Original Research Article

An Alternative Low Cost Multiplication Technique: Natural Substances for Regenerating Plantlets from Mini tubers of Sweet Potato (*Ipomoea batatas, L.*)

ABSTRACT

Aims: This study was conducted specifically to develop a low cost rapid plantlet multiplication process easily affordable for farmers to enable them speedily generate plantlets for their farms from mini tubers of sweet potato variety, TIS 87/0087

Study design: The experimental design was a completely randomised design with three replications. Analysis of variance was used (P=.05) to test treatment effects in a completely randomised design and mean comparison was by LSD.

Place and Duration of Study: The study was carried out between February and April 2019 in the plant culture laboratory of the Department of Plant Science and Biotechnology, Rivers State University, South-South, Nigeria.

Methodology: Mini tubers of sweet potato (*Ipomoea batatas* L) were preconditioned by soaking in a 1:10 ratio of natural substance to water for 12 hours before planting in soil. The natural substances comprised coconut water, grapefruit juice and honey with water as control.

Results: Mini tubers primed with grapefruit juice and coconut water sprouted significantly earlier (P=0.05) (7days and 10days respectively) than the control (water) which sprouted after 19days. However, there was no significant difference in time to sprouting between mini tubers primed with honey (16days) and the control (19days). Mini tubers primed with grapefruit juice, coconut water and the control did not differ significantly (P=.05) in the number of initial plantlet sprouts but mini tubers primed with honey had significantly lower initial sprouts than others. On average, coconut water primed mini tubers produced significantly higher (P=.05) total number of regenerated plantlets compared to the other treatments and continued regeneration of plantlets longer than other treatments. Coconut water regenerated almost 3times the number of plantlets regenerated by mini tubers soaked in water. Mini tubers primed with honey stopped sprouting after 29days which was significantly earlier (P=.05) than those primed with grapefruit juice (38days), water (46days) and coconut water (51days).

Conclusion: This study found that mini tubers of sweet potato after a preconditioning treatment by soaking in dilute coconut water (1:10 coconut water:water ratio) for 12hrs before planting regenerated almost 3times the number of plantlets regenerated by mini tubers soaked in water for the same period.

Keywords: [Sweet potato,Mini tubers, Natural substances, Preconditioning, Regeneration}

