Procedures for the safe movement of sugarcane germplasm

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Many important diseases of sugarcane can be transmitted in vegetative propagation material. These include all viral and phytoplasmal diseases, most bacterial diseases and several diseases caused by fungal pathogens. Sugarcane germplasm is still mainly exchanged between countries in the form of stalk pieces (setts). Therefore, unless precautions are taken, the exchange of varieties can provide a means of spreading diseases between countries (CROFT *et al.*, 1996).

Risks from germplasm exchange and basic control measures

The quarantining of imported germplasm has been widely practised for many years. Until recently security was mainly based on the recognition of disease symptoms in glasshouse-grown plants. Before the causal agents of certain diseases had been identified and before modern methods of diagnosis had been developed, the exchange of germplasm was probably instrumental in the world-wide spread of various pathogens, particularly those that cause non-specific symptoms, such as ratoon stunting disease (RSD), or can remain latent, such as leaf scald. There is good evidence that the organisms that are associated with yellow leaf syndrome (YLS) have recently been spread in this manner. The hazards presented by germplasm exchange are now well appreciated and most countries importing germplasm apply strict quarantine procedures (CROFT *et al.*, 1996; GILLASPIE, 1989).

Because of the risk of transmitting diseases by the exchange of germplasm, the avoidance of material from high-risk areas, where the health standard of the material is uncertain, can be an important security measure for the importer.

An important aspect of quarantine security is thermotherapy, applied mainly as the treatment of setts in hot water. Appropriate hot water treatments (HWT) eliminate all seed cane-borne fungal pathogens and certain bacterial pathogens. Additional security is provided by molecular and serological tests for an increasing number of pathogens. Currently these include the causal agents of gumming, leaf scald, ratoon stunting (RSD), Fiji disease, mosaic, streak and yellow leaf syndrome (YLS). The exchange of diagnostic protocols and materials among organizations that exchange germplasm is encouraged. Newer more accurate diagnostic procedures must be continually developed, evaluated and adapted to improve the quarantine process.

Because diagnostic tests are not available for all pathogens, and because not all importers are equipped for modern diagnosis, the basis of quarantine security remains frequent inspections by experienced diagnosticians.

Recommendations for the safe movement of germplasm

The following is a summary of the recommendations published in detail by Frison and Putter (1993). This publication should be consulted for further information on quarantine security and for information on precautions against specific diseases.

Exchange of setts – actions by the exporter

Although coping with the risks associated with variety exchange is mainly the responsibility of the importer, the exporter should take precautions to ensure, as far as possible, that the material is disease and pest-free.

Setts for export should be taken from propagation plots that were established with seed cane that was subjected to HWT and inspected during growth and found free from symptoms of systemic diseases. Where possible the source plots should be in areas not subjected to hazardous diseases, and the source plants should be indexed for pathogens.

At least two 3-budded setts per variety should be carefully stripped of all trash, washed clean, treated in water at 50°C for 30 min (short HWT), and dipped in a general fungicide and in a general insecticide.

The names of the clones should be written directly on the rind of the setts, the extremities of which are then dipped in low melting point paraffin wax.

The setts should be wrapped in dry paper and protective packing and dispatched by air freight or courier service.

Exchange of setts – actions by the importer

On receipt, parcels should be unpacked in a secure facility, the setts inspected for rotting and insect damage and given a short HWT.

The setts should be planted in sterilized potting medium in an insect-proof growth facility (glasshouse or screenhouse). Ideally, imported sugarcane should be grown in an environment isolated from any other sugarcane growing area. The French international sugarcane quarantine is located in the South of France where no sugarcane is cultivated (Rott *et al.*, 1998). When the quarantine is located close to sugarcane fields, the facilities should be built with the highest security standards such as those of the quarantine station of SASEX in South Africa (Bailey and Bechet, 1988).

Two growth cycles are recommended, setts being cut from the first planting, subjected to a long, cold soak HWT (24–48 h in cold running water followed by 2–3 h at 50°C), and re-planted in fresh sterilized medium. Note: The application of the 50°C treatment for 3 h may reduce germination of some wild germplasm (*Erianthus* etc.) or sugarcane cultivars in certain locations. The possible duration of the 50°C treatment should, therefore, be tested before application to new material or in a new location.

Once the second planting is established and provided the ratoon regrowth remains free of disease symptoms, the first planting should be destroyed.

During both growth cycles, the plants should be inspected frequently by a skilled sugarcane disease diagnostician and any diagnostic tests available for virus and prokaryote diseases should be applied.

Provided the plants remain free of disease symptoms and diagnostic tests are negative, at the end of the second cycle, setts can be cut, subjected to a long, cold soak HWT, and planted in post-quarantine isolation for further propagation.

Pre-export quarantine

A number of organizations have bilateral agreements regarding germplasm exchange. The main feature of these agreements is that the exporter agrees to process material through a quarantine process, which includes diagnostic tests, before dispatch. The main benefit is to improve quarantine security and to reduce the onus on the importer for a strict, lengthy quarantine process. The

latter is of particular benefit when the exporter is better equipped technologically than the importing organization. As an example, the quarantine unit of CIRAD in Montpellier, France, can be cited. This quarantine unit is located within an international research centre on tropical agronomy and exports varieties to West African countries that do not have the laboratory facilities to test sugarcane for diseases (ROTT *et al.*, 1998).

Tissue cultured plant exchange

Increasing use is being made of tissue cultured plants (*in vitro* cultures) as a safe method of exchanging germplasm, but it requires the exporting organization to have appropriate technological expertise.

Setts are prepared from suitable source plants (selected to the same standards as for the exchange of setts). These are planted in sterilized medium in a containment facility.

After several months' growth, shoot tip cultures are prepared from the apical bud and upper lateral buds, excised from young stalks under aseptic conditions. These are cultured on agar medium under an appropriate light, nutrient and temperature regime.

When the buds have started to produce roots, they are excised and transferred to agar growth medium in transparent containers for further growth.

The young plantlets can be exported, growing on agar medium, after tillers have started to develop and when of a suitable size (Figure 1).

On receipt, the plantlets should be planted into sterilized potting medium in a containment facility and grown for at least one cycle, during which diagnostic tests for pathogens should be applied.

Once the plants are determined to be free from pathogens, setts are prepared for further propagation outside quarantine.

Feed-back of information

To encourage the improvement of standards, importing organizations should routinely inform exporters if any diseases are detected in exchanged material.

References

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Figure 1. Tissue cultured sugarcane plantlets (P. Rott).