Morphological Changes in Chilling Injured Sweet Potato Root

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The structure and function of mitochondria prepared from chilling injured sweet potato are irreversibly deteriorated. For example, respiratory activity, cytochrome c content or lipid-p amount per protein-N amount, in the mitochondrial fraction is decreased during chilling storage and the extremely swollen mitochondria are observed by electron microscope. On the other hand, it is expected that the structures of not only mitochondria but also other cell particles and membranes are changed in chilling-injured sweet potato root tissue. This paper shows the morphological changes in sweet potato root tissue suffering from chilling injury.

Each sweet potato root (Ipomoea batatas, variety Okinawa No. 100) was cut perpendicularly into halves. The cut surfaces were covered with vaseline and with two sheets of paraffin paper in order to shut off air. The samples were divided into two groups: one group was stored for 14 days at 0~1°C as chilling injured sweet potato; the other was stored for 14 days at 10~14°C as healthy sweet potato. Corresponding blocks of the same roots were cut into small cubes (1 mm thick), which were submerged in 0.2 M K-phosphate buffer, pH 7.2, containing 5% glutaraldehyde and 0.15 M sucrose for 3 hr at 0°C. The fixed materials were three times washed with the same buffer without glutaraldehyde, left in 0.2 M phosphate buffer, pH 7.2, containing 1% OsO4 and 4.6% sucrose for 3 hr at 0°C, and dehydrated in graded concentration of ethanol and acetone. The samples were submerged in the Epon mixture with absolute acetone for 30 hr at room temperature, burried in the Epon mixture, and left at 37°C for 24 hr, next at 45°C for 8 hr, then at 60°C for 8 hr. The section was cut with a glass knife using a JUM 5A ultra microtome made by Japan Electron Opticus Lab., and was stained in uranyl acetate and was observed with a Japan Electron Opticus Lab. EM-TS-7 microscope.

As shown in Fig. 1a, in the morphological observation of healthy sweet potato root tissue indicates starch granules surrounded by membranes, large vacuolar spaces and cytoplasmic regions. There were observed some differences in the structure of mitochondrial cristae and in the size of cells between parenchymal tissue and fibrovascular bundle (Fig. 1a and b). Cells in the latter were very small in size as compared with the former cells, contained no starch granules, and had tubular structure. Furthermore, the structure of mitochondrial cristae in the latter cells was vesicular or tubular (Fig. 1b), while that in the former cells was reticulate (Fig. 1c). While, the cytoplasmic membranes and vacuolar membranes were preserved without their degradation and they surrounded completely cytoplasmic matrix in both parenchymal tissue cells and fibrovascular bundle cells (Fig. 1a and b).

On the other hand, in the morphological observation of chilling injured sweet potato root tissue (Fig. 2a and b), membranes surrounding starch granules and cytoplasmic membranes appeared to be preserved without damage in the most parts, however the vacuolar membranes were degraded and disappeared at some parts. Consequently, it was observed that mitochondria, some of other particles or vesicular membrane-like broken vacuolar membranes were present in the vacuolar spaces, because of streaming out with cytoplasmic matrix. Furthermore, Fig. 2c showed that cristae of mitochondria in parenchymal cells of chilling injured tissues were developed in larger reticular form than those of healthy sweet potato root tissue.

From these observation, it is noticed that the structure of mitochondria in intact tissue was changed during 14 day chilling storage. The previous paper reported that extremely swollen mitochondria were isolated from 14 day chilling stored sweet potatoes. It is considered that such swollen mitochondria occurred during preparation, owe to structural changes of mitochondria caused in chilling injured intact tissue. Furthermore, the vacuolar membrane was sensitive to chilling as well as mitochondrial membrane and has been broken at some parts during chilling storage. Therefore, it is clear that the structural changes in mitochondrial membrane and vacuolar membrane are closely connected with the irreversible physiological deterioration by chilling storage.

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REFERENCES

FIG. 1a, b and c. Electron Microscopy of Healthy Sweet Potato Root Tissue.
Sweet potato tissue was fixed by 5% glutaraldehyde for 3 hr, then by 1% OsO₄ for 3 hr. M: mitochondria; CW: cell wall; CM: cytoplasmic membrane; VM: vacuolar membrane; ST: starch granule; and V: vacuolar space. a and c: parenchymal tissue; b: fibrovascular bundle.
FIG. 2a, b and c. Electron Microscopy of 14 Day Chilling Stored Sweet Potato Root Tissue.
The methods of fixing and staining were the same as in the legend of Fig. 1. M, CW, CM, VM. SG and V: The same as in the legend in Fig. 1. DVM: degraded vacuolar membrane.