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BIOFERTILIZER IMPACTS | CASSAVA (*MANIHOT ESCULENTA CRANTZ)* CULTIVATION CROP YIELD AND REGENERATIVE AGRICULTURE

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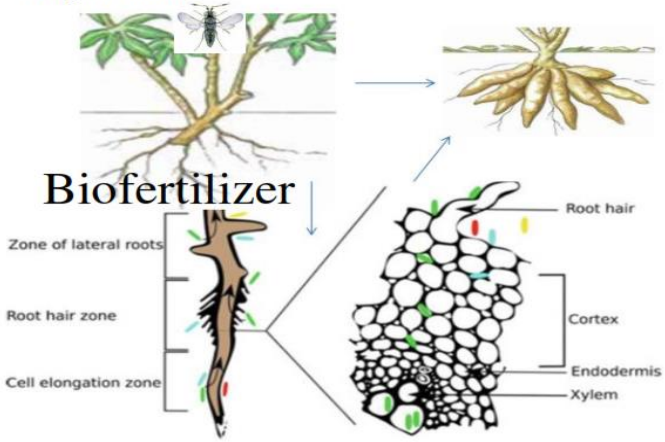
HIGHLIGHTS |

- Biofertilizer Production
- Biofertilizer Cassava Inoculants
- NPK + Biofertilizer Application
- Nutrient Use Efficiency (NUE)
- Soil Health and Quality
- Regenerative Agriculture
- Soil Microbiome Engineering
- Agroecosystems
- Different Microbial Biofertilizers
- Cassava Genetic Transformation
- Cassava Diseases and Pests
- Value Chain Development

Graphic Abstract



ABSTRACT | Cassava (*Manihot Esculenta Crantz*) demand by 700 million people and cultivated in 105 countries between Tropic of Cancer and Tropic of Capricorn within 2300m elevations globally for food security and the cultivation impact on biodiversity require biofertilizer to mitigate climate challenges, crop sustainable development and regenerative agriculture. Nigeria is the world largest producer with a global average yield of 11.80 t/ha, cassava yields can reach 80 t/ha, compared to the current world average yield of just 12.8 t/ha. Biofertilizer solves the traceability problem of chemical farm inputs, suitability and nutrient use efficiency as an integral function of the rhizosphere microbiome via plant microbe interactions for improved soil health quality and crop degradation management. Cassava cultivation with biofertilizer will reduce hydrogen cyanide (HCN) levels in the crop as an integral bioavailability of soil organic matter and nutrient use efficiency. Plant growth-promoting rhizobacteria (PGPR) in the biofertilizer will ameliorate plant abiotic stress and bio-control diseases management. Easily accessed agrobacterium inoculant in biofertilizer has potential for transgenic cassava cultivar development for improved yield and nutrient biofortification. This book chapter encapsulate the case studies trilogy research article papers on biofertilizer impacts on soil microbiome during cassava cultivation for crop yield, soil health, regenerative agriculture, value chain development, food and nutrition security.

KEYWORDS | Soil microbiome, Biofertilizer, Inoculant, Cassava, NPK fertilizer, Biocontrol, Soil health and quality, Hydrocyanic acids (HCN), PGPR, Value chain development, Transgenic Cassava.

INTRODUCTION

The soil microbiome, including bacteria, archaea, fungi, viruses, and other microbial eukaryotes, has crucial roles in the biogeochemical cycling of nitrogen (N), the maintenance of soil fertility, and the plant nitrogen use efficiency (NUE) in agro-ecosystems (Fierer, 2017). Recent advances in omics-based technologies (e.g., metagenomics, meta-transcriptomics, and metaproteomics) have expanded our understanding of the soil microbiome and their controls on specific N-cycling processes (Fierer, 2017; Thompson *et al*., 2017; Trivedi *et al*., 2017). The crop NUE in modern agro-ecosystems is notoriously low, as more than 50% of N fertilizer applied is lost to the environment through ammonia volatilization, nitrate leaching, and emissions of nitrous oxide (N_2O), the third most important greenhouse gas (Hu *et al*., 2015; Coskun *et al* ., 2017). Conventional agricultural practices mainly rely on agronomic measures and chemical inputs to improve NUE, which could lead to soil degradation and loss of biodiversity, with detrimental consequences for soil health and ecosystem functioning (Hu *et al*., 2017). For example, long-term use of synthetic fertilizers, herbicides, and pesticides can negatively influence bacteria and fungi that create organic matter essential to plants. Propelled by re-generative agriculture, there are growing interests focused towards the manipulation of the soil microbiome to reduce soil erosion, to enhance plant growth and disease resistance in agro- ecosystems, and to promote the remediation of heavy metal- contaminated soils (Fierer, 2017; Trivedi *et al*.,2017). Plants have developed intimate relationships with their interacting soil microbiomes and the environment (termed as the 'phytobiome'reported by Leach *et al*., 2017). Some plant and crop roots (e.g. *Fallopia spp. and Brachiaria humidicola*) can exudate organic compounds to inhibit the ammonia monooxygenase (enzyme capable of oxidizing NH_3 to NH_2OH) and hydroxylamine oxidoreductase (enzyme capable of oxidizing NH_2OH to NO_2) of ammonia oxidizers (Subbarao *et al*., 2009) or to inhibit the metabolic activity of denitrifiers (Bardon *et al*.,2014).

Screening agricultural crops with similar traits may greatly enhance our ability to improve crop NUE by using them directly for *in situ* microbiome engineering. Emerging microbial biotechnology tools are proposed to precisely manipulate the soil microbiome *in situ*, by adding or withdrawing chemicals (Sheth *et al*., 2016) to regulate Nitrogen (N) transformation processes under various conditions. Multidisciplinary approaches, especially genome engineering can contribute to microbiome-based biotechnologies to sustainable management of the N cycle. This book chapter encapsulate the case studies trilogy papers (Figure 1) on cassava (*Manihot Esculenta Crantz*) potential of the soil microbiome impacts (an integral function of the broad spectrum microbial innoculants in the biofertilizer production) on the macronutrients and micronutrients bioavailability for the integrated soil nutrient management during the cultivation of cassava crop, Figure 1. Cassava is regarded as a food security crop in many developing countries, with a potential to provide off-season calories even on low-nutrient soils (Nweke *et al*., 2004).The crop is native to tropical regions of South America; but is now a staple crop in many African countries (Allem, 2002). The field cassava case study (Igbariam, Eastern-Nigeria, Africa) of the impacts of biofertilizer on the soil microbiome during cassava cultivation using OBD-Biofertilizer (biofertilizer production visit www.academia.edu/video/l BboEl) applied alone and in combination with inorganic fertilizer NPK (15:15:15) to cassava crop yield and growth components; integrated soil nutrients management, soil health and quality and ultimately re-generative agriculture.

Physicochemical approaches to manipulate the soil microbiome

Physicochemical agricultural N losses through manipulating the abundance, structure and activities of soil N-cycling microorganisms or controlling the amount of N resources available to microorganisms (Figure 2). Some practical tools utilized in agro-ecosystems to improve NUE include: (1) use of synthetic nitrification inhibitors (microbial OBD-biofertilizer) to inhibit the activity of ammonia oxidisers and reduce the loss through N_2O emission and nitrate leaching NPK + OBD-Biofertilizer) affirmed by scholars (Shi, X. *et al.*,2016) and illustrated by Figure 2; (2) use of urease inhibitors (e.g. N-(n-butyl) thiophosphoric triamide (NBPT)) to inhibit the expression of genes encoding ureases that catalyze urea hydrolysis (Shi *et al.*, 2017); (3) manipulation of soil properties (e.g. soil pH, C:N ratio, and moisture) by biofertilizer (Table 3) application that enhance the diversity and structure of soil microbiomes during cultivation; (4) incorporation of plant residues (biowaste carrier for the inoculants broad spectrum biofertilizer to enhance microbial N immobilization and reduce the amount of inorganic N available to soil microbes (Fisk *et al*.,2015); and (5) integrated nutrient management practices to better synchronize N supply and crop N demand and reduce N available to soil microorganisms. association (Singh and Trivedi, 2017).

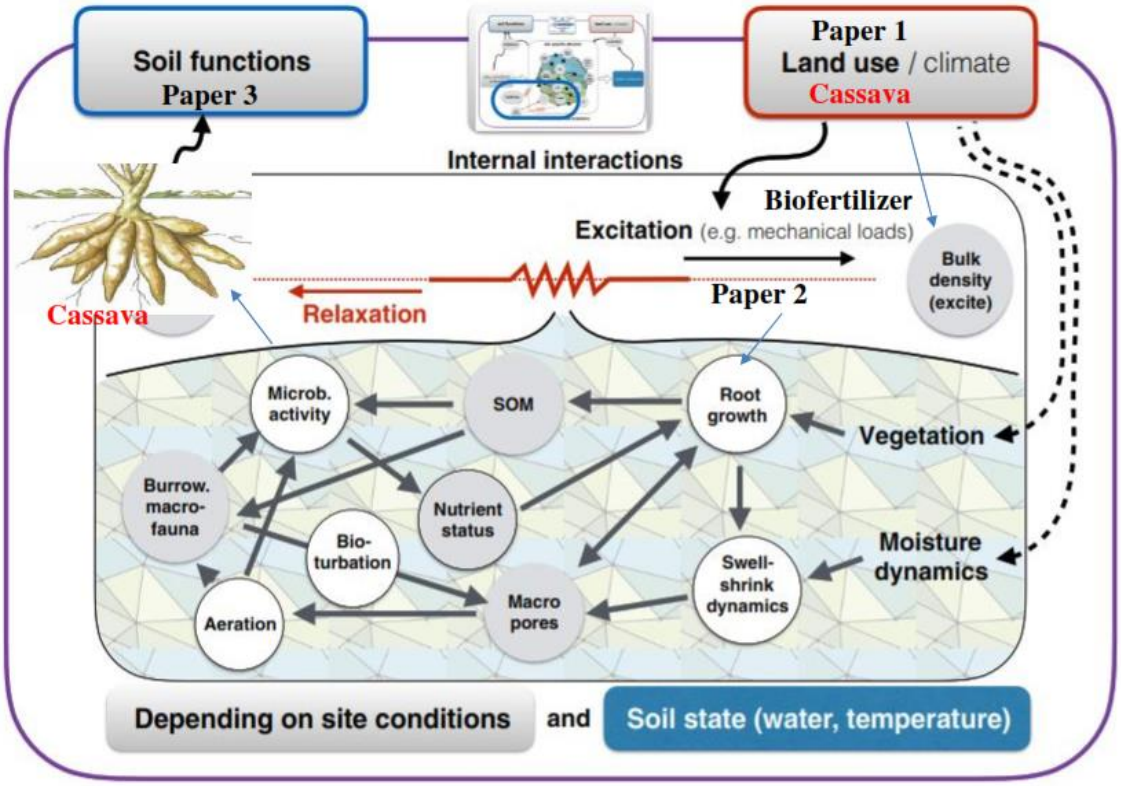


Figure 1 | Adapted from Vogel, Hans-Jörg *et al.*, 2018 as a framework for biofertilizer application to the cassava cultivation soil system is functional characteristics of the impacts on rhizosphere considered for diagnostic tests of 'Biofertilizer rhizosphere holistic soil function'. Where indicated by grey [Paper 1, Otaiku *et al*., (2019a)] circles while the connecting processes are in white [Paper 2, Otaiku *et al*., (2019b)] and sustainable soil management [Paper 3, Otaiku *et al*., (2020)].

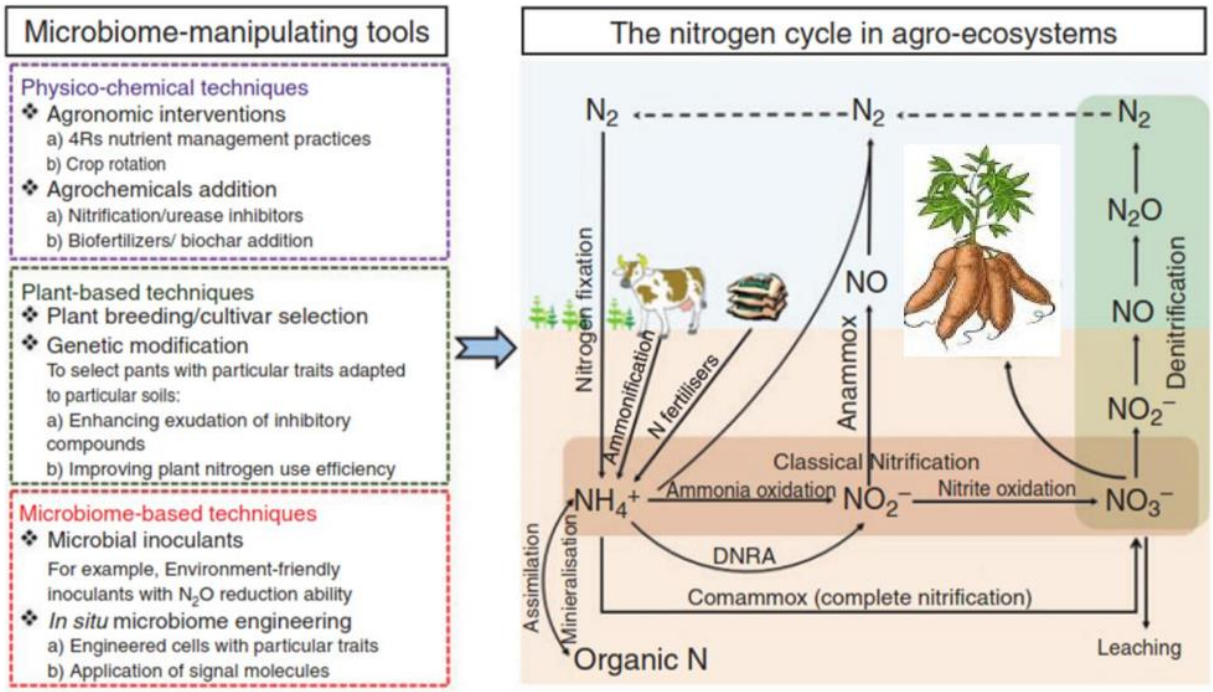


Figure 2 | Adapted from Hu, Hang-Wei and He Ji-Zheng (2018). Schematic overview of the microbiome manipulating tools that can be used for managing the nitrogen cycling processes in agro-ecosystems. DNRA, dissimilatory nitrate reduction to ammonium; Anammox, anaerobic ammonia oxidation.

Tools (1) and (2) are direct practices that impact soil microorganisms while the other three tools are indirect practices and the focus of this book chapter. The outcomes of these physicochemical technologies are variable across soils, primarily owing to their largely unknown impacts on soil microorganisms. Long-term use of chemicals has detrimental environmental impacts, resulting in the accumulation of residues in fields, loss of beneficial microorganisms, and disruption of the plant and soil microbiota (Singh and Trivedi, 2017).The aim of the book chapter was trilogy of three published research articles (Figure 1) from doctoral thesis to determine the bio-fertilizer impacts on cassava on crop yield and Growth Components; Improved Soil Health and Quality; and Soil Microbiome Engineering, Genetic and Sustainable Agroecosystems, Igbariam, Nigeria (Table 11).

CASSAVA (*MANIHOT ESCULENTA CRANTZ*)

Cassava (*Manihot Esculenta Crantz*) is an introduced crop, native of South America (Okigbo, 1980). Okigbo (1989) found that crop wastes such as those of legumes, rice and maize increased yield of cassava as mulch. However, farmers rarely use chemical fertilizer due to scarcity and cost, hence the dependence on cheap organic sources of nutrients. These reasons necessitate research on increasing effectiveness of organic manures and suitable rate of application. The effect of digester effluent was compared with pig and cattle manure (Weite *et al.*, 1998) and it was found that bio-digester effluent gave higher biomers, yield and protein content of cassava. These necessitate the biofertilizer production using agriculture wastes inoculated with broad spectrum microorganism accelerated composed in an

anaerobic bio digester (biofertilizer production, (www.academia.edu/video/lBboEl). One of the factors responsible for low yield is declining soil fertility. In the past, soil fertility has been sustained through long fallowing (Agboola and Unamma, 1994). OBD-Biofertilizer production, visit website (https://www.youtube.com/watch?v=Hi_OpgVcFcg biofertilizer) from biowaste in anaerobic digester inoculated with beneficial microbes that exhibit differing metabolic capabilities (Tables 1 and 2). Biofertilizers applied as soil inoculants, multiply in nutrient cycling and benefit crop productivity (Singh *et al*., 2011). Microorganisms inoculated in the biofertilizer production produce microbial enzymes and metabolites, which mimic the multiplicity of biocontrol mechanisms, set up by microorganisms (Huang and Chen, 2008; Karasuda *et al*., 2003). This category includes microbial secondary metabolites and hydrolytic enzymes as glucanases, proteases, lipases, and chitinases. These molecules can be used alone or, better, in combination with NPK chemical fertilizers reducing impacts on the ecosystems (Berini *et al*., 2018).

Yadav *et al.*, (2000) obtained consistently higher crop yield with NPK fertilizer mixed with organic manure over NPK inorganic fertilizer alone, Ano and Emehute (2004) also obtained higher ginger rhizome yield with organic manure mixed with inorganic fertilizer over inorganic fertilizer alone. Complementary use of organic manure and inorganic fertilizer improves the soil resource base. The effect of biofertilizer on cassava microbiome and phytobiome is unknown or under investigation. This book chapter reports results application of accelerated OBD-Biofertilizer applied alone and in combination with inorganic fertilizer NPK (15:15:15) and its impacts on the cassava crop yield and growth components (Figure 1). Also, reviewed literature on crop cultivation constraints, impacts of pests and diseases on cassava crop and the implications of application of biofertilizer to the integrated nutrient managements of the crop quality and soil health.

Cassava Cultivation Constraints

Many diseases are caused by pathogens, whose damage symptoms appear on the leaves, stems and storage roots (Miskito *et al*., 2000). The common diseases of cassava are: cassava *mosaic* disease, cassava bacterial blight, cassava *anthracnose disease*, cassava bud *necrosis* and root rot. Some of these diseases attack the leaves and stems of cassava plants while others attack the storage roots, Tables 6 and 7 (Olugbenga *et al.*, 2011). Cassava mosaic disease is caused by the African cassava mosaic virus which occurs inside the leaves and stems and causes yield reductions of up to 90 percent (IITA, 2008). Economical damage by diseases, pests and weeds of cassava is relatively moderate, although white flies can be a menace in some regions, if the problem is not identified early, and remedial action not implemented in a timely manner (Figures 3 and 4). Correct identification of the pest and an understanding of its behaviour, including its most vulnerable stages would provide insights into its management. Care must be then taken if pesticide application is contemplated, since there is the likelihood of high residual levels remaining in the product after harvest if an inappropriate formulation is used. Biopesticides can exert fungicidal, insecticidal, or nematicidal action, a combination of them and possibly other auxiliary functions such as bird and mammal repellents or herbicides. According to recent classifications (Czaja *et al*., 2015; Olson, 2015).

Biocontrol action is due to multiple synergic mechanisms, generally including: i) production of antibiotics and other secondary metabolites (e.g., phenazines by *Pseudomonas spp*., lipopeptides by *Bacillus spp.*,

and hydrocyanic acid by Rhizobia); and ii) secretion of lytic and defense enzymes (e.g., chitinases, glucanases, peroxidases, polyphenol oxidases, and phenylalanine ammonia lyases produced by *Trichoderma, Fusarium, Rhizoctonia, Serratia, Streptomyces and Bacillus strains*) (Leahy *et al.*, 2014; Parnell *et al.*, 2016). The drawback of using living microorganisms is that their efficacy is often unpredictable under changing field conditions, and their fitness is reduced by the presence of an indigenous microbiota difficult to displace by non-native microorganisms (Neeraja *et al.*, 2010a; Parnell *et al*.,2016). Additionally, the antagonistic interactions occurring in formulations containing more than one microbial species limit their potential in integrated pest management strategies (Gadhave *et al*., 2016; Xu *et al.*, 2011).

REGENERATIVE AGRICULTURE | CASSAVA

Organic 3.0 encapsulate regenerative agriculture (RA), refers to a set of agricultural techniques that improve soil health by increasing soil organic matter and the carbon content of soil. The critical goal for Organic 3.0 is to inform consumers and reconnect health, environment and product quality as essential elements of general human well-being as well as of the agri-food system (Rahmann *et al.*, 2017). RA seeks to combine the best conventional, organic, and biological farming practices into a system that improves productivity while enhancing ecosystem services with different mindset and different management strategies. It seeks to address the root cause of production problems rather than simply treating the symptoms. RA takes into consideration the environmental and societal implications of our food production systems. Conventional agricultural practices of the past half century have produced abundant food but have done it at tremendous environmental and socioeconomic cost. These practices often relied on 'mining' the soil rather than improving it and have led to degraded soil, lost future production potential, and shrinking rural communities. RA not only improves soil health, productivity, and resilience to weather extremes, raising farm yields and income while strengthening regional food security in the face of a changing climate, but can also form part of a region's broader climate strategy. Under improved management, soils have the potential to absorb hundreds of millions of tons of atmospheric CO_2 more than they do today.

