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# **Application of Genomics and Phenomics in Plant Breeding for Climate Resilience**

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## **Author's contribution**

*The sole author designed, analysed, interpreted and prepared the manuscript.*

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## **ABSTRACT**

Advances in the fields of genomics and phenomics are currently creating significant foundations for the sustainable intensification of plant breeding initiatives targeting climate resilience. Genomics is a biological study that focuses on architecture, function, editing, mapping, and evolution of genomes. It can be applied extensively in climate resilience breeding for cost-effective, rapid, and high-through put genotyping, phenotyping, and trait mapping. The efficacy of genomics-assisted breeding (GAB) is strongly hinged on the high resolution and robustness of Next Generation Sequencing (NGS) and CRISPR/Cas9-based Gene Editing systems. The integration of genomics and phenomics in crop improvement can upscale the efficiency of breeding systems targeting climate resilience and hasten cultivar release cycle. Phenomics is an interdisciplinary field that focuses on the enhanced measurement of plant performance, growth, and composition. Similarly, phenomics has revolutionized the efficacy of plant breeding off-trial initiatives established to phenotypically characterize and study diversity levels of collected germplasm. Field phenomics tools such as the phenonet, phenomobile, and phenonetnetwork have proven to be efficient in capturing large sums of multiscale and multidimensional experimental data. The main purpose of this review article is to present a summarized account of the probable applications of integrated systems of genomics and phenomics in plant breeding for climate resilience in major crops.

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## 1. INTRODUCTION

Climate resilience is integral in the sustainability of livelihoods and cropping systems in drought-prone and poverty-stricken communities. Currently, the levels of malnutrition and food insecurity are skyrocketing at a pace that requires our undivided attention. A report compiled by the Intergovernmental Panel on Climate Change (IPCC) highlighted that the projected climate change will reduce the yield margins of maize, wheat, and rice a rate of 2% per decade starting from the year 2030 [1]. The agricultural industry, therefore, requires a 2<sup>nd</sup> green revolution to cater for the increased food, fibre, fuel and fodder needs of the growing global population [2,3] while being more climate-resilient on finite land, nutrient and water resources. Plant breeding possesses an enormous potential to curtail the forecasted biotic and abiotic constraints in crop production. However, reliance on conventional or traditional breeding approaches has proven to be time-intensive and ineffective in providing immediate solutions to the negative influences of climate change on crop productivity. Ndlovu et al. [4] asserted that, climate change and its associated negative effects call for the adoption and application of result-oriented and novel scientific innovations. Genomics and phenomics are bridging the gap and providing efficient tools for the direct selection of yield and climate resilience traits derived from large germplasm collections through software integrations [2,5]. It is, therefore, imperative to invest more on the advocacy and implementation of these technologies for upscaling the efficiency of crop improvement systems.

Genomics is a facet of science which deals with the structure, evolution, function, editing, and mapping of genomes [6,7]. On the other hand, crop phenomics is a multidisciplinary study of the accurate and high-throughput acquisition and analysis of phenotypes at a multidimensional and field-wide scale throughout the development of the crop [8]. Plant phenomics adoption over the last decade has been a worldwide phenomenon, as exhibited by a burgeoning number of 'plant phenomics centres' that are generating next-generation solutions to phenotyping for plant breeding [9,10]. Additionally, genomics and phenomics are yielding significant successes and can be utilized in tandem to further heighten the

efficiency of breeding systems targeting climate resilience.

The adaptation of crop production systems to global climate change and its associated ecological pressures require wild plant relatives, novel trait combinations, and formerly unknown phenotypic traits to be deployed into climate-resilient crop improvement initiatives [11,12]. Furthermore, climate forecasting frameworks that use high resolution and robust models should be incorporated to act as a guiding framework in crop growth pattern simulations targeting the influence of biotic and abiotic stresses. At the present moment, limited data has been generated to ascertain the effect of biotic and abiotic stressors on cultivated crop species under climate change [13,14,15]. Bridging the existing knowledge gap can create a stable platform that can sustain future breeding for climate resilience. Furthermore, such information will ensure that improved varieties stay relevant for a significant period under shifting climatic conditions.

