, 7, 21. https://doi.org/10.1186/s13578-017-0148-4

Mao, Y., Zhang, H., Xu, N., Zhang, B., Gou, F., & Zhu, J. K. (2013). Application of the CRISPR/Cas system for efficient genome engineering in plants. Molecular plant, 6(6), 2008–2011. https://doi.org/10.1093/mp/sst121
Media springer nature. Retrieved from: https://media.springernature.com/lw685/springer-static/image/art%3A10.1186%2F1759-8753-5-7/MediaObjects/13100_2013_Article_94_Fig1_HTML.jpg

Menchaca, A., Dos Santos-Neto, P. C., Mulet, A. P., & Crispo, M. (2020). CRISPR in livestock: From editing to printing. Theriogenology, 150, 247–254. https://doi.org/10.1016/j.theriogenology.2020.01.063

Mishra, R., & Joshi, R. K. (2019). Base editing in crops: current advances, limitations and future implications. Plant Biotechnology Journal, 18(1) 20 – 31. https://doi.org/10.1111/pbi.13225

Moro, L. N., Viale, D. L., Bastón, J. I., Arnold, V., Suvá, M., Wiedenmann, E., Olguín, M., Miriuka, S., & Vichera, G. (2020). Generation of myostatin edited horse embryos using CRISPR/Cas9 technology and somatic cell nuclear transfer. Scientific reports, 10(1), 15587. https://doi.org/10.1038/s41598-020-72040-4

Nagoya – Kuala Lumpur Supplementary Protocol on Liability and Redress (NP) (2011). Retrieved from https://www.cbd.int/abs/doc/protocol/nagoya-protocol-en.pdf

Nishida, K., Arazoe, T., Yachie, N., Banno, S., Kakimoto, M., Tabata, M., Mochizuki, M., Miyabe, A., Araki, M., Hara, K. Y., Shimatani, Z., & Kondo, A. (2016). Targeted nucleotide editing using hybrid prokaryotic and vertebrate adaptive immune systems. Science (New York, N.Y.), 353(6305), aaf8729. https://doi.org/10.1126/science.aaf8729

Oishi, I., Yoshii, K., Miyahara, D., Kagami, H., & Tagami, T. (2016). Targeted mutagenesis in chicken using CRISPR/Cas9 system. Scientific reports, 6, 23980. https://doi.org/10.1038/srep23980

Park, T. S., Lee, H. J., Kim, K. H., Kim, J. S., & Han, J. Y. (2014). Targeted gene knockout in chickens mediated by TALENs. Proceedings of the National Academy of Sciences of the United States of America, 111(35), 12716–12721. https://doi.org/10.1073/pnas.1410555111

Petersen, B., & Niemann, H. (2015). Advances in genetic modification of farm animals using zinc-finger nucleases (ZFN). Chromosome research : an international journal on the molecular, supramolecular and evolutionary aspects of chromosome biology, 23(1), 7–15. https://doi.org/10.1007/s10577-014-9451-7

Podevin, N., Davies, H. V., Hartung, F., Nogué, F., & Casacuberta, J. M. (2013). Site-directed nucleases: a paradigm shift in predictable, knowledge-based plant breeding. Trends in Biotechnology. 31, 375–383. doi: 10.1016/j.tibtech.2013.03.004

Proudfoot, C., Carlson, D. F., Huddart, R., Long, C. R., Pryor, J. H., King, T. J., Lillico, S. G., Mileham, A. J., McLaren, D. G., Whitelaw, C. B., & Fahrenkrug, S. C. (2015). Genome edited sheep and cattle. Transgenic research, 24(1), 147–153. https://doi.org/10.1007/s11248-014-9832-x

Ran, Y., Liang, Z., & Gao, C. (2017). Current and future editing reagent delivery systems for plant genome editing. Science China. Life sciences, 60(5), 490–505. https://doi.org/10.1007/s11427-017-9022-1

Retrieved from https://media.springernature.com/lw685/springer-static/image/art%3A10.1186%2F1759-8753-5-7/MediaObjects/13100_2013_Article_94_Fig1_HTML.jpg

Ribarits, A., Brüller, W., Hartmann, J., Hochegger, R., Mechtler, K., Peterseil, V., Söllinger, J., Stepanek, W., Widhalm, I., and Wögerbauer, M. (2014). Use of novel techniques in plant breeding and practical consequences concerning detection, traceability, labeling, and risk assessment. AgBioForum, 17, 183-190.

Richter, C., Chang, J. T., & Fineran, P. C. (2012). Function and regulation of clustered regularly interspaced short palindromic repeats (CRISPR) / CRISPR associated (Cas) systems. Viruses, 4(10), 2291–2311. https://doi.org/10.3390/v4102291

Rinaldo, A.R., & Ayliffe, M. (2015). Gene targeting and editing in crop plants: a new era of precision opportunities. Molecular Breeding, 35, 40. https://doi.org/10.1007/s11032-015-0210-z

Roberts, R. J., & Macelis, D. (1997). REBASE-restriction enzymes and methylases. Nucleic acids research, 25(1), 248–262. https://doi.org/10.1093/nar/25.1.248

Ryu, J., Prather, R. S., & Lee, K. (2018). Use of gene-editing technology to introduce targeted modifications in pigs. Journal of animal science and biotechnology, 9, 5. https://doi.org/10.1186/s40104-017-0228-7

Saner, Marc. 2007. What is Public Involvement? Paper presented during the High Level Policy Dialogue Workshop on Public Perception of Agricultural Biotechnology. Lima, Peru, October 4-5, 2007. Public involvement continuum also available at http://www.hc-sc.gc.ca/ahc-asc/public-consult/res-centre/process_e.html

Sarah M Schmidt, Melinda Belisle, Wolf B Frommer, (2020). The evolving landscape around genome editing in agriculture, EMBO Reports 21, 1-4, DOI10.15252/embr.202050680

