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Research Paper

Involvement of Plant Growth Regulators and Varieties in the Multiplication of Cassava Planting Materials: A case study of Rwanda

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ABSTRACT

Cassava (*Manihot esculenta* Crantz) is widely grown in tropical and sub-tropical regions of the world primarily for its starch-rich roots and, in some cases, its nutritional foliage. The overarching goal of this paper was to generate a comprehensive information on the status of the involvement of plant growth regulators and varieties in the multiplication of cassava planting materials with particular reference to Rwanda. This information will provide a platform and further strengthen the knowledge base on the application of plant growth regulators (PGRs) by stakeholders in producing clean, high-quality cassava plantlets. We demonstrated the interactive effect between different phytohormones and varieties. The study showed the classical plant response to hormones with an increase in triggering an optimal value before inhibition sets in. Phytohormone, 1-Naphthaleneacetic acid (NAA), and benzyl adenine (BAP) were reported as being crucial in root and shoot growth, respectively:- with 10 mg/l being optimal doses. The outcome is crucial in recommending users the choice of hormone and specific varietal combinations and points to the fact that various PGRs concentration levels can be conveniently utilized successfully in the multiplication of different cassava varieties. Since the hormonal response has been shown to trigger differential reactions between different varieties, adopting such technologies needs to be considered with caution in consultation with researchers and policymakers to avoid adverse effects. In conclusion, the rapid multiplication of cassava planting materials depends on the PGRs concentration levels and their admixtures, the genetic makeup of the varieties evaluated, subculture type/ number, and shoot tip/ nodal segment used.

HIGHLIGHTS

- Cassava is mainly grown primarily for its starch-rich roots and, its nutritional foliage.
- Whereas 1-Naphthaleneacetic acid plays a key role in triggering root growth, benzyl adenine is crucial in enhancing shoot growth.
- For rapid multiplication of cassava planting materials, the optimal doses for these phytohormones ought to be 10 mg L^{-1}.

Keywords: Planting medium, supplementation, propagation, varieties, hormonal concentrations, root, shoot

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Cassava (*Manihot esculenta* Crantz) is a perennial woody shrub that belongs to the family *Euphorbiaceae* (Sessou *et al.* 2020). It is a rain-fed crop grown widely in tropical and sub-tropical countries of Latin America, Africa, and Asia. It is a staple food for over 800 million people globally, providing a cheap source of carbohydrates (Burns *et al.* 2010). The crop has gained economic importance as a raw material for the industrial processing of foods, ethanol, and starch (Kilwinger *et al.* 2021). It is also used as a cash crop in various cassava-growing regions (Spencer and Ezedinma, 2017; Munganyinka *et al.* 2018). El-Sharkawy, 2004) stated that the yield potential of cassava under optimum conditions is about 90 tons of fresh roots per hectare, equivalent to 30 tons of cassava dry matter per hectare.

In Rwanda cassava ranks 3rd after banana and sweet potato as a staple crop, with an annual production of approximately 1,124,090 MT per annum (MINAGRI, 2011). The crop is grown mainly in the southern and eastern provinces of the country and occupies 22% of the area under cultivation. More than 700, 000 households (i.e. 42% of all family farms) grow cassava annually (MINAGRI, 2011). The crop grows well on soils with relatively low fertility and is also relatively drought and acid tolerant, making it an ideal crop for smallholder farmers in unfavorable upland environments (Malik *et al*. 2020; Howeler and Aye, 2014).