Why Regenerative Agriculture Biotechnology?

Cassava cultivation could contribute significantly to climate change adaptation in Africa, based on the crop's wider tolerance range for moisture availability than other staple crops (such as maize, millet, sorghum, rice, and beans) Jarvis *et al.*, 2012. Later, it was observed that cassava production will benefit immensely from future climate change in the eastern part of Nigeria (Mbanasor *et al.*, 2015). Studies have indicated that from 2050 to 2100, the average temperature across Nigeria will increase, with a corresponding decrease in rainfall at the central and southern sub-regions [Ogunrinde *et al.*, 2019; Shiru *et al*., 2019a; Shiru *et al*., 2019b; Adeniyi and Oyekola, 2017]. Therefore, cassava cultivation could benefit from the vast lands that will no longer be suitable for cocoa cultivation in the current cocoa belt (Schroth *et al*., 2016) Elsewhere, the dominant influences of soil fertility status, fertilizer application, and genotype on cassava yield in the tropics were recently reported (Phanupong, *et al.*,2015) Aliyu *et al*., 2019; Biratu *et al.*, 2019 a, b; Biratu *et al.*, 2019a, b; Senkoro,2018) ; Ezui *et al.* ,2016). Soil organic matter (SOM) [A] refers to plant and animal matter in soil in various stages of decomposition as well as the cells and tissues of soil microbes. Biofertilizers application that contain living microorganisms is one of the management practices that can help to maintain or increase the content of organic matter

and improve soil fertility in arable soils. Bio-fertilizers field application on cassava have not been documented especially within the humid tropics and their potential effects on soil properties and processes, especially but scholars have affirmed the otential impacts (Mayer *et al.,* 2010; Dinesh *et al.,* 2010; Khaliqetal.2006; Piotrowskaetal.2012; Wuetal.2005; Zhaoetal, 2005). Acceleration of the humification of fresh organic matter that is, introduced into the soil influence of biofertilizers was reported by scholars (Valarini *et al.,* 2003; Fatunbi and Ncube, 2009; Piotrowska *et al.*, 2012). The application of specially composed biofertilizer (OBD) is very important in order to accelerate the transformation of the biomass [https://www.academia.edu/video/jEepAj]. The positive influence of organic matter on soil functionality is sustained (Lal, 2011; Krasowicz *et al*., 2011).

The increasing global demand for food can be met by agricultural expansion (e.g. clearing forest land for crop production) or intensification (e.g. increasing yields from existing crop and grassland) (Tilman *et al*., 2011). 50 years ago, the dominant form of agricultural development has been intensification with low consideration of the environmental effects (Pretty *et al.,* 2018). To counter this, "sustainable intensification" is promoted as an approach for increasing food production from existing farmland whilst placing less pressure on the environment and without undermining future production (Godfray and Garnett, 2014). The approach is goal, rather than means-orientated, with the most appropriate form of farming dependent on the context (Garnett *et al.*, 2013). The regenerative organic certification scheme builds on USDA's certified organic standards and has three pillars relating to soil health, animal welfare, and social fairness. Years of analysis of the complexity of soil ecosystems has allowed the scientific community to understand that microbial communities form the basis of highly fertile as well as carbon-rich soils. [B, C] Evidence shows that it is possible for healthy microbial communities to produce sufficient nutrients for high crop yields, as well as promote biodiversity on farmland, which acts as a natural pest control system. (Sharanaiah *et al.*, 2018). Tillage any form of ploughing or disrupting the soil demonstrably leads to the oxidation of soils, damage to mycorrhizal fungi networks and ultimately to loss of organic carbon, and therefore of fertility (Zahangir, 2005). The use of chemical inputs of any kind is [D] not consistent with the goals of maintaining [E] and enhancing soils' capacities to sustain the food system in the long-term (Miller and Krusekopf, 2018), as it requires a transition to no-till systems and an elimination of chemical inputs, the implementation of the Regenerative Agriculture paradigm throughout regions and corporate supply chains would require a holistic restructuring of the way societies produce food. This process has begun with developing sets of practices that need to be customized for each specific context in the broad spectrum of farming ecosystems. The adoption of a regenerative approach is stimulated by a recognition of input costs that could be saved by restoring ecosystem fertilization and irrigation processes (LaCanne and Lundgren, 2018).

A. Bot, Alexandra, and JoséBenites. (The Importance of Soil Organic Matter: Key to Drought-Resistant Soil and Sustained Food Production. Food and Agriculture Organization of the United Nations, 2005. Library of Congress ISBN, http://www.fao.org/3/a-a0100e.pdf.
B. Soilquality.org.au Soil Biological Fertility Fact Sheets http://www.soilquality.org.au/factsheets/soil-biological-fertility..
C. "Institute National de la Recherche Agronomique" (INRA).
D. World Bank, Climate Smart Agriculture. http://www.worldbank.org/en/topic/climate-smart-agriculture.

The Global Soil Organic Carbon Map (GSOCmap) is a recent breakthrough by the GSP that will act as a "consultative and participatory process involving 110 countries" for measuring the soil-carbon impact of agricultural reform practices. [F] Regenerative agriculture could potentially become the dominant paradigm as a strong coalition of actors, including scientists, farmers, consumers, and decision-makers is growing at the national and international levels. The Regenerative Agriculture movement advocates for the adoption of agroecological practices currently used by millions of smallholder farmers throughout the world. [C] Internationally, RA is promoted by a broad coalition that includes scientists (scientific advisory councils like INRA, CGIAR), [G] governments (notably the French Government), and NGOs, including the International Federation of Organic Agriculture Movements (IFOAM), [H] Via Campesina, Nature Conservancy, Oxfam [C] and Regeneration International.

RATIONALE AND SIGNIFICANCE

The development of the technology of planting one stake of cassava per hill or stand using 1m x 1m spacing, by the National Root Crops Research Institute (NRCRI) Umudike, Nigeria, was a major breakthrough in cassava-based farming system practices. This technology encourages the production of larger roots, high yield per stand and makes other farm operations such as weeding and fertilizer application easy. The reasons advanced for the low adoption was low cassava population, low yield per unit area and weed growth in farms. Therefore, farmers are demanding for an increase in the number of cassavas to be planted per hill or with little or no chemical pesticides applied during cultivation. Cassava yields are compromised by pests such as whiteflies, mites, and weevils, which cause significant crop losses through the spread of viral disease and direct damage to plants (Table 6) within the global crop cultivation zones (Figure 1.1). Whiteflies are vectors for viral diseases such as cassava mosaic disease (CMD) and cassava brown streak disease (CBSD), which can reduce yields by up to 40% (Legg and Fauquet, 2004). Studies shows the enhanced by the production of bioactive substances having similar effects as that of growth regulators besides nitrogen fixation through biofertilizer leading to greater dry matter production was reported by Ramanandam *et al*., 2008 and similar findings were reported by Subbiah (1994). The higher dry matter production is attributed to the cumulative effect of progressive increase in the growth attributes, viz., plant height, stem girth and number of leaves per plant. Similar results have been reported earlier in cassava (Ammanullahkhan, 1997). The review on cassava microbiome ecology summarizes and discuss data on putative mechanisms of cassava crop resistance to environmental challenges via mobilization of a hidden reserve (dormant endophytic inhabitants). An endophytic microbiome shaped by extrinsic and intrinsic factors like the microbiome ecology, impact on plant functions, bacterial endophytes in plant disease control and the biofertilizer characteristics (Tables 6 and 7).

E. OXFAM issue briefing, Building a New Agricultural Future: supporting agro-ecology for people and the planet, April 2014.https://www.oxfam.org/sites/www.oxfam.org/files/ib-building-new-agricultural-future-agroecology-280414-en.pdf.
F. Today, 75% of the world's food is produced by family farms which use very limited amounts of chemical inputs if at all, and whose large majority are smaller than one hectare (2.5 acres). http://www.fao.org/news/story/en/ item/260535/icode/
G. IFOAM, Organics International at the Regeneration International Conference, 10/06/2015.https://www.ifoam.bio/en/ news/2015/06/10/ifoam-organics-international-regeneration-international-conference.
H. FAO, Soil Organic Carbon Map. http://www.fao.org/global-soil-partnership/pillars-action/4-information-and-data/ global-soil-organic-carbon-gsoc-map/en/

The key point of the Book Chapter is that inoculation (biofertilizer) on a plant (cassava) by beneficial microbe's bacteria and fungi will be successful, if it possesses beneficial community members that can be activated by specific bioagents or abiotic stressors (Tables 10 and 14). The success of inoculation may depend to a large extent on the endophytic microbial community structure and activity, being one of the multiple plant-host-genotype- based intrinsic factors that influence plant performance. Understanding of the processes that lead to enrichment of specific endophyte species due to environmental stress will offer great benefits for crop yield, post-harvest operations, bio-protection, soil quality and health.

Microbiome Ecology

The rhizosphere is a key habitat documented to contain vast microbial diversity (Egamberdiyeva *et al.*, 2008; Mendes *et al.*, 2011), where soil functions as the medium in which complex signaling occurs among microbes and plants, accomplished by exudates interaction between roots and the adhering/surrounding soil and influenced by climatic factors, the rhizosphere in turn impacts the plants and microbiota that utilize this habitat as an information highway (Bais *et al.*, 2004). Moving closer into proximity with the plant, the next habitat is the rhizoplane, which refers to the surface of the plant tissues in contact with the soil (roots and rhizomes). Microbes that can exist in an adherent form to the plant tissues are termed epiphytes. Endophytes refer to the microbial genomes located inside plant tissues (Bulgarelli *et al.*, 2013). It is important to understand that microbial lifestyles are complex and many microorganisms are not restricted in their interactive potential, thus enabling them to exist as facultative epiphytes and endophytes as dictated by other biotic and abiotic factors. Cassava cultivation using biofertilizer enhance the epiphytes and endophytes microbes in the rhizosphere. Ecosystem services are intricately linked to plant functional traits, of which several are likely mediated by microbes including soil formation, decomposition of organic matter, nutrient mineralization, and primary productivity (De Bello *et al.*, 2010). The impact of the rhizosphere microbiome on plant productivity has not escaped those that are familiar with crops, where modulating soybean (*Glycine max*) cultivars have been historically manipulated to enhance yield through alterations to their interactions with various.

Rhizobium microbial partners (Harris *et al.*, 1985; Heath and Tiffin, 2009; Kiers and Denison, 2008). The plant growth promoting activities that many rhizosphere-dwelling prokaryotes provide to plants include nitrogen (N_2) fixation (James, 2000; Martinez-Romero, 2006), phosphate solubilization, and production of plant-growth hormones (Hardoim *et al.*, 2008). Whether from a macro (plant-centric) or a micro (microbiome-centric) perspective, the plant microbiome can exert influences on plant trait expression through top-down and bottom-up interactions (Figure 2). Microbial exudates comprise much of the basis of modern antibiotics and antifungals. The excretion of antimicrobial substances is a common feature found in prokaryotic and eukaryotic microorganisms, and only a miniscule fraction of these organisms is cultivable (Piel, 2011), making the development of meta-transcriptomics and metabolomics research now imperative. In addition to exuding antimicrobials that assist in plant immunity, microbes also exude low-molecular-weight effector molecules detectible to plants through what are known as pathogen- or microbe-associated molecular patterns in plants that trigger the immune response (Boller and He, 2009).

Impact on plant functions

Plants can actively construct the rhizosphere microbiome and that this community by regulating or recruited by the plant to serve as protection from pathogens (Bernedsen *et al.,* 2012; Friesen *et al.,* 2011). An example of this is found in the bacterial production of enzymes that degrade N-acyl-L-homoserine lactones (AHLs), commonly found among several prokaryotic genera, which inhibit quorum sensing. Plants were capable of recruiting beneficial bacteria expressing high levels of AHL-degrading enzymes when exposed to a pathogen, thus suppressing virulence gene expression (Reading and Sperandio, 2006). Plants also recruit activity within the microbiome specifically to stimulate AHL degradation (Teplitski, Robinson and Bauer, 2000). Microbe like *fluorescent pseudomonads*, many of which are capable of producing the antimicrobials 2, 4-diacetylphloroglucinol (2, 4-DAPG) and derivatives of phenazine (Phz). These bacteria are common to the rhizosphere of a diverse array of plant species (Mavrodi *et al.,* 2011). Both antimicrobials are broad spectrum and provide protection against a wide range of plant pathogens, many of which are fungal (Raaijmakers *et al.,* 2009). 2,4-DAPG and Phz derivatives also serve to reduce mineral content in the rhizosphere, perform plant regulatory and signaling functions that include alterations to exudate profiles, and play a role in the induction of plant systemic resistance (Doornbos *et al.,* 2012; Matilla *et al.,* 2007; Mavrodi *et al.,* 2011).

The dual growth-promoting traits (i.e., phosphate solubilization and ACC deaminase production (Baig *et al.,* 2012); an example of multiplicity of microbial function as a complementary aspect of functional redundancy. Plant microbiome is informative about plant health, at both the individual and community level (particularly in monoculture crops), where a healthy versus diseased state of the plant community can be reflected in the composition of the rhizosphere microbiome (Burdon and Thrall, 2009). PGPR stimulate induced systemic resistance (ISR) in plants, marked by priming of jasmonic acid-inducible genes in leaves (Van Wees *et al.,* 2008), effectively stimulating immunity conferring resistance to a broad range of pathogens (Pineda *et al.,* 2010; Van der Ent *et al.,* 2009; Van Oosten *et al.,* 2008). In turn, plants with activated ISR display increased or enhanced defence signaling aboveground (Ahmad *et al.,*2011) and belowground (Neal *et al.,* 2012; Neal and Ton, 2013). The microbiome located inside plant tissues reflects the microorganisms living in an endophytic lifestyle (Bulgarelli *et al.,* 2013). Many plant species have been shown to harbour endophytes (Compant *et al.,* 2008; Hallmann *et al.,* 1997; James *et al.,* 2002), and wild cultivars, including invasive species (Rout and Chrzanowski, 2009).

Alterations to root architecture have important implications for overall plant health. Take, for an example, border cells in roots; increased numbers of border cells conferred increased resistance to fungal pathogen infection (Chen *et al.,* 2012), likely due to the function of border cells in extracellular trapping of microbes that has been shown to provide a defence for the root tip (Curlango-Rivera *et al.,* 2013). Influencing plant architecture through enhancing root growth is a primary motivation behind mining the microbiome for this Plant Growth Plant trait, which is of interest to agribusiness. Besides assisting plants in niche expansion (or invasion spread) through enhanced root and rhizome production, the microbiome can impact plant ecology through influences exerted on plant–plant competition (Klironomos, 2002), pollination (Cahill *et al.,* 2008), herbivory, and defence (Friesen *et al.,* 2011). Extreme environments, such as deserts, are another area where plant microbiome research efforts have correlated mechanisms of the microbiome permitting the plant to tolerate drought stress (Kaplan *et al.,* 2013). Given the complexity of chemical communication capabilities within the microbiome, and

phytohormones, the link between microbial mediated ecosystem process of nutrient cycling and plant productivity lies embedded in the rhizosphere matrix. The complexities of the defined endophyte community structures indicate that they are not random guests in the plant habitat, but potentially play essential roles, interacting with the plant host and influencing plant physiology (Gaiero *et al.,* 2013), endophytes can significantly affect the physiology of the plant host (Pirttilä *et al.*, 2004; 2005).

Bacterial Endophytes in Plant Disease Control

The plant microbiome is currently attracting a lot of research interest due to its ability to buffer plant hosts against abiotic and biotic stress, facilitate nutrient uptake and nutrient use efficiency, and promote growth (Hardoim *et al*., 2015; Turner *et al*., 2013; Santoyo *et al*., 2016; Gaiero *et al*., 2013; Brader *et al.,* 2014; Truyens *et al.,* 2015; Nair and Padmavathy, 2014); Ma *et al.,* 2011; Iqbal *et al.*, 2017; Blain *et al.,* 2017). Endophytic bacteria can be used to improve plant productivity and stress tolerance in the absence of pesticides and inorganic fertilizers, and to facilitate phytoremediation heavy metals and hydrocarbons, but more research is needed on how to best inoculate plants in field settings (Busby *et al.,* 2017). Nigeria is the largest cassava producer in the world (Figure 3) with 8.76 t/ha average cassava yield (Figure 4) significantly lower than the global average yield of 11.1 t /ha, and much lower than India (34.2 t /ha) FAO (2011) and Laos (32.1 t /ha) (FAO, 2019). Africa, crop production in Nigeria is mainly determined by the climatic conditions (Adefisan and Abatan, 2015; Omotosho et al., 2013). African farmers, was reported 2019 will benefit from climate change by 2100 (Seo *et al.,* 2009) specifically with cassava cultivation could contribute significantly to climate change adaptation in Africa, based on the crop's wider tolerance range for moisture availability than other staple crops (such as maize, millet, sorghum, rice, and beans). Jarvis *et al.,* 2012. Later, it was observed that cassava production will benefit immensely from future climate change in the eastern part of Nigeria (Mbanasor *et al*., 2015).

Although, studies have indicated that from 2050 to 2100, the average temperature across Nigeria will increase, with a corresponding decrease in rainfall at the central and southern sub-regions (Ogunrinde *et al.,* 2019; Shiru *et al.,* 2019a; Adeniyi *et al.,* 2017; Abatan *et al.,* 2016; Abiodun *et al.,* 2013). Cassava cultivation could benefit from the vast lands that will no longer be suitable for cocoa cultivation in the current cocoa belt (Schroth *et al.,* 2016). Dominant influences of soil fertility status, fertilizer application, and genotype on cassava yield in the tropics were reported (Ogunrinde *et al.,* 2019; Shiru *et al.,* 2019ab; Adeniyi *et al.,* 2017; Abatan *et al.,* 2016; Abiodun *et al.,* 2013; Aliyu *et al.,* 2019; Biratu *et al.,* 2019; Biratu *et al.,* 2018; Senkoro *et al*.,(2018). Endophytic bacteria can be used to improve plant productivity and stress tolerance in the absence of pesticides and inorganic fertilizers, and to facilitate phytoremediation heavy metals and hydrocarbons, but more research is needed on how to best inoculate plants in field settings (Busby *et al*., 2017). Endophytes are microorganisms that live within the plants' tissues without causing any damage to the host (Lodewyckx *et al*., 2002). Entophytes could be classified as fungi, bacteria or algae (Schulz and Boyle, 2006). Endophytes primarily assist in promoting the growth of plants that they inhabited as shown Figure 5. Facultative endophytes grow outside its host plant. Meanwhile, obligate endophytes are dependent on their host plant for their growth and survival (Hardoim *et al*., 2008). Endophytic bacteria are correlated with the enhanced plant growth by the production of hormones that increase accessibility of nutrients, such as nitrogen, potassium and phosphorus (Glick, 2012), Table 2. While induced disease resistance activities are allied with the abilities to produce secondary metabolites,

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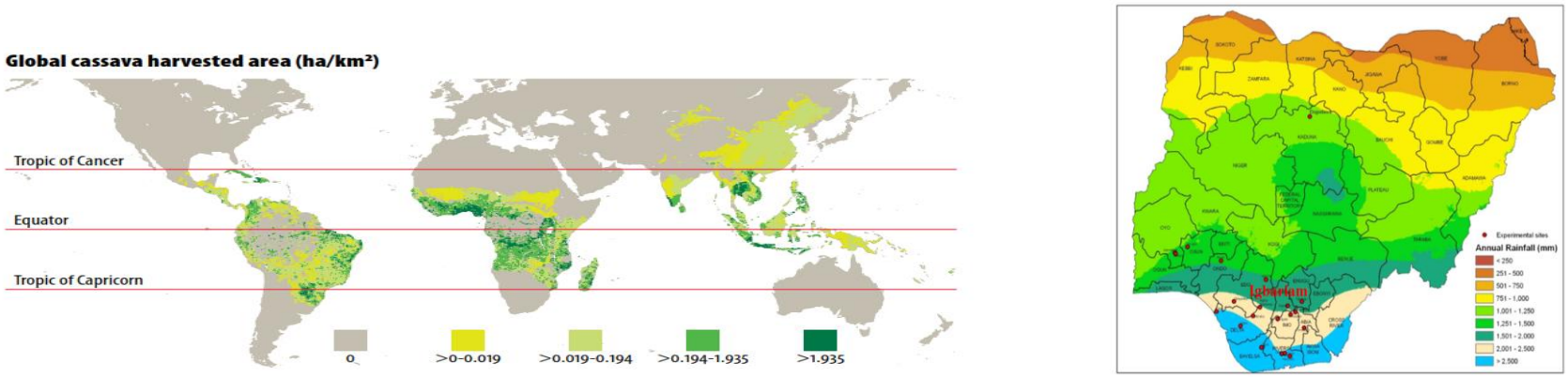

Figure 3 | Geographic distribution of crop areas, yields and physiological types [I] and cassava field trial site Agro ecology humid forest [J] (Igbariam, Anambra state, Nigeria).

