

Full Length Research Paper

Banana as Natural Plants to Artificial Culture Media

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Accepted 24th October, 2016.

Plants are the most living things which sustain the life by production of respiratory oxygen and also play an important role for soil fertilization acting as manure. For banana plants, they are so important in a way that their application is becoming broad, if you consider their chemical content; It show much mineral content that can act as culture media during in vitro culturing, with that ability, the research aims to provide the alternative way to supply natural ingredients that can reduce the cost of the currently used culture media. The research examined currently used Murashige and Skoog (MS) nutrients and then collected information about local plants that met the condition to support the growth of in vitro life. Banana was chosen from other plants examined for its nutrients. The predominant elements found in banana plant pseudostem ash were Na, K, Ca, Mg, Fe and P classified as macro-elements added to present micro-elements. Considering minerals distribution within banana ashes vitamins and hormones were absent, bolus complement was added as compliment both rich in hormones and vitamins. The mixture has been prepared, added agar to yield the full media. Then followed by planting mother plantlets inside the media, the experiment has used two parallel media, natural and MS media have been planted with the same plantlet variety of potato for easy assessment, they transferred them into the growth room. The first two days; there was no significant change for both natural and MS media barquettes. At the fourth day, there was formation of callus where root will rise and appearance of shoot from nodes. At the sixth day, the plantlets have both shoot and root. At the end both have shown the ability to have supported the growth of plantlet, the difference was that the natural media become older early but plantlet were fleshier than MS media, but other features were the same.

Keywords; Macro-elements, micro-elements, callus, in vitro culture, minerals, pseudostem, vitamins, plantlet.

INTRODUCTION

The Rwandan economy depends mainly on the production of the primary sector, where agriculture plays a vital role as the main sector that employs 91% of the country's people. Poor performance observed in this essential sector of economy aggravates poverty in rural areas, as agriculture is the most important source of income. The way to face this challenge is to improve the agriculture production and to add value to agricultural product by improving post-harvest processing, introducing new technology and other research so as to be competitive on regional and internal markets(Office of UN Resident Co-Ordinator, 2002).

However, it was suspected that potato demand is highly increasing while there is a lack of good planting materials as the most challenge which hinders the agriculture production and cause the perpetuity of

hunger in Rwanda(Africa, 2002b). It is for the fulfillments of its mission of training sufficient technicians on the level A0 to the point that their number and skills meet the standard requirements in the field of agriculture and to contribute to ongoing poverty reduction and food security programs through an increased crop production resulting from training, research and application of significant technology. It is with that cases that the government has bring potato program to help secure Rwandans the food,(Africa, 2002a). Again potato have been introduced as the fast growing plant to which does not take long period for harvesting,(Vinterhalter, Dragiüeviü, and Vinterhalter, 2008).

For that purpose, the government of Rwanda has implemented the macropropagation techniques termed 'in vitro culture' which is a techniques used to multiply

and develop much seeds in short period of time which are disease free. In vitro culturing techniques has emerged nowadays to become more usefully than ever (Venkatasalam, Sood, Pandey, Thakur, and Sharma, Ashwani K. and Singh, 2013), this techniques utilize the artificial media acting as soil and provide satisfactory growth of the plant so as to generate high yield in a short period of time. (Saad and Elshahed, n.d.). As agriculture is advancing for eradicating poverty, mass production as well increase. Culture media plays a vital role as the starting point for this in vitro culturing. There exist many types of culture media, including; Murashige and Skoog (MS) medium, Linsmaier and Skoog (LS) medium, Gamborg medium and Nitsch and Nitsch (NN) medium, although we do have all those types of culture media different studies have been done to see how one of the used ingredient, like sugar can be substituted by another thing in a manner of reducing the expensiveness of the culture media. We can give an example of the use of charcoal in the culture media as carbon source (Peng, Wang, and Li, 2012), others have tried to find a new gelling agent that can replace the one used mostly agar, corn starch have been essayed to see whether it can work as a gelling substance, it was examined and showed that the gelling increase in response to the amount of sugars present (Sun, Xing, Qiu, and Xiong, 2014).