Conventionally, cassava can propagate through seeds and stem cuttings. However, cassava seeds are typically diverse because of the cross-pollination nature of the crop. Generally, any particular cassava clone is highly heterozygous (Ceballos *et al.* 2004). Seed viability, dormancy, irregular blooming patterns, and poor seed set are all characteristics that restrict the use of seeds as a viable source of propagation. As a result, stem cutting is the primary mode of propagation (Leihner, 2002). However, this approach has drawbacks such as a low rate of multiplication, ten cuttings per plant every year (1:10), which is difficult and time-consuming, sluggish and resulting in delayed dissemination of new better cultivars, bulky to transport, and insufficient planting supplies for large-scale plantations (Demeke *et al.* 2014). Furthermore, disease build-up throughout the vegetative cycle, high distribution costs, and poor storage quality of the planting material are some of the drawbacks of utilizing stem cutting as a propagation material in cassava (Escobar *et al.* 2006).

Another drawback is diseases such as Cassava Mosaic Disease (CMD) and Cassava Brown Streak Disease (CBSD), which are currently the most threatening biotic stresses to cassava production in East and Central Africa (Alicia *et al.* 2007; Hillocks and Thresh, 2000; Legg and Raya, 1998; Legg *et al.* 2001; Tumwegamire *et al.* 2018). The two diseases cause devastating effects on root quantity and quality, with field and storage losses ranging from 30% to 100% (Kawuki *et al.* 2016; Okonya *et al.* 2019; Patil *et al.* 2015). Both diseases spread via a whitefly vector (*Bemisia tabaci*) and the use and exchange of infected planting material (Legg et *al.* 2011). This implies that farmers' use of local susceptible varieties and recycling of stem cuttings from the previous crop can aggravate the impact of the diseases. Therefore, the introduction of resistant varieties and the availability of clean planting material is of high importance (Night *et al.* 2011) for successful and sustainable cassava production.

Moreover, the newly released cassava varieties, and the scarcity of high-quality and true-to-type planting material are among the main limitations for their widespread commercialization and increased production (Escobar *et al.* 2006). It is, therefore, imperative to consider new technologies for the rapid multiplication of disease-free planting material, which will be a significant step towards achieving adequate, true-to-type, high-quality cassava planting materials. Santana *et al.* (2009) acknowledged the plant tissue culture technique as a powerful tool for studying and solving fundamental and applied problems of cassava production. Moreover, Loyola-Vargas *et al.* (2006) described the plant tissue culture technique as a quicker and less space-consuming technology compared to conventional methods of preparing cassava cuttings. Similarly, Le *et al*. (2007) established that the tissue culture technique is one of the most realistic and efficient means of supplying large volumes of true-to-type clean planting materials of cassava within a limited period. This is also an advantage for producing large populations as required in mutation breeding. In addition, plant tissue culture has many applications, such as clonal propagation and somatic embryogenesis, germplasm exchange, embryo rescue, genetic transformation, etc. (Rao,

1996). This work aims to generate a comprehensive information on the status of the involvement of plant growth regulators and varieties in the multiplication of cassava planting materials, with specific reference to Rwanda as a representative of other developing countries.

Methodology

To acquire information relevant to this work, we used the Google Scholar search engine to Identify relevant peer-reviewed research articles published in high-impact journals.

A total of 58 articles were used that ranged from experimental, reviews, and policy papers. Keywords used in the Google Scholar search engine were Rwanda, Plant growth regulators (PGRs), cytokinins, Kinetin, benzyl adenine (BAP), Thidiazuron (TDZ), Zeatin, 1-Naphthaleneacetic acid (NAA), varietal differences, clean planting material, the government of Rwanda, Basal supplementation, Casava, Seedling regeneration, Murashige and Skoog, varietal interactions with phytohormones. The paper is focused broadly on various methodical strategies used in cassava to improve multiplicative rates using phytohormones in different varieties and plant species in Rwanda and other parts of the world.

