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RAPID MULTIPLICATION OF TANNIA (*Xanthosoma sagittifolium* Schott): AN ADVANCED TECHNIQUE FOR DEVELOPING COUNTRIES

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ABSTRACT

A rapid multiplication technique of tannia (tanier, cocoyam, *Xanthosoma sagittifolium*) is suggested for developing countries, in particular, the Windward Islands of Dominica, St. Lucia, St. Vincent and Grenada, where *Pythium* root rot or tannia rapid yellowing disease (TRYD) caused by *Pythium myriotylum*, has been seriously affecting tannia production. The rapid clonal multiplication of disease-free plant material is done through a two-phase production system - a combination of *in vitro* propagation and subsequent *in vivo* mini-setting. The Murashige and Skoog (1962) basal medium was used for meristem initiation and then amended by omitting benzylamino-purine (BAP), but including 0.1 mg/l 1-naphthaleneacetic acid (NAA) for prolific calli formation and plant regeneration. The technique described can improve tannia germplasm stock and at the same time provides a system that is practical and economically affordable to developing countries.

INTRODUCTION

Tannia [tanier, cocoyam, *Xanthosoma sagittifolium* (L.) Schott] is one of the crops, along with banana, coconut, dasheen (taro), of great importance to the economy of Dominica, both as a food staple and as a valuable foreign exchange earner (Central Statistics Unit, 1987). The crop is not prone to the ravages of hurricanes and fits well into the small-holding farming system in Dominica and the rest of the Windward Islands (St. Lucia, St. Vincent and Grenada).

A decline in tannia production in Dominica was observed in the 1970s. Estimates showed a significant reduction in production by over 65% (CARDI, 1984), which was partly attributed to a fungal disease - Pythium root rot caused by *P. myriotylum*, and more commonly known as "burning disease" or "tannia rapid yellowing disease" (TRYD).

As a result of research done by CARDI during 1982-86, a technological package (tech-pack) was developed to control the fungal disease not only in Dominica, but also in the rest of the Windward Island group (CARDI, 1986). A very important element of the control strategy was the use of clean, disease-free

material. However, it was soon realized that the availability of disease-free, healthy plants with high vigor was a major constraint to materialize the production potential of tannia in the Windward sub-grouping.

Tannia, like all edible aroids in the tropics and sub-tropics, is grown by vegetative propagation, which lends itself to the easy spread of several pathogens such as viruses, bacteria and fungi (Zettler et al., 1970).

Plant regeneration from tissue and cell cultures is important in achieving rapid clonal multiplication, recovery of pathogen-free plants, preservation of valuable germplasm and induction of chromosomal and genetic variation (Gupta, 1985). Tissue culture is not only useful as a method of eliminating pathogens from aroid planting stock but has also proven an invaluable tool for rapid clonal propagation. The developing countries must be able to exploit this technology to enhance the availability of improved germplasm, but this must be at an economical cost. Several workers have developed techniques for micropropagation of tannia (cocoyam) (X. sagittifolium) (Hartman, 1974; Gupta, 1985; Liu et al, 1988).

This paper describes a technique and suggests an approach using in vitro culture of apical meristems of tannia in combination with in vivo propagation to provide sufficiently large amounts of healthy planting material of suitably adaptable and high yielding cultivars. The ultimate objective of this two-phase approach is to enable Dominica and the other major tannia growing islands (St. Lucia, St. Vincent and Grenada) to increase production over a relatively short period to satisfy not only the domestic food needs but also to supply the markets in Europe, Canada and elsewhere and earn valuable foreign exchange.

MATERIALS AND METHODS

Apical meristems were obtained for clonal propagation from corms of 9-month-old tannia mother plants (Rabess cv. and St. Lucia cv.) known to be disease-free and with a record of high yields. Shoot tips from all plants were trimmed to <1.5 cm and rinsed in flowing tap water, submerged for 10 minutes in 0.52% sodium hypochlorite with two drops of "Tween 20", trimmed further and submerged again for 5 minutes in 0.26% sodium hypochlorite with two drops of "Tween 20". The outer layers of the sterilized shoot tips were removed under a dissecting microscope in a laminar flow chamber. Each shoot was then trimmed until only the apical dome and two leaf primordia 0.2 to 0.4 mm long remained.

Each excised meristem with the two leaf primordia was then transferred aseptically with the cut surface downwards to the surface of 15 ml of solidified culture medium in a 50 ml screw-cap tube.