While induced disease resistance activities are allied with the abilities to produce secondary metabolites, such as antibiotics or chitinase enzyme, which can inhibit growth of plant pathogens and act as bio-control agents (Christina *et al*., 2013; Wang *et al*.,2014) Figure 8 below for narrative. Endophytic bacteria have the capacity to cope with phytopathogenic fungi with induced systemic resistance (ISR) (Pieterse *et al.,* 2014). Due to their beneficial function such as plant growth promotion and disease control, endophytes can be used in the form of bio-formulations (seed treatment, soil application and seedling dip) in agriculture (Table 4). Endophytic bacteria can also induce seedling emergence and stimulate plant growth (Chanway, 1997) under stress conditions (Bent and Chanway, 1998). The genera of Bacillus and Pseudomonas are identified as frequently occurring bacteria in agricultural crops (Seghers *et al.,* 2006; Souza *et al*., 2013). It has been reported that most of Gram-negative endophytes act as agents of biological control (Kobayashi and Palumbo, 2000), while among the Gram-positive bacteria, the dominant endophytic species are Bacillus species (Gupta *et al*., 2002; Bacon and Hinton, 2007). The root exudates contain that colonize different bacterial genera and they differ normally according to plant species (Bisseling *et al*., 2009)

[I.] Monfreda *et al*., 2008
[J.]. Yomeni *et al.*, 2010

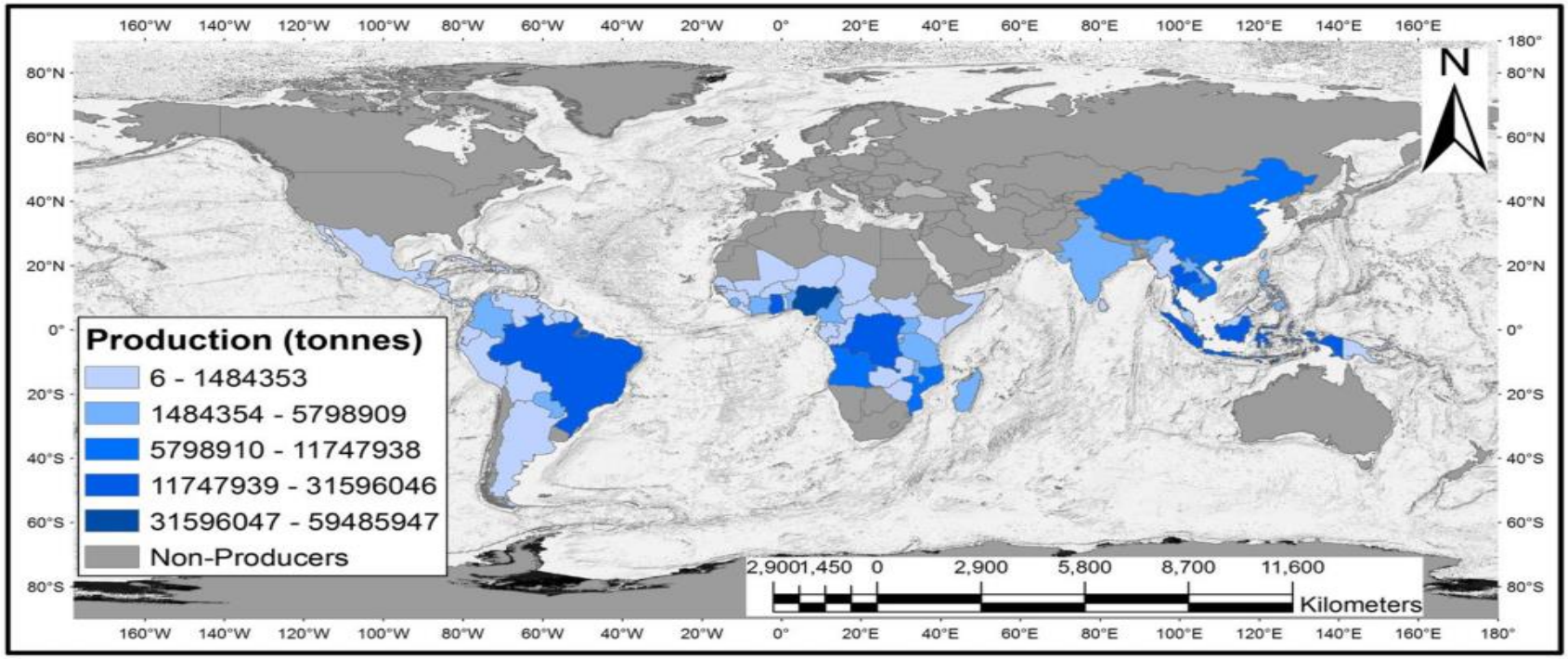

Figure 4 | Global cassava production quantity for 2017, indicating the dominance of Nigeria. (FAOSTAT data was processed in the ArcGIS environment) adapted from Akinwumiju *et al*., 2020.

The apical root zone having thin-walled surface of root cells includes cell elongation and the root hair zone (zone of active penetration), and the basal root zone with small cracks are the preferable sites of bacterial attachment and subsequent entry caused by the emergence of lateral roots

(zone of passive penetration) Figure 5. Root colonization or rhizospheric beneficial microorganisms are familiar bio-control agents and plant growth promoters. Innumerable compounds such as hydrocyanic acids (HCN), DAPG, phenazines, pyrrolnitrin, enzymes and phytohormones to protect plant from toxic effect of fungal pathogens are considered as the significant products to help endophytes to be colonized in rhizosphere (Castro-Sowinski *et al.*, 2007; Ramette *et al.*, 2011; Jousset *et al.*, 2011), Table 18.

The apical root zone having thin-walled surface of root cells includes cell elongation and the root hair zone (zone of active penetration), and the basal root zone with small cracks are the preferable sites of bacterial attachment and subsequent entry caused by the emergence of lateral roots (zone of passive penetration) Figure 5. Root colonization or rhizospheric beneficial microorganisms are familiar bio-control agents and plant growth promoters. Innumerable compounds such as hydrocyanic acids (HCN), DAPG, phenazines, pyrrolnitrin, enzymes and phytohormones to protect plant from toxic effect of fungal pathogens are considered as the significant products to help endophytes to be colonized in rhizosphere (Castro-Sowinski *et al.*, 2007; Ramette *et al.*, 2011; Jousset *et al.*, 2011), Table 18. Bacteria are able to trigger signaling pathways to produce extracellular metabolites with higher toxicity for other microorganism led to destruction of higher pathogen, called induced systemic resistance (ISR). Myriad of bacteria has been documented for beneficial effects, alleviation of several abiotic and biotic stresses. *Pseudomonas* and *Bacillus spp*. have been studied as potential candidate to provide ISR to plants (Chakraborty *et al*., 2006), In Table 6, P in the soil in lesser quantities (Khan *et al.*, 2006). However, plants are well adapted to uptake of P from low concentration soil solution (Jungk, 2001). Therefore, it is presumed that the supply and availability of P to the root surface is influenced by the root and microbial processes by PGPR (Figure 9). Different mechanisms can be broadly studied under (1) Biofertilization, and (2) Biocontrol of pathogens. Biofertilization encompasses: (a) N_2Fixation, (b) Siderophore production, (c) P inorganic solubilization by rhizobacteria. Biocontrol involves: (a) Antibiosis, (b) Secretion of lytic enzymes, and (c) Induction of Systemic Resistance (ISR) of host plant by PGPR, Figure 8 below.

BIOFERTILIZER

A key advantage of beneficial microorganisms is to assimilate phosphorus for their own requirement, which in turn available as its soluble form in sufficient quantities in soil. *Pseudomonas, Bacillus, Micrococcus, Flavobacterium, Fusarium, Sclerotium, Aspergillus and Penicillium* have been reported to be active in the solubilization process (Pindi and Satyanarayana, 2012). A phosphate-solubilizing bacterial strain Micrococcus sp. has polyvalent properties including phosphate solubilization and siderophore production (Dastager *et al*., 2010). Similarly, two fungi *Aspergillus fumigatus* and *A. Niger* were isolated from decaying cassava peels were found to convert cassava wastes by the semi-solid fermentation technique to phosphate biofertilizers (Ogbo, 2010) *Burkholderia vietnamiensis*, stress tolerant bacteria, produces gluconic and 2-ketogluconic acids, which involved in phosphate solubilization. Potassium solubilizing microorganisms (KSM) such as genus *Aspergillus, Bacillus* and *Clostridium* are found to be efficient in potassium solubilization in the soil and mobilize in different crops (Mohammadi *et al*., 2012). Mycorrhizal mutualistic symbiosis with plant roots satisfies the plant nutrients demand (Kogel *et al*., 2006) which leads to enhance plant growth and development, and protect plants from pathogens attack and environmental stress (Lamabam *et al.*, 2011). Pseudomonas aeruginosa has been shown to withstand biotic and abiotic stresses (Pandey *et al*., 2012). Paul and Nair (2008) found that P. fluorescens MSP-393 produces osmolytes and salt-stress induced proteins that overcome the negative effects of salt. Microbial inoculants genera in the OBD-Biofertilizer are isolated using the growth media in Table 1 from different agro biowaste and inoculated into the composted biofertilizer, Table 3.

Biowaste Recycling to Biofertilizer

Agriculture biowaste materials composted by anaerobic digester (AD) to biofertilizer (https://www.academia.edu/video/lBboEl) and inoculated with broad spectrum inoculants OTAI AG® (Tables 1 and 2) and Oso Bio-Degrader (OBD-Plus®) called microbial inoculants. During the

anaerobic ingestion process (Figure 6) and see video available for the biofertilizer production, Abeokuta, Nigeria. www.youtube.com/watch?v=pG2ODAx3ICY. It might be anticipated that a

measurable increase in the proportion of readily available N would occur in these materials, as a result of the digestion process. In addition to nutrient impacts, a number of benefits are claimed to accrue as a result of AD, including a reduced risk of odour nuisance and a reduction in viable pathogenic organisms (Sood, 2006). Inoculated beneficial microbe's direct analysis of metabolites in situ has been achieved for antibiotic lipopeptides from several Bacillus subtilis and for pyrrolnitrin, 2,4-diacetylphloroglucinol and phenazine-1-carboxylic acid from *Pseudomonas fluorescens* strains (Raaijmakers *et al*., 2012).

MATERIALS AND METHOD

OTAI AG® is PGPR and beneficial microbial inocula (Table 4) an easy-to-use and economical carrier material (composted) biofertilizer production with industrial standardized process of production can be defined (Schmidt, 2005). Biofertilizers price is not the same as composts and have been tested as growth media for PGPR (Schmidt, 2005; Vidyarthi *et al.*, 2000). PGPR and/or arbuscular mycorrhizal fungi (AMF) (Jeffries *et al*., 2003) combine inoculation often resulted in increased growth and yield, compared to single inoculation through improved nutrient uptake (Bashan *et al*., 2004; Belimov *et al*., 1995) and resultant interaction of bacteria and AM fungi have beneficial functions related to nutrient uptake, particularly when PGPR (Barea *et al*., 2002 ; Vassilev *et al*., 2001) and N_2-fixing bacteria (Biro *et al*., 2000; Secilia and Bagyaraj, 1987) are involved. Survival of the PGPR (Tables 3 and 4) is important both during the storage period of the bioproduct and after being introduced into the soil (Trzcin´*et al.*,2001) for solid carriers, powder or granules. Standard sizes of the powder material may vary from 75 µm to 0.25 mm (Smith,1992) and application methods depend on the kind of crop concerned can be inoculated by broadcasting the inoculum over the soil for regenerative agriculture. [K]

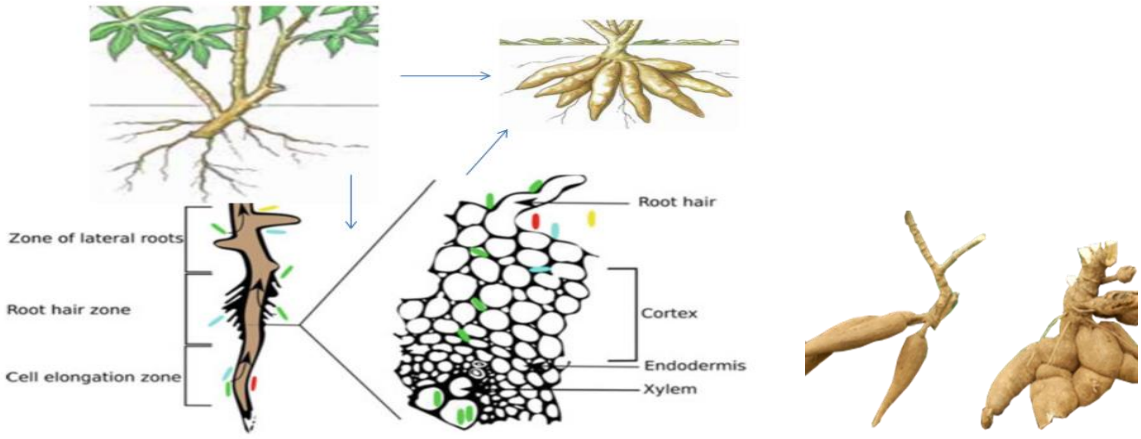

Figure 5 | Endophytic bacterial colonization in cassava plants modelled. Bacteria can enter a plant at several root zones as indicated above (Adapted from Maheshwari and Annapurna, 2017). Endophytes can either remain at the site of entry (indicated in blue) or move deeper inside or occupy the intercellular space of the cortex and xylem vessels (indicated in green). Red and yellow represent rhizospheric bacteria which are unable to colonize inner plant tissues.

K. Regenerative Agriculture https://www.academia.edu/44877314/Regenerative_Agriculture_4_0_Tool_Box.

Table 1. Microbial culture techniques of beneficial microorganisms present in OBD-Biofertilizer.

S.N.	Growth media	Microbes	References
1	Ammonium mineral salt	Methylotrophs	Holland et al. (2000)
2	Congo red yeast mannitol	Rhizobium	Yumoto et al. (2002)
3	DSMZ-97, DSMZ-823, DSMZ-1184; OS	Halophilic archaea	Yadav et al. (2015c)
4	Jensen agar	N2-fixing bacteria	Jensen (1965)
5	King's Bagar	Pseudomonas sp.	Mishra et al. (2009)
6	Luria Bertani agar	Endophytic bacteria	Ventosa et al. (1982)
7	Nutrient agar	Heterotrophic bacteria	Ramesh and Lonsane (1987)
8	Potato dextrose agar	Fungus	Sehgal and Gibbons (1960)
9	Soil extract agar	Soil-specific microbes	Shivaji et al. (1988)
10	Tryptic soy agar	Arthrobacter	Shivaji et al. (1989)

broadcasting the inoculum over the soil surface, alone or together with seeds, or by in-furrow application, seed dressing, or coating; tree crops can be initially inoculated by root dipping or seedling inoculation (Muresuet *et al*., 2003). Chemical analyses: Total Nitrogen Kjeldahl procedure (Rhee, 2001; Available P Olsen's method (Olsen *et al*., 1954); Available K by Flame photometric method (Jackson, 1967); pH (Piper, 1967) Electrical Conductivity by Walkley Black method (McLeod, 1973); Micronutrients (Zinc, Iron, copper, Manganese) ppm atomic absorption Spectrophotometeric method using DTPA (Diethyl Triamine Penta Acetic Acid) by Lindsay and Norvell, 1978.

Bioferilizer Physico-Chemical Properties

Otaiku *et al.,* 2019a reported the nutrient content of the OBD-Biofertilizer indicated the followings: **Macroonutrients**: pH in water (5.8); %N (0.95); %P (3.1); % Ca (13.05); %Mg (0.79) %K (0.35); ppm Na (40.74); ppm Mn (456.78); ppm Fe (460.01); ppm Cu (17.47); ppm Zn (95.5); %Carbon (35.68) and C/N ratio (35.56).

Table 2. PGPRs Biosulfactants presents in OTAI AG® Inocula (Otaiku *et al*., 2019a).

N/S	Biosurfactants	Microorganisms	Economic importance	References
1	Rhamnolipids	Pseudomonas aeruginosa	Antimicrobial	Jadhav et al., 2011
			biocontrol properties	
2	Viscosin	Pseudomonas fluorescens,	lipopeptides	Banat et al., 2010
3	Ornithine lipids	Agrobacterium sp.	Bio-emulsifiers	Desai and Banat, (1997)
		Pseudomonas sp.	Bio-emulsifiers	
		Thiobacillus thiooxidans	Bio-emulsifiers	
4	Carbohydratel Liipid	P.fluorescens	Bio-emulsifiers	Nerurka et al.,
5	Protein PA	P.aeruginosa	Bio-emulsifiers	Hisatsuka et al., 1999
6	Whole cell	Cyanobacteria	Bio-flocculent	Levy et al., 1990
7	Surfactin/Iturin	B. subtilis,	Antimicrobial	Arguella et al., 2009
8	Subtilisin	B. subtilis	Antimicrobial properties	Sutyak et al., 2008
9	Aminoacids lipids	Bacillus sp	Antimicrobial properties	Cotter et al., 2005
10	Lichenysin		Enhance oil recovery	Yakimov et al., 2009

Micronutrients: Molybdenum (Mo) 20 ppm; Boron (B) 30ppm; Copper (Cu) 17.47ppm; Manganese (Mn) 456.78 ppm; Zinc (Zn) 95.5 ppm; Iron (Fe) 460.01 ppm; Sodium (Na) 40.74 ppm. **Essential Nutrients:** Humic acid 2.1. Beneficial micro-organisms present are in the OBD-Biofertilizer (Table 3) : *Azotobacter spp, Clostridium spp, Bacillus spp, Esherichia, Rhizobium spp*. Fe (iii) reducing Bacteria *(Shewanella putrefaciens),* Phosphate Solubilizing Fungi *(Actnomycetes),* Potassium Solubilizing Bacteria *(Bacillus mucilaginous),* Phosphate Solubilizing Bacteria *(Rhizobium, Agrobacterium),* Sulphur Oxidising Bacteria *(Thiobacillus thioxidans).*

Materials | Shea cake and Poultry waste (SPW), Swine waste (SW), Wood ash (WA)
Ratios | SPW 12: SW 12.5: WA 1
Percentages | PW 47.06; SW 49.02; WA 3.92
Microbial Inoculants | OBD-Plus ® and OTAI AG® [Tables 2 and 4]

The trial was conducted at the National Root Crops Research Institute's, substation in Igbariam in 2012/2013 cropping seasons, Anambra state, Nigeria. The soil was an Ultisol and had a pH in water of 5.0, 2.06% organic matter, 0.14% total nitrogen, 5.8 mg /kg Bray 2 P, exchangeable calcium, Mg and K of 4.60 cmol/kg, 2.50 cmol/kg and 0.12 cmol/kg respectively. The field was slashed ploughed, harrowed and ridged, thereafter plots each measuring 5 m x 5 m were marked out. Cassava stakes each measuring about 25 cm were then planted at 1m x 1m spacing on top of the ridges on 16th November 2012. Cassava Mosaic Disease (CMD) resistant varieties used was from the International Institute of Tropical Agriculture's (IITA) fields. The treatment was arranged in a randomized complete block design replicated three times. The treatments were as follow:

1. 300 kg/ha NPK 15:15:15 + 1.0 t/ha OBD- Biofertilizer
2. 300 kg/ha NPK 15:15:15 + 2.0 t/ha OBD-Biofertilizer
3. 300 kg/ha NPK 15:15:15 + 3.0 t/ha OBD-Biofertilizer
4. 300 kg/ha NPK 15:15:15 + 4.0 t/ha OBD-Biofertilizer
5. 2.0 t/ha OBD-Biofertilizer
6. 600 kg/ha NPK (15:15:15)
7. Control (no application).

The treatments were applied on 9th May, 2013. Harvesting of cassava was carried out in November 2013 and the fresh root yield was measured. The data obtained was subjected to analysis of variance. Significant treatment means were separated using Fischer's least significant difference (F-LSD) at 5% probability and see Plates 1 and 2 below.