1. INTRODUCTION

Sweet potato (*Ipomoea batatas* L) is a vegetatively propagated crop which is commonly propagated by farmers from the root tuber, or from the vines and by breeders from the seed. While developing countries account for over 95% of the world's sweet potato production [1,2], in Europe, the biggest producers are Portugal, Spain, Italy and Greece [3,2]. Although sweet potato is a source of food, animal feed and industrial raw material for production of sugar syrups, ethanol and flour, etc for confectionaries [4,5] one major constraint facing sweet potato growers worldwide is shortage of clean planting materials at the beginning of every planting season. It is cultivated by recycling planting materials from old fields; is locally sourced, and must be fresh to be viable and therefore is not sold in local markets or by traders like seed propagated crops. The problem is more severe for farmers in the drought prone and high disease pressure areas. For instance, it was reported [6] that farmers in Bukedea and Soroti districts in Uganda normally experience 3–4 months of dry weather between mid-November and March. During this dry period, vegetation of the sweet potato crop is completely desiccated, leading to difficulties in securing vines as planting material at the onset of the rains. About 58% of the farmers interviewed in Soroti claimed that their failure to plant was due to lack of planting material. In some sub-Saharan African regions, the dry season extends even more to 5-7 months later, especially in those with prolonged drought periods further compounding the problem [7]. Elsewhere, it was stated [8] that availability of quality planting material on a sustainable basis is a major challenge for the farming communities of Odisha state of east India. In order to overcome these challenges, farmers in many areas try to solve this problem by conserving planting materials near water sources, or in the home garden, or by storing roots which they sprout at the onset of rains, or do sequential planting immediately after onset of rains to get enough materials for field expansion [8,6]. Most farmers lose upwards of 4-6 weeks or more of the growing season at the beginning of the rains while they re-establish sufficient vine production for planting. They obtain initial limited planting material from residual plants, re-sprouting roots, or secondary growth of harvested fields, thereby limiting the sweet potato production areas [7]. The immediate obvious solution to the problem of limited planting material of course, is rapid multiplication either by tissue culture, sand hydroponics or aeroponics techniques [9,10]. However, the main disadvantages of these rapid multiplication techniques in developing economies are that they require special skills, extra manpower, sophisticated equipment, high capital outlay and high production costs [11,12] that are not readily available to farmers. Plant tissue culture is essentially rapid multiplication of tiny shoot tips/apical meristems, axillary buds, and sometimes of somatic embryos and cell clumps, etc in suspension cultures and bioreactors. What developing economies need is a technique to do the same at minimal cost. Besides, it has been stressed time and again that in the long-term, agriculture needs to be sustainable, use little or no crop-protection chemicals, have low energy inputs and yet maintain high yields, while producing high quality material, and also saving land resources [13]. Bearing these in mind therefore, an alternative cheaper, efficient, effective and simple rapid multiplication technique with low energy inputs designed to provide planting material for mass propagation of sweet potato other than by use of tissue culture technique has become imperative. This means that farmers must be able to apply the technique themselves while avoiding high technology and building of complex infrastructure especially during the release of improved varieties. Such a technique must not be too complicated, nor labour intensive and must be within a short duration in limited spaces and should not require farmers to learn multiple new tasks so it can be readily adopted by resource poor farmers. In fact, some farmers may choose to specialise solely in the production of planting materials in desired quantities timed to match peak demands and thus establish a new line of business. One of such techniques is the use of mini-tubers as has been done for yams through use of mini setts [14]. The other is by a preconditioning of the mini tubers in natural substances to act as growth primers for regenerating plantlets. Both techniques combined could provide a

substantial increase in the number of plantlets regenerated for planting and expand farmers' fields beyond the practices earlier described. To the best of my knowledge, there is no scientific literature available on the use of the following natural substances; grapefruit juice, honey and coconut water as growth primers in a preconditioning technique for rapid regeneration of sweet potato plantlets from mini tubers.

Grapefruit juice contains in addition to myo-inositol also known as inositol, carbohydrates, proteins, fat, and vitamins B1, B2, and B9, vitamin C, and vitamin P (bioflavonoid). It has also a plethora of minerals like iron, iodine, potassium, calcium, cobalt, magnesium, manganese, copper, sodium, phosphorus, fluoride and zinc [15]. Most of these are essential for plant cell and tissue growth [16]. Honey is composed of sugar (about 76% - fructose, glucose and sucrose), water (18%) and minerals: potassium, chlorine, sulphur, calcium, sodium, phosphorus, magnesium, silicon, iron, manganese and copper; proteins, acids and vitamins: vitamin C and some B complex vitamins - riboflavin, pantothenic acid, pyridoxine, biotin, nicotinic acid (niacin) (6%), and inositol [17,18,19,20,21]. The constituents of coconut water are water 94%, sugars such as glucose, fructose and sucrose around 5%, proteins around 0.02% and lipids only about 0.01%. It is rich in minerals such as potassium, calcium, magnesium and manganese, and low in sodium. Amino acids include glutamic acid, asparagine, proline, and glycine; and organic acids particularly malic acid [22,23,24,25,26,27]. In addition to inositol [28] coconut water contains auxin, various cytokinins, and gibberellins [29,30] which are all plant growth hormones that support cell division and promote rapid growth.

The main objective of this combined mini tuber and natural substance primers is to produce elite planting material irrespective of season, for the rapid regeneration of sweet potato plantlets as planting materials for farmers. The method would also be useful for introduction of new improved varieties to farmers' fields. The ease of application of the method at relatively low cost and the avoidance of any use of chemicals is an added advantage.

