BIO-DETERIORATION OF CASSAVA (*Manihot esculenta)* ROOTS, CAUSES AND APPLIED MANAGEMENT TECHNIQUES: *A REVIEW.*

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ABSTRACT
Every crop is subject to some form of natural degradation over time under the right enabling conditions. A fresh cassava root is the least stored crop among the tuberous root family. Cassava root deterioration is related to two processes; physiological (primary) deterioration and microbiological (secondary) deterioration. The physiological deterioration is due to an endogenous phenomenon called Post-harvest Physiological Deterioration (PPD). Its losses are estimated to be as high as 40-60% within the first seven days after harvest. The PPD induces both quality and quantity loss of starch and renders the cassava roots unmarketable and inedible. Symptoms include blue/black vascular streaking, brownish occlusions followed by discoloration of the tissues and an unpleasant odour. Oxidative stresses that may be associated with alternative respiratory pathways and potentially cyanide production are factors that cause PPD. Techniques to delay bio-deterioration are; mechanical techniques (rapid processing, pruning, traditional technique and modern storage technique), conventional breeding (improved varieties, genetic manipulation, speed breeding) and using transgenic approaches. Efforts to breed delayed PPD in cassava had met with little success. It will be of interest to focus on traits for PPD and disease resistance, nutrition, reduced cyanogenic compounds, and postharvest longevity, in delivering quality storage roots for the future.
Keywords: Cassava, bio-deterioration, biotechnology, roots.

INTRODUCTION
Cassava (*Manihot esculenta* Crantz.) is a major staple food crop grown in Africa, Latin America, Oceania, and Asia, feeding more than 800 million people each day. The root, which is the major edible portion of the plant, is an important source of dietary energy and comprises more than 80% starch (Lyer *et al*., 2010; Harris and Koomson, 2011; Uarrota *et al*., 2015). Cassava plays an important role in food security due to its vigorous nature with traits such as drought tolerance and its ability to grow in marginal environments where many other crops perform poorly (Lyer *et al*., 2010; Morante *et al*., 2010). Similarly, this crop remains viable below ground for up to 36 months which provides an addition to food security and a vital subsistence crop for small-scale farmers globally(Rosenthal and Ort, 2012; Uarrota*et al*., 2015). In addition to its role in food security, cassava starch in a large scale is being used as a source of ethanol and biofuel crop in many countries, including China, Thailand, and Brazil (Zidenga *et al.*, 2012). Globally, in terms of caloric intake, cassava has been rated as fifth most important crop overall (Rosenthal and Ort, 2012). However, due to the short shelf life of cassava after harvest, its subsistence and commercial utilization is affected from a rapid postharvest physiological deterioration (PPD) process, which renders the root unpalatable within 72 hrs of harvest (Owiti *et al*., 2011). The deterioration in cassava is more rapid than other tuber and tuberous root crops such as yam and sweet potato. Physiological deterioration occurs two to three days after harvesting, followed by microbial deterioration three to five days after that (Karim *et al.*, 2009; Zainuddin *et al*., 2018). This process, known as postharvest physiological deterioration (PPD), begins at the wounded root terminal and is influenced by the cultivar as well as environmental conditions (Salcedo and Siritunga, 2011). PPD induces both quality and quantity loss of starch and renders the cassava roots unmarketable and inedible. Symptoms include blue/black vascular streaking, brownish occlusions, and chemical deposits from wound sites, followed by discoloration of the storage tissues and an unpleasant flavour and odour (Reilly *et al.*, 2007). Significant quantities of cassava root are also damaged or rot during transportation to markets or processing facilities (Wenham,1995; Harris *et al*., 2015). Postharvest physiological deterioration (PPD) has become one of the primary limiting factors in the production and utilization of cassava (Uchechukwu-Agua *et a*l., 2015) because PPD is triggered by unavoidable physical injury of cassava roots during harvesting (Salcedo and Siritunga, 2011; Saravanan *et al*., 2016). Therefore, it is paramount to investigate suitable and sustainable preservation techniques to delay or inhibit the cassava PPD.
Strategies for delaying PPD consist of pre-harvest, harvest, postharvest, and plant breeding methods. Pre-harvest pruning has been found to delay PPD by decreasing the starch/sugar ratio content in the storage