Basal media supplementation with plant growth regulators on cassava seedlings regeneration

Growth regulators (PGRs), especially Cytokinins are among the most critical components that play a significant role in micro shoots regeneration (Abu-Romman *et al.* 2015; Garland and Stoltz, 1981; Huy *et al.* 2019; Lane, 1979). Specifically, the use of growth regulators for growth initiation from the meristem culture of different cassava varieties is recommended by Razdan (2005). Cytokinin could interact with other growth regulators to stimulate the vegetative growth of plants (Maxwell and Keiber 2004). This is because, in the plant physiology process, cytokinin influences cell division to broaden the area of the tissues and plantlet height (Davies 2004). Kinetin, benzyl adenine (BAP), Thidiazuron (TDZ), and Zeatin have been used in cassava micropropagation (Konan *et al.* 1997; Faye *et al.* 2015; Kabir *et al.* 2015; Opabode, 2017). However, the most commonly used cytokinin for inducing shoots in cassava is either BAP alone or in combination with NAA (Cacaï *et al.* 2012; Faye *et al.* 2015; Sesay *et al.* 2012) (Fig. 1). Besides, Auxins are essential factors involved in rooting because they promote the formation of adventitious roots in most species (De Klerk, 2002).

Use of plant growth regulators to enhance shoot multiplication

Several authors maintain that BAP is the best phytohormone for shoot initiation and shoot multiplication in cassava (Opabode *et al.* 2017; Pita *et al.* 2001; Onuoch and Onwubiku, 2007; Guohua, 1998; Trigiano and Gray, 2000; Roca, 1984). In support of this, Konan *et al.* (1997) observed the effect of BAP on the formation of multiple shoots from enlarged axillary buds of cassava cultivar TMS 30555, and the variable rates of shoot proliferation were observed 5-6 weeks after culture initiation. We have shown schematically how the mechanism works (Fig. 1).

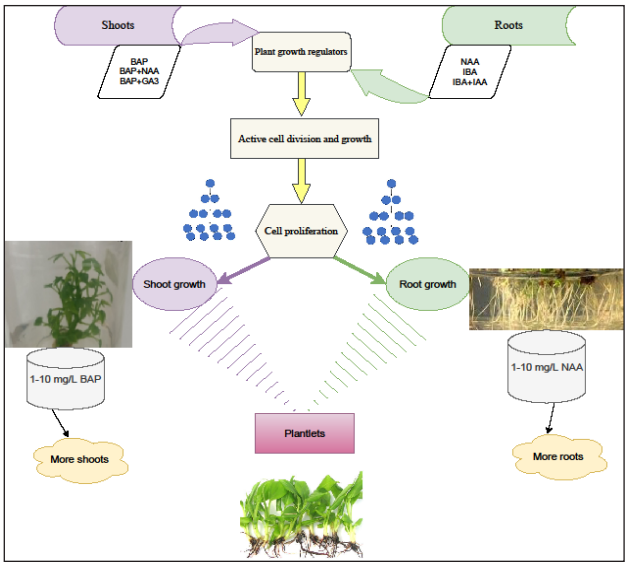

Fig. 1: Schematic representations of plant growth regulators on shoot and root growth for enhanced planting material multiplication. Different hormones have differential triggers on roots and shoots, but they work within a range with specific optimal values. In the diagram, the BAP is shown to have a better influence on shoot regeneration up to 10mg/l after which there is inhibition. A similar trend is shown for roots, though with NAA

The highest number of shoots per explant was obtained with BAP at 10 mg/1 BAP where the percentage of explants forming shoots was shown to be about seven times higher than with 15 mg/1 BAP (73% vs. 9%). On the contrary, Fan *et al.* (2011) reported that BAP 0-2.0 mg/L was effective on shoot regeneration. Furthermore, Konan *et al.* (1997)

Table 1: The response of cassava shoots cultured on Murashige and Skoog (MS) media supplemented with plant growth regulators