This study was therefore conducted specifically to develop a low cost rapid plantlet multiplication process easily affordable for farmers to enable them speedily generate plantlets for their farms from mini tubers of sweet potato.

2. MATERIAL AND METHODS / EXPERIMENTAL DETAILS / METHODOLOGY

This study was carried out in the plant culture laboratory of the Department of Plant Science and Biotechnology, Rivers State University, Port Harcourt, south-south, Nigeria.

Preparation of Natural Substances as Primers

The following natural substances were obtained: fresh coconut water, fresh grapefruit juice, fresh honey and water (control). These were prepared as primers by adding 50ml of each substance to 500ml of water in a 1:10 ratio of natural substance to water.

Preparation of propagules (mini tubers)

The tubers of purple skinned/white fleshed sweet potato (*Ipomoea batatas* L)variety TIS 87/0087 (TIS 87/0087-registered variety from the National Root Crop Research Institute, Umudike, Abia State, Nigeria) were washed under running water after which the tubers were cut into pieces (mini tubers) of approximately 7cmX7cm size or 25g weight.

Treatment Applications (Preconditioning Technique) and Experimental Design

Each mini tuber was soaked in the prepared natural substance primers for 12hours (soaking beyond 12hours caused the mini tubers to rot) before planting in soil in a plastic germination

tray with watering done as required for multiplication of plantlets. Treatments were the four natural substance primers described earlier with water as control in a completely randomised design with 3 replications. The regenerated plantlets were transplanted from the mini tubers to the field every 2weeks.

Data Collection and Statistical Analyses

(1) Number of days taken from planting to first sprouting of plantlets from mini tuber;
(2) Number of initial plantlets sprouted from mini tuber;
(3) Total number of plantlets regenerated from mini tuber
(4) Length of time the mini tuber continued to produce plantlets until the last plantlet;
(5) Physical appearance and quality of sprouted plantlets

The treatment effects were subjected to analysis of variance (ANOVA) using the GLM procedure of Statistical Analyses Software (SAS) version 9.1 [31] and any effects found to be significant were tested at a significance level of 5% while means were compared using LSD at P = 0.05.

3. RESULTS AND DISCUSSION

Effects of preconditioning on time taken to first sprouting of plantlets from mini tubers

The effect of the natural substance primers on the time taken to first sprouting in each treatment is shown in Figure 1. Grapefruit juice and coconut water treated mini tubers produced the first plantlets 7days and 10days after priming respectively while the control (water) sprouted after 19days. Thus grapefruit juice and coconut water treated mini tubers sprouted significantly earlier (*P=.05*) (almost 3times and 2times faster respectively) than the control. However, there was no significant difference in time to sprouting between mini tubers treated with honey (16days) and the control (19days). Coconut water, grape fruit juice and honey are natural sources of myo-inositol also known as inositol, a plant growth factor although grapefruit juice has a much higher content than coconut water and honey [28,32]. The high viscosity of honey may have made its low inositol content less effective. In addition, coconut water contains auxin, various cytokinins, and gibberellins [29,30] which are all plant growth hormones that support cell division and promote rapid growth. These growth promoters could account for the early sprouting by mini tubers treated with grapefruit juice and coconut water.