Varshney et al. [16] underscored that, in the context of breeding for climate-resilient crops, it is paramount to identify, mine and incorporate elite alleles adapted to adverse and extreme climatic conditions through resequencing and phenotypic characterization of numerous crop accessions. Next-generation sequencing and gene editing can mine and incorporate superior alleles conferring various traits of interest (e.g. drought tolerance). Integrating genomics with phenomics in plant breeding can enhance the efficiency of phenotypic characterization procedures. Varshney et al. [16] alluded that, climate-resilient crop development with accelerated genetic gain requires the integration of different technologies and disciplines. The integration reduces the long gestation period required in the transfer of elite genes during inter specific crosses or introgression procedures. Furthermore, a genomics and phenomics merger in crop improvement can catapult our response to climate change effects in crop production systems. The objective of this review are to give a brief outline of the applications and associated challenges of genomics and phenomics in breeding for climate resilience in major crops, and to assess the possibility of integrating genomics and phenomics tools in crop breeding for climate resilience.

## 2. APPLICATION OF GENOMICS IN PLANT BREEDING FOR CLIMATE RESILIENCE

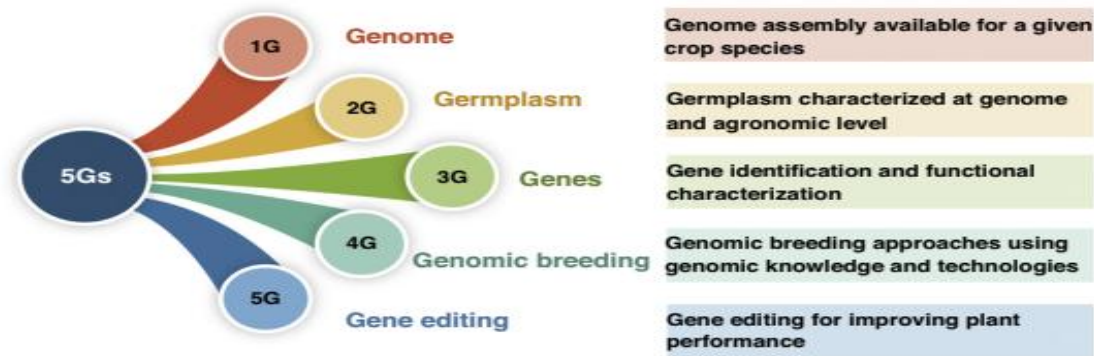
Genomics defines a wide variety of tools, methods, and technologies utilized in studying and manipulating genomes. The science of genomics can be divided into two broad groups namely: structural and functional genomics. Structural genomics seeks to gain an insight into the structural organization of the whole genome using computational modelling or experimental approaches for accurate identification and characterization of genes [17]. Structural genomics encompasses sequencing and mapping, genome organization and comparative genomics. Conversely, functional genomics is a field of molecular biology that focuses on understanding gene expression through assessments on the existing relationship between the genome and phenotype of the organism. Climate resilience-oriented breeding initiatives should therefore place more emphasis on exploring the application of genomics-assisted systems in selecting for various traits of interest. Structural and functional genomics can produce relevant data on gene morphology, location, and expression patterns which are of greater importance in improving desirable traits (for instance drought, salinity, and heat stress tolerance). The generated information can be utilized in making timely and informed trait selection decisions.

Sanger Sequencing was amongst the first-generation sequencing technologies to be employed in assembling several plant genomes. However, its wide adoption was thwarted by the associated high cost in operation and its low throughput attributes. The recent introduction of NGS has revolutionized genomics studies, made high-throughput sequencing of the whole-genome possible and less cost-intensive for many breeding schemes [10,18]. Advances in NGS and genotyping technologies have also promoted the generation of large-scale genomic resources, for instance, molecular markers, BAC-end sequences (BESs) and transcript reads in semi-arid tropic leguminous crops such as chickpea, groundnut and pigeon pea [19,20]. Moreover, the reinforcement of NGS with state-of-the-art OMICS technologies and latest genome editing tools are opening new avenues of possibilities in precise genome engineering and paving the way for targeted mutagenesis [21,22]. However, the application of these systems in breeding for climate resilience is still on the low side.

The availability of genomics-oriented systems needs to be capacitated with necessary breeding platforms or plans to ensure successful executions in climate resilience breeding. Varshney et al. [23] proposed a 5G breeding approach for accelerating and promoting disruptive shifts to crop improvement programmes (shown in Fig. 1). The 5Gs are Genome assembly (GA), Germplasm characterization (GE), Gene function identification (GFI), Genomic breeding (GB), and Gene editing (GE). The availability of crop genome assemblies and characterized germplasm accessions is crucial in the identification of superior haplotypes and marker-trait accessions in any breeding programme. Genome assembly developments in NGS technologies packaged with improved assembly algorithms have facilitated the *de novo* assemblies of greater than 264 genomes, including crops such as maize, wheat, rice, barley, cotton, sorghum, soybean, tomato, chickpea, pigeon pea, and groundnut [23]. Breeding for climate resilience needs to adopt these five elements and incorporate them as a guide to the release of improved varieties under genomics-assisted breeding schemes.