RESULTS AND DISCUSSION

The effect of OBD-Biofertilizer applied alone and in combination with inorganic fertilizer on cassava root yield is shown in Table 9 and Figure 7. Application of either inorganic fertilizer, OBD-Biofertilizer alone, OBD-Biofertilizer in combination with inorganic fertilizer gave significantly higher cassava root yield ($P< 0.05$) than the control (no application). Inorganic fertilizer applied at the recommended rate of 600 kg NPK (15:15:15) / ha also gave higher cassava root yield than OBD-Biofertilizer applied either alone or in combination with inorganic fertilizer (Figure 7). OBD-Biofertilizer applied at the rate of 4 t/ha mixed with 300 kg NPK (15:15:15) /ha gave the highest cassava root yield of 31.2 t/ha which was however not significantly higher than 30.0 t/ha obtained with OBD-Biofertilizer applied at the rate of 3 t/ha

mixed with 300kg NPK (15:15:15)/ ha (Tables 9.10 and 11).Complementary use of OBD-Biofertilizer and inorganic fertilizer is therefore more beneficial than OBD-Biofertilizer alone in cassava production. This result is in agreement with Ano and Ikwelle (2000) and Mokwunye, (1978). Soils of the experimental site and indeed most Nigerian soils are highly weathered and have low activity clays (Ano, 1990) and therefore require application of soil amendment for high crop yield to be obtained. This explains why inorganic fertilizer NPK 15:15:15 applied at the recommended rate of 600 kg/ha or inorganic fertilizer applied at 300 kg/ha mixed with OBD-Biofertilizer gave significantly higher yield than the control, Figure 7.

Biofertilizer - Mechanism of action

The absence of a population of degrading microorganisms can be overcome by the inoculation of the plant rhizosphere with pollutant degrading strains and biosulfactants (Tables 1 and 2). This approach successful in reducing the levels of benzene, ethylene, toluene xylenes, hydrocarbons, polychlorinated biphenyls and pesticides in polluted environments (Johanson *et al.*, 2004; Yousaf *et al.*, 2010a; Germaine *et al.*,2006). The rhizosphere is defined as the volume of the soil over which roots have influence, and which is shared with soil bacteria. Plants release exudates in the rhizosphere likely to serve as carbon source for microbes (Gregory, 2006; Olson *et al.*, 2003). Consequently, rhizosphere microbes can promote plant health by stimulating root growth via production of plant growth regulators, enhance mineral and water uptake. Some bacteria, especially fluorescent pseudomonads, produce siderophores that have very high affinities for iron as compared to fungal siderophores (O'Sullivan and Gara,1992) and can sequester this limited resource from other microflora thereby preventing their growth (Kuc,1995). Earlier reports have demonstrated the importance of *P. fluorescens* siderophores in disease suppression (Costa and Loper, 1994; Leong and Expert, 1989) was illuminated in Figures 6 and 7.

However, many endophytic bacteria are facultative plant colonizers and have to compete well in the rhizosphere before entering the plant (Compant *et al.*, 2010) and might be therefore equipped with a rich arsenal of metabolites involved in defense as well as in interaction with the plant. Many bacteria with the capacity of colonizing plants utilize the nutrient niche of root surfaces in the rhizosphere and most of them might even actively switch from root surface to endophytic lifestyles (Rosenblueth and Martinez-Romero, 2006 ; Compant *et al.*, 2010).These bacteria comprise several well characterized species of *Bacillus* and *Pseudomonas* and a number of metabolites, particularly lipopeptides synthesized by non-ribosomal peptide synthesases, have been described to be important for rhizosphere bacteria for antibiosis and for inducing plant defense mechanisms (Figure 9).

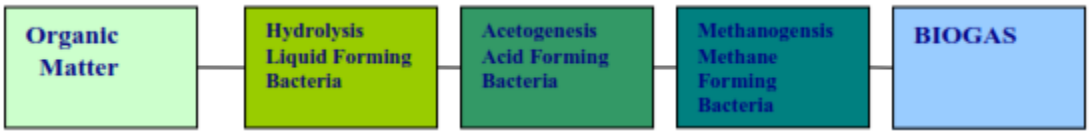

Figure 6 | Anaerobic digestion is a multi-stage process, watch video biofertilizer production | https://www.youtube.com/watch?v=pG2ODAx3ICY). https://www.academia.edu/video/jEe

Table 3. Biofertilizer characteristic as integrated nutrient management during crop cultivation reported by Otaiku *et al.*, 2019b.

N/S	Characterization	Definition	Mechanisms	Crops Application	References
1.	Biofertilizer	A substance which contains live microorganisms which, when applied on the seed, plant surface or the soil, colonizes the rhizosphere or the interior of the plant and promotes growth through increased supply or availability of primary nutrients for the host plant	a. Biological nitrogen fixation. b. Utilization of insoluble forms of phosphorus.	Tubers, Plantain, and Horticulture,	Fuentes-Ramírez and Caballero-Mellado, 2006
2.	Phytostimulator	Microorganism with the ability to produce or change the concentration of growth regulators such as indole acetic acid, gibberellic acid, cytokinins and ethylene.	a. Production of phytohormones (auxins, cytokinins and gibberelins). b. Decreased ethylene concentration (in the interior of the plant)		Lugtenberg *et al.*, 2002; Somers *et al.*, 2004.
3.	Biopesticide or Bio-control agent	Microorganisms that promote plant growth through the control of phytopathogenic agents, mainly for the production of antibiotics and antifungal metabolites.	a. Production of antibiotics (siderophores, HCN, antifungal metabolites) b. Production of enzymes that degrade the cellular wall of the fungi c. Competitive exclusion d. Acquired and Induced systemic resistance		Vessey, 2003; Somers *et al.*, 2004; Chandler *et al.*, 2008.

ECRTD-UK-https://www.eajournals.org/ | Journal level DOI: https://doi.org/10.37745/gjar.2013

Table 4. OTAI AG® Inoculant Characteristics of the PGPRs | Microbial Metabolites Processes related to Plant Nutrient in Biofertilizer.

N/S	Genera	Microbe Species	Contribution	Disease Biocontrol	Crops	Metabolites	References
A	Bacteria	Agrobacterium	increased the NO3 and K uptake	Fusarium solan	Potato	antimicrobial metabolites like siderophores,	Idris et al., 2007;
		Azotobacter sp	consequently, the shoot and root	Botrytis cinerea	Beans ,tomato	antibiotics, cyanides, fungal cell-wall-degrading enzymes	Lugtenberg and Kamilova,2009
		Bacillus sp	dry weights by 22 to 33 percent and	F. oxysporum		and gaseous products including ammonia .	Bertrand et al. 2004
		Pseudomonas sp.	6 to 21 percent, respectively	Alternaria spp	Roots, leaves	Phenazines, pyrrolnitrin, pyoluteorin	Srivastava and Shalini, 2008
		Rhizobium sp		Sclerotium spp	rstop on leaves, oot rot and stem rot	and cyclic lipopeptides like viscosinamide.	
		Streptomyces sp		Colletotrichum lindemuthianum	Beans	Pseudobactin and pyoverdin.	Hillel, 2005
		Enterobacter				Pyoverdine, pyochelin and its precursor salicylic acid	
						chitinase and laminase .	
B.	Bacteria	Pseudomonus putida	Denitrification, methanogenesis,	Hydrocarbon Pollutants	biological remediation	Oxygenease and peroxidases	Prescott et al. 2002
			sulfidogenesis diseases and as	[Benzene, anthracene,	synoptic interaction of fermentative and	pseudomicelle formation	Glazer and Nikaido 2007
			therapeutic agents	hydrocarbons, PCBs]	acetogenic bacteria, with methnogens or		Kapley et al. 1999
		Pseudomonas aerogenosa	Degrade hydrolysable tannins,	Agricultural/agro-industrial wastes	mineralization by amphipathic molecules	Plasmids ,glycolipids, phospholipids, lipoproteins	Bhatta et al., 2012 ; Nitiema et al., 2010,
		Pseudomonas fluorescens	diseases and as therapeutic agents	Antimicrobial activity	Decrease surface and interfacial tension	lipopeptides and polymeric compounds	Ray, 1994; Hamzah et al., 2010
				Biofouling degradation	Decrease surface and interfacial tension	Reduction of interfacial tension	Chaillan et al., 2004
				Antiviral activity		Rhamnolipid	Bhatia and Ichhpujani, 2005
		Bacillus sp	Bacillus cereus	Aromatics, long chain alkanes,	Sulphate reducers	Plasmids , glycolipids, phospholipids, lipoproteins	Cybulski et al. 2003 ; Hamzah et al., 2013
				phenol, cresol .			Cybulski et al. 2003
		Azotobacter sp		Aromatics			Chatterjee et al. 2008
		Mycobacterium sp		Aromatics, branched hydrocarbons			Jogdand ,1995
				benzene, cycloparaffins			Sunggyu, 1995 ; Stanier et al., 1986
		Streptomyces sp		phenoxyacetate, halogenated			Jogdand 1995 , Kuiperet al. 2004
				hydrocarbon , diazinon			Rockne and Reddy 2003, Schlegel, 1995
		Streptococci sp	Degrade the most recalcitrant	Bioaccumulation of heavy metals	Mineralization	Beta-oxidation pathway	Gadd, 1986;
		Aspergillus species		Bioaccumulation of heavy metals	Mineralization	Beta-oxidation pathway	Gadd, 1986

Biofertilizer characteristics (Table 3) and biosulfactants (Table 2) applied in the filed cassava cultivation requires no chemical pesticide. This was as a result of might be cassava plant-associated lifestyle requires adaptation to several niches, in which different metabolites act as signals for interaction (communication) with the plant and host specific plants nutrient and crop protection. Rhizobium and Bacillus were found to synthesize indole acetic acid (IAA) at different cultural conditions such as pH, temperature and in the presence of agro waste as substrate (Sudha *et al*., 2012).

Ethylene, unlike other phytohormones, is responsible for the inhibition of growth of dicot plants (Ansari *et al.,* 2013). It was found by Glick *et al*., 1998 that, PGPR could enhance the growth of plant by suppressing the expression of ethylene. Interestingly, a model was suggested in which it was shown that ethylene synthesis from 1-aminocyclopropane-1-carboxylate (ACC), an immediate precursor of ethylene, which is hydrolyzed by bacterial ACC- deaminase enzyme in the need of nitrogen and carbon source is also one of the mechanisms of induction of conditions suitable for growth, Figure 9. For plant-associated microorganisms introduced as bio-control agents into the rhizosphere or phyllosphere, the population of the microbial bio-control agent declines to background levels (Figure 8) when the supporting plant dies, and it must be applied again with the next planting of that crop with the graphic narrative in Plates 1 and 2 respectively.

Diseases and Pests of Cassava

Cassava anthracnose disease is caused by fungus which occurs on the surface of cassava stems and leaves (Alvarez *et al*., 2012) and appears as cankers (sores) on the stems and bases of leaf petioles, Tables 6 and 7. Cankers weaken the petioles so that the leaf droops downwards and wilts (Yaninek *et al*., 2000). The wilted leaves die and fall causing defoliation and shoot tip die-back or complete death of the shoot. Soft parts of cassava stems become twisted under severe attack by the disease. The disease usually starts at the beginning of the rains and worsens as the wet season progresses (Alvarez *et al*., 2012). Cassava bacterial blight, Leaf spot diseases, Cassava brown streak disease, Cassava root rot diseases, cassava mealybug. The cassava green mite Table 7. The treatment of the cassava disease and pests by disease suppression bio control broad spectrum microbial PGPR (Table 4) inoculants in the biofertilizer formulation with external validity of their efficacy by applied field studies, Table 9

Microorganisms affecting stress tolerance

Bacteria with the potential to act as biostimulants (Table 2) have been isolated from a number of ecosystems with saline, alkaline, acidic, and arid soils. These bacteria belong to several genera such as *Rhizobium, Bradyrhizobium, Azotobacter, Azospirillum, Pseudomonas*, and *Bacillus* (Tables 12 and 15 respectively). Members of these genera have developed strategies to adapt and thrive under adverse conditions (Selvakumar *et al*., 2009; Upadhyay *et al.*, 2009). Amongst these adaptations, alterations to the composition of the cell wall and the ability to accumulate high concentrations of soluble solutes are common. These allow for enhanced water retention and increased tolerance to osmotic and ionic stress. Cell wall composition is altered through enrichment for exopolysaccharides (EPS) and lipopolysaccharide–proteins and polysaccharide–lipids which form a protective biofilm on the root surface (Sandhya *et al.*, 2009; Zahran, 1999). Plant growth-promoting rhizobacteria (PGPR) inoculated (Figure 10) soils can ameliorate plant abiotic stress responses and narrated in details in Figure 9 below. ARATI Biopesticide * microbial consortium strains is the diverse range of modes of action including antibiotic production, cell wall-degrading enzymes, biosurfactants, and volatiles and also induces systematic resistance in plants.The bacterial isolates inhibit the growth of pathogen through different mechanisms like by secreting antibiotics, toxins, and surface-active compounds (biosurfactants), by competition of minerals, and by secreting cell wall-degrading enzyme like chitinase and ß-1, 3-glucanase (Kumar, 2015a).

*ARATI Biopesticide
https://www.academia.edu/41445902/ARATI_Biopesticide_Microbial_Granular_and_Liquid

Table 5. Effect of OBD - Biofertilizer applied alone and in combination with inorganic fertilizer on cassava root yield.

S/N	Treatments	Cassava Root Yield (t/ha)
1	300 kg/ha NPK 15:15:15 + 1.0 t/ha OBD-Biofertilizer	20.5
2	300 kg/ha NPK 15:15:15 + 2.0 t/ha OBD-Biofertilizer	22
3	300 kg/ha NPK 15:15:15 + 3.0 t/ha OBD-Biofertilizer	30
4	300 kg/ha NPK 15:15:15 + 4.0 t/ha OBD-Biofertilizer	31.2
5	5.0 t/ha OBD-Biofertilizer	16
6	600 kg/ha NPK (15:15:15)	35.6
7	Control (no application)	12
	F-LSD(0.05)	3

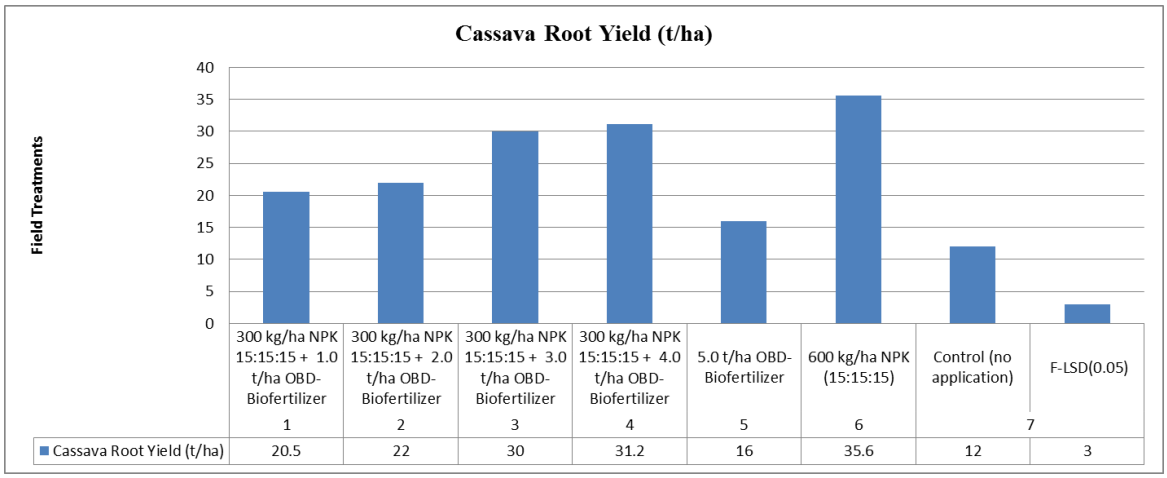

Figure 7 | Field application yield of application of biofertilizer and complementary use of NPK (15:15:15) reported Otaiku *et al.*, 2019a.

Growth, Yield and Root Quality | Biofertilizer

Biofertilizer facilitate the below-ground biological activity of earthworms, bacteria and fungi, and supply a wide range of nutrients, including secondary and micro-nutrients (Tables 16-17). Adoa (2009, Susan *et al*., 2005 reported highest plant height with the application of poultry manure on Nkabom and IFAD cassava varieties. Adjei-Nsiah and Issaka (2013) observed that average fresh tuber yield increase from 13.7 t/ha without amendment to 23.7 t/ha with application of 4 t/ha poultry manure and compared with Table 4 where biofertilizer application at 5t/ha yield 16 t/ha and control yield 12 t/ha (Figure 7) due to the impacts of the beneficial microorganisms, Table 10 and Figure 10 shows that biofertilizer promote the growth of stems and leaves of cassava, increase the chlorophyll content and the photosynthesis of leaves and improve the physiological metabolism of cassava (Luo *et al.*, 2008). The period of maximum rate of dry matter partitioning depends on genotype-by-environmental interaction (Fregence *et al*., 1994). Canopy spread in cassava ensures large surface solar interception for photosynthesis (Lebot, 2009). Nutrient supplied by poultry manure enhances increase in plant height due to increase in cell elongation of plant tissues as a result of steady release and mineralization of nutrients, see Tables 8 and 9 (Sharma and Govil, 1988; Christopher *et al*., 2007).

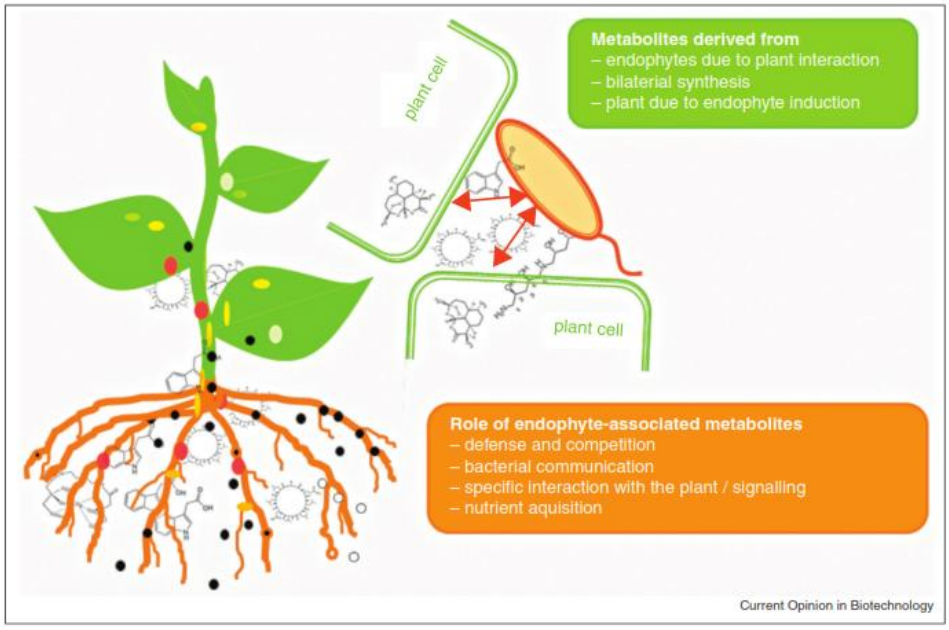

Figure 8 | Schematic overview showing the different types of plant-endophyte interactions leading to the synthesis of metabolites, which are in many cases not produced by the macro- or micro symbiont alone or in different quantities. Furthermore, the different known functions of endophyte-associated metabolites are presented, adapted from Sessitsch *et al.*, 2014.

