CRYOPRESERVATION AS A TOOL FOR LONG-TERM CONSERVATION OF SUGARCANE GERMPLASM.

FLORENT ENGELMANN[1, 2, 3], MARÍA TERESA GONZÁLEZ ARNAO[4, 5], FLORENCE PAULET6 & DANIÈLE ROQUES.

[1]CIRAD, Station de Roujol, F-97170 Petit-Bourg, Guadeloupe, French West Indies (current address).
[2]International Plant Genetic Resources Institute (IPGRI), Via dei Tre Denari 472/a, 00057 Maccarese (Fiumicino), Rome, Italy.
[3]Institut de Recherche pour le Développement, BP 64501, F-34394 Montpellier Cedex 5, France.
[4]Centro Nacional de Investigaciones Cientificas (CNIC), Ave 25 y 158 N0 15202. Cubanacan, Playa. Ciudad de la Habana 12100, Cuba.
[5]Universidad Veracruzana, Facultad de Ciencias Químicas. Prol. OTE. 6, No. 1009, CP 94340, Apartado Postal 215, Orizaba, Veracruz, México (current address).
[6]CIRAD, Avenue Agropolis, TA 70/09, F-34398 Montpellier Cedex 5, France.
E-mail: daniele.roques@cirad.fr

Cryopreservation, i.e. the storage of biological material at ultra-low temperature, usually that of liquid nitrogen (-196oC) is the only technique currently available to ensure the safe and cost-efficient long-term conservation of vegetatively propagated plant species. At this temperature, all cellular divisions and metabolic processes are stopped. The plant material can thus be stored without alteration or modification for a theoretically unlimited period of time. Moreover, cultures are stored in a small volume, protected from contamination, and require a very limited maintenance. Today, cryopreservation protocols have been developed for well over 100 different species and the number of cases where it is routinely used in the genebank context is increasing steadily.

In the case of sugarcane, a cryopreservation protocol has been developed at the beginning of the 90's in the framework of FAO and IPGRI funded collaborative projects between French and Cuban institutes. The protocol uses the encapsulation-dehydration technique, in which apices are encapsulated in beads of calcium alginate, pretreated with high sucrose concentrations, submitted to partial physical desiccation before freezing. This protocol has been successfully applied both in

France and in Cuba to a total of 15 different commercial varieties, with recovery rates after freezing ranging between 24 and 91 %. Biochemical and agronomic studies have not revealed any difference between plants coming from non-frozen and frozen material.

Based on the results obtained, it is considered that the protocol is ready for large scale application in a genebank context, even though it should be compared with a new protocol named droplet-vitrification, which might be easier to implement and produce higher recovery. The implementation of cryopreservation is envisaged in the Cirad Roujol Station in Guadeloupe, for the long-term storage of plants which are difficult to maintain in the field, of released varieties, of virus-free plants and of plants with particularly interesting characteristics.

Another potential application of cryopreservation is cryotherapy, i.e. the elimination of viruses through apex cryopreservation. In the case of sugarcane, this would be particularly interesting for the elimination of the sugarcane yellow leaf virus (ScYLV). A research programme should be implemented to compare the efficiency of cryotherapy with meristem culture.