Schusser, B., Collarini, E. J., Yi, H., Izquierdo, S. M., Fesler, J., Pedersen, D., Klasing, K. C., Kaspers, B., Harriman, W. D., van de Lavoir, M. C., Etches, R. J., & Leighton, P. A. (2013). Immunoglobulin knockout chickens via efficient homologous recombination in primordial germ cells. Proceedings of the National Academy of Sciences of the United States of America, 110(50), 20170–20175. https://doi.org/10.1073/pnas.1317106110

Shan, Q., Wang, Y., Li, J., Zhang, Y., Chen, K., Liang, Z., Zhang, K., Liu, J., Xi, J. J., Qiu, J., & Gao, C. (2013). Targeted genome modification of crop plants using a CRISPR/Cas system. Nature Biotechnology 31(8) 686 – 688. https://doi.org/10.1038/nbt.2650

Sirsingandla, I. (2020). CRISPR Prime Editing: The Next Step in Gene Editing. https://medium.com/@shri.52712.sivako/crispr-prime-editing-the-next-step-in-gene-editing-5d4a47b592b4

Smyth, S. J., McDonald, J., & Falck-Zepeda, J. (2014). Investment, regulation, and uncertainty: managing new plant breeding techniques. GM crops & food, 5(1), 44–57. https://doi.org/10.4161/gmcr.27465

Sugisaki, H., & Kanazawa, S. (1981). New restriction endonucleases from Flavobacterium okeanokoites (FokI) and Micrococcus luteus (MluI). Gene, 16(1-3), 73–78. https://doi.org/10.1016/0378-1119(81)90062-7

Tan, W., Proudfoot, C., Lillico, S. G., & Whitelaw, C. B. (2016). Gene targeting, genome editing: from Dolly to editors. Transgenic research, 25(3), 273–287. https://doi.org/10.1007/s11248-016-9932-x

Tzfira, T., Weinthal, D., Marton, I., Zeevi, V., Zuker, A., & Vainstein, A. (2012). Genome modifications in plant cells by custom-made restriction enzymes. Plant Biotechnology Journal 10(4) 373-389. https://doi.org/10.1111/j.1467-7652.2011.00672.x

Van Eenennaam AL. (2019). Application of genome editing in farm animals: cattle. Transgenic Res., 28(Suppl 2):93-100. doi: 10.1007/s11248-019-00141-6. PMID: 31321690

Wang, X., Niu, Y., Zhou, J., Zhu, H., Ma, B., Yu, H., Yan, H., Hua, J., Huang, X., Qu, L., & Chen, Y. (2018). CRISPR/Cas9-mediated MSTN disruption and heritable mutagenesis in goats causes increased body mass. Animal genetics, 49(1), 43–51. https://doi.org/10.1111/age.12626

Wells KD, Prather RS. (2017). Genome-editing technologies to improve research, reproduction, and production in pigs. Mol Reprod Dev., 84(9): 1012-1017. doi:10.1002/mrd.22812

Wendy C. (2016). Guidance for a fit for purpose regulatory framework for GMOS Posted to the Biosafety Clearing-House https://bch.cbd.int/database/record.shtml?documentid=115012

Whelan, A. I., & Lema, M. A. (2015). Regulatory framework for gene editing and other new breeding techniques (NBTs) in Argentina. GM crops & food, 6(4), 253–265. https://doi.org/10.1080/21645698.2015.1114698

Whelan, A. I., Gutti, P., & Lema, M. A. (2020). Gene Editing Regulation and Innovation Economics. Frontiers in bioengineering and biotechnology, 8, 303. https://doi.org/10.3389/fbioe.2020.00303

Xu, J., Yu, L., Guo, J. et al. (2019). Generation of pig induced pluripotent stem cells using an extended pluripotent stem cell culture system. Stem Cell Res Ther, 10, 193 https://doi.org/10.1186/s13287-019-1303-0

Yamano, T., Nishimasu, H., Zetsche, B., Hirano, H., Slaymaker, I. M., Li, Y., Fedorova, I., Nakane, T., Makarova, K. S., Koonin, E. V., Ishitani, R., Zhang, F., & Nureki, O. (2016). Crystal Structure of Cpf1 in Complex with Guide RNA and Target DNA. Cell, 165(4), 949–962. https://doi.org/10.1016/j.cell.2016.04.003

Yu, B., Lu, R., Yuan, Y., Zhang, T., Song, S., Qi, Z., Shao, B., Zhu, M., Mi, F., & Cheng, Y. (2016). Efficient TALEN-mediated myostatin gene editing in goats. BMC developmental biology, 16(1), 26. https://doi.org/10.1186/s12861-016-0126-9

Yu, S., Luo, J., Song, Z., Ding, F., Dai, Y., & Li, N. (2011). Highly efficient modification of beta-lactoglobulin (BLG) gene via zinc-finger nucleases in cattle. Cell research, 21(11), 1638–1640. https://doi.org/10.1038/cr.2011.153

Yum, S. Y., Youn, K. Y., Choi, W. J., & Jang, G. (2018). Development of genome engineering technologies in cattle: from random to specific. Journal of animal science and biotechnology, 9, 16. https://doi.org/10.1186/s40104-018-0232-6

Zetsche, B., Gootenberg, J. S., Abudayyeh, O. O., Slaymaker, I. M., Makarova, K. S., Essletzbichler, P., Volz, S. E., Joung, J., van der Oost, J., Regev, A., Koonin, E. V., & Zhang, F. (2015). Cpf1 is a single RNA-guided endonuclease of a class 2 CRISPR/Cas system. Cell, 163(3), 759–771. https://doi.org/10.1016/j.cell.2015.09.038

Zetsche, B., Heidenreich, M., Mohanraju, P., Fedorova, I., Kneppers, J., DeGennaro, E. M., Winblad, N., Choudhury, S. R., Abudayyeh, O. O., Gootenberg, J. S., Wu, W. Y., Scott, D. A., Severinov, K., van der Oost, J., & Zhang, F. (2017). Multiplex gene editing by CRISPR-Cpf1 using a single crRNA array. Nature biotechnology, 35(1), 31–34. https://doi.org/10.1038/nbt.3737

Zhang, D., Li, Z., & Li, J. F. (2016). Targeted Gene Manipulation in Plants Using the CRISPR/Cas Technology. Journal of genetics and genomics = Yi chuan xue bao, 43(5), 251–262. https://doi.org/10.1016/j.jgg.2016.03.001