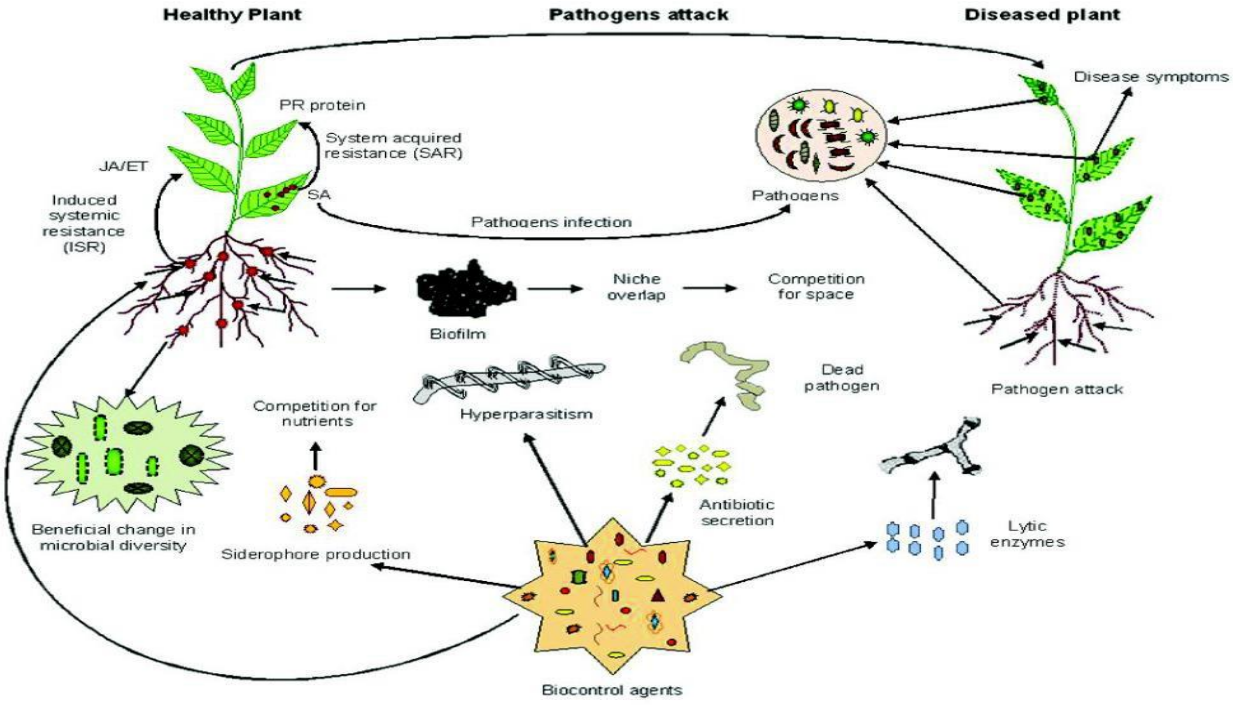

Figure 9 | Mechanism of actions implemented by bio-control agents for management of plant diseases

adapted from Weller, 1984; Cook *et al.,* 1991. Otaiku *et al.,* 2019b.

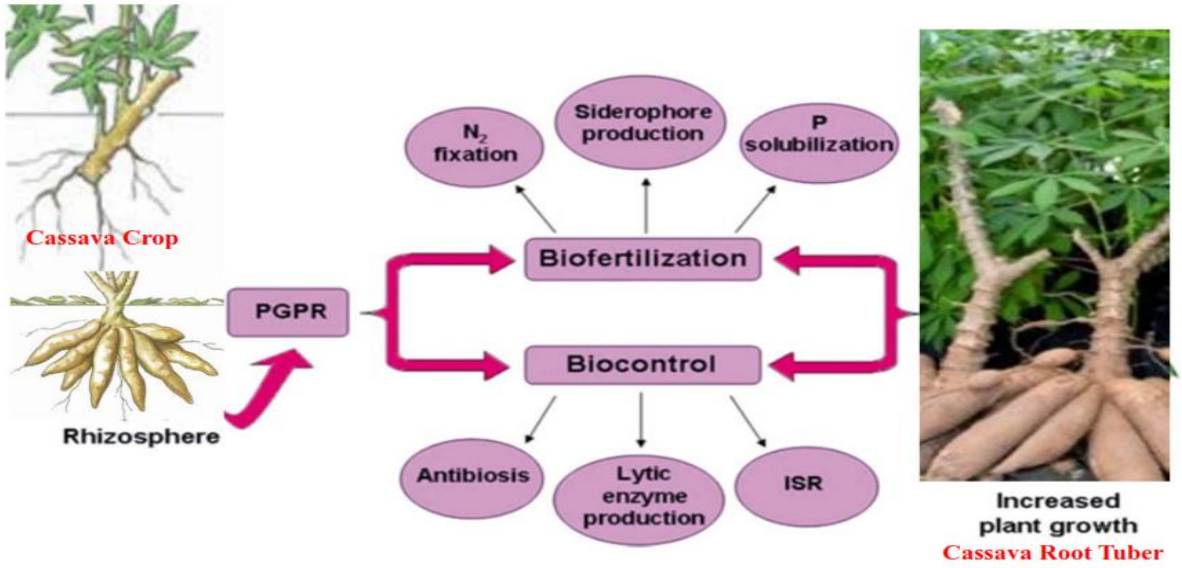

Figure 10 | Emphasized on the mechanisms of action of PGPRs on cassava crop impacts on integrated crop management adapted from Maheshwari, 2011, Otaiku *et al.,* 2019.

Amanullah *et al.,* 2006; Parkes *et al.*, 2012 observed that the number of roots per plant was significantly influenced by organic fertilizer treatment steady availability of nutrients throughout the crop growth period favorable changes in soil, such as loose and friable soil conditions, enabling better root formation (Figure 10) and mode of action Figure 9. An increase in the number of storage roots per plant in response to organic fertilizer application has been reported by Kasele (1980) and Pellet and El-Sharkawy (1997). Leo and Kabambe (2014), observed a significant increase in number of roots per plant, and tuber diameter having a positive correlation with fertilizer treatment. Manure application has resulted in higher root yields of cassava (Wilson and Dufour, 2002; Agbaje and Akinlosotu, 2004; Issaka *et al*., 2007; Ojeniyi *et al.*, 2012). Manure application enhances the cooking quality (mealiness) of cassava (Adoa, 2009). Various observations have been made of a positive correlation between dry matter content and cooking quality of cassava. (Safo-Kantanka and Asare, 1993; Safo-Kantanka and Owusu Nipa, 1992).

Biofertilizer and Inorganic fertilizer

The use of mineral fertilizer in combination with poultry manure has shown an increase yield as much as 60 t/ha of cassava roots (CSIR- AGRA, 2012). The fertilizers supplied the bulk of the macronutrients needed by the plants, while the organic sources provide secondary and micronutrients which are only needed in very small quantities and improve the soil's physical conditions (FAO, 2012; Ojeniyi *et al*., 2012) as confirmed in the Table 4 and Figure 7 where treatments with 600 kg/ha NPK 15:15:15 had a yield of 35.6 t/ha and 300 kg/ha NPK 15:15:15 + 3 t/ha OBD-Biofertilizer had a yield of 30 t/ha using Fischer's least significant difference (F-LSD) at 5% probability due to the impacts of microbial metabolic processes related to Plant nutrition in the

biofertilizer (Table 10).Nutrients contained in organic manures are released more slowly and are stored for a longer time in the soil, thereby ensuring a long residual effect (Tisdale *et al*., 1993). A combined use will increase synchrony and reduce losses by converting inorganic N into organic forms (Kramer et al., 2002).

The resultant impacts are integrated nutrient management programme with increase cassava yield through improving soil productivity, higher fertilizer use efficiency, reduces the environmental problems that may arise from the use of sole inorganic fertilizers and improves the microbial properties of the soil and sustain maximum crop productivity and profitability (Belay *et al.,* 2001; John *et al.,* 2004; Ayeni, 2008; Ayoola and Makinde, 2007; Santhi and Selvakumari, 2000). Endophytes are also of special interest for their high number of microbial niches and environments they may inhabit and provide therefore a high potential as a less exploited resource. In Table 13, AGPase, ADP-glucose pyrophosphorylase; bar, bialaphos resistance gene; GAP, beta-glucuronidase analysis positive; hpt, hygromycin phosphotransferase gene; ipt, isopentenyl transferase gene; luc, luciferase gene; NAP, Northern analysis positive; nptII, neomycin phosphotransferase II gene; pat, phosphinothricin acetyl transferase gene; pmi phosphomannose isomerase gene; RAP, reverse transcription–polymerase chain reaction analysis positive; SAP, Southern analysis positive; SE, somatic embryogenesis; SO, shoot organogenesis; TGE, transient gene expression; uidA, beta-D-glucuronidase gene; uidAint, uidA with intron; WAP, Western analysis positive.

SIGNIFICANCE OF BIOFERTILIZER ON CASSAVA CULTIVATION

Alves (2002) stated that cassava was a subsistence crop, grown by resource poor, small-holder farmers, who plant it preferably as an intercrop to reduce the risk of crop failure, while maximizing returns to land and labour. Cassava is also thought to require less labour than other crops and to be grown without inputs (Leihner, 2002). There is the need to take advantage of less labour cost and apply recommended fertilizers to increase crop yield, without necessarily increasing income to farmers and improves soil health and yield (Table 10). Cassava is frequently cultivated on marginal soils (Dixon *et al.,* 2002). Hillocks (2002), suggested that the observed increase in acreage is related to declining soil fertility levels in Africa (Figures 2 and 3). According to FAO (2006), average cassava yields in Africa have gradually increased from 6 to 10 t/ha over the past five decades. At present, the average African farmer harvests approximately 20% less cassava per hectare than the world average 12.2 t/ha due to no or low fertilizer inputs.

There is the need to apply supplementary nutrients for sustainable crop production (Asare *et al*., 2009) and see Tables 16, 17 and 18. Howler (1990) earlier stated that large bulk of foliage are created by the action of nitrogen and consequently an extensive assimilating area, a prerequisite for the good development of the roots (Figure 5, Tables 3 and 4). The trend in number of roots per plant is attributed to the observation that manure promotes (Table 9) the photosynthetic organs in the plant to produce and make available more assimilates to the root and increase the yield of cassava (Zhongyong *et al*., 2006; Kasele, 1980; Pellet and El-Sharkawy,1997). The cassava crop plants inoculated beneficial microorganisms significantly improve plant growth based (Figure 7) microorganisms in the biofertilizer inoculated (Table 17) to elaborate mechanisms of action (Figure 9 and Table 16). In Figure 5, bacteria (orange) and fungi (purple), can colonize the internal tissues of the plant (middle panel). Once inside the plant, the endophytic bacteria and fungi interact intimately with the plant cells and with surrounding microorganisms (large panel), Figure 12. Table 20. Transgenic cassava cultivars reported since 2010 for which genes expressing traits of interest for producers and/or consumers, other than marker and selectable genes, have been introduced. The Genetic Transformation Platform

at CIAT used *E. adhaerens* strain OV14 with plasmid pCAMBIA5105 to transform *cassava cv*. 60444, based on the protocol reported by Zúñiga-Soto *et al.* (2015) for rice. Three transgenic independent lines were confirmed by Southern blot as having one insert (two lines) or four copies of the T-DNA (Figure 15) and adapted from Chavarriaga-Aguirre *et al.*, 2016.

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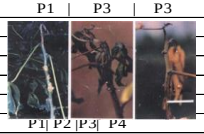
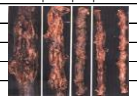
Table 6. Diseases and Pests of Cassava cultivation.

			Table 6. Diseases and Pests of Cassava				
	The largest number of Cassava diseases is found in Sub-Saharan Africa, South Asia, Latin America and the Caribbean						
N/S	**Crops Components**	**Disease Infection**	**Pictures**	**PGPR + Fungi Treatments Etcs**	**Biofertilizer**	**Impacts**	**References**
1	Leaves and stems	Bacterial blight		Bacillibactin	Paenibacillus	Destroyed after root harvest	Alvarez et al., 2012 ; Singh et al., 2009
				Diffusible metabolites as elicitors	*P. polymyxa*	yield losses of 20 to 100 %	IITA, 2004 ; Phi et al., 2010; Wen etal., 2011
				Viruses		Fungicides and insecticides	FAO, 2012 ; Kumar el al., 2016
	Integrated Nutrient Management						
				Giberellin (GA) production	OBD-Biofertilizer	Enhancing seed germination rate	Goswami et al., 2016
				Insecticidal toxins	Bacillus, Pusedomonas,	Insecticidal Protein Production	Pechy-Tarr et al. 2013 ; Roh et al., 2007
				Pseudomonas and Bacillus species	PGPRs	Biocontrol agent called lipopeptide (LP)	De Bruijn et al. ,2007; Raaijmakers et al., 2010
						bio-surfactantsFacilitate root colonization	Bais et al., 2004 ; Khabbaz et al.,2015
			P1 | P3 | P3				
2	Stems / bases of leaf petioles	Anthracnose disease		Volatile organic compounds (VOCs)	PGPRs	Leaf droops downwards and wilts	Farag et al., 2013 ; Von Der Weid et al., 2005
	A			Antimicrobials by Paenibacillus		Defoliation and shoot tip die-back	Yaninek et al., 2000
				P. polymyxa		Microbial inhibiting agents	Abriouel et al, 2011 ; Phi et al., 2010
				ISR against pathogenic bacteria		Indole acetic acid production	Lee et al., 2004 ; Lee et al., 2012
			P1| P2 |P3| P4				
	B						
				Bacilius subtilis	PGPRs	PGPRs rhizobacteriamediated ISR	Choudhary & Johri , 2008 ; Zhao et al., 2007
						Antifungal and antibacterial metabolites	Sessitsch et al., 2002a ; Sturz et al., 2000
						Phosphate solubilisation	Wakelin et al., 2004
						Supply of essential vitamins to plants	Pirttila et al.,2004
	Integrated Nutrient Management			*Trichoderma sp*	OBD-Biofertilizer	Microbes within the rhizosphere modify	Tucci et al., 2011
	Biofertilizer field application			*P.Fluorescens*	PGPRs	modify root exudate composition Enhance growth	11ta 2008 ; Hastuti et al., 2012
						induce systemic resistance to subsequent pathogen attack	FAO, 2013
						Phytohormones used in plant defense, ijasmonic acid, ethylene,	
3	Leaf	Leaf spot diseases		*Trichoderma Sp*		and salicylic acid, Infected leaves become yellow,	Kumar and Legg, 2009 ; Kalita et al., 2012
		White leaf spot, brown		*P.Fluorescens* (for Vector)		dry and die prematurely	FAO, 2013 ; Hastuti et al., 2012
		leaf spot, and leaf blight		*Bacillus Subtilis*		Spots sometimes have purplish borders	IITA, 2008 ; Sivakumar et al., 2007
							Yaninek et al., 2000
	Integrated Nutrient Management			Pseudomonas antibiotics likes:	OBD-Biofertilizer	Bacterial ureases can control/kill the insect host	Salvadori et al., 2012 ; Nakkeeran et al., 2006
				2,4-diacetylphloroglucinol (DAPG	Pseudomonas genus	Produce antibiotics kill the growth of target pathogen	Glick et al., 2007 ; Santoyo et al.,2012
				Pyoluteorin (Plt) ; pyrrolnitrin (Pm)	PGPRs	Antibiosis relies on the secretion of pathogenic molecules	Glick , 1995 ; Ahmad et al. , 2008
				phenazine-1-carboxylic acid (PCA)		Antibiotic has antifungal, antibacterial	Raaijmakers et al., 2002 ; Cronin et al., 1997
				Protein-type (bacteriocins)		and antihelmintic properties	Loper and Gross, 2007; Velusamy et al.,2006
				Hydrogen cyanide (HCN)			Thomashow and Weller ,1988
4	Leaf	Brown streak disease		Plant plasticity	PGPRs	Brown streak disease appears on the leaves	Miskito et al., 2000 ; Hillocks, 2002
		(BSD)		*Bacilius subtilis*		stems and storage roots of cassava plants.	Goh et al., 2013 Zhao et al., 2007
				Pseudomonas sp		Resistance to fungal pathogen infection	Chen et al,, 2012; Vasudvan et al., 2002
				Cassava Mosaic Disease-resistant		damages	Mbanzibwa et al., 2011 ; Maruthi et al. ,2005
				cultivars.		harvest quality	Navas-Castillo et al., 2011 ; Omongo et al., 2012
	Integrated Nutrient Management			Plant antioxidant system	PGPRs Endophyte	Degrade organic pollutants	Viñaset al.,2005, Taghavi et al.,2011
					Pseudomonas	Secondary metabolites for plant defense and communication	Kirby and Keasling, 2009

5	Root	Root rot diseases		Exopolysaccharides (EPS)	PGPRs	leaves on cassava plants affected by root rot disease turn brown,	IITA, 2008 ; FAO, 2013
				lipopolysaccharide–proteins		Defence for the root tip	Curlango-Rivera et al., 2013
				Polysaccharide–lipids		Protective biofilm on the root surface	Zahran , 1999 ; Morris and Djordjevic, 2006
				Plant root architecture		Wilt and the plant appears scorched.	Olugbenga et al., 2011
						Post-harvest control	Allard et al., 2014
	Integrated Nutrient Management			Siderophore Production	OBD-Biofertilizer	Catalyst in enzymatic processes, oxygen metabolism,	Aguado-Santacruz et al., 2012
				Plasticity in a given environment	PGPRs	electron transfer and DNA and RNA syntheses	Friesen et al., 2011, Spaepen, et al., 2008
				Rhizosphere microbiome		Function in root disease suppression	Martinez-Viveros et al. 2010; Neilands 1995
				Plant root architecture		Competitive fitness of plant growth-promoting	Barton and Abadia 2006 ;Weber, 2005
				Phytobiome /Microbiome		Sequester iron and forms complexes with other essential elements,	Bellenger et al., 2008 ; Braud et al., 2009a, b
							Albrecht-Gary and Crumbliss 1998
							Burdon and Thrall, 2009
6	Pests : Stems and leaves	Mealybug		*Apoanagyrus (Epidinocarsis) lopezi*		Reduces the lengths of the internodes and causes the leaves	LoÈ hr et al., 1990; Pieterse et al., 2014
		Phenacoccus manihoti		Effective parasitoids		to clump together into 'bunchy tops'	Mavrodi et al., 2011 ; Sang el al., 2014
						Reduce leaf and root yield, sometimes by as much as 80 %	Antonopoulos et al., 2008 ; Bellotti et al., 2012
						Yield loss of 60 percent of the roots and leaves 100 percent	Tjamos et al., 2005 ; Gkizi el al., 2016
						100 percent of the leaves.	Neuenschwander , 2003
	Integrated Nutrient Management			Hydrogen Cyanide Production	OBD-Biofertilizer	biocontrol of certain plant pathogens	Martinez-Viveros et al. , 2010 ; Gupta et al., 201
				hcnAB genes detecting	PGPRs	Pseudomonas strains producing antibiotic 2,4-DAPG	Haas and Defago 2005 ; Svercel et al. , 2007
				HCN-producing pseudomonas		HCN is also involved in metal sequestration	Wongfun et al. 2013 ; Rijavec and Lapanje 2016
				Bacterial volatile compounds (BVCs)		Trigger plant growth and immunity	Chung et al., 2016 , Mavrodi et al., 2011
				Pathogen suppression		Does not require any established physical contact	Ortfz-Castro et al. 2009
				Paenibacillus kill larvae of pest insects		to trigger growth response .(Paenibacillus).	Sharma et al., 2013 ; Neung et al., 2014
						BVCs such as 2,3-butanediol and acetoin accelerate	Audrain et al. , 2015 ; Ryu et al. , 2003
						plant growth and induce systemic resistance.	
						BVC which strikes the plant's physiology, growth and defence	D'Alessandro et al. ,2014
				Trait plant hormone signalling		Use natural enemies such as predators, parasitoids .	Jang et al., 2008
						and parasites e.g .ladybirds.	FAO, 2013
7A	Pests : Green mite (Leaves)	*Mononychellus tanajoa*				Severe mite attack can result in 13 to 80 % loss in cassava yield	Alvarez et al., 2012
						Lowland areas with a prolonged dry season.	Olugbenga et al., 2011
						Heavily attacked leaves become stunted and deformed.	
						Pest causes tiny yellow chlorotic leaf spots,	
						the size of pin pricks, on the upper leaf surfaces.	
	Integrated Nutrient Management			Trait plant hormone signalling	OBD- Biofertilizer	Plant systemic resistance	Mendes et al., 2011; Hu et al., 2003
					PGPRs		
B	Pests:White flies (Leaves)	*Aleurodicus dispersus*		Genes conferring resistance to CMD			Akano, 2001; Okogbenin et al., 2012
		Spiraling white flies		Cultivars resistance to Cassava BSD		Herbivory, and defence	Friesen et al., 2011 ; Zhang, 2005;
				Develop varieties resistant to both		Damage cassava by sucking sap from the leaves .	Paterson et al., 1995
				whitefly and viruses.		They secrete large amounts of honeydew that supports	Patil, 2011
				Genotypes that show resistance		the growth of black sooty mould on the plant,	Legg et al., 2006 ; Omongo, 2012
				to whiteflies.		causing premature fall of older leaves.	Bellotti, 2012a ; Vanderschuren, 2012
				MEcu 72 in Uganda in 2005.			Bellotti et al., 2012b ; Omongo, 2012a
				38 spider species and 123 insect			Horowitx et al., 2011
				species that are predators of *B. tabaci* .			
	Integrated Nutrient Management			ACC deaminase	OBD- Biofertilizer	Variety of stochastic disturbances	Baig et al., 2012
					PGPRs	Microbiome mediated physiology	van Kleunen, et al., 2010
8	Termites (Chew , eat stem cuttings)			Nematodes			Khan et al., 2012
	Cassava root scale	*Stictococcus vayssierrei*				Causing the cassava to grow poorly and die or rot.	
	Cassava white scale	*Aonidomytil usalbus*				Cause the tubers to be smaller and deformed,	
						and stems to lose a lot of water and die	
	Integrated Nutrient Management			"Induced systemic tolerance (1ST)"	OBD-Bioertilizer	Enhance survivability of stress-affected plants	Yang et al. ,2009
				Bacillus subtilis	PGPRs	modulation of hormonal status in host plant	
				Fluorescent pseudomonads		Increase in stress ethylene level plant In response to	Singh and Jha , 2016
						stress stimuliof (salinity, drought, metal toxicity, etc.	
						VOC produced by Bacillus subtilis confers salt tolerance	Zhang et al., 2008
						reduce the growth inhibitory effects of stressors	Penrose and Glick, 2003