roots (Oirschot *et al*., 2000), but pruning has a negative impact on cassava yield (Ayoola and Agboola, 2004; Liu *et al*., 2019). Although inhibiting PPD in cassava by breeding approach(Tumuhimbise *et al*., 2015), can be challenging (Morante *et al*., 2010). Hot water treatment for 10 min combined with modified atmosphere packaging delayed PPD during storage of cassava roots (Acedo, 2012). Moreover, keeping 10°C and 80% relative humidity contributed to delay PPD for 2 weeks (Sánchez *et al*., 2006; Liu *et al*., 2019). However, these techniques are costly. Chemical treatment can extend cassava root shelf-life, for example, melatonin (Hu *et al*., 2016; Ma *et al*., 2016) and CaCl2 (Hu *et al*., 2018) treatment delayed cassava PPD. Therefore, other suitable and sustainable preservation techniques to delay or inhibit the cassava postharvest physiological deterioration need to be further revealed.

CAUSES OF CASSAVA POST-HARVEST DETERIORATION

Cassava deteriorates much more rapidly than other tuber and tuberous root crops such as yam and sweet potato. Postharvest Physiological Deterioration (PPD) is physiological damage that occurs very quickly from 1 to 3 days after harvest (Hu *et al*., 2016; Liu *et al*., 2017). Physiological damage is characterized by discoloration of the vascular to blue, black, or brown part, then spread throughout the parenchyma part of the cassava. Also, the taste of roots becomes more bitter and emits a less pleasant aroma (Liu *et al*., 2017). General discoloration of the storage parenchyma is the initial symptom and this depends upon the degree of mechanical damage of the roots as well as the genotype and environmental conditions. The factors causing PPD are variable and wide ranging, but scientific research on the processes and pathways leading to PPD seem to be converging to oxidative stresses that may be associated with alternative respiratory pathways and potentially cyanide production (Sayre *et al*., 2011; Xu *et al*., 2013). Cassava root deterioration is related to two separate processes that is physiological (primary) deterioration and microbiological (secondary) deterioration (Acedo and Acedo Jr, 2013; Njoku*et al*., 2013).

Physiological or Primary Deterioration

Primary deterioration also involves changes in oxidative enzyme activities which generate phenols, including catechins and leucoanthocyanidins, which polymerize in later stages to form condensed tannins (Zidenga *et al*., 2012; Sánchez *et al*., 2013).

Oxidative stress and reactive oxygen species (ROS) scavenging mechanism

Wounding of the roots causes oxidative stress after 15 minutes, Iyer *et al*., (2010) observed rapid increase in free radical compounds such as singlet oxygen (O_2), superoxide (O-2), and hydrogen peroxide (H_2O_2), namely "oxidative burst". The ROS concentration increases because the wound causes electrons from oxygen altered into reactive and toxic structures through Haber-Weiss reactions (Reilly *et al*., 2007). The increase in hydrogen peroxide concentration is related to PPD (Djabou *et al*., 2017), and it can be used as a biomarker of deterioration (Uarrota *et al*., 2015). Oxidative stress causes root metabolism changes, leading to stress defence mechanisms, programmed cell death, cell wall remodelling, and triggered signal transduction (Reilly *et al*., 2007; Salcedo and Siritunga, 2011).

Oxidative stress activates plant defence mechanisms (Djabou *et al*., 2017). One of them is by increasing the activity of antioxidant enzymes such as peroxide (POD), superoxide (SOD), catalase (CAT), glutathione peroxidase (GPX), and ascorbate peroxide (APX) to change free radical compounds into non-toxic compounds such as water (H2O) (Xu *et al*., 2013; Hu *et al*., 2016). ROS scavenging mechanism is the main regulator in inhibiting PPD.

