Behaviour of MLO Evoking Potato Witches' Broom in Callus Tissue Culture of Solanum laciniatum AIT. and Nicotiana tabacum L. cv. Samsun

EVA PETRŮ* and MARIE ULRYCHOVÁ**

Institute of Experimental Botany, Czechoslovak Academy of Sciences, Praha

Abstract. The growth of callus tissue cultures and the infectivity of twenty five Solanum laciniatum AIT. plants and of sixteen Nicotiana tabacum L. cv. Samsun plants were investigated. The plants were obtained from callus tissue cultures derived from stem pieces of the respective plants infected with a mycoplasma-like organism (MLO) evoking potato witches' broom. The tissues were cultivated on synthetic nutrient medium with kinetin and IAA. All de novo obtained S. laciniatum plants were healthy. On the contrary twelve of the sixteen reconstituted tobacco plants showed MLO presence.

Summarizing these and previous results, the authors suppose that the most important factor influencing MLO persistence in callus tissues cultivated on the applied nutrient medium may be the callus growth rate and the organogenesis set. Both these conditions are determined by the metabolism of the investigated plant species.

JACOLI and RONALD (1974) and JACOLI (1974) investigated the presence of a mycoplasma-like organism (MLO) in calus tissues derived from carrot root and aster stem. These authors demonstrated the presence of MLO in their electron microscopic studies but they failed to transmit aster yellows agent from callus tissue culture by leafhoppers. They did not investigate the fate of MLO in tissue cultures because they found them especially in primary explants and in subsequent transfers MLO gradually disappeared.

From the growth and infectivity point of view, callus tissue cultures derived from tomato and Nicotiana glauca Gram plants infected with potato witches' broom were investigated by PETRŮ et al. (1971, 1972). Cultivation conditions for MLO persistence in callus tissue cultures of N. glauca are given by PETRŮ and ULRYCHOVÁ (1975). MLO persists in callus tissues cultivated in vitro on a synthetic medium with kinetin and IAA as growth regulators. This nutrient medium is suitable for organogenesis associated with the formation of sieve tubes where these microorganisms are mostly localized. However the nutrient medium alone does not determine MLO persistence in callus tissue cultures and the factor of primary importance may be the host plant. While the tissues derived from N. glauca stem grow on the above mentioned medium very well and the organogenesis appears

Received March 6, 1978; accepted April 11, 1978.

Addresses: * Ul. 1. listopadu 1050, 140 00 Praha 4, Czechoslovakia. ** Na Karlovce 1, 160 00 Praha 6, Czechoslovakia.
relatively soon (Petrů and Ulrychová 1975) the growth of callus tissue cultures derived from Solanum nigrum L. proceeds very slowly and the formation of organs takes place on the same nutrient medium only after more than several months of cultivation (Petrů and Ulrychová 1977).

The trials to evoke organogenesis on the same nutrient medium in callus tissues derived from stems of tomatoes infected with potato witches’ broom were negative in both healthy and infected tissues.

Theendeavour to generalize these partial results of our studies led us to new experiments with additional Solanaceous plants. We chose Solanum laciniatum Ait. and Nicotiana tabacum L. cv. Samsun. S. laciniatum is economically a very important plant for its high content of the steroidal alkaloid solasodine serving as a precursor for the synthesis of steroid drugs and N. tabacum is very often used as experimental material in plant tissue cultures. The reaction on potato witches’ broom infection of the first host plant has been described by Ulrychová and Jokex (1977) and of the second one by Valenta (1958).

Material and Methods

Callus tissue cultures were derived from stems of S. laciniatum Ait. and N. tabacum L. cv. Samsun plants both healthy and infected with potato witches’ broom. The infectivity of S. laciniatum plants was verified by back transmission (by grafting) on tomatoes reacting very sensitively to this disease; S. laciniatum plants are mostly a symptomless carrier of this disease.

The nutrient medium, the technique of callus tissue establishment and of infectivity assays are described in papers of Petrů and Ulrychová (1975) and Ulrychová and Petrů (1975).

Results and Discussion

Callus tissue developed on stem pieces of S. laciniatum cultivated in vitro on the nutrient medium used is compact and there are no differences in growth rate and appearance between the tissues derived from healthy and diseased plants. Organogenesis appears during two to four months. The stems are formed relatively rarely (Fig. 1) and their growth is very slow. On the contrary, organogenesis is visible in callus tissue derived from N. tabacum cv. Samsun in some cases during a fortnight, it is very intense and proceeds very quickly (Fig. 2). Callus tissue originating from healthy and MLO infected plants behave identically.

Reconstituted plants of S. laciniatum were obtained after six months on average, those of N. tabacum cv. Samsun even after two months of cultivation. Rooted plants were further cultivated after transmission into soil in a greenhouse and repeatedly assayed for the presence of MLO by grafting onto tomatoes. Twenty-five reconstituted plants of S. laciniatum were assayed in two or three repetitions. These plants were derived from different stem parts and from cultures established in various seasons. We failed to demonstrate the presence of MLO in all 25 examined S. laciniatum plants derived from diseased individuals. Sixteen reconstituted N. tabacum cv. Samsun plants also obtained from various explants were assayed similarly. Twelve plants demonstrated the presence of MLO in repeated assays, the