### 2.1 Genomics-Assisted Breeding (GAB)

Genomics-Assisted Breeding (GAB) methodologies encompass an array of stages including characterization of germplasm collections; mapping population development; genomic region identification through association or genetic mapping; and application of molecular markers [24,25]. GAB rose to popularity in the field of plant breeding in the past few years partly due to the presence of low-cost NGS and high-throughput genotyping (HTPG) technologies [25] which allowed the rapid identification of superior genes implicated in climate resilience [26,3]. The fight against climate change needs immediate responses to counter the associated negative effects on crop productivity. The advancement of genomics promotes the development of NGS-based trait mapping procedures, which are speeding up trait mapping initiatives from years to months (Afzal et al., 2020; Bekele et al., 2019; [16]). Smallholder farmers are in dire need of more tolerant varieties that can perform better under the prevailing adverse conditions. It is therefore imperative to establish breeding protocols that incorporate the genomics-assisted tools in tackling productivity constraints under the adverse climate change effects.



**Fig. 1. The 5Gs crop breeding approach**

Source: Varshney et al. [23]

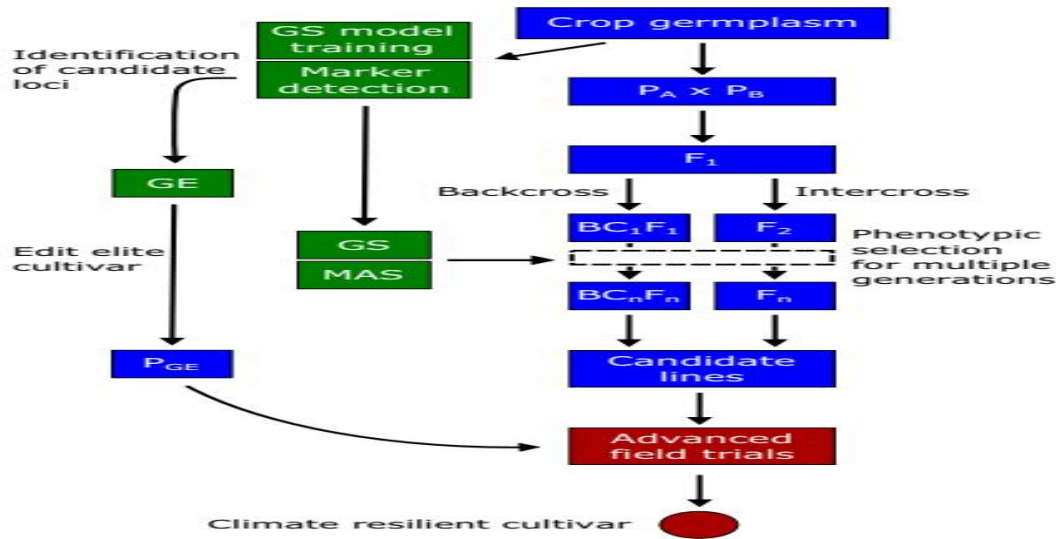
Varshney et al. [25] asserted that, efficient and effective analytical and decision support tools are crucial for the successful implementation of GAB in the evaluation and whole-genome selection of next-generation crops. Furthermore, the large sums of data generated through GAB require efficient systems of informatics and analytics for data visualization and interpretation. Analytical and decision support tools (ADSTs) refer to a wide variety of computer-based tools (algorithms, simulation models, techniques, and/or methods) generated for dataset analysis and selection of superior genotypes in GAB schemes [25]. ADSTs allows breeders and bioinformaticians to conduct genomic sequence comparisons for the identification and characterization of functional genomic regions. The comparisons of re-sequenced data against standardized reference genomes allow for the detection of existing haplotype and allelic diversity across genotypes [27]. The existing genetic or allelic diversity is an integral component of breeding programmes targeting various traits including climate resilience. Moreover, genotyping of numerous genetic samples or selected recombinants from the targeted population either for product development or trait mapping is an ultra-critical step in the identification of superior lines and/or better alleles for climate-resilient crop breeding [16].