Metabolites change during PPD

Wounds such as cuts and abrasions on cassava storage roots show early physiological changes, including increased respiratory rate and water loss (Marriott *et al*., 1978). Wounding also triggers the production of signalling compounds, such as reactive oxygen species (ROS), jasmonic acid, salicylic acid, ethene, and acetone (Reilly *et al*., 2007; Iyer et al., 2010). Increased respiration induces the conversion of starch to sugar (Sánchez *et al.*, 2013). In post-harvest, the starch content of intact cassava storage roots free from wounds gradually decreases over 6 days (Osunsami *et al*., 1989). The secondary metabolite compounds increased at the beginning of the PPD process in response to oxidative stress, including coumarin, flavanones, phenol, and flavan-3-ol(Iyer*et al*., 2010). Hydroxycoumarin compounds are widely analysed since they are assumed to act as precursors of vascular discoloration. It is reinforced by the association between decreasing hydroxycoumarin content and increasing root storage period (Buschmann *et al*., 2000; Uarrota *et al*., 2015). The wounding of cassava roots exposed them to oxygen, resulting in hydroxycoumarin oxidization and causing blue or black discoloration (Buschmann, *et al*., 2000). Scopoletin is the most common hydroxycoumarin compound that changes to an insoluble coloured product that indicates peroxidase activity (Buschmann, *et al*., 2000). However, there was no correlation between hydroxycoumarin or scopoletin compound with the PPD resistant level (Salcedo and Siritunga, 2011; Mahmoud and Al-Ani, 2016). This phenomenon probably due to the reduction of hydroxycoumarin has done before the discoloration symptom fully appears (Salcedo and Siritunga, 2011) .

Ethylene Production

Ethylene is a plant hormone causing metabolic changes in plant tissues. Production of ethylene increases

steadily from 6 hours after harvesting (Hirose *et al*., 1984; Zainuddin *et al*., 2017), as well as emission of other volatile metabolites such as ketones, cyanohydrin, aldehydes and alcohols (Iyer*et al*., 2010). Storage root tissue undergoing PPD emits four times higher ethylene levels compared to non-deteriorating root tissue (Hirose *et al*., 1984). Unexpectedly, pre-harvest pruning, a practice known to delay the onset of PPD, does not affect ethylene production following injury and the application of exogenous ethylene was not sufficient to accelerate PPD (Hirose *et al*., 1984).

Microbiological or Secondary Deterioration

Microbiological deterioration results from pathogenic rot, fermentation, and/or softening of the roots, and generally occurs when the roots have already become unacceptable because of physiological deterioration(Plumbley and Richard, 1991; Wenham, 1995). PPD is much more important economically than secondary microbial deterioration because the visible colouration of the root is used as an indication of its cooking quality making the crop difficult to sell.

Investigations have shown that the number of different species of fungi and bacteria isolated from roots stored under different conditions shows that post-harvest decay of cassava is a complex matter, involving more than a single initial organism. Two distinct specific types of rot have been described by (Acedo and Acedo 2013), a dry rot occurring under aerobic conditions and a soft rot in an anaerobic environment caused by an unidentified Rhizopus sp. and Bacillus sp respectively.

A more detailed investigation (Ekundayo and Daniel, 1973) indicated that soft rot of cassava roots was caused by a complex of fungi; *Lasiodiplodia theobromae* (Pat.) Griff. & Mobl., which is the most important. Others are *Trichoderma harzianum* Rifai, *Cylindrocarpon candidum* (Link) Wollenw., *Aspergillus niger* van Tieghem and *Aspergillus flavus* Link, *Cylindrocarpon candidum*, Wollenw and *Trichoderma harizianum* Rifia. These workers clearly associated the decay with penetration through injury and concentrated on the later stages of decay rather than on the initiation of postharvest deterioration. (Wenham, 1995), who also worked mainly with material that was in an advanced stage of deterioration, isolated *A. niger* together with "*Cylindrium cladostrinum*" (presumably *C. clandestrinium* (Corda) Saccardo) and unidentified *Penicillium* and *Cladosporium spp*.

