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***Growth Medium and Environmental Studies
of Sweet Potato Meristem Culture***

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Abstract

The ability of three New Zealand local sweet potato (*Ipomoea batatas* L.) cultivars 'Toka Toka Gold', 'Beauregard', and 'Owairaka Red' to form plantlets *in vitro* was investigated. Meristematic tips (0.2–0.4 mm) of apical shoots from vines of the three cultivars, and from tubers of 'Owairaka Red' were cultured in modified Murashige and Skoog (1962) medium (MS medium) containing plant growth regulator (s).

Cultivars and organs of explants differed in response to exogenous levels of plant growth regulator(s) and in the rate of proliferation. Optimal regeneration occurred in liquid MS medium supplemented with BA 0.1 mg/l for 'Toka Toka Gold' and 'Owairaka Red' (from vines), and with BA 0.5 + IBA 0.1 mg/l for 'Beauregard'. For 'Owairaka Red' (from tubers), MS liquid medium with BA 0.3 mg/l, and MS liquid medium with GA₃ 20 mg/l (plus other organic compounds) proliferated shoots and plantlets.

Continuous lighting inhibited the proliferation of plantlets in all three cultivars. Regeneration was strongly affected by the age of the shoots from which the explants were excised and the season when cultures were begun. Successful culture was obtained by culturing explants from young shoots in the Spring.

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Chapter One: Brief Introduction to Sweet Potato

1.1 Origin and economic importance

Sweet potato (*Ipomoea batatas* L.), native to tropical America, is one of the world's most economically important crop plants. According to FAO (1995), sweet potato production in 1995 was ranked 8th among crops grown for food with an estimated 122 million metric tons produced throughout the world. China accounts for 84% of the world sweet potato production. The area under sweet potato cultivation throughout the world was estimated at 9234 thousand hectare in 1995. Sweet potatoes are grown in nearly all parts of the tropical and subtropical world, and in warmer temperate regions.

Sweet potato is of particular importance as a food crop throughout subtropical and tropical regions. It is the most important carbohydrate sources for many millions of people, particular those in the developing nations. It is not only an efficient producer of calories but also rich in many nutrients, especially vitamins and minerals (Tsou and Hong, 1992). It is becoming a popular food in the modern diet because of its high dietary fibre content.

Agricultural statistics for 1993 showed that sweet potato be New Zealand's 9th largest crops in terms of production area with 905 hectares planted (Anon.1995). Sweet potato was the most important food crop cultivated by the pre-European Maori (Wood, 1983). In New Zealand, 'Owairaka Red', with a deep red skin and a deep yellow flesh, is the most important cultivar due to market preference for red cultivars and it's high yield. The other main cultivars are 'Toka Toka Gold', 'Caromex', 'Jewel', and 'Beauregard'.

1.2 Growth requirements and vegetative propagation

Growth of sweet potato is best at or above 24°C; when temperatures fall below 10 °C growth is severely retarded (Woolfe, 1992; Onwueme and Charles, 1994). The crop is damaged by frost and this restricts its cultivation in temperate regions to areas with a minimum frost-free period of 4–6 months, and with relatively high temperatures during this period. Sweet potato grows best where light intensity is relatively high, but at the same time both flowering and rooting are promoted by short day lengths. Optimum rainfall is 740–1000 mm per annum, with approximately 500 mm falling during the growing season. A soil pH of 5.5–6.6 is preferred.

1.3 The need for *in vitro* culture of sweet potato

One of the most important limitations to good sweet potato production is the presence of virus diseases which reduce yields by up to 78% (Hahn, 1979). Viruses transmitted by aphids, white flies (Schaefer and Terry, 1976; Beetham and Mason, 1992), and unidentified vectors (Beetham and Mason, 1992) severely affect this crop. Sweet potato feathery mottle virus has been found in all sweet potato growing countries (Clark and Moyer, 1988; Beetham and Mason, 1992). This virus appears to be fully latent in all genotypes of sweet potato. New Zealand varieties, such as 'Owairaka Red' and 'Gisborne Red', have also been infected by viruses (Over de Linden and Elliott, 1971).

Fungal diseases such as black rot (*Ceratoystis fimbriata*), scurf (*Monilochaetes infuscans*), and pink rot (*Sclerotinia sclerotiorum*) are the most significant diseases of sweet potato in New Zealand. These cause the tubers to shrivel and decay both during growth and in storage (Wood, 1983; Stewart, 1989). Presence of scurf significantly reduces product presentation and saleability with potential returns dropping from around \$2.50 per kg to \$1 or less (Tate, 1996). Each of these diseases can be carried over in tubers from one generation to another.

In tropical or subtropical regions, sweet potato is propagated commercially either from vine cuttings or storage tubers. In New Zealand, sweet potato is propagated from tubers (Wood, 1983) due to its short growing season. Unfortunately, vegetative propagation perpetuates viruses and fungal diseases in the progeny plants (Clark and Moyer, 1988). Furthermore, vine cuttings and storage roots are difficult to transport because of their fragility, tendency to spoil and large volume.

In the absence of effective therapeutic chemicals capable of eradicating diseases from infected plants, tissue culture techniques (especially meristem culture) have been employed to eliminate systemic diseases in vegetatively reproduced crops, such as sweet potato (Quak, 1977; Mellor and Stace-Smith, 1977; Beetham and Mason, 1992). Virus-free propagation material provides growers with a high-health crop, superior quality and potentially higher yield (Over de Linden and Elliott, 1971; Wang and Hu, 1980). Virus-free planting material can increase storage root fresh-weight yield of sweet potato by 250% (Beetham and Mason, 1992). Although virus-free material is quickly re-infected by viruses, over two generation yields are still significantly higher.

In addition, to provide a method for development, maintenance, and distribution of specific disease-free clones, micropropagation offers many advantages over conventional methods for clonal propagation independent of climatic conditions and with conservation of space and time. It also allows for germplasm conservation and genetic manipulations.

Meristem (or shoot-tip) culture, callus culture, and somatic embryogenesis culture of sweet potato have been successful (Henderson et al., 1984; Chee et al., 1992). Only meristem (or shoot-tip) culture are phenotypically true to type because of the absence of a distinct or prolonged callus stage, which increases the likelihood of genetic changes. Meristem culture of sweet potatoes has been studied previously (Alconero et al., 1975; Frison and Ng, 1982; Kuo et al., 1985; Marco and Walkey, 1992; Zamora and Gruezo, 1993). However, different cultivars differed in their response to the plant growth regulators. Meristem culture of New Zealand sweet

potato cultivars has not been previously reported in the literature. Although shoot-tip culture of New Zealand sweet potato cultivar 'Owairaka Red' has been studied (Elliott, 1969), the the size of the initiation explants used in the studied were too large to achive disease-free clones and germplasm conservation. And its regeneration rate was limited.

1.4 Objectives of this study

The objectives of this study were to investigate the growth medium and environmental factors for meristem culture of New Zealand sweet potato cultivars 'Toka Toka Gold', 'Beauregard', and 'Owairaka Red'.