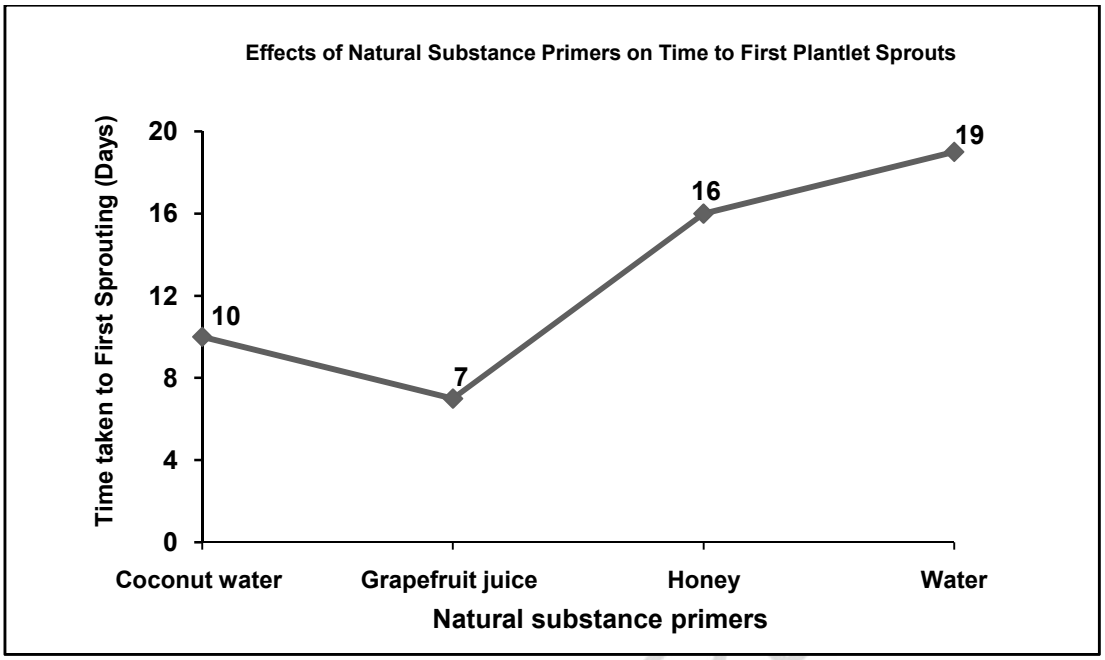

Fig. 1. Effects of natural substance primers on length of time from planting to first plantlet sprouts from mini tubers

Effects of natural substance primers on number of initial plantlet sprouts

In Figure 2, the numbers of initial plantlet sprouts are presented. There were no significant differences (*P=.05*) in the number of initial plantlet sprouts between grapefruit juice and coconut water treated mini tubers and the control. The initial plantlet sprouts from mini tubers treated with honey was significantly less than all other treatments. Honey being more viscous than the other natural substances [17] could have resulted in a slower effect on mini tubers than other liquid substances despite its inositol content compared to water.

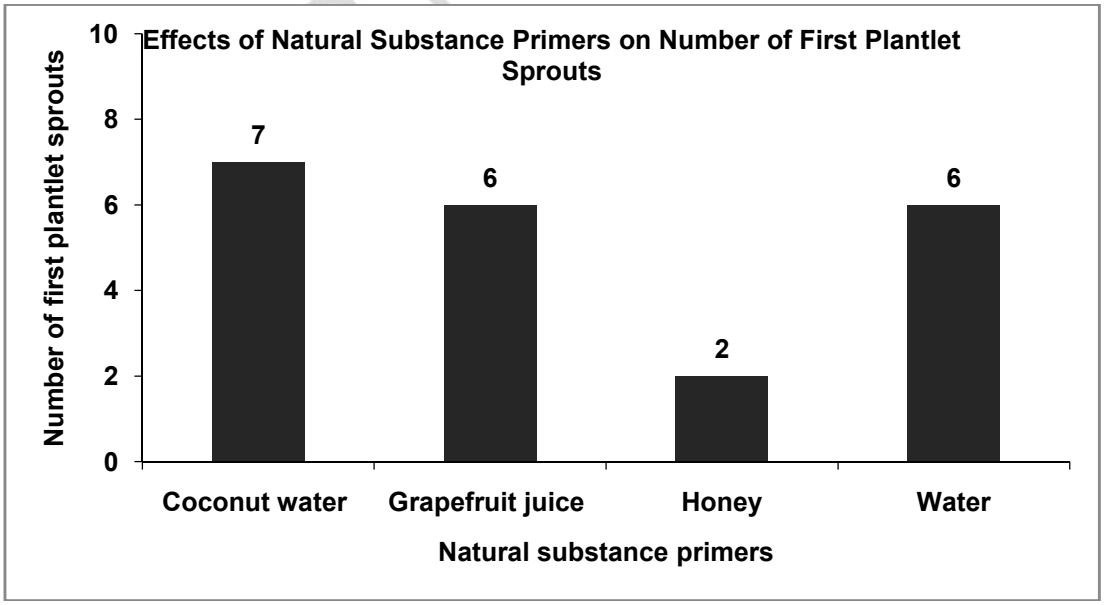

Fig. 2. The effects of natural substance primers on number of first plantlet sprout from mini tubers