Booth, (1976), in a more detailed study on the deterioration of cassava, isolated from the surfaces of cassava roots various species of *Pythium, Mucor, Rhizopus, Penicillium, Aspergillus, Fusarium, Cladosporium, Glomerella, Gloeosporium, Rhizoctonia, Bacillus, Xanthomonas, Erwinia, Agrobacterium* and many saprophytic bacteria. However, Booth was consistently unable to isolate any specific microorganism from the advancing margins of deterioration in the flesh of the rots. It was therefore concluded that the earlier stages of postharvest deterioration in cassava with discoloration manifesting at the vascular tissue were not as a result of microbial attack, and that the later stages were essentially the decay of already dying tissue caused by a wide variety of saprophytes.

APPLIED TECHNIQUES OF BIO-DETERIORATION ON CASSAVA

Cassava roots, suffer a remarkably short shelf life due to a physiological deterioration compared to other tuber crops. The extent of deterioration has been attributed to the degree of mechanical damage by roots as well as the genotype. The drivers of short shelf-life of harvested cassava roots limits market potential and discourages all stakeholders in the value chain (Howeler *et al*., 2013). PPD poses a lot of negative outcome such as reduced incomes for cassava farmers and lowers the reliable supply of cassava as raw material for industry (Salcedo and Siritunga, 2011). PPD inhibition techniques to extend the shelf life of cassava storage roots widely developed, ranging from cultivation, postharvest, plant breeding to genetic transformation and biotechnology. Aside from the effectiveness of inhibiting PPD, the developed methods should also consider the ease and cost of application.

Mechanical Techniques

Storing and transporting the roots in plastic sacks, coating with paraffin wax and freezing (Njoku *et al*., 2013) all ensures the exclusion of oxygen and this techniques are actively and currently used to delay PPD most importantly for the export market. These moderntechniques have been adopted and implemented in developed countries with little success due to the increased cost; however, these methods are not practical in developing countries. Traditional methods have been developed by farmers in African to delay deterioration. In this method, roots are either stored in pits, clamps, trenches, boxes or left in the soil, but these methods are after root harvest until they are consumed, processed or marketed, which makes it easier and cheaper for farmers.Ravi et al., (1996) carried out a novel research using pits in sandy soil to store fresh cassava roots and reported its low-cost as a method for extending the shelf life of fresh cassava roots and reducing PPD. This method was able to prolong the shelf life of cassava for more than two months, with effects on the roots becoming very sweet and having poor cooking qualities leading to its only use as cattle feed. These methods are based on the process of curing, which has been a common method for enhancing the storage life of other root crops. Curing over the time has aid in healing of wounds faster on produce thus limiting deterioration at relatively high temperatures (25 to 40ºC) and high relative humidity (RH; 80 to 85%) (Ravi *et al.,* 1996).

Rapid processing

Several traditional marketing and storage systems have been adapted to prevent root perishability on a small scale. PPD becomes a serious problem considering large scale production, since there is no technique available to store and preserve cassava roots commercially (Aristizabal and Sanchez, 2007; Njoku *et al*., 2013). The systems adapted include establishment of processing centres very close to areas of production to ensure daily supply of raw material, processing into storable forms (by peeling, grating, drying, fermentation) and the traditional trading of small quantities of roots (Westby, 2002).

Pruning

Pruning is another practice used to overcome PPD, thistakes place approximately 2-3 weeks prior before harvest, by removing all the leaves and stems of the cassava plant approximately 40-50 cm above the soil level. Pruning has been associated with the reduction in the time of onset of PPD compared to un-pruned plants (Plumbley and Richard, 1991; Njoku *et al.*, 2013).

Traditional techniques

Cassava roots are known to last in soil up to three years, this is a common way of avoiding losses due to PPD. They are left standing in the soil until they can be immediately consumed, processed or marketed. This common method of storage has its disadvantages because it occupies large areas of land used by the crop, thereby making it unavailable for other agriculture production. Furthermore, cassava roots left to stand in the soil for a long period may increase in size, they become more woody and fibrous, decreasing palatability and increasing the cooking time, respectively, especially if left longer than the optimal harvest time of 10-12 months after planting. Another negative effect of allowing extensive in-field storage of cassava roots increases their susceptibility to attack by pathogens as well as the reduction of extractable starch (Ravi *et al.*, 1996).