Currently the most frequently used culture media is Murashige and Skoog media. (Saad and Elshahed, n.d.), but due to its high market demand it do have high cost. Therefore, the research suggested an alternative way to see how Rwandans can benefit from nonused resources by exploring them to the production of culture media used in the in vitro laboratory, it then examined the MS nutrients content and then collected information about different plants which can supply nutrient to support the growth of potato which was used as the modal for the research, Tobacco leaf was among the plants examined, it was found to be rich in minerals which are affected according to the agriculture, types of soil, weather conditions and plant diseases. (Leffingwe, 1999). It was found to be having the following minerals; volatile bases as ammonia, Nicotine, Ammonia Glutamine as ammonia, Asparagine as ammonia, Nitrate nitrogen as NO_3 , it has Acidic pH, volatile acids as acetic acid, Formic acid, Malic acid, Citric acid, Oxalic acid, Volatile oils, Alcohol-soluble, resins, Reducing sugars as dextrose, Pectin as calcium pectate, Crude fiber, Ash rich in Calcium as CaO , Potassium as K_2O Magnesium as MgO Chlorine as Cl Phosphorus as P_2O_5 and Sulfur as SO_4 . (Leffingwe, 1999). Although the leaf was found to be rich, it was found to be in scarce at the market level, not only that but also the area for cultivation would be a stopping factor.

By that case Banana came as another plant to deal with, it is found to be one of the largest cultivated plant not only in Rwanda but also worldwide. (Mohapatra, Mishra, and Sutar, 2010). From its high cultivation, it's by

product result to be in excess so that they can be explored into profitable products. (Of and Chemical, 2010). To that it was found to be having minerals nearly equal to that of MS. It was found it is rich in minerals that can support the growth of in vitro plantlets. The predominant elements found in banana plant pseudostem flour were Na, K, Ca, Mg and P which are classified as macro-elements. Calcium was found in the highest amount, followed by K, Na, Mg and P (Ho, Noor Aziah, and Bhat, 2012). However, the banana pseudostem flour results obtained from the present study differ from those reported by Selema and Farago (1996), who have reported that the banana pseudo-stem (*Musa Paradisiaca*) possesses a higher amount of K than Ca. (Ho et al., 2012). For allocating what parts is more vulnerable to minerals, it was found that each part is having different mineral enriching from other. For example Bract was found to be rich in potassium, magnesium and iron content, while the leaf was highest in calcium and phosphorus content. (Okareh, Adeolu, and Adepoju, 2015). by then to yield a rich solution, it was recommended to prepare both leaf and pseudostem. Considering minerals distribution within banana, it was found that banana alone cannot satisfy the whole plantlets, as result bolus complement was added as compliment rich in both minerals and vitamins (Sheet, n.d.), this complement was found to be having .

At the last it was the study of what type of medium would be better, the study preferred to use the solid medium which is provided by plant agar.

This research aimed to prepare culture media and to explore non used resources to contribute to the developments. I used potato as an experiment specimen.

MATERIALS AND METHOD

Our research was a laboratory based experiment for assessing the feasibility of new culture media. it took place at Rwanda Agriculture Board/northern agricultural zone division (RAB/NAZD) from June to 10th July 2016.

We used different materials; laminar flow, barquettes, oven, agitator, ph meter, growth room,

To obtain the needed culture media you burn banana leaves and stem collect ash and mix with bamboo flour remained from skewer and mix them with distilled water. You adjust ph by using nitric acid and sodium hydroxide, their after you add agar, lastly you autoclave them for 15minutes at 120^oc.

Below are the procedures.

Ash from pseudostem

Put 1 liter of distilled water in a beaker
Add 9.6 grams of pseudostem ash
Add 25 grams of sucrose.

Add 9 grams of bolus
Put the magnetic bar inside the beaker and put it on magnetic shaker until dissolution is done.
Adjust the ph at 5.9
Transfer the mixture solution in autoclavable bottles.
Add 7 grams of plant agar.
Autoclave the medium at 121⁰c for 15 minutes for sterilization.