Zhang, J., Cui, M. L., Nie, Y. W., Dai, B., Li, F. R., Liu, D. J., Liang, H., & Cang, M. (2018). CRISPR/Cas9-mediated specific integration of fat-1 at the goat MSTN locus. The FEBS journal, 285(15), 2828–2839. https://doi.org/10.1111/febs.14520

Zhang, J., Liu, J., Yang, W., Cui, M., Dai, B., Dong, Y., Yang, J., Zhang, X., Liu, D., Liang, H., & Cang, M. (2019). Comparison of gene editing efficiencies of CRISPR/Cas9 and TALEN for generation of MSTN knock-out cashmere goats. Theriogenology, 132, 1–11. https://doi.org/10.1016/j.theriogenology.2019.03.029

Zhang, Y., Liang, Z., Zong, Y., Wang, Y., Liu, J., Chen, K., Qiu, J. L., & Gao, C. (2016). Efficient and transgene-free genome editing in wheat through transient expression of CRISPR/Cas9 DNA or RNA. Nature communications, 7, 12617. https://doi.org/10.1038/ncomms12617

Zhang, Y., Massel, K., Godwin, I. D., & Gao, C. (2018). Applications and potential of genome editing in crop improvement. Genome biology, 19(1), 210. https://doi.org/10.1186/s13059-018-1586-y

Zhao, H., & Wolt, J. D. (2017). Risk associated with off-target plant genome editing and methods for its limitation. Emerging topics in life sciences, 1(2), 231–240. https://doi.org/10.1042/ETLS20170037

Zhu, B., & Ge, W. (2018). Genome editing in fishes and their applications. General and comparative endocrinology, 257, 3–12. https://doi.org/10.1016/j.ygcen.2017.09.011

Annexure 1. MORE INFORMATION ABOUT THE TECHNIQUES AND APPLICATIONS OF GENOME EDITING

Reference to genome editing application in this policy Guideline will include the use of all five genome editing applications. These are meganucleases, homing nucleases, zinc finger nucleases, TALENS, CRISPR/Cas system nucleases, and ODM. These are summarised as follows:

MEGANUCLEASES /HOMING ENDONUCLEASES (MN/HES)

Meganucleases enzymes are rare-cutting enzymes encoded by introns and inteins. The meganucleases, have been isolated and identified in archaea, bacteria, and eukarya as shown in Figure A and Table A (Belfort & Roberts, 1997). The MN and HEs usually consist of either a single monomer or a dimer. They have a central nuclease domain flanked by two DNA binding domains. The latter have recognition sequences of 12–40 bp (Roberts & Macelis, 1997); Mueller et al, 1993; Lambowitz & Belfort, 1993). The application of meganucleases in genome editing is curtailed by their limited number and low repertoire of naturally occurring recognition sequences. The latter number of recognition sequences has been amplified through creating mutations in their natural DNA recognition domain, or by fusing with sequence domains from different enzymes. Through this approach, a variety of different MN/HEs were developed in recent years (Podevin et al, 2012; Antunes et al, 2012; Gao et al, 2010; Tzfira et al, 2012).

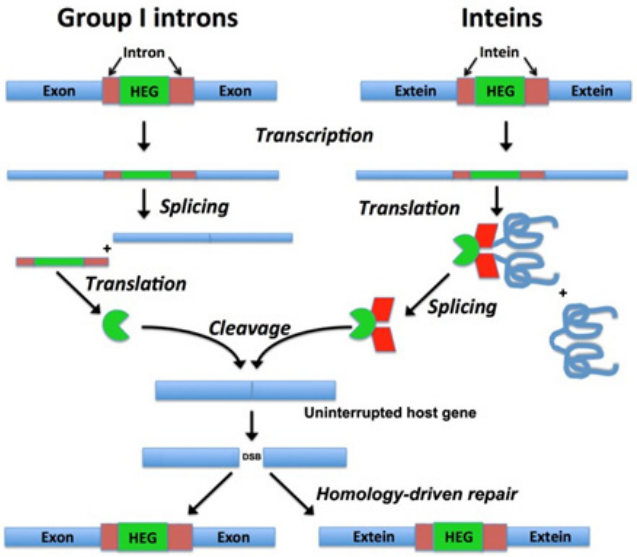


Figure A: Mechanism of the meganucleases enzymes in the rare-cutting process as encoded by introns and inteins (Source: media.springernature.com)

ZINC-FINGER NUCLEASES

Zinc-finger nucleases (ZFNs) are artificial restriction enzymes that are generated by fusing a zinc finger DNA binding domain and DNA cleavage domain. ZFNs are finger-like structures that can recognise the specific stretches of three nucleotides (nt) in a DNA sequence. They are linked together into arrays of 3-4 ZFNs as shown in Table A.

Table A. Summary notes of on genome editing enzymes systems

Enzymes	Source	DNA Binding Domain	DNA Cutting Domain/Cleavage	Remarks
Meganucleases	Archaea, bacteria and eukarya	Two DNA binding domains	One Nuclease domain	Have limited number of recognition sequences 12-40 bp
Zinc-finger nucleases	Artificial restriction enzymes – fusion of DNA binding and DNA cleavage domain	Zinc Finger DNA Binding Domain recognize 3 nt in DNA	DNA cleavage Domain usually endonuclease FokI	
TALENs	Dimeric enzymes nuclease domain fused to DNA binding Domain	DNA-binding domain	Nuclease domain fused	Nucleotide sequence of 30 nucleotides
CRISPR/CAS	DNA cleavage domain fused to Synthetic guide RNAs direct the nuclease activity	DNA binding domain consists of guide RNA	DNA cleavage domain consist of the CAS protein	Most used CRISPR/Cas system most used is CRISPR/Cas9 system

The Zinc finger DNA binding domain can be engineered to target desired, specific DNA sequences. When it is coupled with an endonuclease, usually Fok 1, it enables the latter to cut unique DNA sequences, based on the ZF recognition sites, within complex genomes. Fok 1 is isolated from Flavobacterium okeanokoites, a type II class of restriction endonuclease (Sugisaki & Kanazawa, 1981). Fok 1 binds to the cognate sequence 5′-GGATG-3′ and subsequently cleave the DNA phosphodiester bonds 9 bp away on this strand and the 13 bp away of the complementary strand to yield variable, four nucleotide 5'-overhangs (Li et al., 1992). The modular use of FokI in combination with ZF-domains has led into the development of artificial enzymes with new specificities (Kim et al, 1988; Kim et al, 1996; Kim et al, 1997; Boch and Bonas, 2010).