Scheben et al. [3] illustrated how GAB can be utilized in the development of climate-resilient cultivars on a diagrammatic illustration shown in Fig. 2. The proposed approach follows the conventional breeding format with the utilization of genomics being initiated at the parental selection and gene editing stages. In the available germplasm, genebank curators and plant breeders look for physiological and morphological traits that could improve crop

tolerance under climate variability [5]. The diagrammatic presentation shows the genome-edited cultivars and selected parental cross ( $P_a \times P_b$ ) in blue. The Genomic methodologies that support traditional backcrossing ( $BC_1F_1$  to  $BC_nF_n$ ) and intercrossing approaches ( $F_2$  to  $F_n$ ) are represented in green. In the final stage (shown in red) genome-edited varieties and candidate lines were chosen from successive backcross or intercross generations are screened for wide viability and performance in advanced multi-environment off-site trials to climate-resilient varieties. Additionally, the genotypic assessment may be carried out in the off-season (e.g. in winter nurseries), where yield trials are not normally performed, which helps in speeding up breeding schemes [28]. However, the implementation and utilization of GAB, especially for inbreeding or minor crops, is currently constrained by the associated high costs.

## 2.2 Gene Editing

Gene editing (GE) methodologies can be employed in the modification of genomes with defined quantitative trait nucleotides (QTNs) thereby influencing sizeable phenotypic transformations on cultivars [7]. GE tools such as the transcription activator-like effector nucleases (TALENs), meganucleases (MNs), and zinc-finger nucleases (ZFNs) have enabled crop scientists to successfully manipulate desired crop genes over the years [21]. Moreover, the discovery and utilization of programmed sequence-specific nucleases (SSNs) is facilitating precise gene editing and is currently being applied in the integration of DNA sequences at the desired locus and the production of mutations (such as insertions, replacement, deletions, and substitutions) [21].



**Fig. 2. Genomic-assisted breeding for climate resilience**

Source: Scheben et al. [3]

Abbreviations: Marker-Assisted Selection (MAS), Genomic Selection (GS), Genome Editing (GE), breeding generation products from backcrossing to a parental line ( $BC_1F_1$  to  $BC_nF_n$ ), breeding generation products from intercrosses ( $F_1$  to  $F_n$ )

The knockout of negatively influencing genes (deletions) has been utilized in several crops including wheat and rice to exploit the benefits of target traits [7]. Similarly, climate resilience can be achieved by knocking out host-susceptibility conferring genes using GE technologies. However, the adoption and extensive application of GE approaches in plant breeding is limited by the cost-ineffectiveness and labour-intensiveness of the procedures. Interestingly, the emergence of the Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR/Cas9) system in recent years, has provided an entrancing, cost-effective, versatile and easy-to-design tool for targeted mutagenesis, single base substitution, gene knockouts, multiplex gene editing, and gene transcription regulation in plants [21,23,29]. CRISPR/Cas9 system is a special technology that allows biologists and geneticists to manipulate sections of the genome by adding, removing, or altering parts of DNA (as shown in Fig. 3). In the context of breeding for climate resilience, the CRISPR/Cas9 system can be employed for precise and accurate knockout of genes coding for undesirable and knock-ins for those coding for desirable traits.

Single-celled microbes such as bacteria and archaea utilize the CRISPR-derived RNA and Cas proteins (such as Cas9) to thwart virus and foreign body infections (by destroying the DNA of the foreign organism). The CRISPR technology

was developed following the defense mechanism of these prokaryotes. Pickar-Oliver and Gersbach [30] asserted that our ability to control, identify, picture, and annotate particular RNA and DNA sequences in living cells of various organisms has been transformed by the prokaryote-derived CRISPR-Cas9 GE systems. Wang et al. [31] from Syngenta Biotechnology China successfully used CRISPR sgRNA to delete dense and erect panicle1 (DEP1) gene from line IR58025B of Indica rice. The experiment recorded improvements in dense and erect panicles, yield traits and reduced plant height.

### 2.3 Constraints in the Application of Genomics in Plant Breeding

Despite the efficiency of the CRISPR/Cas9 system, many factors have also been documented which negatively influence its editing ability such as Cas9 codons, GC contents, sgRNA structure, targeted DNA, and expression of sgRNA and cas9 [21]. Furthermore, the non-specific off-targets and cargo-vector inefficiency have also been tabled as limitations to the application of the technology. Moreover, plants have large genomes, high ploidy level, long and complex genetic repetitive regions which demand detailed bioinformatics frameworks for analysis and interpretation [32,33]. The current computational genomics

field is still lagging with regards to the interpretation and storage of large amounts of data generated by these systems. Furthermore, high cost and skilled-labour requirements of genomics-oriented systems limit their adoption in resource-constrained or developing countries. It is therefore imperative to establish financial crowd-sourcing partnerships that target research and knowledge sharing on the application of these technologies in climate resilience breeding.