Solid agar culture medium, MS (Murashige and Skoog, 1962), was used for initiation of apical meristems. Six to eight weeks after initiation those calli cultures which showed slow and little differentiation were transferred to a second solid medium in Magenta GA7 vessels with 50 ml of agar in order to encourage greater callus formation and strong root development. This second medium consisted of MS basal medium but without the cytokinin, benzylamino purine (BAP). The auxin, naphthaleneacetic acid (NAA) 0.1 mg/l was retained in the culture medium, which was adjusted to pH 5.8.

All cultures were maintained at 22-24^{o}C and given 12 hours of daily light at 5000-8000 lux from fluorescent bulbs.

Plantlets which had differentiated from meristems in the standard MS medium through direct organogenesis were excised from undifferentiated calli and transplanted individually to sterilized potting medium consisting of sand:soil:perlite (1:1:1 ratio) in 7.5 cm plastic cups. Covers were provided with same sized cups and plantlets weaned for 7-8 days. These were subsequently hardened in a plastic cloche for 3-5 weeks and afterwards were transferred to the field nursery.

Plantlet regeneration from cultures on the second and amended culture medium with no BAP were eventually weaned and hardened in a similar manner as described for those few plantlets regenerated on the basal medium (Murashige and Skoog, 1962).

All regenerated plants from in vitro propagation at the 3-4 leaf stage were transplanted to a certified nursery and after 5-9 months they were harvested to provide certified disease-free corms. These corms then provided the tannia "setts" for the mini-setting technique. This technique involves the selection of certified disease-free 5-9-month-old plants in an active vegetative growth phase. The corms were washed, all their roots cut off, and then dipped in a fungicidal mixture of 0.29% metalaxyl and 0.025% benomyl to disinfect them. The corms were then cut longitudinally into two segments and the cut surfaces placed downwards under shade provided by dried leaves; banana and coconut branches provided an effective ground sheet and cover. As the need arose, light watering was done to avoid drying out of the corm pieces.

After 2-3 weeks the buds on the corms sprouted, and the sprouting corms were cut into "bits" each about 100-250 g in weight and used as planting material. Depending on the size of a corm, about 10-12 plants can be derived in less than a month after mini-setting.

RESULTS AND DISCUSSION

Differentiation of meristem cultures on the standard Murashige and Skoog (1962) medium was slight with poor plantlet development and only slight calli formation after 13 weeks. Similar results with the MS medium were reported by McDonald and Royer in 1989. Greening of meristem cultures became apparent within 1 week after initiation. Shoot induction was observed after 6 weeks of culture maintenance, but roots were few in number in all cultures in the standard MS medium.

In contrast, differentiation of meristems was very much enhanced on the second solid agar medium in Magenta vessels amended by omitting BAP but with 0.1 mg/l NAA included, after initiation of meristems on the MS basal medium. Calli formation was prolific and over 80% differentiated with the regeneration of plantlets. Increased plantlet regeneration from these cultures was quadrupled. All plantlets regenerated from calli and axillary shoots had strong roots.

The two-step system used in the rapid multiplication of a herbaceous monocotyledonous crop such as tannia (cocoyam, X. sagittifolium) was found to be efficient and adaptable. Other workers (Gupta, 1985; Hartman, 1974; Liu et al, 1988) reported improvement to the standard MS medium for rapid multiplication of tannia (cocoyam). Amendments were made to the basic formula by supplementing various combinations of growth hormones: kinetin, indoleacetic acid, naphthalene acetic acid, etc. However, the present state of the art of in vitro commercial propagation of aroids allows for production only by organogenesis and proliferation of axillary shoots; hence the production is labor-intensive and expensive (Zettler and Hartman, 1987). In addition, edible aroids such as tannia are known to give a lower cash return than their ornamental counterparts. It therefore becomes necessary for developing countries to be innovative and to exploit in vitro propagation (micropropagation) in such a manner that the technology becomes affordable and practical.

Here then it becomes pertinent to combine in vitro propagation with in vivo propagation as far as possible so that the entire production system and a crop germplasm improvement program becomes realistic in small developing states such as Dominica.

The suggested technique herein described utilizes both technologies (Plate 1) for the rapid multiplication or mass production of disease-free plants. Phase I consists of in vitro culture and Phase II utilizes the production technique of "mini-sett". A similar method of rapid multiplication of X. sagittifolium (cocoyam) has proven successful in Cameroon (Acquah and Hbehnchow, 1989).