Table 7. Cassava Biology and Pathology

N/S	Picture Description Cassava Disease and Pest	Infection Cause	References
1	Water soaked lesions on plant leaves	Humidity conditions favour growth and reproduction .	Fokunang et al., 2000a
2	Cassava anthracnose disease (CAD) shoot symptom	*Collerorrichum gloeosporioidss f.sp.manihotis.*	Jeffries et al., 1990 ; Muimba, 1982 ; Theberge, 1985
A.	P1 = Young cassava plant with shoot die-back symptoms	Oval pale-brown shallow depressions bearing a spot.	IITA, 1999 ; Hahn et al., 1989b
	P2 = Cassava seedling with shoot die-back and wilt symptoms	Insect vectors *Pseudotheraptus devastans, dist.*	Boher et al.,1983 ; Legg, 2009 ; Navas-Castillo et al., 2011
	P3 = Seedlings,arrow shows point of infected leaf defoliation	Fungus in the development and spread of the disease.	Chadrasekharan-Nair et al., 1 979 ; Lozano et al.,1981
B.	P1 = Large deep expanding cankers	Blocking transportation of vital materials.	Van der Bruggen and Maraite, 1987
	P2 = Crocodile-like deep anthracnose cankers	Growth potential of the fungus.	Fokunang et al., 1995 ; Fokunang et al., 2000b
	P3 = Superficial invading cankers	*Pseudotheraptus devastans dis.t*	Hahn and Keyser, 1985 ; Fokunang et al., 2000a
	P4 = Non expanding superficial cankers	Less severe on the approach of the dry season.	Fokunang et al., 2001 ; Muyolo, 1984
3	Plant with white leaf spot symptom	Fungal pathogen can differ inherited characteristics.	Tjamos et al.,1993
		Fungal pathogen can arise by mutation in somatic cells	Hahn et al., 1989b; Suresh et al.,1990
4	CAD infected stem cuttings	50-75% loss in seed viability ; 40-60% germination rate.	lkotun and Hahn,1992 ; Hahn et al.,1989a
		Leaves, the disease appears as patches of yellow areas .	Boher et al.,1983
	Cassava root rot diseases	Fungi living on the root or in the soil.	Miskito et al., 2000
5	a. Cassava mealybugs *Phenacoccus manihoti*	*Apoanagyrus (Epidinocarsis) lopezi,* parasitoid	Neuenschwander, 2001
6	b. Leaf distortion caused by cassava mealybugs	Biologically reasonable mechanism to use,	Lohr, 1990
	a. Cassava green mite	Pests.	Gerling et al., 2001
7A.	b.Speckled appearance on cassava leaves caused by cassava mites	Affects photosynthetic pathway .	El-Sharkawy et al., 1984c, 1985
	a. Adult whiteflies	Pests.	Asiimwe et al., 2007
7B.	b.Whiteflies feeding on the underside of cassava leaf	Pests.	Night, 2011
	c.Chlorosis and sooty mould on cassava leaves	Numerous small circular sunken spots on the Ieaf Iamina	Boher et al.,1983
	as a result of whiteflies feeding.	affected by genotype and environment.	Cock et al., 1979; Irikura et al., 1979

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Table 8. Comparative analysis of Treatments Materials for Cassava Cultivation.

	Treatment Materials	Control yield/Ton	Treatment/Yield /ton	Impacts on Crop Component	References
1	Poultry manure	13.7 t /ha	23.7 t/ha	Cell elongation of plant tissues as a result of steady release	Sharma and Govil, 1988;
				and mineralization of nutrients	Christopher et al., 2007; Adoa ,2009
				Highest nutrient content	Susan et al., 2005
	IFAD cassava varieties.				
	4 t/ha	13.7 t / ha	23.7 t/ha	Increase the chlorophyll content in stem/leaves and	Adjei-Nsiah and Issaka , 2013
				the photosynthesis and physiological metabolism	Luo et al., 2008
				Dry matter partitioning depends on genotype-by-environmental	Fregence et al., 1994 ; Lebot, 2009
				interaction	
				Higher root yields of cassava	Wilson and Dufour, 2002; Agbaje and Akinlosotu, 2004; ssaka et al., 2007;
					Ojeniyi et al., 2012
				Enhances the cooking quality (mealiness)	Adoa , 2009
				Positive correlation between dry matter content and	Safo-Kantanka and Asare 1993;
				cooking quality	Safo-Kantanka and Owusu Nipa, 1992
2	Biofertilzer				
	OBD-Biofertilizer			Contents Biocontrol microbes, PGRs and PGPR	Crawford, et al., 1993 , Bressan, W.2003
	5 t/ha	12t / ha	16 t/ha	Nitrogen fixing bacteria (NFB)	Madhaiyan et al., 2010
				Phosphate Solubilizing Bacteria	Xie H et al., (1996) ; Chanway and Holl (1993)Vyas and Gulati 2009
				Potassium Solubilizing Bacteria	Loon Van et al., (1998)
				PGRs and PGPR are expensive when compared to manure	Khan et al. 2017
				and chemical fertilizer	
				Improved yield under drought environment	Ergen and Budak 2009
				PGPR improve the availability of micro-nutrients to host plant and	Khosravi et al. 2018;Kumari et al. 2018
				improving growth pattern of roots	
				PGPR direct mechanisms involved synthesis of phytohormones	Glick 1995; Lugtenberg and Kamilova 2009)
				or increase in the uptake of certain nutrients from the environment	
				Creating antagonistic substances or by inducing re- sistance to patho	Sood et al. 2018 ;Beneduzi et al. 2012
				PGPR produces exopolysaccharides to protecti plant from desiccati	Pal et al. 1999 ; Salazar et al. 2009 ;Czarnes et al. 2000
				pollutant degradation and maintenance of primary cellular functions	Zhuang et al. 2007 ; Bahat- Samet et al. 2004
				PGPR like Azospirillum conserve water by producing cyst formation	Somers et al. 2004 ; Chenu1993; Vu et al. 2009
				around the roots by synthesis of poly-hydroxybutyrate and	
				production of melanin	
				mycorrhizal fungi act as a strong sink for photosynthate and	Kohler et al. 2007; Wu et al. 2005a, b ; Barea et al. 2002; Artursson et al. 2006
				improved soil aggregation	
				PGPR involved in the synthesis of phytohormones	Karlidag et al. 2007
				Improve root area, thus fas- cinate nutrients uptake, and	Compant et al. 2005; Kloeppe et al. 1999 ; Adesemoye et al. 2010
				tempt plant productivity	
				Guard plants from the lethal effects of environmental stresses	Glick et al. 1997
				Water conservation and endure diverse biotic and abiotic stress cond	Vessey 2003; Wang et al. 2012

Table 9. Combine Organic/Chemical Fertilizers Application to Cassava crop.

	Treatment Materials	Control yield/Ton	Treatment/Yield /ton	Impacts on Crop Component	References
1	Mineral fertilizer/Poultry manure		60 t / ha	Chemical fertilizers supplied the bulk of the macronutrients	CSIR- AGRA, 2012
				needed by the plants	FAO, 2012
				Organic sources provide secondary and micronutrients	
				Good soil fertility management strategy.	Ojeniyi et al., 2012
				Higher fertilizer use efficiency.	Santhi and Selvakumari , 2000
2	Biofertilizer				
	OBD-Biofertilizer			Integrated nutrient management programme	Ayoola and Makinde (2007)
	Urea/NPK 15:15:15/OBD-Biofertilizer Formulatiom			Slow release fertilizer ensuring a long residual effect	Tisdale et al., 1993
	300 kg/ha NPK 15:15:15 + 1.0 t/ha OBD-Biofertilizer	12 t/ha	20.5 t/ha	Reduce losses by converting inorganic N into organic forms	Kramer et al., 2002
	300 kg/ha NPK 15:15:15 + 2.0 t/ha OBD-Biofertilizer		22 t/ha	Improves the microbial properties of the soil	Belay et al., 2001
	300 kg/ha NPK 15:15:15 + 3.0 t/ha OBD- Biofertilizer		30 t/ha	Sustain maximum crop productivity and profitability	Ayeni, 2008
	300 kg/ha NPK 15:15:15 + 4.0 t/ha OBD- Biofertilizer		31.2 t/ha	Minimizing environmental impact from nutrient use	
	600 kg/ha NPK (15:15:15)		35.6 t /ha		

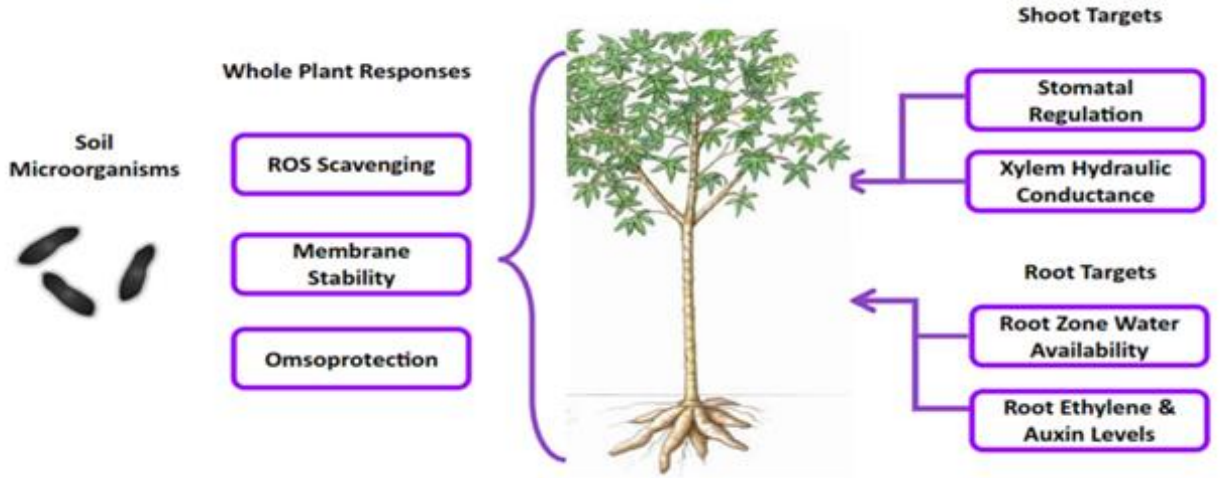

Figure 11 | Summary of main key mechanisms targeted by microorganism-based biostimulants in Table 17 and reactive oxygen species detoxification (ROS detox) enzymes might also ameliorate the plant-induced stress (Van Oosten *et al.*, 2017).

Table 10. Soil Indicators for Soil Heath and Soil Quality.

References	**Index used/proposed**
Andrews et al. ,2002	Indices based on parameters related to entrance of water and plant growth
Bastida et al, 2006	Microbiological index of soil degradation – dehydrogenase, water
	soluble carbohydrates, urease, water soluble carbon and respiration
Beck ,1984	EAN – more enzyme activities (dehydrogenase, phosphatase, protease and amylase)
Dilly and Blume, 1990	As many as ten parameters
Doran and Parkin,1994	Index based on sustainable production, environmental quality and
	human and animal health
Doran and Parkin, 1994	Soil quality index = function of (food and fibre production, erosivity,
	groundwater quality, surface water quality, air quality and food quality)
Kandeler and Eder ,1993	Simple indices – quotients between enzymatic activity and microbial biomass
Kang et al , 2005	Microbial index of soil (CHECK) based on microbial biomass C and N,
	potentially mineralisable N, soil respiration, bacterial population, mycorrhizal infection, and dehydrogenase and phosphatase activities
Karlen et al.,1994	Soil quality index based on four sour functions : ability of soil to
	accommodate water entry, retain and supply water to plants, resist degradation and support plant growth
Klein and Paschke,2000	Total/active funagal and bacteria ratio – the ratio of total total to active
	fungal plus bacterial biovolumes is divided by the ratio of the active fungal to bacterial biovolume
Parr et al.,1992	Soil quality index based on different functions: soil properties, potential
	productivity, environmental factors, human and animal health, erodibility, biological diversity, food quality and safety and management inputs
Parr et al. ,1992	Soil quality index = function of (soil properties, potential productivity,
	environmental factors, human/animal health, erodibility, biological diversity, food quality/safety and management input
Puglisi et al.,2005	Soil alteration index
Stefanic et al., 1984	Biological index of soil fertility based on activity of two enzymes –
	dehydrogenase and catalase
Trasar-cepeda et al.,1998;	Indices/equations based on prameters that reflect the total content of N or organic C
Harris et al. ,1996	Soil quality index based on three soil functions: ability to resist soil
	erosion, provide plant nutrients and provide a favourable root environment
Velasquez et al.,2007	General indicator of soil quality based on abundance of 17 groups of macrofauna, eight soil chemical properties
	(extractable P, total P, exchangeable K, Mg, Ca, Na and pH, six physical properties
	(bulk density, real density, porosity, moisture content, shear strength, penetration resistance, soil morphological features and organic C fractions

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Table 11. Categorization of general goals for agro-ecosystems.

Goal type	General goal	Key controlling variables
Economic viability	High productivity	Genetic potential, weather, soil, management, economics
	Low cost of production	Yield potential*, input requirements*, input costs
		Market variation, production variation*
Stewardship	Preservation of productive land	Soil, climate, management
	Healthy animals	Feed quantity and quality*, disease
	High quality food and fiber	Chemical or microbial contamination*, composition*
Social	Viable local communities	Population size, economic viability, economic, diversification
		Profitability, size and resilience of industry
Environment	Clean water	Climate, soil, management
	Clean air	Climate, soil, management
	Wildlife habitat	Climate, soil, management

N.B | * Variables also influenced by soil properties.

Table 12. Key microbial metabolic processes related to plant nutrition.

Element	Biochemical process	Microbial genes	Soil enzymology literature	Culture-independent References	Culture-dependent References
Nitrogen	Nitrogen fixation	nifD, nifH, nifK		Reganold et al., 2010; Xue et al., 2013	Bremer et al., 1990
	Protein depolymerization	apr, npr, sub	Mader et al., 2002	Rasche et al., 2014	Kohler et al., 2007
	Urea catabolism	ureA, ureB, ureC	Dick et al., 1988; Bowles et al., 2014	Reganold et al., 2010; Fierer et al., 2012, Xue et al., 2013	Kohler et al., 2007
Phosphorous	Phosphate ester cleavage	phoA, phoD, phoX,	Mader et al., 2002; Garcia-Ruiz et al., 2008	Fraser et al., 2015	Kohler et al., 2007
		ACPase, glpQ, ushA, appA, phyA, phyB			
	Phosphonate breakdown	phnJ, phnX		Bergkemper et al., 2016	Schmalenberger et al.,2008
Sulphur	Sulfate ester cleavage	aslA, asfA	Garcia-Ruiz et al., 2008	Schmalenberger et al., 2008	
	Sulfonate breakdown	ssuD			Kertesz and Mirleau,2004

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Table 13. Progress and current status of cassava genetic transformation (Otaiku *et al*., 2020).

N/S	Explant	Regeneration mode	Gene-transfer technique	Plasmid marker genes	Selection	Target traits	Integration/ expression	References
1	Somatic cotyledon	SE	Agrobacterium	pGV1040(nptII,bar,uidA)	ppt	Herbicide resistance	SAP,GAP	Sarriaetal.2000
2	Suspension	SE	Agrobacterium	pHMG(uidAint,hpt,pmi)	Hygromycin, mannose	Herbicide resistance	SAP,NAP, GAP	Zhangetal.2000b
3	Suspension	SE	Agrobacterium	pCP15GUS;pCP54GUS	Hygromycin,	Root-specific	SAP,NAP, GAP	Zhangetal.2003b
				(uid Aint,hpt)		promoters		
4	Suspension	SE	Agrobacterium	pCASP1(uidAint,hpt)	Hygromycin	Improved protein	SAP,NAP	Zhangetal.2003a
						content	GAP,WAP	
5	Suspension	SE	Agrobacterium	patatin-CYP79D1/D2(nptII)	Paromomycin	Reduced cyanogen	SAP,RAP	SiritungaandSayre2004
						content		
6	Suspension	SE	Agrobacterium	pILTAB9001(nptII)	Paromomycin	CMD resistance	SAP, NAP	Chellappanetal. 2004
7	Somatic cotyledon	SE	Agrobacterium	3D(nptII)	Paromomycin	improvedstarch	SAP,RAP	Ihemereetal.2006
						content		
8	Somaticcotyledon	SE	Agrobacterium	pMAT21;pEXM2;pIPT5(ipt)	Kanamycin	marker-free	SAP,RAP,GAP,GAP	Saelimetal.2009
9	Suspension	S0	Agrobacterium	pCP2	Hygromycin	tissue-specific	SAP,GAP	Beltr´ anetal.2010
						promoter		
10	Suspension	SE	Agrobacterium	pSG529(nptII)	Paromomycin	prolonged leaf life	SAP,RAP	Zhangetal.2010
11	Suspension	SE	Agrobacterium	pILTAB600;pILTAB601(nptII)	Paromomycin	mproved protein	SAP,WAP	Abharyetal. 2011
						content		
12	Suspension	SE	Agrobacterium	p35S::GBSSI-RNAi;p54/1.0	Ygromycin	waxy cassava	SAP,RAP,	Zhaoetal.2011
13	Suspension	SE	Agrobacterium	RNAiFL-CP(nptII)	Paromomycin	CBSVDresistance	SAP,NAP	Yadavetal.2011

Endophytic fungi, represented here as *arbuscular mycorrhizal fungi* (AMF) (lilac), might form specialized structures, called arbuscules, where plant-derived carbon sources, mainly sucrose (Su), are exchanged for fungus-provided phosphate (Pi), nitrogen (NH_4^+), and potassium (K^+) elements (blue). Plant cytoplasmic sucrose is transported to the peri-arbuscular space, where it is converted to hexose (HEX) to be assimilated by the fungus. Hexose is finally converted to glycogen (G) for long-distance transport (Hardoim *et al.*, 2015). Phosphate and nitrogen are transported inside the fungal cytoplasm as polyphosphate granules (Poly-P), which are converted to Pi and arginine (Arg) in the arbuscule. Pi is transported to the host cytoplasm,

whereas Arg is initially converted to urea (Ur) and then to ammonium (NH^+). Fungal and bacterial plant hormones, such as auxins (IAA), gibberellins (GAs), cytokinins (CKs), volatile organic compounds (VOCs), and polyamines (Poly-NH_2), as well as secondary metabolites (SMs), are transferred to the host (violet) Figure 12.

Table 14. Various organic or inorganic substances produced by plant growth promoting rhizobacteria facilitating resource acquisition to stimulate plant growth.