### 3. FUNCTIONAL PHENOMICS FOR CLIMATE-RESILIENT PLANT BREEDING

Phenomics is a set of methods assembled for speeding-up phenotyping through the use of high-technological imaging systems and computing power [34]. Plant phenomics is a multi-scale and multi-source system that encompasses joint data analyses originating from numerous phenotyping fields and platforms [35]. In the context of plant breeding, phenotypic characterization generates meaningful data for crop diversity assessments. Genetic diversity is

the principal component of crop improvement which allows plant breeders to build on mechanistic science and physiological traits in the selection of parental material for hybridization and genetic gain [36,10,37]. York [38] asserted that, the advent of functional phenomics signifies the reintroduction of physiology as a modern science through the utilization of advanced and accurate sensing technologies and big data analytics. Phenomics covers all the facets of plant physiology and facilitates its linkage to plant breeding practices.

The advent of phenotyping technologies can accelerate screening procedures targeting several traits across a large number of genetic materials. Senapati et al. [39] highlighted that, breeding for high yield potential under adverse climatic conditions requires a) knowledge of the physiological basis of crop yield under a changing environment, b) desirable trait selections and c) evaluating the new cultivar under stress conditions. Furthermore, the selection intensity (*i*) of a particular trait is enhanced through screening numerous genotypes per unit area or time [40,16].