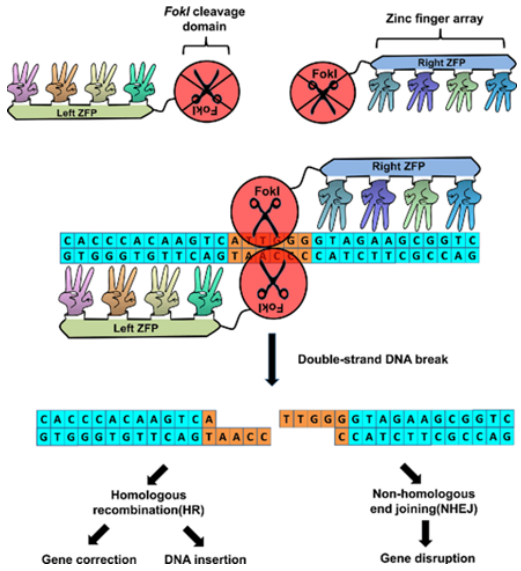


Figure B: The Fok 1 binding to the cognate sequence 5'-GGATG-3′ and subsequently cleaving the DNA phosphodiester bonds 9 bp away from the strand and the 13 bp away of the complementary strand to yield variable, four nucleotide 5'-overhangs (Source: Jo et al, 2015)

TRANSCRIPTION ACTIVATOR-LIKE EFFECTOR-LIKE NUCLEASES (TALENS)

The Transcription Activator-like Effector Nucleases (TALENs) are dimeric enzymes with a structure related to ZFNs. They have a nuclease domain fused to a DNA-binding domain as shown in Table A and Figure C. The FokI is usually used as a nuclease domain while the DNA binding domain consists of the transcription activator-like (TAL). On the other hand, the DNA-binding domain consists of an array of up to 30 modules. These are specific for a particular nucleotide sequence of 30 nucleotides. Due to their longer DNA recognition sites, the TALENs are specific for genomic locations. Thus, this causes fewer unwanted off-target effects than the ZFNs. The TALEN approaches were also applied for modification of plant and animal genomes (Li et al., 2012).

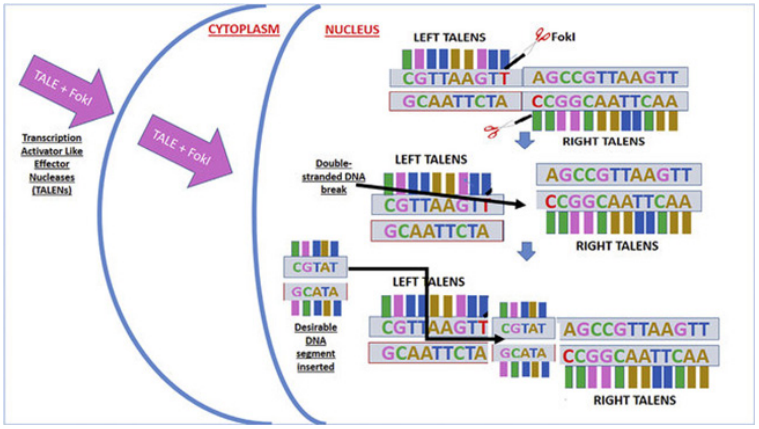


Figure C: The Transcription Activator-like Effector Nucleases (TALENs) as dimeric enzymes with a ZFNs related structure and the nuclease domain fused to a DNA-binding domain and the transcription activator-like (TAL) (Source: Khan, 2019)

CLUSTERED REGULARLY INTERSPACED SHORT PALINDROMIC REPEATS (CRISPR)/CAS NUCLEASE

Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)/Cas nucleases are molecular scissors used in genome editing (Figure D). They have a DNA binding domain that consists of a guide RNA while the DNA cleavage domain consist of a CAS protein. The most utilised CRISPR/Cas system is the CRISPR/Cas9 system (Table A). The Cas9 protein is a DNA endonuclease that utilises a guide RNA to target and cleave DNA. Native Cas9 utilises a guide RNA composed of crRNA and trans-activating CRISPR RNA (tracrRNA). The CRISPR/Cas-based nucleases are also the most used systems for editing in plant and animal genomes (Shan et al, 2013; Mao et al., 2013).

Apart from the CRISPR/Cas nucleases binding to a particular genomic DNA sequence by guide RNAs, the enzyme can also accept specifically designed synthetic guide RNAs modelled on the Cas9 guide RNA. The CRISPR/Cas complex's gRNA sequences can be redesigned to target any specific sequence in a genome to allow for NHEJ-based or HDR-based editing in that site. This versatility allows for a multitude of different target sequences and different genome sites to be targeted. As such, there have been ongoing improvements of the CRISPR/Cas System to improve efficacy and reduce off target effects (Mishra & Joshi, 2019; Anzalone et al, 2020; Anzalone et al, 2019; Sirsingandla, 2020; Zhang et al).