Modern storage techniques

As cassava becomes a more industrial commodity modern techniques such as the use of polyethylene bags, waxing and deep freezing are being applied commercially. The application of these more modern techniques is very limited considering the conditions under which much of the world's cassava is grown (Ravi *et al*., 1996; Njoku *et al*., 2013).

Polyethylene bags have been widely used and adopted in West Africa and South America to store and transport the roots of cassava after harvest, and this has been observed to prevent PPD up to 4 weeks by subjecting the root to high relative humidity inside the bag which reduces transpiration and respiration. Also, putting into consideration the quality of the roots (with minimal damage), protection from sunlight, treatment with fungicide, and packing within three hours after harvest (Ravi *et al*., 1996) yields successful conservation. A more recent modern method of limiting PPD is covering cassava roots with paraffin wax by dipping the root in paraffin wax (at a temperature of 55-65^{o}C for a few seconds) after treatment with fungicide. It has been reported that the use of wax prolongs shelf-life of cassava roots up to 2 months (Aristizabal and Sánchez, 2007). Paraffin wax works probably by cutting of the influx of oxygen needed for oxidative respiration into the harvested root.

Harnessing wild relatives for cassava improvement

The genus Manihot is from the family Euphorbiaceae with about 98 species spread through-out the Central and South America. *Manihot fabellifolia* and *Manihot peruviana* are believed to be the primary gene pool for domesticated cassava (Allem, 1999). Cultivated cassava, *Manihot esculenta* ssp. esculenta, was domesticated from *M. esculenta* ssp. Fabellifolia. Comparative genome-wide analyses between wild relatives and cultivated cassava varieties have revealed that wild cassava relatives have more genetic variation than the cultivated cassava (Mbinda and Mukami, 2022). However, is unfortunate that some of the wild relatives of cassava are on the verge of extinction, and may be extinct in the near future (Nassar et al. 2007). This will deprive the gene reservoir for cassava breeding programs. Natural hybridization occurs among wild Manihot species as well as between them and domesticated cassava varieties. Successful interspecifc hybridization of cassava genotypes and *Manihot walkerae* for delayed PPD (Morante et al. 2010) signifies that genetic improvement for PPD tolerance can be accomplished by using wild relatives.

Conventional Plant Breeding

Plants that are PPD resistance are genetically transferred to the next generation through plant breeding and this has been a long-term solution. PPD has a wide range of heritability values between 52% and 94%. Hence, the character of PPD resistance is controlled strongly by genetic (Tumuhimbise *et al*., 2015; Verturini *et al*., 2015), but environmental variance still needs to be minimized. There have been constant efforts of breeders in improving cassava yield and resistance to stresses of both biotic and abiotic through conventional breeding. However, some limitation to the success of breeding includes lack of resistant genes in existing germplasm, high heterozygosity, poor flowering, the outcrossing nature of cassava and quantitative trait such as PPD most especially. Unfortunately; trait separation has been difficult due to some desirable traits often being recessive and genetically linked to undesirable ones. Research has established that there is a strong genetic link between PPD and high dry matter content (Rahmawati *et al*., 2022), this has made breeding for

PPD tolerance via conventional means a difficult challenge.

Improved varieties

Variety of attempts has been made towards creating PPD resistant cultivars; this has shown some promise for industrial cassava production, however due to the gradual loss of starch during the storage process even within PPD-resistant cultivars, the time for which cassava can be stored is limited. Each day stored cassava loses starch at a rate of about 1% , thereby depreciating the amount of starch during storage even as little as a week, thereby rendering it un-useful for industrial applications (Sánchez *et al.*, 2013).