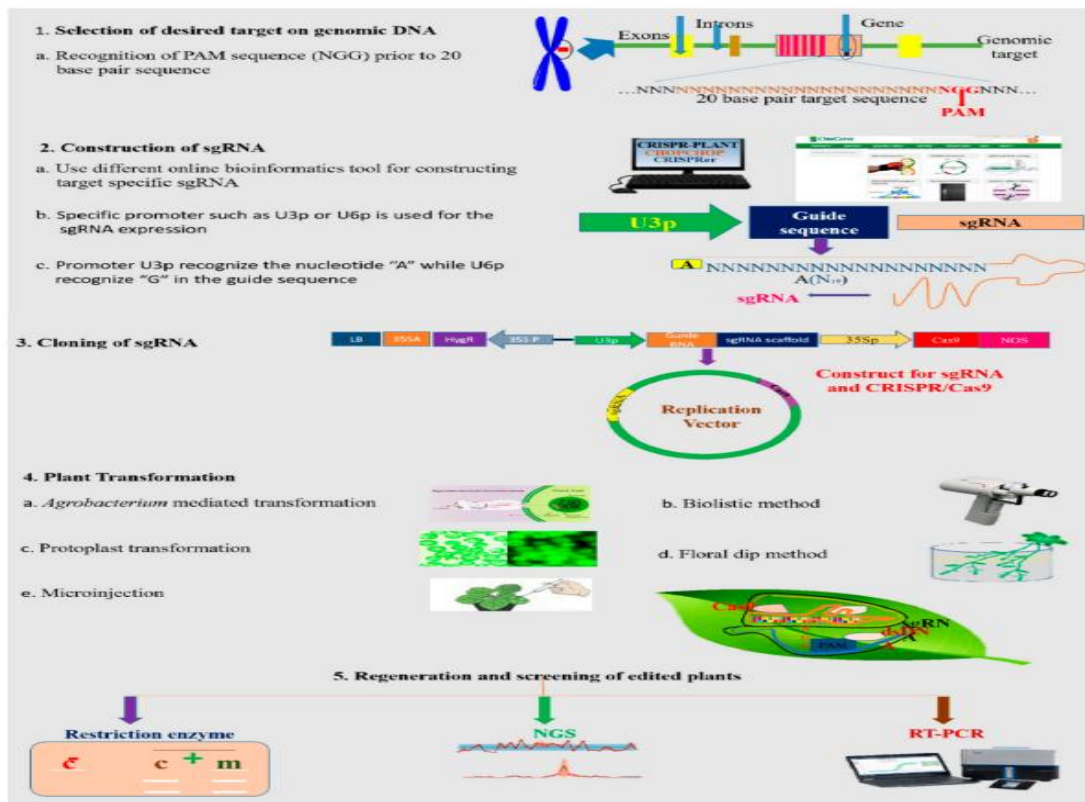


Fig. 3. CRISPR/Cas9-based genome editing system

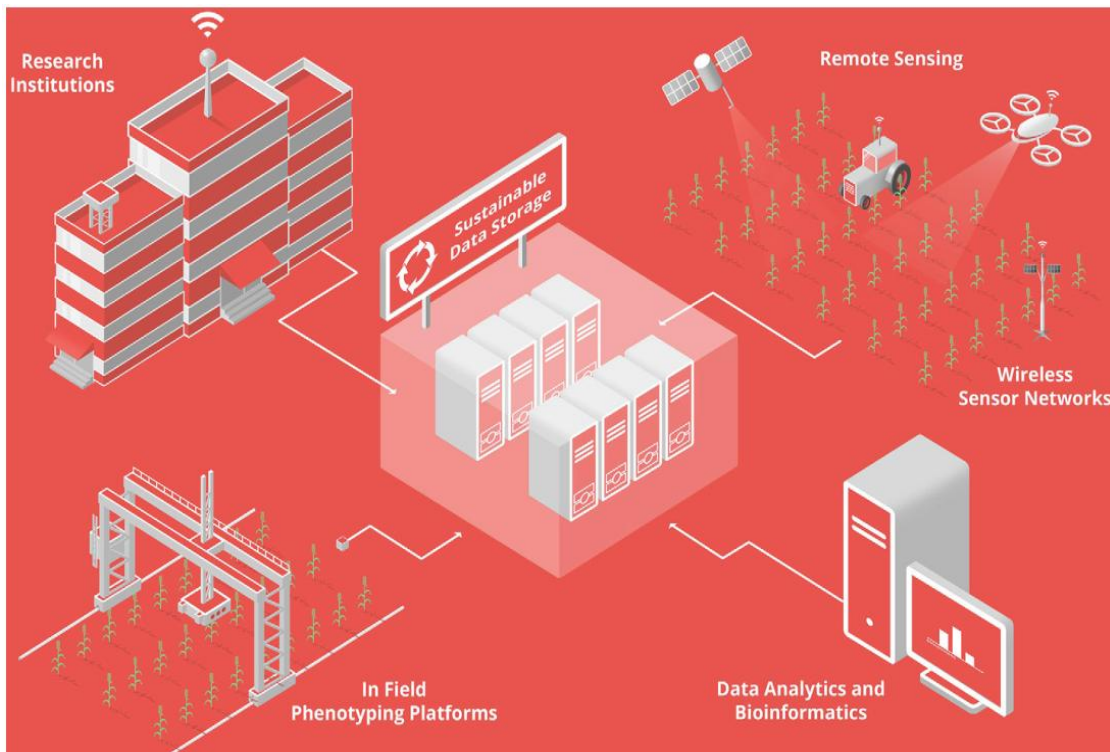
Source: Razzaq et al. [21]

Methodologies that utilize genotypic information and statistical analytic models for generating a genomic estimated breeding value (GEBV) for germplasms at early stages of plant growth hold a great deal of potential in accelerating breeding gains [41]. Heun et al. [42] highlighted that, using electronic and sensor systems for plant trait characterization provide valuable digital inputs which support complex analytical modelling in plant genetics research. Roitsch et al. [43] alluded that, sensor-based crop phenotyping is an integral and essential part of a holistic phenomics methodology in controlling the genotype x environment x management (GxExM) interactions in applied and fundamental plant science studies, germplasm screening in crop improvement programs, precision agriculture and digital farming. Additionally, high-throughput phenomics (HTPs) based on robotics, machine vision, and computing enables plant breeders and physiologists to quantitatively measure previously intractable and complex traits [10]. HTP can observe and record time-series data on functional traits including 3D and top-down models of crop growth and development [2] in multi-environments. Multi-

environmental data testing provides a potential for “stress-impacts” modelling on crops and target populations of specific environments [5].

### 3.1 Types of Phenomics

Phenomics can be divided into two main sections namely forward and reverse phenomics (as illustrated in Fig. 5). Forward phenomics is a phenotyping approach designed to select the ‘best of the best’ amongst the tested genotypes using automated imaging technology [34]. This approach uses high-throughput phenotyping tools to discriminate numerous genotypes with desirable traits. Forward phenomics can accelerate crop improvement cycles targeting drought, heat, and cold stress tolerance in major crops. On the other hand, reverse phenomics is used in the detailed dissection of desirable traits to understand the basis of their superiority [34] and allow for advanced exploitation of these traits in future plant breeding schemes [45,46]. Based on the two approaches, best-performing lines can be selected and advanced to the next improvement phase.