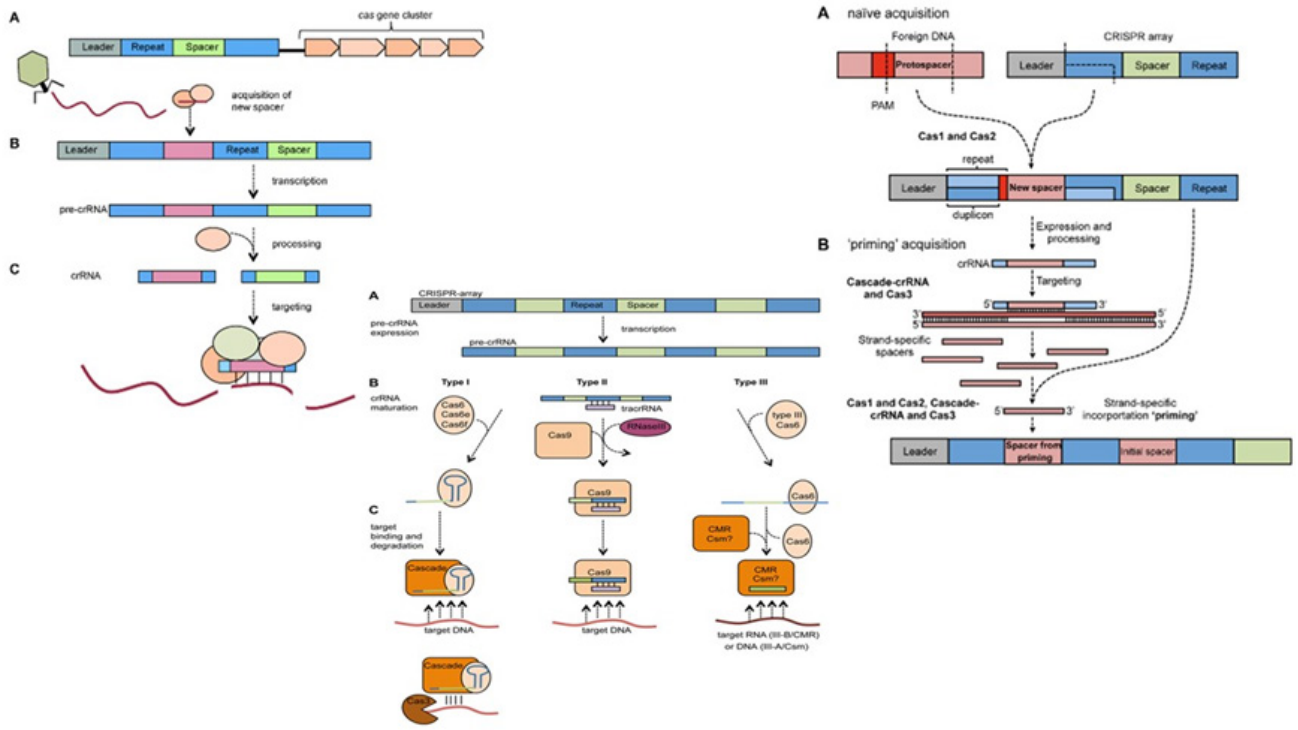


Figure D: Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)/Cas nucleases acting as molecular scissors used in genome editing (Source: Richter et al, 2012)

OLIGONUCLEOTIDE-DIRECTED MUTAGENESIS (ODM)

Genome editing can also be achieved using a process called Oligonucleotide-Directed Mutagenesis (ODM). It has been observed that when a short single strand DNA sequence, called an oligonucleotide is complementary to a specific target DNA sequence in a genome, except for one or two nucleotides, mutation is introduced into a host cell. The host organism will use this short DNA as a template when replicating its own DNA. The sequence of the oligonucleotide will be incorporated into the host DNA together with the mutation. This ODM is another method used for genome editing, specially to make economically important mutation of 1-2 bases.

Annexure 2. RECENT IMPROVEMENTS IN CRISPR/CAS GENOME EDITING

Four recent developments to improve the efficacy of CRISPR/Cas systems for use in genome editing include the use of Base editors, priming editors, DNA free editing, and CRISPR/Cas12a.

BASE EDITING

Base editors are obtained through fusion of catalytically inactive CRISPR/Cas9 domains (Cas9 variants, dCas9 or Cas9 nickase) and a cytosine or adenosine deaminase domain which converts one base to another (Yum et al, 2018). This enables the CRISPR/CAS system to generate precise point mutations in genomic DNA and RNA with and enhanced scope, specificity, and precision. This also helps with an in vivo delivery of base editors. The base editing components from the CRISPR systems, together with other enzymes, directly induce point mutations into cellular DNA or RNA without making double-stranded DNA breaks.

Notably, these genome editing tools comprise of catalytically disabled nucleases fused into nucleobase deaminase enzymes. In some cases, DNA glycosylase inhibitors are utilised (Yum et al, 2018). The base editing tools enable for direct and irreversible conversion of one base pair into another. This is accomplished without the need for introducing the DSBs or template DNA (Yum et al, 2018; Nishida et al, 2016; Gaudelli et al. 2017). Furthermore, base editors, such as the Cytosine base editors (CBEs) and Adenine base editors (ABEs), are mainly composed of a deaminase fused to a Cas9 variant: [nickase Cas (nCas9) or catalytically deactivated Cas9 (dCas9)].

PRIME EDITING

Prime editing uses a catalytically impaired Cas9 endonuclease fused to an engineered reverse transcriptase, programmed with a prime editing guide RNA (pegRNA). This can specifically target the site and encode the desired edit (Yum et al, 2018). Prime editing has higher or similar efficiency, fewer by-products than homology-directed repair (HDR). Furthermore, it has similar strengths and weaknesses when compared to base editing. It can also induce much lower off-target editing than Cas9 nuclease at known Cas9 off-target sites (Yum et al, 2018). The RNA that Prime Editing RNA uses (pegRNA), unlike the CRISPR RNA (sgRNA), uses the guide pegRNA in Prime Editing. This also contains the edited gene that will be inserted as a part of the process (Yum et al, 2018).

DNA FREE GENOME EDITING

DNA free genome editing involves inducing genome changes without introducing the use of any foreign genetic material sequences in an intermediate step. This approach eliminates any chances of unintentionally introducing foreign genetic material sequences into the edited organism, which may have regulatory consequences. This has been demonstrated with transfected preassembled complexes of purified Cas9 protein and guide RNA into plant protoplasts of Arabidopsis thaliana, tobacco, lettuce, and rice. The techniques achieved the intended and targeted mutagenesis in regenerated plants (Yum et al, 2018). The DNA-free delivery has been developed by transfecting preassembled CRISPR/Cas9 ribonucleoproteins into protoplasts4 or in vitro fertilized zygotes5 (Yum et al, 2018). Additionally, the CRISPR–Cas ribonucleoproteins or RNA transcripts have been biolistically bombarded into immature embryo cells or calli to yield highly specific genome editing, albeit at low frequency (Yum et al, 2018).