Breeding for PPD resistance is challenging and complicated due to the multiple chemical pathways and genes that contribute to the resistance, and also the interaction with other desirable quality traits in cassava. For example, high dry matter content in cassava that is typically desired by industrial producers has to PPD to battle with, because PPD is positively (though weakly) associated with dry matter content (Sánchez *et al*., 2006). PPD is also negatively associated with carotenoid content: high carotenoid content may delay PPD by 1-2 days. This positive relationship between the two nutritive effects suggests the potential for breeders to develop PPD-resistant cultivars that will contain both high dry matter and high carotenoid (Sanchez *et al*., 2006). Sayre *et al*., (2011) found that genetically over expression of Arabidopsis alternative oxidase in cassava delayed the onset of PPD by as much as three weeks, and additionally that transgenic plants with elevated β-carotene content had a shelf life of four weeks.

Genetic manipulation to increase PPD tolerance in cassava storage roots

Cassava root responses to PPD have been studied over the years at the ecological, phenotypic, cellular, physiological, biochemical, and molecular levels. Investigation from these studies establishes a firm foundation on measures to mitigate the perishability of cassava roots through genetic alterations. At the expression level, microarray assays (Reilly *et al*., 2007) and total RNA sequencing have been utilized to explicate the differential expressions of RNA transcripts and protein profiles involved in PPD response (Yan *et al*., 2021).

The physiological changes that occur during cassava storage root development are caused by stage-dependent control of metabolic pathways, which is also reported in other plants (Sun *et al*., 2019; López-Ruiz *et al*., 2020). Several researchers have investigated numerous genes that are crucial in dry matter accumulation, cytosolic processes, oxidative mechanisms, stress responses, programmed cell death, cellular metabolism, as well as biosynthesis and activation of protein syntheses and secondary metabolites involved in PPD specific-stage mechanisms (Morante *et al*., 2010; Uarrota *et al*., 2015). Having highlighted these genes, this will help in genetically editing some genes that replaced or over expressed.

Genes conferring PPD tolerance

Studies on different tuber crops such as potato, sweetpotato, and eggplant (Mbinda and Mukami, 2022) have revealed the mechanisms involved in carbohydrate metabolism and starch deposition in tubers, tuber wounding, and postharvest internal browning. He stated the mechanisms underlying these processes in cassava can be drastically different from that of other plant species. Findings from potato and sweetpotato have provided some insights on the possible regulatory mechanisms involved in PPD in cassava. Therefore, various studies have been conducted over the last two decades to unveil various important and essential genes for PPD and its components in cassava. More recently, the mechanisms that govern PPD development and progression in cassava roots were examined using cDNA microarray, proteomics, metabolomics, and large transcriptomic analyses (Mbinda and Mukami, 2022). Reilly et al. (2007) researched towards identifying the full set of genes expressed during cassava post-harvest physiological deterioration, he used cDNA microarray technology to characterise genes that show significant change in expression during the PPD response. 63 out of the 72 non-redundant expressed sequence tags were induced which showed altered regulation during the post-harvest period, whilst 9 were down-regulated. Many of the up-regulated and PPD-specific expressed sequence tags were predicted to play a role in cellular processes including reactive oxygen species turnover, cell wall repair, programmed cell death, ion, water or metabolite transport, signal transduction or perception, stress response, metabolism and biosynthesis, and activation of protein synthesis.

Speed breeding

Shortening plant generation periods have been the aim breeders using the new breeding technologies, thereby increasing genetic acquisition by reducing breeding cycles. Speed breeding technology, or rapid generation advancement, have been established to shorten the breeding cycle. This is successfully carried out by adjusting environmental conditions such as temperature, prolonged photoperiod humidity control, plant growth regulators such as gibberellic acid, and use of haploids to maximize plant development rate and flowering thereby, quickening the genetic gain (Ghosh *et al*., 2018). Speed breeding has been effectively applied in some crops of long-day such as barley, wheat, canola, chickpea (Ghosh *et al.*, 2018), and some few short-day crops plants such as soybean (Nagatoshi and Fujita, 2019) and peanut (O'Connor *et al*., 2013). Till date, no speed breeding technique has been applied for cassava. Therefore this call is an urgent need to formulate a cassava genotype-independent high-density planting to

trigger early flowering for speed breeding. For crop research and breeding, accelerated breeding of cassava will be instrumental, particularly in introducing traits in response to emerging threats and needs.