**Fig. 4. Crop Phenomics system**

Source: Shakoor et al. [44]

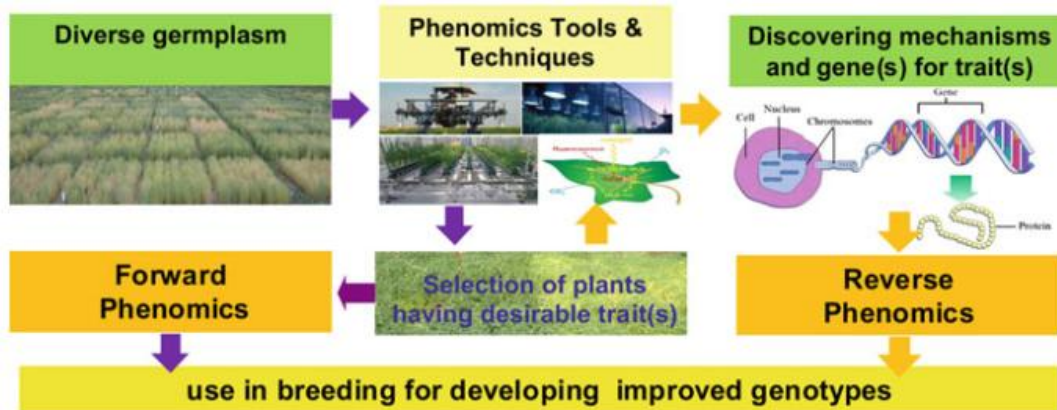


Fig. 5. Forward and reverse phenomics

Source: Kumar et al. [45]

### 3.2 Crop Phenomic Tools

Several phenomic tools are currently being used in plant phenotyping, these include:

- i. **Three-dimensional (3D) imaging** - captures digital photos of the sides and top (of shoot mass, shape, leaf number, leaf health and colour) and combine them into a 3D image.
- ii. **Far-infrared (FIR) imaging system** - utilizes light in the FIR region (spectrum: 15 $\mu$ m to 1mm) to study the temperature differences and their effects on salinity tolerance, photosynthesis efficiency, water usage, etc.
- iii. **Near infra-red (NIR) imaging system** - uses the NIR region spectrum to measure water content and its movement patterns in leaves and soil.
- iv. **Fluorescence imaging system** - uses a computer program to convert the fluorescence generated when an object absorbs light into false-colour signals for plant health analysis.
- v. **Magnetic resonance imaging (MRI) system** - employs a magnetic field and radio waves to capture 3D geometry images of roots [34].

### 3.3 Field Phenomics Equipment

Phenomics employs several pieces of equipment in recording field data. These include phenonet sensor network, phenomobile, and phenotower (shown in Fig. 6). Phenonet sensor network is a pipeline of connected data loggers (such as weather sensors, far infrared thermometer, soil

moisture meter, and thermistor) that collects data from the crop fields and sends it to researchers at the lab through a mobile-based phone network [34]. It is an algorithm that integrates networks and pathways associated with diverse plant phenotypes. The phenomobile is an adjusted golf buggy that manoeuvres through a crop field at a speed of 3-5 km/hr, taking measurements (such as leaf temperature, biomass volumes, leaf area, height, and plant density) in the process [34]. The system can be adjusted electronically to suit the height of the crop. Lastly, the phenotower is employed in capturing images of plants that reach a height of 16m above the ground level [34]. It uses two cameras - an infrared and a standard camera.

### 3.4 Challenges of High-throughput Phenotyping

Despite the ongoing development in computational and imaging systems, extracting quantitative traits reliably from time-series imagery of numerous plants remains a challenge [2]. Furthermore, the broad range of high-throughput phenotypic data is presented in a variety of databases, spreadsheet files, and lab books, thereby making traceability, and access to trial results and associated metadata complicated [35]. However, a research article published by Neveu et al. [35] presented the Phenotyping Hybrid Information System (PHIS) which is a suite of methodologies, synthesized in an open source for sharing and integrating multi-scale and multi-source data. PHIS has the capacity to address the aforementioned constraints and is available for both controlled and field installations by the public community.



However, in crops such as rice and wheat, where large germplasm is available and numerous organizations operate, the applicability of PHIS is constrained. Furthermore, due to the high costs involved in purchasing and maintenance, of these facilities, world-wide accessibility and utilization is only limited to a few operations sites.

#### 4. INTEGRATING GENOMICS AND PHENOMICS IN PLANT BREEDING FOR CLIMATE RESILIENCE

High anticipation levels are rooted in the integration of several technologies and disciplines to deliver success on the breeding of



Fig. 6. Field phenomics equipment  
Source: Roshni and Prajwala [34]