Cassava genotype	Country	Propagation media	Explant	Treatment that gave the best results	References
Agric-rouge, Atinwewe, Agblehoundo.	Kenya	MS	Nodes	10 uM BAP	Sessou *at al.* (2020)
Darul Hidayah, Malang-6 and Adira-4	Indonesia	MS	Auxiliary shoots	1 mg/L BAP + 0.1 mg/L TDZ	Sukmadjaja and Widhiastuti (2011)
92/0326, 95/0289, I-30572	Nigeria	MS	Stem	a) 0.01 mg/L NAA; 0.05 mg/L BAP b)0.02 mg/L NAA; 0.10 mg/L BAP	Mapayi *et al.* (2013)
MAUS 2, MAUS 4	Australia	MS and 2.5 uM IBA	Axillary buds	1.0pM BAP and 0.25 uM NAA	Smith *et al.* (1986)
4/72-NR: 44/72-NW.	Ethiopia	MS	Nodal buds	BAP and Kin each at 0.75 mg/L	Demeke *et al.* (2014)
Sree Padmanabha.	India	MS	Apical node	0.3 mg/L NAA	Shiji *et al.* (2014)
TMS 98/0379; TMS 98/0581	Nigeria	MS	Stem	0 mg/L BAP	Onuoch and Onwubiku, (2007)
Slicass 6, Slicass 11 and Cocoa	Sierra-leone	MS	Stem	1.0 mg/L NAA	Janatu *et al.* (2018)
		MS	Stem	0.1 mg/L BAP	Janatu *et al* (2018)
CM6740-7 cassava cultivar	Venezuela	MS; 2 g/L Hydro Agri´s fertilizer (12-11-18/3 (MgO-EDTA)	Stem	0.027 M NAA, 0.011 M IBA, 0.027 M IAA)	Santana *et al.* (2009)
Qulle and Kello	Ethiopia	MS	Stem	0.5 mg/L BAP; 1 mg/L GA_3; 0.01 mg/L NAA	Beyene *et al.* (2010)
Arara, Caturra, Cacau Vermelha and Roxinha.	Colorado do Oeste	MS	Shoot petiole	MS - BAP addition,	Menegazzo *et al.* (2019)
Cultivar TMS 30555, ipin Valenca, MBra 769, CMC 76, MCub 51, MCub 58, MMex 55, MPar 133, TMS 60444 Red, TMS 30395, TMS 50395, TMS 60142, TMS 83350, TMS 84537, TMS 90059, TMS 90853, and Mpira Red	—	MS	Nodes	10mg/L BAP	Konan *et al.* 1997

reported a maximum of 25 shoots per explant in a solid Murashige and Skoog (MS) medium with 10 mg/l BAP. Similarly, Sessou *et al.* (2020) showed that BAP at 10 µM contributed to the highest number of micro shoots/explant (3.60) though the response was cultivar and concentration-dependent. Although in an exciting, and contradicting report, Beyene *et al.* (2010) pointed out that increased supplementation of BAP concentration from 5 mg/L to 10 mg/L, as a multiplication medium, caused shoot dwarfing and resulted in abnormal morphological appearance of stems and leaves, which probably depended on variety.

Sessou *et al.* (2020) reported that BAP beyond 10 µM significantly reduced the number of micro shoots/explant and their lengths. They also observed that the multiple shoots regenerated from media supplemented with BAP were stunted compared to those obtained with kinetin (Table 1). This might be a result of the supra-optimal amount of the hormone. This response is in agreement with the results of Berrie (1984), Onuoch and Onwubiku (2007), who observed that high BAP concentrations had a negative effect on the plantlet's physiology, such that there was an inhibitory effect on shoot growth at high concentrations. Moreover, Shiji *et al.* (2014) observed that MS medium with NAA alone produced better results in terms of the response of

explants to shoot initiation, intermodal elongation/ shoot length, number of nodes, number of leaves and number of roots when compared to the other treatments.