PGPR in the Biofertilizer to Cassava	**PGP Traits**	**References**
Rahnella aquatilis	ACC deaminase*	Mehnaz, Baig and Lazarovits , 2010
Acinetobacter sp., Pseudomonas sp.; Enterobacter sp.	ACC deaminase*	Indiragandhi et al., 2008) Kumar et al., 2008
Pseudomonas jessenii	ACC deaminase	Rajkumar and Freitas , 2008
Pseudomonas aeruginosa	ACC deaminase*	Ganesan, 2008
Achromobacter xylosoxidans A551,	ACC deaminase*	Belimov et al., 2005
Rhizobium hedysari ATCC 43676	ACC deaminase*	Ma et al., 2003
Pseudomonas marginalis DP3	ACC deaminase*	Belimov et al., 2005
Mesorhizobium loti	ACC deaminase*	Sullivan et al., 2002
Rhizobium leguminosarum	Indole-3-acetic acid	Ahemad and Kibret , 2014
Azotobacter sp. ; Pseudomonas sp.	Indole-3-acetic acid	Ahmad et al., 2006 ; Roesti et al., 2006
Bacillus sp, Paenibacillus sp.	Indole-3-acetic acid	Beneduzi et al., 2008
Rhizobium leguminosarum b. Trifolii ACCC18002	Indole-3-acetic acid	Jin et al., 2006
Streptomyces strains C	Indole-3-acetic acid	Sadeghi et al., 2012
Enterobacter aerogenes NII-0907, Enterobacter aerogenes NII-0929,	Indole-3-acetic acid	Deepa, et al., 2010
Pseudomonas tolaasii ACC23, Pseudomonas fluorescens ACC9,	Indole-3-acetic acid	Dell'Amico et al., 2008
Mesorhizobium loti MP6 ; Enterobacter sp., Klebsiella	Indole-3-acetic acid	Chandra et al., 2007 ; De Santi Ferrara et al., 2013
Pseudomonas aeruginosa, Pseudomonas fluorescens, Ralstonia metallidurans	Siderophores	Braud et al., 2009
Proteus vulgaris ; Enterobacter sp.	Siderophores	Rani et al., 2009 ; Kumar et al., 2008
Azotobacter sp., Mesorhizobium sp.	Siderophores	Ahmad et al., 2008
Mesorhizobium ciceri, Azotobacter chroococcum	Siderophores	Wani et al., 2007
Pseudomonas, Bacillus ; Pseudomonas jessenii	Siderophores	Wani et al., 2007 ; Rajkumar and Freitas ,2008
Bacillus sp. PSB10 ; Paenibacillus polymyxa	Siderophores	Wani et al., 2007 ; Ahemad and Kibret , 2014
Pseudomonas aeruginosa4EA ; Enterobacter asburiae	Siderophores	Naik and Dubey , 2011 ; Ahemad and Khan , 2010

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Table 15. Different microbial biofertilizers available in market and their application.

	Microbial biofertilizers	**Trade names**	**Application**
1	Azospirillum lipoferum,	Biospirillum, Green Plus,	1) For normal and acidic soils
	Azospirillum brasilense, and different strains of Azospirillum	Bio-N, Azo-S, ROM, and	and dry soils. (2) For paddy and other crops
2	Rhizobium, Azospirillum, Azotobacter, Bacillus sp.,	ARATI BAJA | liquid formulations *1	All crops
	Acetobacter, Fungi sp.		
3	Enterobacter, Flavobacterium, Klebsiella, Mesorhizobium,	ARATI NAWOZ | liquid formulations	All crops
	Micrococcus, Fungi sp.		
4	Agrobacterium, Azospirillum, Azotobacter, Arthrobacter,	OBD-Biofertilizer	All crops
	Bacillus, Enterobacter, Fugi sp.		
5	Pseudomonas, Rhizobium, Rhodococcus, Bacillus, Fungi sp.	Gateway Biofertilizer *2	All crops
6	Azotobacter chroococcum,different strains of Azoto-	Bioazoto, Bhoomi Rakshak, Kisaan Azotobacter culture	For all crops like wheat,
	bacter (non-symbiotic)	,and Azonik	sorghum, barley, maize, paddy, mustard, sunflower,
			sesamum, cotton, sugarcane, banana, grapes, papaya, water-
			melon, onion, potato, tomato, cauliflower, chilly, lady finger,
			rapeseed, linseed, tobacco,mulberry, coconut, spices,
			fruits, flowers, plantation, crops, and forest plants
7	Gluconacetobacter : diazotropicus	Sugar-Plus	For sugarcane
8	Rhizobium strains (symbiotic, nitrogen fixing)	Biobium, Rhizo-Enrich,Kisaan Rhizobium culture,	Pulses (gram, peas, lentil, moong, urd, cowpea, and
		Rhizoteeka, Green Earth Reap	arhar), oil legumes (groundnut and soyabeans), fodder
		N4, and Rhizonik	legumes (barseem and lucerne), and forest tree
			legumes (subabul, shisam, and shinsh
9	Phosphorus-solubilizing and Phosphorus-mobilizing	Biophos, Get-Phos, MYCO-RISE, Kisaan P.S.B. culture,	For all crops
	microbes like Bacillus , megaterium, mycorrizhal fungi, etc.	MycoRhiz, Reap P, and Phosphonive	
10	Potassium-mobilizing or	BIO-NPK, Bharpur,	For all crops
	potash bacteria like Bacillus	BioPotash, Potash-Cure, and	
	mucilagenosus	Green Earth Reap K	
11	Sulfur-solubilizing microbes like Thiobacillus thioxidans	Biosulf, Sulf-cure, Sulphonik, S Sol B®, Siron,	For cereals, millets, pulses, oilseeds, fiber crops, sugar
		and MicroS-109	crops, forage crops, plantation crops, vegetables, fruits
			spices, flowers, medicinal crops, aromatic crops,
			orchards, and ornamentals
12	Zinc-solubilizing microbes	Biozinc, Zinc-Cure, Zinc activator, Zinc extra	For crops like paddy, wheat,
		and MicroZ-109	pulses, citrus, pomegranate, ginger, etc.
13	Silica-solubilizing microbes	BioSilica, Silica-Cure, and	For crops like cereals, sugar cane, onions, leafy greens,
		Silica-109	legumes, cucumber, pumpkin, and gourd.

N.B | Visit website for material safety data sheet (MSDS)
* https://www.academia.edu/43310069/ARATI_BAJA_-Liquid_Biofertilizer_Integrated_soil_fertility_management_ISFM_
*2 https://www.academia.edu/42632817/Gateway_Organic_Fertilizer_Biofertilizer_Gateway_Biofertilizer_Organic_3.0

Table 16. Cassava crop inoculants: Biofertilizer and Biostimulator Phosphate Solubilizing Bacteria.

Phosphate solubilizing bacteria	**Plant growth promoting traits**	**Cassava Inocula**	**References**
Pseudomonas sp.	ACC deaminase, IAA, siderophore	OBD-Biofertilizer	Poonguzhali et al., 2008
Bacillus subtilis	IAA, siderophore, antifungal activity	OTAI AG®	Singh et al., 2008
Pseudomonas fluorescens	ACC deaminase	OBD-Biofertilizer	Shaharoona et al., 2008
Acinetobacter sp.,	ACC deaminase, IAA, antifungal activity,	OTAI AG®	Indiragandhi et al., 2008
Pseudomonas sp.	N2- fixation	OBD-Biofertilizer	
Enterobacter sp.	ACC deaminase, IAA, siderophore solubilization	OTAI AG®	Kumar et al., 2008
Pseudomonas jessenii	ACC deaminase, IAA, siderophore, heavy metal solubilization	OTAI AG®	Rajkumar et al., 2008
Pseudomonas aeruginosa	ACC deaminase, IAA, siderophore	OBD-Biofertilizer	Ganesan, et al., 2008
Pseudomonas sp.	ACC deaminase, IAA, siderophore, heavy metal solubilization	OBD-Biofertilizer	Rajkumar et al., 2008
Azotobacter sp., Mesorhizobium sp.,	IAA, siderophore, antifungal activity, ammonia	OBD-Biofertilizer	Ahmad et al., 2008
Pseudomonas sp.,	production, HCN	OTAI X ®	
Bacillus spp.	IAA, siderophores, ammonia production, HCN,	OBD-Biofertilizer	Wani et al., 2007a ; Wani et al., 2007c ;
chromium reduction, metal solubilization			Ahmad et al., 2008
Bacillus subtilis	IAA	OTAI AG®	Zaidi et al., 2006
Pseudomonas sp., Bacillus sp.	IAA, siderophore	OTAI AG® , OTAI X®	Rajkumar et al., 2006
Pseudomonas putida	antifungal activity, siderophore, HCN	OBD-Biofertilizer	Pandey et al., 2006

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Plate 1. Control field Casaava experiment.

Plate 2. Treated field cassava experiment using Biofertilizer.

Table 17. Soil microbiome engineering inoculants in biofertilizers applied in cassava cultivation.

	Biofertilizer Types Group	**Micro-organisms**	**Cassava inocula**	**Applications**	**References**
	Biofertilizer for micronutrients				
1	N2-fixing biofertilizers	Azotobacter, Nostoc	OBD-Biofertilizer	Biofertilizer	Amutha et al. 2014 ; Otaiku et al., 2019a
a.	Free-living	Rhizobium	OTAI X ®	Biofertilizer	Meena et al. 2015f, 2016b; Otaiku et al., 2019a
b.	Symbiotic	Azospirillum, Gluconacetobacter diazotrophicus	OBD-Biofertilizer	Bioremediation	Jaiswal et al. 2016 ; Murumkar et al.2016
c.	Associative symbiotic	Associative symbiotic	OBD-Biofertilizer	Xeonbiotic Biodegrader	Otaiku and Alhaji, 2019 ; Otaiku and Alhaji c
2	P-solubilizing biofertilizers				
a.	Bacteria	Bacillus megaterium var. Bacillus subtilis	OTAI AG®	Biofertilizer, Biopesticide	Kumar et al. 2017a; Meena et al. 2015a;
		Bacillus circulans, Pseudomonas striata	OBD-Biofertilizer	Xeonbiotic Biodegrader	Bahadur et al. 2016b; Das and Pradhan 2016).
				Biocontrol	Otaiku et al., 2019a ;Otaiku et al., 2019b
b.	Fungi				
	P-mobilizing biofertilizers	Penicillium sp., Aspergillus awamori	OTAI AG®	Biofertilizer, Biopesticide	Meena et al. 2015b, e; Teotia et al. 2016;
			OTAI X ®, ,OTAI AG ®	Xeonbiotic Biodegrader	Otaiku and Alhaji, 2019 ; Otaiku and Alhaji a, b ,c
	Biofertilizer for macronutrients				
a.	Arbuscular mycorrhiza	Glomus sp., Gigaspora sp., Acaulospora sp.,	OTAI AG®	Biofertilizer	Verma et al. 2017b; Kumar et al. 2017b
		Scutellospora sp. and Sclerocystis sp.	OBD-Biofertilizer	Mycorrhizosphere	Raghavendra et al. 2016; Zahedi 2016
	Sulphur oxidizers	Thiobocblus thioxidans	OTAI AG®	Mycoremediation	Sharma et al. 2012 ; Vaid et al. 2014
a.	Zinc solubilizers	Bacillus sp., Pseudomonas sp., Aspergillus niger	OBD-Biofertilizer	Biofertilizer	Sharma et al. 2012 ; Vaid et al. 2014
b.	Potassium and silicate solubilizers	Pseudomonas sp., Bacillus sp.,	OTAI X®	Xeonbiotic Biodegrader	Nath et al. 2017; Sarkar et al. 2017; Otaiku and Alhaji a ,c
			OTAI AG®	Bioremediation	Rawat et al. 2016; Yasin et al. 2016
c.	Manganese solubilizers	Penicillium citrinum	OBD-Biofertilizer	Bioremediation	Lovley, 2000 ;Ehrlich & Newman, 2009
	Plant growth-promoting rhizobacteria				
a.	PGPR				
	Bacteria	Pseudomonas fluorescens, Bacillus sp.,	OTAI AG®	Biofertilizer , Bioremediation	Yadav and Sidhu 2016 ; Saha et al. 2016b
		Azotobacter, Klebsiella, Enterobacter, Azospirillum,	OBD-Biofertilizer	Xeonbiotic Biodegrader	Meena et al. 2016d; Otaiku and Alhaji., 2020
b.	Fungi				
	Biofilmed biofertilizers	Fungal-bacterial biofilms (FBB),	OBD-Biofertilizer	Biofertilizer , Bioremediation	Hettiarachchi et al. ,2014 ; Amundson et al., 2007
		fungal-rhizobial biofilms (FRB); Bacillus cereus	OTAI AG®	Bio-control	Verma et al. 2014, 2015b; Meena et al. 2013c, 2014a;
					Trivedi et al. ,2011 ;Otaiku et al., 2019a

Table 18. Activities of soil borne bacterial functions regulated by Quorum Sensing (QS) signals, adapted from Raghavendra (2017).

	Taxonomic class	Genus or species	Cassava Inocula	QS signals produced	Known regulated functions	References
1	Actinobacteria	Streptomyces sp.	OBD-Biofertilizer	Gamma-butyrolactones	Antibiotic compound synthesis,	Chater ,1993 and Shaaban et al. , 2016
					differentiation	
2	Alpha- proteobacteri	Agrobacterium tumefaciens	OTAI AG®	OOHL	Ti plasmid transfer, virulence	White and Winans , 2007 ; Otaiku et al., 2019a
3	Rhodopseudomonas		OTAI AG®	pCHL	Chemotaxis	Schaefer et al. 2008
	palustris					
4	Mesorhizobium loti		OBD-Biofertilizer	OHHL, OHL, DHL, dDHL	Nodulation	Yang et al., 2009; Otaiku et al., 2019a
5	Gammaproteobacteria	Pseudomonas aeruginosa	OTAI AG®	BHL, OdDHL	Biofilm, elastase, lipase, alkaline protease,	Braeken et al. ,2008
			OBD-Biofertilizer		HCN, pyocyanin, exotoxin A, swarming,	Ferluga et al. ,2008
					lectins, rhamnolipids, virulence.	Otaiku et al., 2019a
				PQS	Elastase, pyocyanin synthesis, LecA lectin,	Dubern and Diggle , 2008
					biofilm, AHL signaling, motility + intrinsic	
					functions (antibiosis, iron chelation)	
				DKPs (e.g., cyclo (D-Ala-LV al))	Unclear, cross-linked to AHL signaling	Holden et al. , 1999
6		Pseudomonas fluorescens	OTAI AG®	OHHL, OHL	Biofilm formation, wheat rhizosphere	Wei and Zhang , 2006
					colonization, biocontrol ability.	Otaiku et al., 2019a

N.B |
BHL = N-butyryl-homoserine lactone | dDHL = N-dodecanoyl-homoserine lactone
DHL = N-decanoyl- homoserine lactone | OdDHL = 3-oxo-N-dodecanoyl-homoserine lactone
DKP = diketo-peptides | OHL = N-octanoyl-homoserine lactone | OOHL = 3-oxo-N- octanoyl-homoserine lactone.

Table 19. Lists of some beneficial plant growth promoting traits in the OBD-Biofertilizer.

N/S	Trait	Role	Microbe	References
1	Phosphate	1. Organic acid production	*Bacillus licheniformis; B. amyloliquefaciens; Penicillium sp.*	Chen et al., 2006 and Wakelin et al.,2004
	solubilization.			
		2.Phytase production	*Bacillus mucilaginosus; Aspergillus niger*	Vassilev et al., 2007 ; Ryu et al., 2005
		3. Phosphatase production	*Serratia marcescens*	Ryu et al., 2005 and Unno et al., 2005
2	Nitrogen fixation.	1. Symbiotic	*Vesicular-arbuscular mycorrhizal fungi*	Shah et al., 2010
		2. Non-symbiotic	*Gluconacetobacter diazotrophicus*	Bhattacharyya and Jha., 2012
3	Phytohormone	1. IAA production	*Bacillus licheniformis; Penicillium sp.*	Goswami et al., 2016 and Waqas et al., 2012
	production.	2. Cytokinin production	*Bacillus megaterium*	Castro et al. , 2008
		3. Gibberellin production	*Acetobacter diazotrophicus, Penicillium sp.*	Basti et al.,1998 and Waqas et al..2012
4	Biocontrol.	1. Extracellular enzyme production		
		(a) Chitinase	Enterobacter agglomerans	Nielsen and Sorensen ,1999
		(b) Glucanase	*Bacillus cepacia*	Compant et al., 2005
		2. Antibiotic production	*Pseudomonas fluorescens ; Trichoderma koningii*	Thomashow and Weller ,1988 ; Xiao -Yan et al., 2006
		3. Siderophore production	*Pseudomonas aeruginosa*	Braud et al., 2009 a , b
		4. HCN production Production	*Pseudomonas chlororaphis*	Nandi et al., 2015
5	Potassium	Production and excretion of	*Bacillus mucilaginosus*	Ullman et al., 1996
	solubilization	organic acid and inorganic acid		
	Induced systemic	1. ACC deaminase production	*Trichoderma , Asperellum; Penicillium citrinum*	Mayak et al., 2004, Viterbo et al. , 2010 ; Jia et al.,2000
	tolerance.	2. Exopolysaccharide production	Oceanobac illus	
		3. VOC production	*Bacillus amyloliquefaciens*	

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Table 19. Continuation

Cassava Genotypes	Traits of interest (genes)	References
60444	Biofortified β-carotene (crtB)	Welsch et al. (2010)
60444 Biofortified	β-carotene(crtB, crtI, crtY)	Bonilla (2010)
60444	Leaf retention (senescence-inducible ipt)	Zhang et al. (2010)
60444	Waxy starch (RNAi GBSSI)	Zhao et al. (2011)
60444	CBSVD (RNAi FL-CP)	Yadav et al. (2011)
60444	Protein content/cyanogenic content (HNL)	Narayanan et al. (2011)
60444	RNAi CMD (ACMV/EACMV); CBSD (n.d.)	Taylor et al. (2012)
60444z	Iron biofortification (FEA1)	Ihemere et al. (2012)
TME7 (Oko-Iyawo)	CMV and CBSV resistance (RNAi-CBSV coat protein)	Vanderschuren et al. (2012)
Adira4	Waxy starch (RNAi-GBSSI)	Koehorst-van Putten et al. (2012)
60444	UCBSV resistance (siRNA-UCBSV coat protein)	Ogwok et al. (2012)
60444	Biofortified β-carotene (crtB and DXS)	Failla et al. (2012)
60444	UCBSV resistance (RNAi-UCBSV) coat protein)	Odipio et al. (2014)
KU50z	Resistance to Sri Lankan CMV (AV2 and AV1 coat proteins)	Ntui et al. (2015)
TME 204	Resistance to CBSV and UCBSV, increase	Chauhan et al. (2015)
TME7, 60444	carotene content in roots	
60444	Biofortified vitamin B6 (AtTDX1.1 and AtTDX2)	Li et al. (2015)

Various bacterial structures, such as flagella, pili, secretion Total Cyanogenic Glucosides concentration depends on cultivar, environmental condition, cultural practices and plant age (McMahon *et al.*, 1995). Cassava roots contain the glycoside linamarin which is converted into hydrogen cyanide (HCN) by the enzyme linamarinase. HCN is toxic to man and hence much of the processing of cassava tubers is to promote release and removal of the HCN prior to consumption. When linamarin is hydrolysed, it releases hydro cyanide, a volatile poison (Cooke and Coursey, 1981); but some cyanide can be detoxified by the human body (Oke, 1983). In some varieties of cassava (Figure 20), the interior of the roots (parenchyma) contains only a small amount of cyanide. This is called sweet cassava, which may be boiled and eaten, as is normal in the South Pacific (Bradbury and Holloway, 1988). The linamarin content of cassava flour was reported to be more than system machineries (e.g., TIV SS and SEC), and lipopolys-accharides, as well as bacterium-derived proteins and molecules, such as effectors (EF), auto inducers, and antibiotics, are detected by the host cells and trigger the induced systemic, resistance (ISR) response (red). ACC, the direct precursor of ethylene (ET), is metabolized by bacteria via the enzyme ACC deaminase (ACCd), thus ameliorating abiotic stress (light green). A range of reactive oxygen species detoxification (ROS detox) enzymes might also ameliorate the plant-induced stress (orange). Diazotrophic bacterial endophytes are capable of fixing atmospheric nitrogen (N) and might actively transport NH^+ and nitrate (NO) to the host (dark green). Bacterial processes of siderophore production (Sid) and uptake (Fe) that are involved in plant growth promotion, bio-control, and phytoremediation are shown in brown (Figure 12).