CRISPR/CAS12A GENOME EDITING

Cpf1 is a class 1, type-V CRISPR/Cas effector endonuclease can exhibit gene-editing using a single RNA-guided approach. The Cpf1 has been reported to cause fewer off-target cleavages when compared to Cas9 (Yum et al, 2018). To exert sequence-specific endonuclease activity, the Cpf1 is functional through a single crRNA and without an additional tracrRNA (Zetsche et al, 2015; Fonfara et al, 2016; Zetsche et al, 2017). The Cpf1 protein can interact with the pseudoknot structure formed by the 5′-handle of crRNA (Yamano et al, 2016; Gao et al, 2016). A guide segment, composed of a seed region and 3′ terminusi, possesses complementary binding sequences with the target DNA sequences. This protein–RNA complex recognizes a T-rich protospacer-adjacent motif and leads to a staggered DNA double-stranded break (Yamano et al, 2016). This kind of techniques have found applications in genome editing and biotechnology.

Annexure 3. MECHANISM OF GENOME EDITING USING SITE DIRECTED NUCLEASES

The goal of Site Directed Nuclease Technology is to take advantage of the targeted DNA break and the host cell's endogenous DNA repair mechanisms to induce specific changes at the site of the DNA break that are of economic importance. The change can either be a small deletion, a substitution, or the addition of several nucleotides. Such targeted edits result in a new and desired characteristic, such as enhanced nutrient uptake or decreased production of allergens. The initial applications of SDNs are divided into three categories: SDN1, SDN2 and SDN3 (Box A). While this was an initial classification scheme, there are some new applications of genome editing that do not fit into one of these categories (e.g. chromosomal inversions and epigenetic changes). Understanding the different mechanisms and products obtained from the three SDN applications and the technical difficulties and degree of ambiguity in classification is important in defining the regulatory approach to adopt.

SITE DIRECTED NUCLEASE 1 (SDN1)

For SDN1, the SDN is introduced into the cell without addition of a DNA template (Figure E). The DNA binding domain of the programmed SDN searches and binds to a complimentary DNA sequence and the DNA nuclease domain makes a DSB at the target site. The cell's own endogenous repair mechanism joins the broken ends by a process called non-homologous end joining (NHEJ) that is error prone. The spontaneous repair of DSB can lead to a mutation, causing gene silencing, gene knock-out, or a change in the activity of a gene. This SDN1 technology is the more widely used today to improve crops, animals, and microorganisms of economic importance (CropLife).

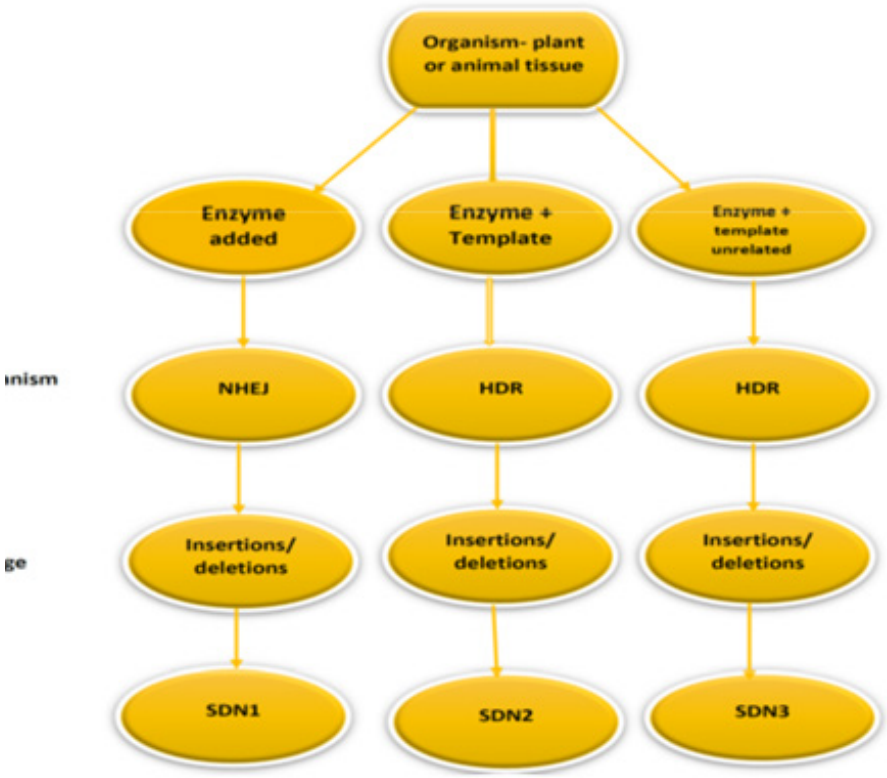


Figure E: The processes of Site Directed Nucleases (SDN) activity showing – SDN1, SDN2, and SDN3 because of Non-Homologous End Joining (NHEJ) and Homology Directed Recombination (HDR)

SITED DIRECTED NUCLEASE 2 (SDN2)

In SDN2 type edits, the engineered SDN is introduced into the recipient cell together with a DNA repair template that is complementary to the target sequence of the recipient except for a few bases. The DNA binding domain of the SDN binds to the complementary DNA in the host cell, while its DNA cleavage domain makes double strand DNA breaks at the target sequence. The cell's endogenous DNA repair process repairs the DNA break using the introduced DNA repair template through a mechanism referred to as HDR. Since the complementary template is used to repair the DNA DSB, a designed short mutation is generated in the target gene including the desired sequence changes. This results in specific genetic changes at the target site in the recipient organism to impact the function of the targeted sequence in a desired way. It is important to note that this approach of using a longer repair template can also introduce cisgenes which may be similar to the genetic variation that could be obtained with conventional techniques. This mechanism is somewhat used for developing crops, animals, and microorganisms of economic importance.