Effects of natural substance primers on total number of plantlets regenerated

The total numbers of regenerated plantlets in all treatments are shown in Figure 3. Coconut water produced significantly higher (*P=.05*) total number of regenerated plantlets than all other treatments. The vitamins, minerals, and amino acids present in coconut water [22,23,24,25,26,27] and the auxin, various cytokinins, and gibberellins which are all plant growth hormones that support cell division and promote rapid growth [29,30] all working in synergy could explain the sustained regeneration of plantlets that almost quadrupled, tripled and more than doubled the number of plantlets produced by mini tubers primed in coconut water compared to the honey, water (control) and grapefruit juice respectively [33,34].

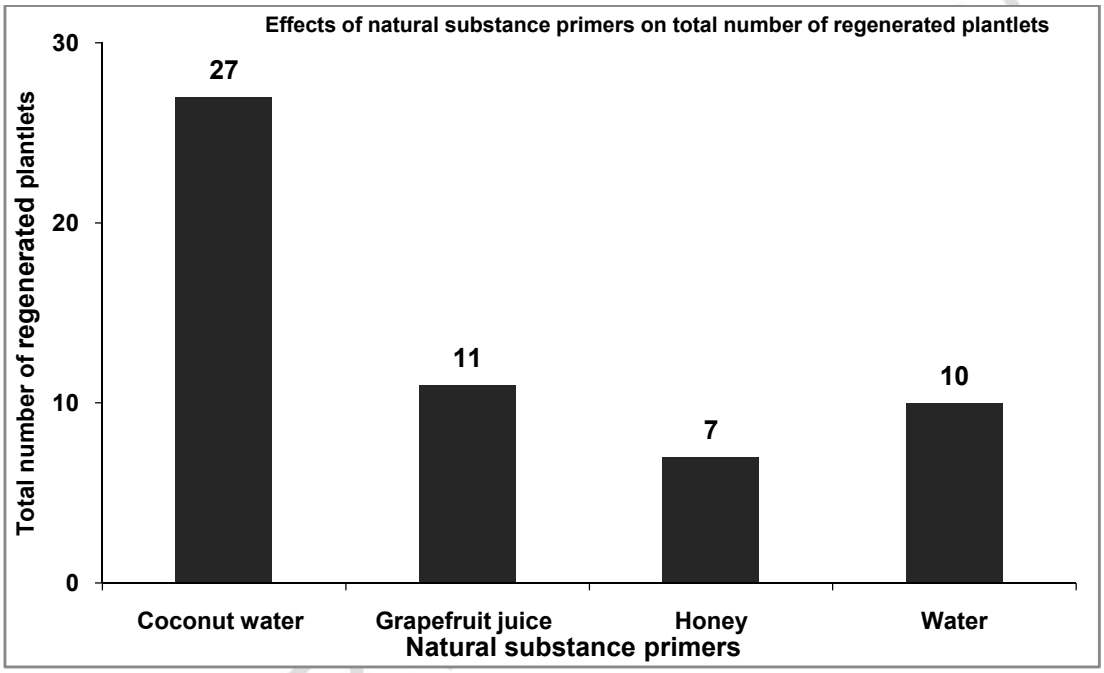

Fig. 3. Effects of natural substance primers on the total numbers of regenerated plantlets from mini tubers

Effects of natural substance primers on time taken to last sprouting of plantlets from mini tubers

In Figure 4, the number of days to last sprouting in each treatment is shown. Mini tubers treated with honey stopped sprouting significantly earlier (*P=.05*) than those primed with grapefruit juice (38days), water (46days) and coconut water (51days). Coconut water primed mini tubers continued regeneration of plantlets longer than other treatments. Perhaps the high content of vitamins, minerals, amino acids and plant growth hormones in coconut water could explain the prolonged healthy shoot regeneration over a longer period of time than the other substances.