CASSAVA NUTRITION, GENETICS AND ECONOMICS

Crop yields are greatly reduced by low soil fertility. Cassava is grown throughout the tropic and could be regarded as the most important root crop, in terms of area cultivated and total production (Ano, 2003), see Figures 3 and 4. Because cassava roots are very low in protein content (values range among cultivars from 5 to 19 g/kg dry matter, based on an average conservative Kjeldahl nitrogen-to-protein conversion factor of 2.49 - 3.67 (Yeoh and Truong,1996). Cassava leaves are also consumed and constitute an excellent source for protein supplement (leaf crude protein contents on a dry basis range among cultivars from 21% to 39% (Yeoh and Chew, 1976), minerals and vitamins for the human diet in many African and Asian countries, as well as in certain regions of Brazil (Lancaster and Brooks, 1983; Montagnac *et al.*, 2009; Djuikwo *et al.*, 2011). All cassava organs, except seeds, contain Cyanogenic Glucosides (CG). Cultivars with < 100 mg kg/1 fresh weight (FW) are called sweet ‘while cultivars with 100-500 mg kg/1 are bitter cassava (Wheatley *et al*., 1993) double during drought (Cardoso *et al*., 2005), which leads to outbreaks of konzo; most recently there were more than 100 cases in Nampula and Zambezia Provinces due to drought in 2005 (Muquingue *et al*., 2005). Consumption of cassava and cassava products containing large amounts of cyanide can cause acute intoxication, with symptoms of dizziness, headache, nausea, vomiting, stomach pains, diarrhoea and sometimes death (Mlingi *et al*.,1992) high-cyanide cassava roots containing >100ppm cyanide are normally bitter and are called bitter cassava pains, diarrhoea and sometimes death (Mlingi *et al*.,1992). high-cyanide cassava roots containing >100ppm cyanide are normally bitter and are called bitter cassava. Cassava (Manihot Esculenta Crantz), a Euphorbiaceae native to the Amazon region bordering Venezuela (Cagnon *et al*., 2002), is one of the main energy foods required by more than 700 million people in at least 105 countries. The most widely exploited product of the crop is starch, both for its nutritional importance and its use in the textile, pharmaceutical, food, and paper industries (Nunes *et al.*, 2009)

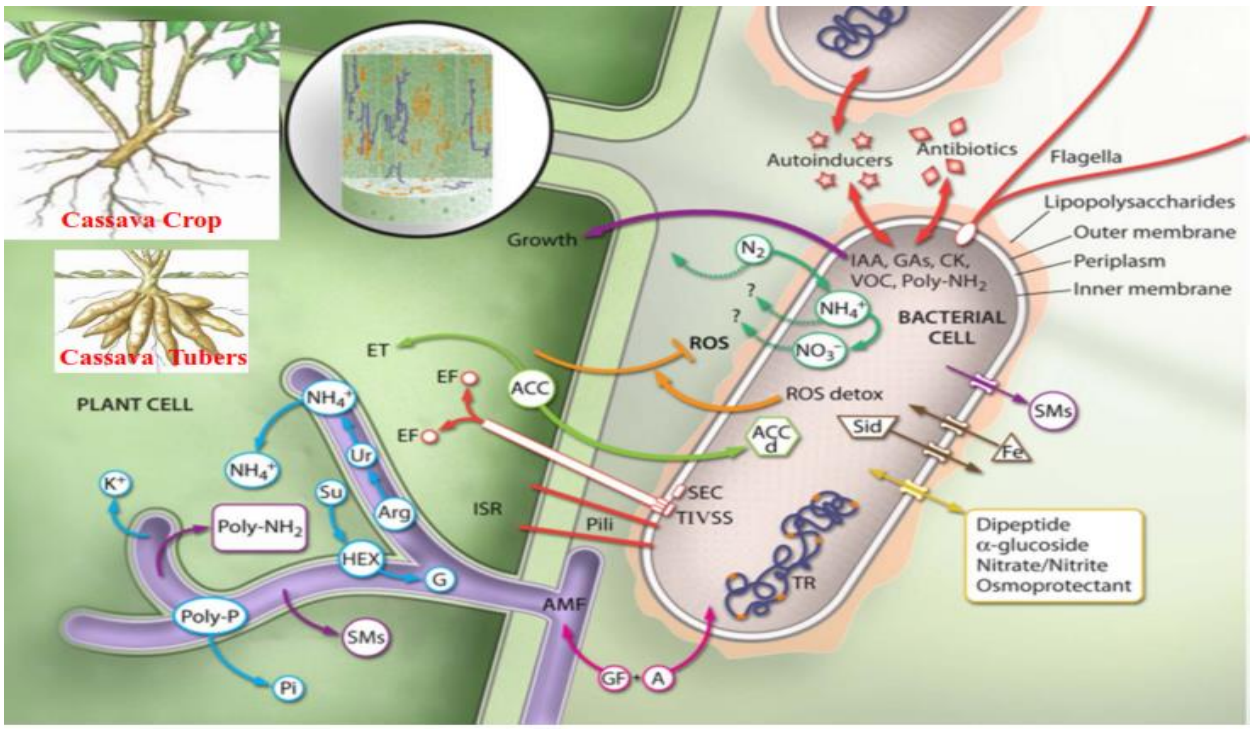

Figure 12 | Beneficial properties of endophytes on the cassava crop adapted from Hardoim *et al*., 2015. Endophytes high potential as a less exploited resource in sustainable agriculture.

In Brazil, cassava is cultivated on 1.5 million hectares, with a production of 22.8 million tonnes and an average yield of 15.2 Mg/ ha (IBGE, 2016). In the world ranking, Brazil occupies the 21th position for cassava yield, with India in first place with a yield of 36.5 Mg /ha (Faostat, 2015). In general, this low yield is due to the use of few inputs in managing the crop, and its cultivation in marginal areas. Research to improve traditional cassava processing methods for Garri production (machinery, skills or reduce the HCN in Garri, in Nigeria in the early 1950s (Idowu, 1990) Figure 13. Because of this and the high demand for cassava products, the use of nutrients through fertilization is inevitable in the near future (Adjei-Nsiahe Sakyi-Dawson, 2012). CGIAR, the Consultative Group on International Agricultural Research breeding strategy goals centered on selecting and developing cultivars with adequate and stable yields, and able to adapt to a wide range of biotic and abiotic stresses (Kawano, 2003; Hershey and Jennings,1992; Jennings and Iglesias, 2002). This strategy was stimulated by cassava's inherent capacity to tolerate adverse environments, particularly those where other major staple food crops such as cereals and grain legumes would fail to produce.

Cassava Cyanogenic Glucoside Production Implications |

Cyanogenic glucosides (Figure 14) naturally occur in cassava and are an important nutritional quality determining factor in its edible parts of cassava. High amounts of cyanogenic glucosides ingestion expose humans to cyanide intoxication, with detrimental effects on their health. Levels of cyanogenic glucosides need to be very low in fresh cassava roots or in cassava products, if these foods are to be considered innocuous and safe for consumption. Cassava roots generally have a high moisture content, which can differ with variants. The average moisture content of cassava roots, flour and starch usually ranks in the color sequence: yellow = orange > cream > white (Aniedu, and Omodamiro, 2012; .; Vimala *et al.*, 2009; La Frano *et al*., 2013; Maziya-Dixon

et al., 2005; Ukenye *et al*., 2013; Onitilo *et al.*, 2007).

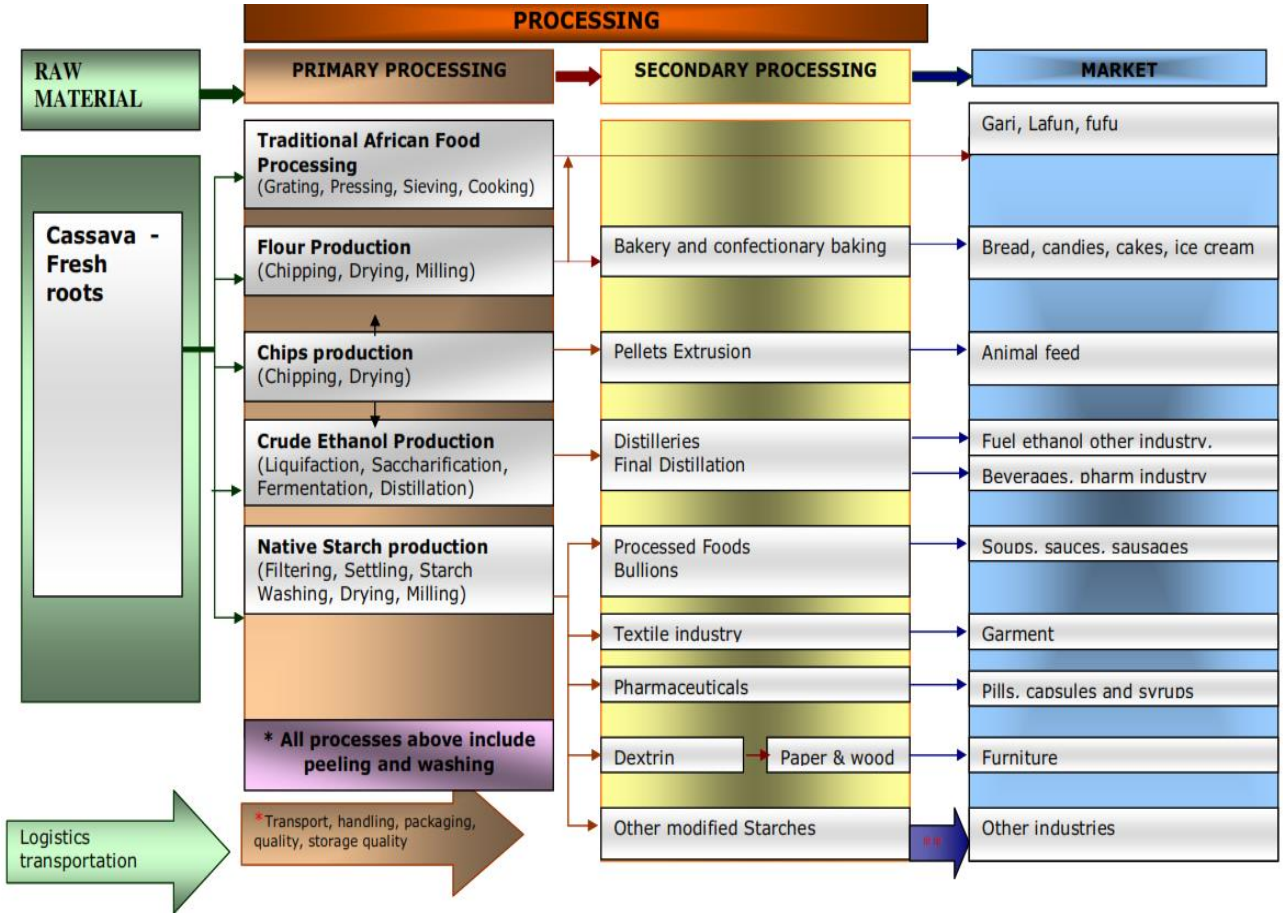

Figure 13 | A Simplified Example of the Cassava Value Chain., adapted from Cassava Master Plan March, 2006, the Presidential Initiative on Cassava, p, 14.

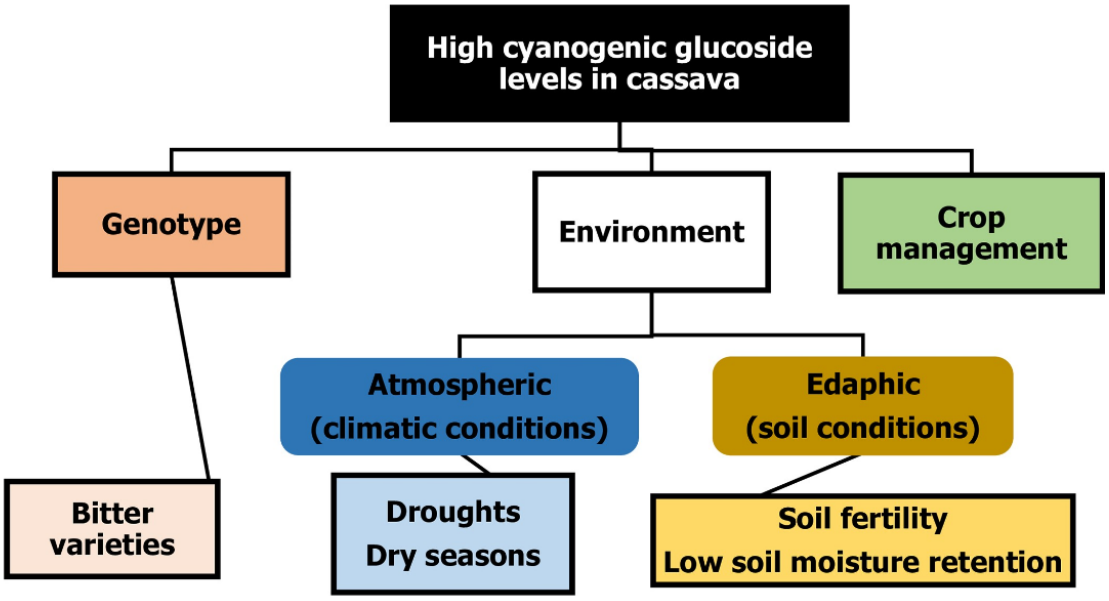

Figure 14 | Commonly known and less known agronomic reasons for high cyanogenic glucoside levels in cassava. Adapted from https://doi.org/10.1371/journal.pone.0216708.g001 and Imakumbili *et al.*, 2019.

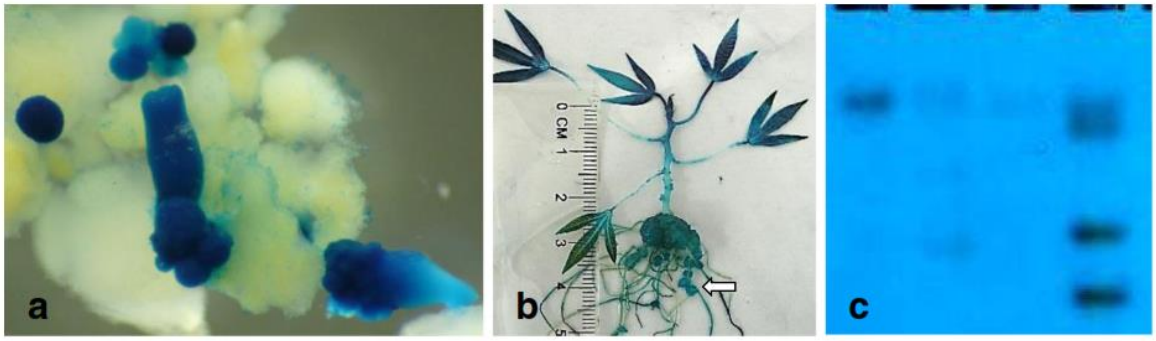

Figure 15 | (a) Transgenic somatic embryos and (b) plant of cassava cv. 60444 transformed with *Ensifer adaherens* OV14, expressing GUS. Note the formation of nodules on roots (arrows). This event was one of three obtained for which a Southern blot (c), confirmed the presence of single copy insertions (first and third lanes) as well as multi-copies of the T-DNA (second lane; fourth lane is control transgenic plant).

Cassava starch is characteristically low in fiber (0.10–0.15%) and lipids (0.11–0.22%) reported by Moorthy et al., 1994; Moorthy et al., 1996. Acidic and basic amino acids such as glutamic, aspartic and arginine are, however, relatively plentiful in cassava roots (Gil and Buitrago, 2002). Cassava root is relatively poor in other nutrients such as proteins, lipids, and vitamins (Talsma,2014). The Codex Alimentarius Commission currently recommends total hydrogen cyanide (HCN) levels (a measure of cyanogenic glucoside content) of less than 50 mg/kg in fresh cassava roots, as safe for consumption. Cases of cassava cyanide intoxication in cassava dependent communities, have often resulted in a health disorder called konzo (spastic paraparesis), which causes an irreversible paralysis of legs in affected individuals (Bradbury and Denton, 2010 ; Nzwalo and Cliff, 2011) A number of sub-Saharan African countries have been affected by konzo, and the disorder is reported as persistent in very deprived areas of Mozambique, the Democratic Republic of Congo (DRC), Tanzania Banea *et al*., 2012, Central African Republic (Tylleska and Legue,1994; Mbelesso *et al*., 2008) and in eastern Cameroon (Cigleneki *et al*., 2011; Agbor *et al*., 2014).The low organic carbon (OC) levels in all (100%) crop fields together with the low use of fertilizers, explains why N (Nitrogen) was low in these soils. Soil OC (organic carbon) is the main source of N for crops grown without fertilizer application. With regard to cyanogenic glucoside production, some studies have reported that an improved supply of N, on N deficient soils, is able to reduce cassava root HCN levels (Mohankumar *et al*., 1988; and Cadavid, *et al*., 1998). Other studies have similarly reported reductions in cyanogenic glucosides with improved N (Nitrogen) supply (Agbede, 2018; Pooja and Swadija, 2018). Improving the supply of N on these N deficient soils could thus be beneficial for reducing cassava root HCN levels. A moderate supply of N would however be better, as a high supply of N could increase cyanogenic glucoside levels (Pooja and Swadija,2018). Some studies have however shown no effects on cyanogenic glucosides with an improved supply of Potassium (Cuvaca *et al.,* 2015). Reduced root HCN levels are expected in cassava with an adequate supply of Ca (Gosh and Nair,1984; Mohankumar *et al.*, 1988). Like soil Ca, the adequate supply of Mg and Zn in soils, is beneficial for reducing cyanogenic glucosides in cassava (Mohankumar *et al.,* 1988; Susan ,2005). This is observed in the reduction of cassava root HCN levels, with the application of ash, which is rich in K, Ca and Mg (Susan,2005).

Transgenic Cassava |

In the past 25 years genetic transformation of cassava (*M. esculenta Crantz*) using Agrobacterium

tumefaciens or particle bombardment as gene-delivery systems with a new frontier globally where it has been possible to obtain transgenic plants of cassava expressing marker and selectable genes, as well as genes of agronomic interest. Agrobacterium-mediated transformation (Agrotrans) of cassava has been the technology of choice because it is more easily accessed by national agricultural research

programs (NARPs) in developing countries, where ultimately, transgenic cassava landraces with novel traits are most needed. The first genetic transformations (Table 13) of cassava using Agrobacterium were published in 1996 (Li *et al*., 1996; Raemakers *et al.,* 1996; Schöpke *et al.,* 1996), much work was done prior to these reports, especially at the International Center for Tropical Agriculture (CIAT) and at the Vrije Universiteit Brussel. The pioneering experiments that culminated with the production of the first transgenic cassava calli, expressing selectable and useful genes, were developed towards the end of the 1980s by (Calderon-Urrea, 1988). Currently, Table 20 cassava has been transformed with bacteria different than *Agrobacterium* named *Ensifer adhaerens* OV14. It contains chromosomal genes homologous to virulence genes of *Agrobacterium* (Rudder *et al.,* 2014) and was identified in 1982 as a gram-negative, predatory bacterium, inhabiting the rhizosphere with the ability to transfer genes into several plants, i.e., potato, tobacco, Arabidopsis, Solanum, and rice (Casida 1982; Wendt *et al.,* 2012; Soto *et al.,* 2015). Apparently, *Ensifer* seemed to be less virulent and pathogenic than *Agrobacterium* and therefore was considered an ideal vector to produce clean and unique insertions into plants (Rudder *et al*., 2014; Zúñiga-Soto *et al*., 2015). Transgenic cassava cultivars reported since 2010 for which genes expressing traits of interest for producers and/or consumers, other than marker and selectable genes, have been introduced. The Genetic Transformation Platform at CIAT used *E. adhaerens* strain OV14 with plasmid pCAMBIA5105 to transform cassava cv. 60444, based on the protocol reported by Zúñiga-Soto *et al.* (2015) for rice. Three transgenic independent lines were confirmed by Southern blot as having one insert (Two lines) or four copies of the T-DNA (Figure 12).

CONCLUSIONS

The study soil microbiomes during cassava cultivation as a component of integrated soil fertility management and a potential for climate mitigation cum crops sustainable development; and confirmed in the report of Yomeni *et al.,* (2010). Poor quality planting material is often associated with marginal growth and productivity of cassava. Microbial inoculants have paramount significance (Table 15) in integrated nutrient management systems to sustain agricultural productivity and healthy environment (Adesemoye and Kloepper, 2009). Application of 300 kg/ha NPK 15:15:15 + 3t/ha OBD -Biofertilizer gave high cassava root yield and is recommended for cassava production on soils having similar characteristics as the soil of the field experimental site (Table 4 and Figure 7). Environmental stresses are becoming a major problem and productivity is declining at an unprecedented rate (Figure 8). Our dependence on chemical fertilizers and pesticides has encouraged the thriving of industries that are producing life-threatening chemicals and which are not only hazardous for human consumption but can also disturb the ecological balance (Table 9). Biofertilizers can help solve the problem of feeding an increasing global population at a time when agriculture is facing various environmental stresses. It is important to realize the useful aspects of biofertilizers and implement its application to modern agricultural practices (Table 13). The success of the science related to biofertilizers depends on inventions of innovative strategies related to the functions of PGPRs (Figure 10) and their proper application to the field of agriculture. The major challenge in this area of research lies in the fact that along with the identification of various strains of PGPRs and its properties it is essential to dissect the actual mechanism of functioning of PGPRs for their efficacy toward exploitation in sustainable agriculture (Table 12). Future cassava development of cultivars (Table 20) should integrate the knowledge of the phytobiome for improved yields, and able to adapt to a wide range of biotic and

abiotic stresses and other ecosystem functions.

Conflict of Interest Statement

The author declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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