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Frontline remote sensing tool to locate hidden traits in root and tuber crops

Root and tuber crops (RTCs) play an important role in alleviating poverty, improving health and nutrition, and providing excellent opportunities for improving the livelihoods of smallholder farmers in countries like sub-Saharan Africa. Starch roots and tubers are edible starch materials that are stored in the subterranean stems, roots (cassava and sweet potatoes), rhizomes (canna and arrow roots), corms (taros and cocoyams), and tubers (potatoes and yams) (Chandrasekara and Kumar, 2016). RTCs are second in importance to cereal crops in terms of calories, food security, and industrial purposes. Therefore, it is apparent that understanding the role of root system architecture in RTCs for resource mobilization is a vital step in building the foundation for next-generation crops. In cereal crops, the advancement in high-throughput phenotyping seems to lead the way in breeding and selecting traits for climate resistance, improved yield, and quality. Yet, in RTCs, there is no suitable model system to monitor storage root formation and bulking growth patterns underlying the development of improved traits, which leave behind the response to the next green revolution. Therefore, we discuss the current and future directions of high-throughput phenotyping methods for RTCs with Cassava as a case study for locating the hidden traits.

Cassava (*Manihot esculenta* Crantz), long known as a famine-reserve crop, can thrive under adverse climatic conditions, including prolonged drought and poor soils, and it has a wide harvest window, allowing farmers to harvest as and when needed (Nweke et al., 2002). While smallholder subsistence farmers conduct most cassava cultivation, its role in economic development through commercial farming to provide raw materials for starch, high-quality flour, ethanol, and other industrial products has increased significantly. The global cassava processing market trend had reached ~300 million tons, demonstrating the international importance of the crop (Research and Market, 2021).

The sustainability of agricultural production in the face of increasing demands and production constraints relies heavily on modernizing cassava breeding programs. Due to market demands, cassava genotypes that bulk faster and yield profitable dry matter in fewer months would greatly benefit the farmers. In recent years, cassava breeding has benefited from high-potential technologies like genomic-assisted breeding. The choice of breeding traits is determined by the multiple production traits and end uses of the roots: high dry matter content for industry and animal feed; low amylose cassava for the food industry; tolerance to post-harvest deterioration; resistance to major pests and diseases; biofortification, and adaptation to climate change.

In root crops such as cassava, final yield and quality are evaluated upon maturity, usually 12–24 months after planting. Early storage root bulking is a crucial trait required by not only subsistence but also commercial farmers for its attribute to achieve early maturity and escape late-season threats—pests and diseases, drought, production costs—that compound when the plants are exposed to multiple annual cropping seasons. Despite the apparent importance of early bulking, assessment of this trait has thus far been based on destructive harvesting at regular intervals. This cumbersome, labor- and resource-intensive activity can only be applied to a few genotypes. Furthermore, there are no above-ground morphological indicators that are correlated with early bulking to enable indirect selection. Likewise, no model system nor specialized analytical software exists for real-time monitoring of tuber and storage root development (Duque and Villordon, 2019). The limitation in high-throughput phenotyping represents a significant bottleneck in realizing the full potential of the crop's improvement efforts. Therefore, the tools and approaches used to monitor the root development in cereal crops can be modified to account for RTCs.

CHARACTERIZATION OF ECONOMICALLY IMPORTANT ROOT TRAITS UTILIZING ABOVE-GROUND PRECISION PHENOTYPING

The use of high-throughput phenotyping allows continuous monitoring of plants through imaging the dynamic changes of all activities in plant canopy growth, enabling the characterization of phenotypes–linking traits with associated genes. Above-ground (canopy) field-based phenomics systems, including hand-pushed platforms, tractor-based systems, autonomous platforms, unmanned aerial vehicles (UAVs) with RGB sensors, thermal cameras, light detection and ranging sensors, multispectral/hyperspectral cameras, etc., have been used to predict the above-ground biomass.