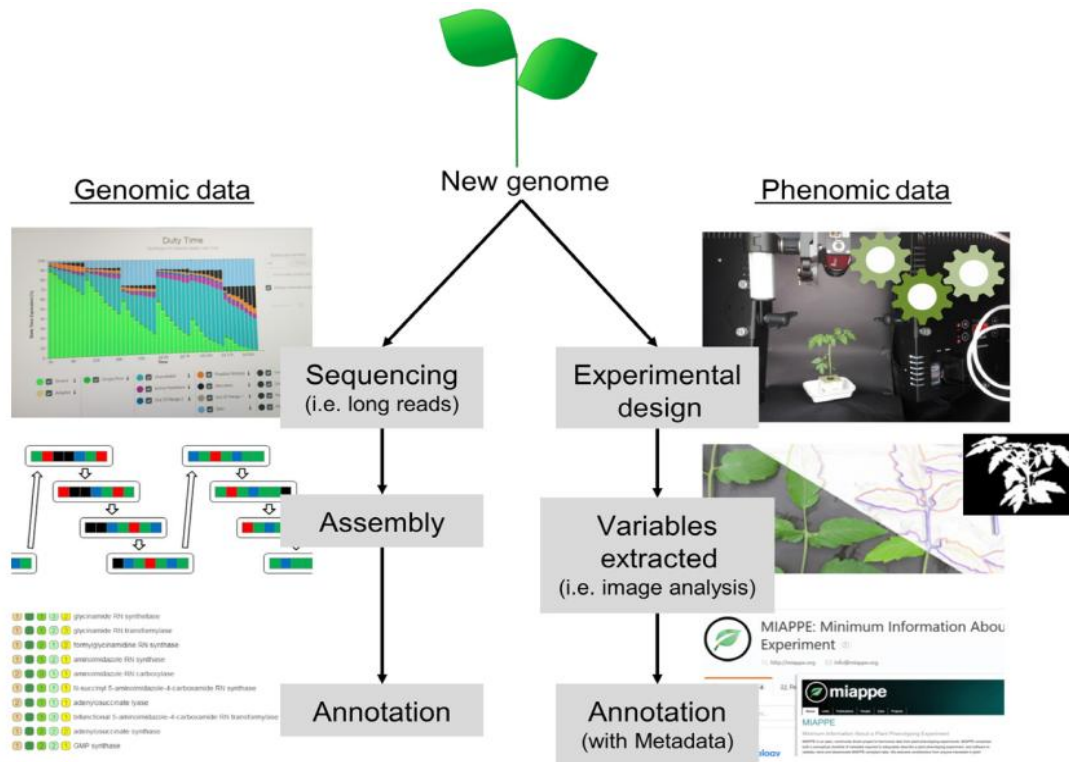


Fig. 7. Genomics plus phenomics data system  
Source: Bolger et al. [51]

climate-resilient crops in less time [16]. Integrating the merits of genomics with advanced phenomics will enhance the breeding efficiency and reduce the time required in cultivar development [47,48,49,50]. The comprehensive and multi-dimensional phenotyping properties of phenomics permits specific hypotheses about genotypic and environmental influences to be successfully tested across a large number of plants and then associate it to the recorded whole-genome sequence variation [2]. An illustration presented by Bolger et al. [51] showed a simplified protocol that can be used in integrating data from genomics and phenomics in a breeding programme (Fig. 7).

Precise phenotypic evaluations and effective biometric analysis can help to classify the particular reactions of a group of genotypes at a given physiological level as determined by changes in weather patterns [5]. Trait capturing across environments and throughout development from multi-dimensional phenotypes can be achieved by applying Genome-Wide Association Studies (GWAS) in identifying causal genes and existing variation and functional structural plant models (FSPMs) in predicting crop growth and reproduction in specific environments [2]; (Xavier, 2019). GWAS data can also be aligned with computer network systems through various phenomics-oriented systems. Cloud-enabled GWAS on crop growth variation can be easily performed during an experimental trial allowing for real-time results and improve quantitative trait loci (QTL) detection [2]. Additionally, molecular markers can also be utilized as proxies for specific phenotypic traits thereby allowing selection to be done in early generations and/or on young plants [16]. The use of trait-linked markers in plant breeding enhances the selection accuracy ( $r$ ) which then promotes off-season selections in any location. However, associating phenomics data with the genomic resources data requires advanced bioinformatics (Choubey et al., 2019), [52]. It is therefore imperative to establish integrated systems that house genomics, phenomics, and bioinformatics in breeding for climate resilience.

## 5. CONCLUSION

The sole task of plant breeding initiatives is to create pathways fostering the development of diverse plant varieties adapted to the changing environmental space by combining traditional and modern technologies. The positive impact made by the genomics revolution on plant

science is unquestionable and can still provide sustainable solutions to the effects of climate change on crop production systems. The research landscape has also witnessed the growth of phenomics in crop production systems. However, a disparity in the use and availability of integrated genomics and phenomics-assisted breeding tools remains. The integration of genomics and phenomics can revolutionize the plant breeding schemes and aid in the development of climate-resilient varieties.

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## COMPETING INTERESTS

Author has declared that no competing interests exist.

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