Work by Beyene *et al.* (2010) revealed the highest number of shoots per explants (where 27 and 21 shoots for 'Qulle' and 'Kello' varieties, respectively) that were obtained in multiplication medium with 0.5 mg/L BAP in combination with 1 mg/L GA_3 and 0.01 mg/l NAA (Table 1). This might be due to the combined effect of the three growth regulators (Staden *et al.* 2008). Further, Konan *et al.* (2006), working with nodal explants of cassava with axillary meristems cultured on MS medium supplemented with 0.1 mg/1 NAA, 1 mg/1 BAP, and 0.1 mg/L GA_3, reported that a combination of these phytohormones was superior in producing multiple shoots. Smith *et al.* (1986) in their report from *in vitro* propagation of cassava using nodal culture, recommended the use of 1.0 mg/l BAP, supplemented with 0.25mg/L NAA-induced multiple-shoot formation (Table 1). In addition, Kabir *et al.* (2015) found that among the media components used, MS + 2.0 mg/l BAP + 0.1 mg/l NAA achieved the best for multiple shoot formation at 3rd subculture, in which 90% of explants produced multiple shoots. Consistently, the average number of shoots per explant was 6.30, the average length of the shoot was 10.30 ± 0.70 cm, and the average number of nodes per explant was 6.25 in the same medium. Furthermore, in their study, Kabir *et al.* (2015) revealed that BAP in combination with NAA performed comparatively better in terms of multiple shoot initiation, which suggests that the combination of growth regulators has a good impact on *in vitro* shoot proliferation of cassava. Similar results were observed by many authors using different concentrations and combinations of BAP + NAA in other plants, including sweet oranges (Roy and Kabir, 2006), apples (Roy and Kabir, 2006), cabbage (Munshi *et al.* (2007), sugarcane (Roy and Kabir, 2007). In addition, Alla *et al.* (2013) reported that the maximum number of performed shootlets per explants (5.67) was achieved on MS medium supplemented with 1.0 mg/L BAP plus 0.05 mg/L NAA (proliferation medium).

Murashige and Skoog medium has also been supplemented with other phytohormones such that Mapayi *et al.* (2013) reported that the best survival rate (100%) of cassava shoots was, generally, obtained in MS medium supplemented with a low concentration of NAA and BAP hormones and high concentration of sucrose (Table 1). Sukmadjaja and Widhiastuti (2011) reported that the highest number of shoots from three elite cassava cultivars were obtained on media supplemented with a combination of BAP and thidiazuron (TDZ) (Table 1). According to Lu (1993), the addition of TDZ in media containing BAP could increase the explant's ability to produce shoots. Medium supplemented with GA_3 only and in control recorded the lowest number of shoots per explant. This is because GA_3 concentration increases the shoot height rather than multiplying the number of shoots. Similar results were obtained by Acedo (2006). According to Moshkvo *et al.* (2008) this could be due to the physiological effect of the GA_3 hormone that causes stem elongation and inhibits the formation of adventitious root and shoot formation. Although Beyene *et al.* (2010) observed that combined supplementation of BAP and GA_3 resulted in shoots with very good morphological appearance (reasonable shoot height, stem thickness, and leaf structure in comparison with the other multiplication medium in combination), the effect might be related to the combined reaction of the two PGRs. In addition, Kane (2005) reported cytokinins, BAP/Kinetin (0.01-10 mg/L) as the most widely used and effective plant growth regulators for shoot multiplication. Further, Faye *et al.* (2015) found that kinetin gave better results than BAP in regenerating micro cassava shoots from nodal explants. Sessou *et al.* (2020) also observed that the multiple shoots regenerated from media supplemented with BAP were stunted compared to those obtained with kinetin. Kinetin has also been found to be superior to other cytokinins in *Tacca leontopetaloides* (Martin *et al.* 2012).

