

RECENT IMPROVEMENTS IN PROCEDURES TO IMPROVE SECURITY AND EFFICIENCY OF SUGARCANE QUARANTINE IN SOUTH AFRICA

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The Plant Breeding Department at the South African Sugar Association Experiment Station imports sugarcane clones from other countries primarily for crossing purposes to introduce new genes and gene combinations to their breeding programme. Sugarcane is primarily multiplied by planting setts, and many important diseases caused by exotic viruses, bacteria and phytoplasmas can be transmitted in this way. These diseases may occur symptomlessly or be latent, and may present a hazard to production of sugarcane in South Africa.

To ensure that imported sugarcane does not carry disease it is placed under quarantine where it is inspected and tested for the presence of pathogens. Until recently security was mainly based on the recognition of disease symptoms in plants grown in the quarantine glasshouse. However, biotechnological and new serological methods have recently been introduced to improve the sensitivity and efficiency of diagnostic tests in quarantine; for example, the polymerase chain reaction (PCR) is used for the detection of sugarcane mosaic virus, the Yellow Leaf Syndrome (YLS) phytoplasma, leaf scald disease, ratoon stunting disease (RSD), the streak virus and Fiji disease. The tissue blot immunoassay is used for the detection of the YLS luteovirus. A set of universal primers was developed to make it possible to detect any unknown bacterial pathogens that might be present in imported plants. In addition shoot tip cultures are now being used to eliminate the YLS virus from important breeding material.

Another improvement in the quarantine glasshouse is that local varieties are now pre-quarantined to provide disease-free cuttings that are exported to other sugar-producing countries of the world. A number of organizations that include the South African Sugar Association Experiment Station (SASEX) and the Australian Bureau of Sugarcane Experiment Stations, have bilateral agreements regarding germplasm exchange. The main feature of these agreements is that the exporter puts material through the quarantine process, which includes diagnostic tests, before dispatch. The main benefit of this is to improve quarantine security. Four new cubicles have been added to the quarantine glasshouse facility at SASEX to provide room for local varieties to go through a pre-quarantine process before export. This procedure ensures that the planting material that is being exported to other countries is of a high phytosanitary standard. The quarantine facility situated at SASEX also provides a user pays quarantine service for other African countries (for example Swaziland) should they want to import varieties from a country such as Mauritius.

Quarantine procedures

First growing cycle

Varieties to be quarantined are sent directly by the source country. On arrival the setts are hot water treated at 50°C for 30 minutes and dipped in a fungicide (difenoconazole and carbendazim -Eria, 0.5 ml/l) and an insecticide (permithrin, 1 ml/l) before being planted in sterilised soil in 25 litre pots. Up to four setts of one variety are planted in each pot and up to 12 varieties can be grown in one compartment. Each consignment is grown in a separate compartment. The plants are maintained in the glasshouse and sprayed with insecticide (mercaptotion) several times during their growth cycle. The cane is inspected regularly and several biotechnological and new serological tests are conducted as well as isolation on selective media. Varieties found to be infected with a hazardous disease are destroyed.

Second growth cycle

The sugarcane stalks are cut after a nine to twelve month growth cycle in quarantine. The plant crop stool, roots and any associated living material in the soil are destroyed by autoclaving. The stalks are cut into setts which are then soaked in cold water for two days. The setts are then heat treated for 3 hours at 50°C, dipped in fungicide and insecticide and planted in sterilized soil. In recent years the potting medium used in the quarantine glasshouse has been changed from a 1:1 mixture of sand and filtercake, to composted pine bark. The plants are inspected frequently during the second growth cycle that lasts for a further nine to twelve months.

Post quarantine cycle

At the end of the second cycle, single-budded setts are hot water treated for 10 minutes at 52°C with the addition of Eria at a concentration of 0.5 ml/l. The setts are then planted into transplant trays. The transplants of the imported varieties are planted outside the glasshouse in a designated area for disease inspection and bulking at the Experiment Station. The varieties are normally released from quarantine for planting either in early or late summer (October or March) and are cultivated for two more growth cycles. It is now normal practice to use transplants instead of setts, as this gives the variety a better start and if gapping up is necessary, there are always extra transplants at hand. Each variety is planted into a five-metre row using 21 transplants spaced at quarter metre intervals.

Periodic inspections are carried out by the Pathology Departments' disease inspection team. The local diseases found in the imported varieties growing in the post quarantine bulking plots over the last two years (March 1999 to 2001) are given in Table 1.

Discussion and Conclusions

Approximately 60 varieties are imported every two years from countries such as Australia, Mauritius, United States of America and Zimbabwe. Most quarantine cycles occur without any pathogens being detected. However, RSD, the YLS phytoplasma and the YLS luteovirus have been detected in plants in the 1999/2000 growing cycle. The YLS virus was diagnosed in one plant originating from Zimbabwe and in 13 plants from Florida. The phytoplasma was detected in one variety from Mauritius, while RSD was detected in 12 varieties from Zimbabwe. Plants infected with pathogens are destroyed and removed from the quarantine glasshouse. However, RSD infected plants can be cured by treating the cane setts with the 3-hour hot water treatment after the first growth cycle.

In the 1999/2000 growing season several varieties in the post-quarantine area became infected with local diseases through

lack of resistance. The main diseases observed were pokkah boeng, mosaic and YLS. This situation illustrates the importance of quarantine. If cane varieties are imported illegally they may not only be infected with diseases common in the country of origin, but they may also be susceptible to local diseases. The outbreaks of a foreign disease (e.g. Fiji from Australia and grassy shoot from India and Sri Lanka) in our local susceptible varieties would be extremely serious. Likewise, growing illegal infected varieties susceptible to our local diseases could also have serious consequences if the variety became widely grown.

The presence of the YLS virus in both imported and local varieties in the quarantine glasshouse was a major concern because the diseased material had to be destroyed. To prevent the loss of valuable breeding material, a tissue culture programme was started to produce healthy plants. *In vitro* sugarcane cultures were established by using shoot tip cultures which successfully removed the virus from the plant material. These virus-free local varieties will now be used for epidemiological studies and to determine the effect of the virus on yield.

Table 1. Most common diseases found on imported varieties in the post-quarantine area at SASEX.

YEAR PLANTED	COUNTRY	VARIETY	DISEASE
1999	Zimbabwe	ZN88-1485	Mosaic
2000	Zimbabwe	ZN88-1243, ZN88-1485, ZN88-2453, ZN88-2453, ZN88-2471, ZN89-2591, ZN94-132, ZN94-147	Mosaic
	Mauritius	M469/84	Mosaic
	Queensland	Q141, Q159	Mosaic
	Mauritius	M1162/82	Smut
	Queensland	Q158	Smut
	Zimbabwe	ZN89-3254	Pokkah boeng
	Zimbabwe	ZN94-129	Rust
2001	Queensland	Q155, Q155	Mosaic