In above-ground phenotyping, estimating the canopy traits is essential in predicting the root yield, dry matter, leaf area, and plant architecture (Mulualem and Ayenew, 2012). However, measuring the canopy metrics is laborious and time-consuming; hence, using multispectral imagery with UAVs offers convenient operation for high spatial and temporal resolutions (Van der Meij et al., 2017). Thus, it can estimate the variability within the micro plot at different time points of the growth cycle on a large scale. Vegetation indices and canopy metrics (canopy height, canopy cover, canopy volume) derived from UAVs could provide quantitative data for novel traits defining the genotype-specific

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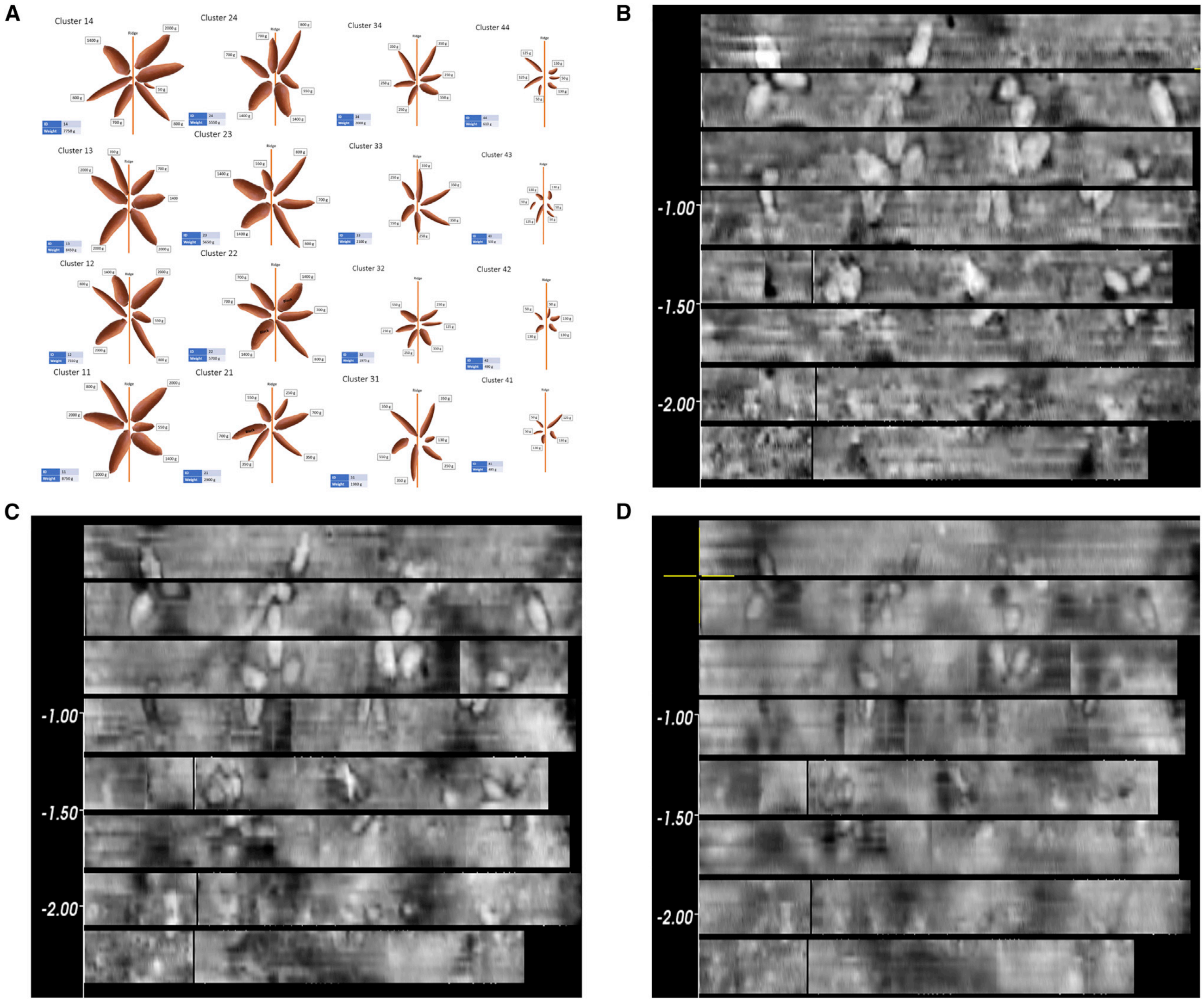