Use of plant growth regulators in root multiplication

In the work of Medina *et al.* (2007) it was apparent that 0.54 mM NAA was most effective in stimulating root formation. To support this argument, Demeke *et al.* (2014) used 0.5 mg/L NAA and reported the production of 6.14 roots within 4 weeks (Table 2), while Cacaï *et al.* (2012) used 0.1 mg/L NAA and reported the production of 5.2 roots. Furthermore, Fan *et al.* (2011) reported that NAA ranging from

Table 2: The response of cassava roots cultured on Murashige and Skoog (MS) media supplemented with plant growth regulators

Cassava genotype	Country	Propagation media	Explant	Treatment that gave the best results	References
92/0326, 95/0289, I-30572	Nigeria	MS	Stem	a) 0.01 mg/l NAA, 0.05 mg/l) BAP and b) 0.02 mg/l NAA; 0.10 mg/l BAP	Mapayi *et al.* (2013)
MAUS 2, 4, 7	Australia	MS and 2.5 Um IBA	Auxiliary bud	1.0pM BAP and 0.25 uM NAA	Smith *et al.* 1986
Sree Padmanabha.	India	MS	Apical node	0.1 mg/l NAA,	Shiji *et al.* 2014
Slicass 6, Slicass 11 and Cocoa	Sierra-leone	MS	Stem	1.0 mg/L (NAA)	Janatu *et al.* 2018

0-2.0 mg/ L was effective on root development. In addition, Kane (2005) reported that auxin NAA (0.01-10 mg/L) was the most widely used and effective plant growth regulator (PGR) for root induction. In consistence, Shiji *et al.* (2015) (Table 2) and Opabode (2017) observed that NAA was the best supplement for rooting in cassava.

There is more research on hormones and elite cassava varieties. Sessou *et al.* (2020) found that IBA performed better than NAA in rooting the micro shoots from the 3 elite cassava cultivars. Similar results were reported by Kabir *et al.* (2015), who concluded that cassava micro shoots rooted better in MS media supplemented with IBA compared to NAA and IAA. In the same breadth, the effectiveness of IBA for rooting over other auxins has also been reported by Naranjo and Fallas (2017) in cassava. Additionally, Alla *et al.* (2013) ascertained that MS medium supplemented with 2.0 mg/L IBA achieved the maximum number of root formations (10.2), with a 100% rooting percentage. Similar observations have also been made in many other *in vitro* cultured plants. For instance, Sadeghi *et al.* (2015) achieved 100% *in vitro* rooting of *Prunus empyrean* in MS medium with IBA, and Singh *et al.* (2016) reported IBA as the best auxin for rooting in *Santalum album*. Further, Acedo and Laban (2008) reported that IAA at 0.02 mg/L and IBA at 0.04 mg/L were the best for rooting short-maturing cassava genotypes, and 0.06 mg/L IBA was the best for long-maturing genotypes. Smith *et al.* (1986) also showed that the use of 2.5 mg/l of IBA effectively improves the root initiation of cassava plantlets. On the contrary, Beyene *et al.* (2010) found that half and full-strength MS at 0, 0.01, and 0.1 mg/L of IBA had less rooting frequency, and roots were long and fragile with very few secondary roots. Nevertheless, at high concentrations of IBA (0.5 and 1 mg/L IBA) roots become short and thick without secondary roots.

Furthermore, Sessou *et al.* (2020) observed that increasing the concentration of IBA from 10 to 20 µM significantly reduced the mean number of roots in plantlets derived from the media supplemented with the three cytokinins. Further, Mushiyimana *et al.* (2011), Yandia *et al.* (2018), and Faye *et al.* (2015) proved that MS medium without exogenous auxins is the best in cassava micro rooting. Rooting in a medium without growth regulators has been reported in *Yucca glauca* Bentz *et al.* (1988) and *Gentiana dinarica* Beck plant (Vinterhalter *et al.* 2012). A possible explanation could be that there is a high level of endogenous auxins.