SITE DIRECTED NUCLEASE 3- (SDN3)

The SDN3 is like SDN2 except that the repair template added could contains longer inserts, even complete genes from unrelated organism, i.e., cis/transgenes. Both SDN2 and SDN3 use HDR mechanisms. The cell's natural repair process then utilizes this template to repair the break resulting in the introduction of the genetic material. Unlike SDN1 and SDN2, the SDN3 can be used to introduce foreign genetic material. However, SDN1, SDN2, and SDN3 can raise variable biosafety and concomitant regulatory considerations as will be elucidated later.

Annexure 4. METHOD OF DELIVERY OF SDNS INTO RECIPIENT ORGANISMS

The methods used to introduce SDNs into organisms vary among plants, animal species, microorganisms, and even the tissues into which SDNs are introduced.

METHODS OF INTRODUCING GENOME EDITING IN PLANTS

In plants, SDNs can be introduced through polyethylene glycol DNA uptake, Agrobacterium DNA transformation, or biolistic methods for delivering DNA or Protein/RNA complexes.

METHODS OF INTRODUCING GENOME EDITING IN ANIMALS AND TARGET TISSUE

In birds, the avian primordial germ cells are the most preferred target tissue for genome editing and are genome edited by direct transfection through microinjection (Han and Park, 2018). In cattle, genome edited target tissues include somatic cell nuclear transfer (SCNT), bovine foetal fibroblast (BFF) and sometimes zygotes while methods of delivering genome editing sequences include electroporation, cytoplasmic injections (CPI), nucleofection (Yum et al, 2018; Yu et al, 2011). In sheep, key target tissues include zygotes and to lesser extent, SCNT while methods of delivering genome editing sequences include CPI, electroporation, nucleofection, pronuclear injection (Proudfoot et al, 2015). For goats, tissue used for genome editing include zygotes, SCNT, goat foetal fibroblast while methods used include CPI, electroporation, and nucleofection (Yu et al, 2016). Pigs have been genome edited using somatic cell nuclear transfer using Talens, ZFN and CRISPR/CAS 9 (Ryu et al, 2018), while in rabbits' successful genome editing has been reported (Hammer et al, 1985).

Annexure 5. EXAMPLES OF GENOME EDITING APPLICATIONS IN ANIMALS

GENOME EDITING APPLICATIONS IN PIGS

Pigs are the preferred large animal models in biomedical research and considered a better choice over the rodent models. Furthermore, the transplantation studies using pig pluripotent stem (PS) cell derivatives have demonstrated a better testbed for safety and efficacy prior to human trials (Xu et al., 2019). However, the pigs have limited true embryonic stem cells. Thus, genome editing is accomplished through somatic cells and somatic cell nuclear transfer (Ryu et al, 2018). Notably, the pigs' genome editing has been accomplished through genome editing tools such as ZFNs, TALENs, and the CRISPR/Cas systems (Ryu et al, 2018). The three SDN applications have been successfully achieved in pigs using genome editing tools such as ZFN, TALENS, and CRISPR/CAS9.

The ability to directly manipulate the pig genome through genetic engineering for research purposes has advanced from the haphazard insertion of foreign genetic material, through techniques such as pronuclear microinjection, sperm mediated gene transfer, and integration of mobile genetic elements, to the manipulation of endogenous genes. This is accomplished through homologous recombination in somatic (vegetal) cells followed by somatic cell nuclear transfer. In recent years, newly designed genome editing nucleases have facilitated the development of techniques that can provide efficient ways to introduce foreign genetic material (Wells and Prather, 2017). Consequently, this is enabling scientists modify endogenous genes in eggs, zygotes, and somatic cells. Together, these genome editing technologies have essentially enabled genome manipulation in swine. Even though the regulatory environment remains uncertain for agricultural applications, genetic engineering of pigs is envisaged to advance biomedicine and biology. As such, the genome editing techniques are now sufficient, simple, and efficient to efficiently advance agricultural basic and applied research without limitations on funding.

GENOME EDITING APPLICATIONS IN BIRDS

Birds are an important biological and agricultural species, and their genome editing has gained significant interest for scientists. The genome editing applications are primarily performed by using a primordial germ cell (PGC)-mediated method. This is because the pronuclear injection is not practical in the avian species. Notably, this is different from the approach used in genome editing in mammals (Han & Park, 2018). In this method, the PGCs are isolated, cultured, genetically edited in vitro. Thereafter, they are injected into the recipient embryo to yield a genome edited offspring.

For example, genome edited quails have been produced by utilising the newly developed adenovirus-mediated method. As such, a direct injection of adenovirus into the avian blastoderm in the freshly laid eggs was undertaken without technically employing in vitro procedures of the PGC-mediated method. This resulted into the generation of germ-line chimera and genome edited offspring. Notably, as more approaches are made available in avian genome editing, the avian research in various fields is expected to progress rapidly (Lee et al., 2020).

The primordial germ cells were first discovered by Waldeyer et al, (1870) as cited in Ryu et al, (2018). They were later reported to originate from an endodermal region called the germ cell wall (Ryu et al, 2018). The three genome editing systems known as ZFNs, TALENs, and CRISPR)/Cas9 (Ryu et al, 2018) have potential applications in birds. For instance, the development of avian genome editing has scientific and industrial applications for genome edited avian species. These include the removal of the myostatin gene and single amino acid deletion in myostatin propeptide to enhance muscle mass for chicken and quail.

Further to this, other genome editing techniques have been undertaken to improve the chicken's resistance to leucosis virus sub-group, exhibition of gray feather color, expression progenesis for sexing, reduction of abdominal fat deposition, and production of human interferon beta (β) in the egg white, among others. In addition, the genome edited progenitor cells of gametes have been reported in chicken using HDR (Schusser et al, 2013), TALENS (Park et al, 2014), and CRISPR/Cas systems (Oishi, 2016; Han & Park, 2018).