Ash from leaves

Put 1 liter of distilled water in a beaker
Add 9.6 grams of pseudostem ash
Add 25 grams of sucrose.
Add 9 grams of bolus
Put the magnetic bar inside the beaker and put it on magnetic shaker until dissolution is done.
Adjust the ph at 5.9
Transfer the mixture solution in autoclavable bottles.
Add 7 grams of plant agar.
Autoclave the medium at 121⁰c for 15 minutes for sterilization
After autoclaving, the media is obtained,
Then pour the media into barquettes and wait for five hours for solidification

RESULTS

After obtaining the media, it was poured in the barquettes for solidification that takes one full day. Then you start planting your mother plantlets inside the media, for easy assessment of the feasibility of our media, I planted together the MS media and the new media with the same plantlet variety for comparison.

After planting, the barquettes contains 36 daughter plantlets, each barquettes have been labeled by their mother varieties, date, and responsible name of cutting for easy identification, and they have been transferred in the growth room. this room possess air conditioner and light that provide environment similar to the outside.

The first two days (figure1); there was no significant change for both banana media and MS media barquettes.

With the fourth day (figure2), there was formation of callus where root will rise and the appearance of the shoot from the node.

At the sixth day (figure3), the plantlets do have both shoot and root.

for the growth of in vitro plantlet, each must be followed daily so as to sustain the environment and both light and dark. the new media have shown to be having very green plantlet, as the figure 4 and 5 shows, after a month the plantlet are ready for replanting.

Below are images at different days after plantation in media.

The first two days after plantation

No slight change



Figure 1

- After The second two days (4th day)
- Appearing of the shoots from nodes and roots.



Figure 2

- Above the fourth day, plantlet grow in size and roots at high rate.



Figure 3

Remember that each day bring new thing to the growth and media become weak, the weakening of media leads to the oldness of the plantlets.

After a month depending on a certain varieties of plant most of them they are ready for producing the next generation.

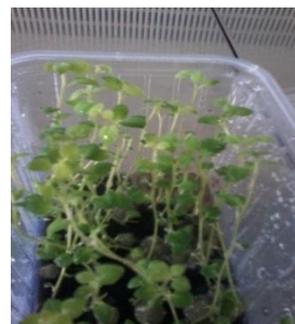


Figure 4



figure 5

CONCLUSIONS

As the results shows our culture media is able to support the life of in vitro. banana derivatives, pseudostem and leaves were all rejected as waste and little was used to feed cows. The high amount was rejected instead of exploring them, let us protect them and create a new way to dispose them. The study has made a study to different comparison, it started from testing between leaves and stem what region is more vulnerable to minerals and see what changes would results if both are mixed. The mixture showed much success than both leaves and stem. we even though the experiment succeeded but the pathway was not easy and the preparation of reagents was in a traditional way. So me and you are able to make it to the point that we no longer stuck on the use of expensive Murashige and skoog culture media only let us keep working and see how we can reach the final stage, again our agriculture is going to be improved not only for the seeds but also for the crops and harvest. this new media is too cheap comparing to the ones used because the main ingredients are the currently non explored resources.

RECOMMENDATIONS

If there is a way to progress the experiment, we highly demand to improve the way reagents were prepared. And to make sure that reagents to be used are well sterilized.

Again we used bolus complement which was recommended to be used for cattle, we demand that next study can focus on the substitution of this bolus.

ACKNOWLEDGEMENT

I thank the almighty God for his abundant blessings and protection during my study. My special thanks goes to the board of RAB (Rwanda Agriculture Board) who supported me during my work. My deep sense of gratitude is due to my supervisor Mr. NTIZO SENKESHA, research coordinator of potato program, for his constant constructive criticism and all possible help rendered for the successful completion of laboratory experiences and the whole research activity. My sincere thanks goes to prof. Beth KAPLIN for her meaningful contribution, advice and all support that encouraged me to work hard at all sector.

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