Varieties and their interactive effect with growth regulators on the multiplication of different plantlets

Marked differences are observed among the plant varieties and their interaction with PGRs on the shoot and root rapid multiplication. A comparison of *Solanum tuberosum* varietal effect showed that Cardinal produced the highest number of shoots explant^{-1} (1.98) followed by Dheera (1.83), whereas the lowest number of shoots explant^{-1} (1.71) was obtained from Diamant (Hossain *et al.* 2013). Similarly, Nagib *et al.* (2003) reported that Cardinal was the best, and Diamant was also found to be more responsive than Multa and Lal Pakri. Comparing both studies, the Cardinal variety emerged to be the best. The result is also supported by other findings of Millar *et al.* (1987) where differential responses of different potato

varieties due to genetic makeup towards *in vitro* shoot multiplication and their development was reported. Combined effects of variety and plant growth regulator revealed that Cardinal gave the maximum number of shoots explant^{-1} (2.43) with 1.0 mg/L BAP followed by Granula with 1.5 mg/L BAP (2.40) (Hossain *et al.* 2013). Similar results were also obtained by Nagib *et al.* (2003), where Cardinal gave the maximum number of shoots explant^{-1} with 0.5 mg/L BAP than with 2.0 mg/L and without BAP.

Working with French beans under water-stressed conditions, Kalawa *et al.* (2018) demonstrated the interactive effects of growth regulars in alleviating drought stress and hence increasing yield, particularly compared to the non-stressed and also with control without phytohormones. Several researchers (Nduwimana *et al.* 2020; Ochieng *et al.* 2021; Gweyi-Onyango *et al*. 2009; Munene, 2017) working with different types of nitrogen in soils as well as in culture media were able to report varietal responses to different N forms, particularly nitrate-treated plants. In these cases, the nitrate acted through a phytohormonal transduction cascade by eliciting more root and shoot divisions and hence increased multiplicative rates and not necessarily playing a role of nutrient *per se*. Working on superior cassava clones, i.e. Darul Hidayah *et al.* (2011) revealed that the highest number of shoots were 4.93 Darul Hidayah, 4.20 Malang-6, and 7.20 Adira-4, although this was recorded from different MS medium supplementation that is 1 mg/L BAP+ 0.1 mg/L thidiazuron, 1 mg/L BAP, and 1 mg/L BAP + 0.1 mg/L thidiazuron respectively. Similarly, this was observed in root multiplication such that Darul Hidayah and Adira-4 showed an increase of both root number and length resulting from adding IAA to the nutrient media. However, no significant effect of IAA and NAA were obtained on the number and length of roots in Malang-6 variety. This difference could be attributed to naturally occurring endogenous phytohormones that differ among genotypes and are coupled with their response to the exogenous growth regulators added to the culture medium (Rahimi *et al.* 2022). Popular cultivars among farmers, namely Victoria, Kachpot 1, and Kinigi differed as influenced by a combination of GA_3 and NAA in shoot height (Nuwagira *et al.* 2015). The shoot height and the number of buds, roots, leaves, and nodes were significantly different for varieties and hormonal combinations (Awati *et al.* 2019). Furthermore, shoot multiplication of *Solanum tuberosum* L.Gudiene and Belete varieties differed as influenced by growth hormones. The combination of 1.5 mg/l BAP and 3.0 mg/LNAA proved best in Gudiene, whereas 1.0 mg/L BAP and 2.0 mg/l NAA produced more shoots in Belete (Hajare *et al.* 2021) (Fig. 1). Furthermore, Sakha *et al.* (2019) showed varietal differences in sweet potatoes' production and regeneration of planting materials.

CONCLUSION

By and large, the study showed that the multiplication of planting materials is dependent on the genotype, growth medium composition, its supplementation with individual PGRs and their mixtures, the genetic makeup of the varieties evaluated, subculture type/ number, and shoot tip/ nodal segment used. Moreso, it is necessary to alter the composition and or concentration of growth regulators in the culture medium depending on the genotype, origin of the explants, and culture conditions. If used through consultation with scientists, government agencies, and policymakers, the technology can enormously contribute to the enhanced and sustainable production of clean cassava planting materials in a short period in Rwanda.

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