Biotechnology

Biotechnological techniques have been promising strategies to regulate gene expression in the storage root to increase cassava root shelf-life. There are various genes that have been found to be up- or down-regulated during storage (Hu *et al.*, 2016), and their modulation at either genomic, transcriptomic, or proteomic level can effectively delay PPD. Overexpression of alternative oxidase (Zidenga *et al*., 2012) and co-overexpression of proteins for superoxide dismutase and catalase (Xu *et al*., 2013), have effectively slowed down PPD responses. Using RNAi technology, Liu *et al*., (2019) have completely inhibited expression the feruloyl CoA 6′-hydroxylase gene in cassava, concomitantly blocking scopoletin biosynthesis, a critical metabolite in PPD development. Recently, Beyen *et al*., (2020) have employed virus induced gene silencing to modulate PPD development in Cassava roots. These findings put together demonstrate the great potential of biotechnological tools in mitigating the effects of PPD in cassava roots.

Genome editing

These technology utilize various endonucleases such including meganucleases, zinc finger nucleases (ZFN), transcription activator-like effector nucleases (TALENs), and CRISPR/Cas (Mbinda and Mukami, 2022), for targeted genome modifications. The RNA-based CRISPR/Cas system has a comparative advantage in precision, simplicity, and affordability and has emerged as more robust and convenient than protein-based techniques. CRISPR/Cas technology's ability to perform multiplex genome editing, (simultaneous targeting of multiple related or unrelated targets) is greatly appealing, especially for complex and multigenic traits such as PPD. For example, four genes were simultaneously edited *Setaria viridis* in multiplexed CRISPR/ Cas9 (Weiss et al. 2020). CRISPR/Cas technology could therefore be the most significant advancement in plant breeding since the Green Revolution; it will most certainly become the standard tool of crop breeding in future. CRISPR/Cas genome editing approaches also facilitate the modification of traits in plants that are difficult to acquire through conventional breeding approaches such as cassava, leaving no traces of foreign gene(s).

Transgenic approaches

All successful transgenic strategies to reduce PPD in cassava storage roots have so far been enzyme based that reduce oxidative stress or scopoletin biosynthesis. It has been suggested that cyanogenesis during the onset of PPD (Uarrota *et al.*, 2015) triggers ROS production by inhibition of mitochondrial respiration (Zidenga *et al*., 2012). Transgenic roots of cassava had a 10-fold reduction in ROS accumulation, expressed arabidopsis cyanide-insensitive mitochondrial alternative oxidase (AOX) and significantly lower PPD scores compared to wild-type roots up to 21 days after harvest (Zidenga *et al.,* 2012). Hydrogen cyanide (HCN) content and PPD score in 1374 cassava genotypes have no correlation(Chávez *et al*., 2005). This suggests for PPD susceptibility, HCN content of cassava roots cannot be used as a proxy, and therefore the mechanism triggered by AOX to reduce PPD remains unclear.

CONCLUSION AND FUTURE PROSPECTS OF MITIGATING PPD

Post-harvest physiological deterioration of cassava is a major problem for cassava farmers and processors and remains to be a major constraint for expansion of cassava cultivation. Cassava growers yarn to supplying fresh and high quality roots with longer storability. Having such varieties will be highly beneficial not just to small holder farmers but to the industrialists and scientists. Despite the immense potential of cassava, postharvest quality traits have not been prominently factored in breeding programs. The considerable germplasm and molecular tools that are present gives so much hope towards making an expeditious advancement in cassava PPD breeding. It will also be of interest to focus on traits for PPD and disease resistance, nutrition, reduced cyanogenic compounds, and postharvest longevity, in delivering sustainable, high quality storage roots for the future.

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