

Virus-elimination of potato breeding lines

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Introduction

The incidence of virus infection is an ever increasing problem annually, resulting in the loss of thousands of rands in income to seed growers. The importance of starting with virus-free material cannot be stressed enough. Potatoes are vegetatively propagated, and if a plant is virus-infected, the progeny from such an infected plant will also be infected. (See Fig 1.).

Virus-elimination program

The ARC-Vegetable and Ornamental Plant Institute (ARC-VOPI) virus-elimination program on potato breeding lines has been linked to the potato breeding program for many years now. The team working on this virus-elimination program comprises Ms Sanette Thiar as the project manager and Ms Bela Vcelar as the technical scientist.

Unfortunately in the case of plant viruses, one cannot just spray a chemical and the disease will be eliminated. The process of virus-elimination is a rather time-consuming and expensive operation.

In vegetatively propagated crops, a number of methods can be used to eliminate viruses from explant material. These methods can be used individually or in combination.

The methods include; a) heat therapy and b) chemotherapy. It is commonly accepted that virus particles do not occur in the actively dividing region of the meristem tip. This meristematic region has yet to differentiate into xylem and phloem, which forms the vascular system of the plant. The usual mode of transport is through the phloem, but cell to cell movement cannot be excluded.

By dissecting out the meristem and some of the surrounding tissues, the likelihood

of escaping the virus is good. Dissecting of the meristematic tissue is done with the aid of a dissecting microscope. (See Fig. 2.).

Heat Therapy

During the virus-elimination process of potato material, the infected material is multiplied and then exposed to gradually increasing temperatures over a period of time.

This is called heat therapy. The temperature to which the plants are subjected starts at 25°C, which is then gradually increased over a period of approximately 6 weeks up to a maximum of 37°C, under a 16 hour day, 8 hour night cycle. Meristems are dissected out before the plant collapses.

Chemotherapy

The dissected meristem tip is approximately 0.1 to 0.5mm in diameter, and consists of the meristematic dome and



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Figure 1: Plant infected with *Potato Leaf Roll Virus (PLRV)*. The arrow indicates the typical rolling of the leaves (Photo: Sanette Thiar).

one or two leaf primordia with some stem tissue. These meristems are then placed on Murashige and Skoog basal medium containing the anti-viral compound, Virazol®. Virazol®, with the active ingredient Ribavirin, is supplemented into the growing medium onto which the meristems are cultured.

These tiny growing points (meristem tips) are therefore exposed to a chronic treatment with the anti-viral compound. This is the chemotherapy part of the virus-elimination process. The meristems that survive the heat- and chemotherapy will grow into mature plants, which can be transferred to a glasshouse for the production of tubers.

It is possible that the virus concentration in these plants is reduced to a level that is too low to be detected by the indexing system used. It is therefore important to maintain replicates of the plants in the *in vitro* gene bank, while the progeny is tested for virus up to a third or fourth time.

Once the material tests negative after three tuber phases, it can be declared "virus-free". All material that tests positive during the process is destroyed.

From the virus-elimination program, 11 breeding lines have been released as virus-free in 2006. Once these breeding lines are released as cultivars, virus-free



Fig. 2. Bela Vcelar busy dissecting out meristems under the dissecting microscope. (Photo: Sanette Thiar).

planting material becomes available.

Although starting with virus-free material for the production of quality seed is extremely important, it is just as important to practice good crop management in order to keep the level of re-infection as low as possible.