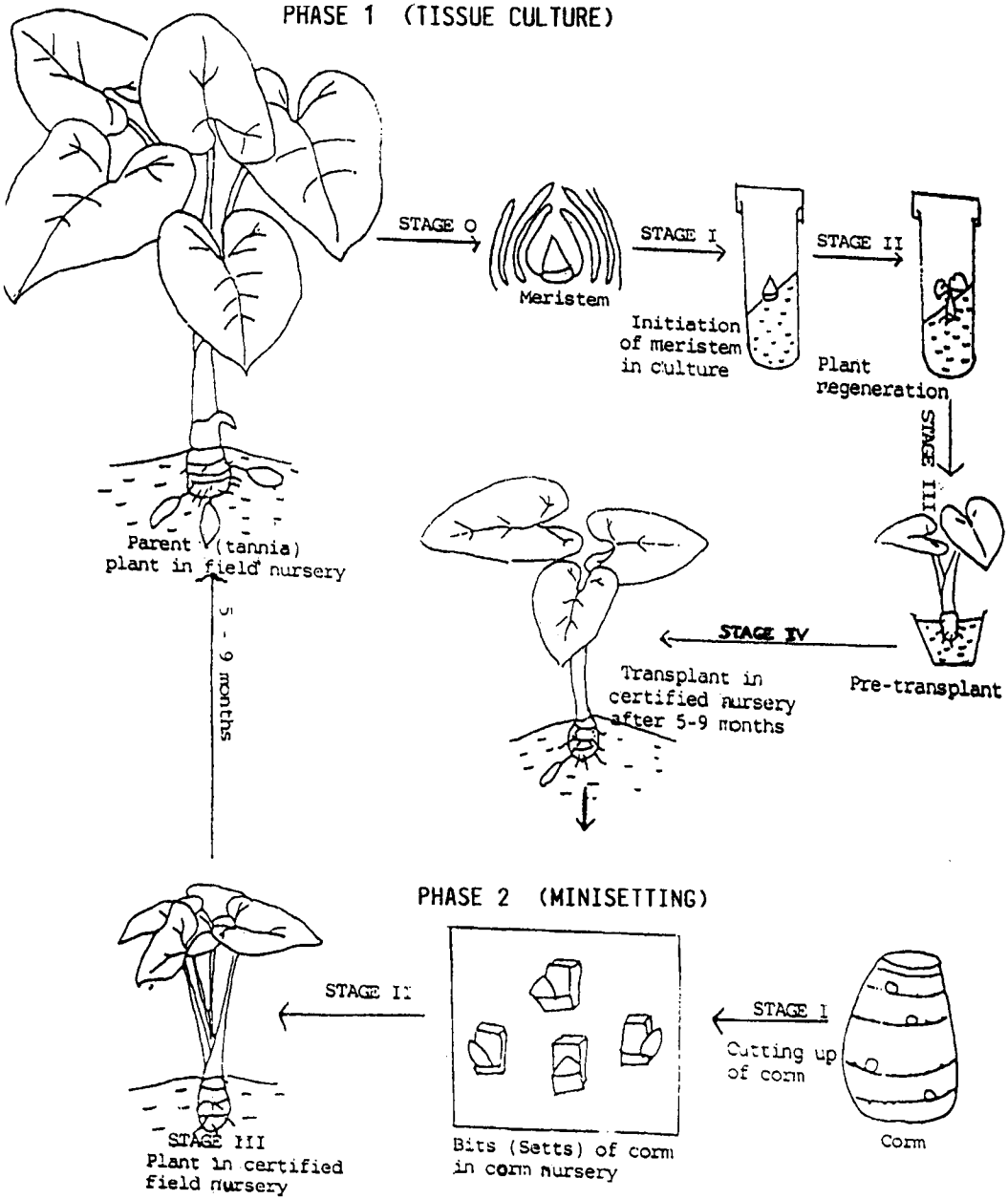

Plate 1. A schematic representation of a module for rapid multiplication of tannia using tissue culture (phase 1) and mini-setting (Phase 2).

The multiplier effect in this combined technique for rapid multiplication of tannia material can yield hundred-fold more plant material than the traditional method, and over a much shorter period.

It has been stated that the best way to improve stocks of plant material in developing countries is via a combination of cultivation in vitro and the subsequent propagation in vivo of the disease-free juvenile material (Jordens-Rottger, 1987). The approach and technique utilizes two levels of technology for rapid (mass) production of disease-free, vigorous material, in dire shortage in small developing economies. The approach also intends to bridge the gap between high technology and intermediate and appropriate technology in developing countries; it has been said by Hardy (1985) that the impact of biotechnology in the field of agriculture will not be felt in developing countries until the year 2008.

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