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USE OF ISOENZYMES FOR THE DETERMINATION OF GENETIC VARIABILITY FROM TISSUE CULTURE AND BETWEEN CULTIVARS OF TARO, COLOCASIA ESCULENTA

(Utilisation des isoenzymes pour la détermination de la variabilité génétique à partir de culture de tissus et de cultivars de taro, Colocasia esculenta)

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SUMMARY

A frequent problem with plants produced from tissue culture is the presence of genetic alterations. This is a particular problem where such procedures are undertaken for the purpose of clonal propagation of material. While some of these changes are morphologically visible, it is possible for minor changes in the primary sequence of a protein to leave the whole plant largely unaffected. These changes could, in many cases, be detected by alterations of protein migration in an electrophoretic system. Starch gel electrophoretic techniques have been adapted to monitor the changes in several isoenzyme groups in plants from tissue cultured taro, *Colocasia esculenta*. These techniques are also useful in the development of isoenzyme profiles for specific cultivars of taro. This data is important in producing characterizations of taro gemplasm collections ; particulary where similar cultivars are found in two or more collections. The reliability of isoenzyme analysis for tissue culture and cultivar distinction is discussed.

RESUME

Un problème fréquent chez les plantes issues de cultures de tissus est la présence d'altérations génétiques, en particulier lorsqu'il s'agit de propagation clonale du matériel. Si certains de ces changements sont morphologiquement visibles, des changements mineurs dans la séquence primaire des protéines peuvent dans beaucoup de cas, être détectés par l'altération de la migration d'une protéine en électrophorèse. Des techniques d'électrophorèse sur gel d'amidon ont été adaptées au contrôle de la variation de plusieurs groupes d'isoenzymes chez des plantes provenant de culture de tissus du taro, Colocasia esculenta. Ces techniques rendent également services dans l'établissement

des profils enzymatiques de cultivars particuliers du taro. Cette donnée est d'importance pour la caractérisation des cultivars appartenant à deux ou plusieurs collections. La fiabilité de différents systèmes d'isoenzymes est discutée.

INTRODUCTION

Taro, *Colocasia esculenta*, is a major subsistence crop of the Pacific and an important secondary crop of Asia, West Africa, and many parts of the humid tropics. As is common to clonally propagated crops, taro is in need of research to develop techniques which will allow establishment of genebanks of important material. Also like many clonal crops, the most practical method for storage of taro is by tissue culture. Techniques have been published which allow for tissue culture of many taro cultivars (ARDITTI and STRAUSS, 1979). However, none of these has addressed the problem of possible genetic changes occurring during the culture process. Further, no mechanism exists for monitoring the presence of such possible change, apart from grow-out of cultured materials.

Soon after tissue culture was applied to clonal propagation of plants it became obvious that not all plants resulting from a culture were phenotypically identical to the original. Such variation is not unusual to vegetatively propagated crops and can result from somatic mutation (BEAUCHESNE, 1982 ; EVANS, SHARP and MEDINA-FILHO, 1984 ; HUSSEY, 1978 ; SCOWCROFT and LARKIN, 1982 ; SKIRVIN, 1978). These mutant plants are usually rogued out or, rarely, saved for an improved character they display. Other mutations may bring about morphological or physiological changes not readily obvious. Thus, it is suggested that such crops consist of a mixture of slightly differing clones (HUSSEY, 1978). Long term culture and repeated subculture, particularly of callus, increases the likelihood of genetic changes occuring. Polyploid or aneuploid lines often result from callus cultures (CHALEFF, 1983 ; HUSSEY, 1978, 1980 ; MURASHIGE, 1977 ; SKIRVIN, 1978). Consequently, it is necessary that any method for propagating plant material be designed to minimize induced genetic alterations. Since minor biochemical changes may have little effect on growth characteristics or yield of the crop, general phenotypic variation should be of first consideration. However, a mechanism for characterizing of even subtle changes will allow for detection of genetic changes at an early stage.

Isoenzyme analysis has been applied to a number of crop species (TANKSLEY and ORTON, 1983). A large amount of the variation seen in these isoenzymes has been attributed to genetic change (SCANDALIOS and SORENSON, 1977). Often the biological properties of the molecule are unaltered by small primary sequence changes. Consequently, these changes may be undetected at the phenotypic level. This study was initiated to investigate the use of isoenzymes to monitor genetic stability of <u>in vitro</u> maintained taro cultivars. It is part of a larger study to develop techniques of tissue