GENOME EDITING APPLICATIONS IN CATTLE

Milk and meat from cattle and buffaloes have contributed about 45% of global animal protein supply. These are followed by chickens (31%), and pigs (20%). As such, in 2016, the global cattle population of 1.0 billion head generated approximately 6.5 billion tonnes of cows' milk, and 66 million tonnes of beef (Van, 2019). In some regions of the world, the recent cattle breeding programmes have critically bolstered the yield per animal with a resultant decline in the emissions intensity per unit of milk and beef. Consequently, the genome editing research in cattle has primarily focused on disease resistance such as tuberculosis and production focusing mainly on myostatin knockout and production of all-male offspring. The genetic editing has also focused the elimination of allergens such as beta-lactoglobulin knockout and enhancing welfare traits such as hornlessness.

Modeling has also exposed efficient methodologies of genome editing that are capable of introducing beneficial alleles into cattle breeds. This could maintain and further fast-track the rate of genetic gain that can be attained by conventional breeding programmes. The genome editing approach can hasten the lengthy process of introgression of the same alleles from distant breeds, i.e., the transfer of genetic information from one species to another through the hybridisation process and repeated backcrossing. As such, genome editing can accurately introduce useful alleles into the organism. For example, they can improve heat tolerance and disease resistance of animals, and haplotypes into native and locally altered cattle breeds. This can subse-

quently improve their productivity. However, employing these genome editing approaches in cattle genetic improvement programmes should depend on robust and comprehensive continental and global decisions on regulatory and governance frameworks of genome editing for food animals.

Genome editing in cattle is based on micro-injection of somatic nuclear cell transfer cells (SNCT) due to absence of stem cells (Yum et al, 2018). The three genome editing methods used are ZFNs (Yu et al, 2011), TALENs (Proudfoot et al, 2015), and CRISPR)/ Cas9 (Tan et al, 2016; Ryu et al, 2018) with the latter being the most preferred method. Somatic nuclear cell transfer cells substituted the previous DNA micro-injection of zygotes, and subsequently transferred into recipient reformed severe mosaicism. This technique has a low efficiency (Yum et al, 2018).

GENOME EDITING APPLICATIONS IN SHEEP, GOATS, RABBITS, AND HORSES

Sheep, goats, and rabbits are valuable livestock species that are reared for their meat, milk, fiber, and other by-products. Because of their appropriate sizes, short gestation period, and abundant production of milk, sheep and goats have received tremendous importance in model animals for agricultural, pharmaceutical, and biomedical research. As such, genome editing has been widely employed in sheep and goat research. On the other hand, rabbits have also gained interest for their supply of protein. Moreover, horses are raised for entertainment and transportation. For example, the pronuclear injection and somatic cell nuclear transfer has represented primary procedures for genetic editing in sheep and goats. Further to this, assisted tools have also been developed to improve the proficiency of genetic editing. This is further simplifying the generation of genetically modified founders. These genetic editing tools include sperm-mediated gene transfer, viral vectors, RNA interference, recombinases, transposons, and endonucleases.

Gene-edited sheep and goats that are generated using the four classes of site-specific endonucleases (meganucleases, ZFNs, TALENs, and CRISPRs) are enabling valuable investigations on gene functions, advancing animal breeding, generating pharmaceuticals in milk, and enhancing animal disease resistance. In addition, some derivative tools of CRISPR systems have appeared such as base editors which facilitate the introduction of single-base alterations without any prerequisites for homology-directed repair and DNA donor (Kalds et al, 2019). These precise editors are beneficial in enabling advantageous phenotypes and rectifying genetic diseases controlled by single bases.

In goats, successful genome editing based on TALENS (Yu et al, 2016) and CRISPR/CAS systems has been reported. Both TALENS and CRIsPR/CAS 9 have successfully gene edited goats and DNA delivery was through electotransfections and nuclear transfer of somatic cells (Zhang et al, 2019; Zhang et al, 2018). In addition, in goats CRISPR/CAS introduced through microinjection has been used successfully (Kalds et al, 2019; He et al, 2018). Although genetic modification of large animals is recalcitrant, microinjection has been used for genome editing of sheep (Hammer et al, 1985) using CRISPR/Cas9(Crispo et al, 2015), and CRISPR/CAS of somatic nuclear cell transfer (Han et al, 2014; Wang et al, 2018).
In rabbits (Hammer et al, 1985; Liu et al, 2018) successful genome editing is based on CRISPR/CAS and use of improved CAS with base editors (Liu et al, 2018). There are also precedents of genome editing in horses as the application of new technologies for genome editing in horses may allow the generation of improved sportive individuals. The aim was to knock out the myostatin gene (MSTN), a negative regulator of muscle mass development, using CRISPR/Cas9 and to generate edited embryos for the first time in horses (Moro et al, 2020).

GENOME EDITING APPLICATIONS TO IMPROVE FISHERIES AND AQUACULTURE

Aquaculture is rapidly growing and becoming the primary source of seafood for human diets. Thus, selective breeding programmes are facilitating genetic improvement of essential economic traits such as disease resistance. However, progress remains limited because of the heritability of the traits and generation interval of the species. As such, new breeding techniques, such as genome editing using CRISPR/Cas9, can potentially accelerate sustainable genetic enhancement in aquaculture. Genome editing is potentially introducing essential and beneficial variations to the genome. These includes fixing alleles at existing trait loci, designing de novo alleles, and introducing alleles from other strains or species. The high productiveness and external reproduction of most aquaculture species can accelerate genome editing for research. This can also enhance application capacity to enhance aquaculture farming (Gratacap et al, 2019).

Genome editing using CRISPR/Cas has been successful in many aquatic species including atlantic salmon, tilapia, channel cut fish, trout, among others (Gratacap et al, 2019; Zhu & Ge, 2018). The genetic editing methods used included injection of the CRISPR/Cas9 complex into newly fertilised eggs at the one-cell stage of development (Zhu & Ge, 2018). The target traits have included sterility and disease resistance (Zhu & Ge, 2018).

Notes

AUDA-NEPAD
AFRICAN UNION DEVELOPMENT AGENCY

AUDA-NEPAD
230, 15th Road, Randjespark
Midrand, Gauteng
South Africa
Telephone +27 (0) 11 256 3600
www.nepad.org