Figure 1. The layout of root features that were scanned with a prototype GPR from IDS GeoRadar in a test pit (A), the associated scanned output at three depth slices showing scanned root features at 0.05 m below surface (B), 0.10 m below surface (C), and 0.15 m below surface (D).

responses to environmental stress throughout the growth period of cassava, as such help breeders elect novel lines (Selvaraj et al., 2020). Such phenotyping methods aid in improving crop yield and open new avenues to complement the current genomic selection projects that could be transformative for cassava breeding by allowing the rapid selection of progeny and high-performing clones for industry and food security purposes.

Knowledge of cassava storage root formation and regulation has increased over time. However, root bulking dynamics are less known, such as the time and stage when cassava accumulates starch in storage organs. Several studies have reported different non-destructive approaches to phenotype cassava root systems under controlled conditions. Image-based phenotyping methods are increasing becoming popular in plant segmentation. An image-based phenotyping system using machine learning methods predicted early bulked storage roots of aeroponically grown cassava at 91% accuracy (Atanbori et al., 2019). The time and mode of differentiation from fibrous to storage roots were almost identical in aeroponics and field situations (Selvaraj et al., 2019). Aeroponics is an efficient method for in-depth analysis of storage root and starch accumulation; however, the system still lacks interaction with the soil and biological factors that affect the root development over time. Hence, the general interest in developing controlled environment cassava phenotyping platforms to predict field performance will open access to the non-destructive root architecture, thereby revolutionizing the ability of cassava breeders to select high-yielding, early bulking varieties favorable for food security and industrial purposes.

GROUND-PENETRATING RADAR: A PATH TO NEXT-GENERATION ROOT PHENOMICS

Ground-penetrating radar (GPR), a novel non-destructive, high-throughput, three-dimensional imaging method, is one such tool designed to predict root yield more accurately by combining

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the above- and below-ground time series information. GPR automates the images through image acquisition hardware with specialized software to achieve the best quality images to screen large genotypes in large multi-location trials (Figure 1). Several studies have defined the ability of GPR to determine root diameter and root dimensions, which has laid a strong foundation for present and future root biomass estimation. Delgado et al. (2017) derived a prediction model for GPR to capture the early onset of starch accumulation or root bulkiness. GPR offers tremendous potential to predict the envisioned changes in root development affected by diseases like frog skin disease, where the plants will not indicate any symptoms in the above-ground part of the plant but affect the root development and pose a potential threat to other healthy plants. Such facilities are possible only if the tool is sensitive enough to detect the subtle differences in root development and biomass that facilitate disease prediction. Cassava generally follows a sinusoidal growth pattern, meaning that changes in root bulking occur over time. Therefore, studies are underway to develop a sinusoidal model to determine the bulking rate plateau and threshold of cassava. Thus, GPR models could be manipulated to study cassava growth and development, providing a greater chance to improve productivity.

In future studies, the extensive GPR arrays optimized for cassava breeding programs can therefore be aimed at phenotyping diverse CGIAR's (Consultative Group for International Agricultural Research, a global research partnership for a food-secure future) germplasm collection for the early bulking trait and novel climate-smart traits for increased cassava productivity, initiating the next "green" revolution. There is a lot of excitement around the root and tuber crop communities to identify a major gene controlling storage root initiation and bulking. Thus, a better understanding of morphological variation in cassava roots will extend our knowledge to other RTCs like potato, yam, and sweet potato. In recent years, there have been efforts to develop analytical pipelines to allow automated or semi-automated extraction of above-ground features representing crop phenotypes. One of the critical aspects of GPR phenomics is automatic data processing and analytics. With hardware/software technologies and computing developments, onboard image processing and real-time analysis will become a reality. Therefore, this data flow needs to be integrated with genomics, metabolomics, and proteomics to speed up the rate of genetic gain. These advancements will only be possible through collaborative efforts across disciplines, private-public partnerships, and other global breeding programs. We anticipate that RTCs breeders can widen the breeding efforts through a GPR revolution with AI-powered decision-supporting tools.

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