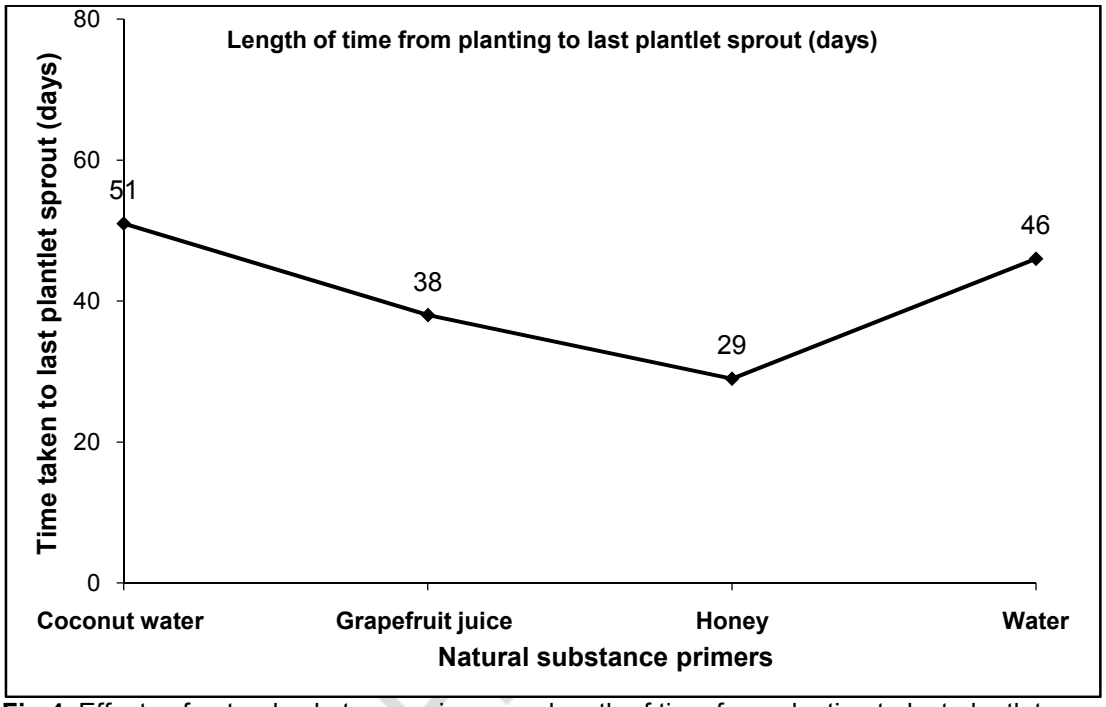

Fig 4: Effects of natural substance primers on length of time from planting to last plantlet sprout from mini tuber

Physical appearance and quality of sprouted plantlets

Plantlets from the mini tubers primed with coconut water, grapefruit juice and honey were normal and of good quality not requiring any hardening period and easily withstood transplanting to the field without any adverse effects as is often needed for tissue cultured plantlets. However, plantlets produced with water priming had small juvenile leaves, with pale yellow colour and reduced photosynthetic capacity. This meant that they took much longer to recover when transplanted to the field than plantlets of other primers.

4. CONCLUSION

This study found that mini tubers of sweet potato after a preconditioning treatment by soaking in dilute coconut water (1:10 coconut water :water ratio) for 12hrs before planting regenerated almost 3times the number of plantlets regenerated by mini tubers soaked in water for the same period.

CONFLICT OF INTEREST

Author has declared that no conflict of interests exists

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