The Resistance to Anoxia and the Mitochondrial Fine Structure of Rice Seedlings

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Summary

Under anoxia, rice coleoptiles have a remarkable capacity to grow and to preserve undamaged mitochondrial structure and functions. The transfer of aerobically grown intact seedlings to anaerobic conditions resulted in the appearance of unusual mitochondria in coleoptiles as well as in leaves and roots. These mitochondria become filled with stacks of extended cristae, but, obviously, are not affected structurally (only in the root cortex the cells are damaged after a longer period without oxygen). On the contrary, the mitochondria and other organelles of excised coleoptiles, roots and leaves disintegrate after a relatively short exposure to an oxygen-free environment. The degeneration can be avoided if the excised organs are supplied with glucose. Then the mitochondrial fine structure resembles that of intact plants kept under anaerobic conditions. The observations suggest that the capacity of rice coleoptiles to grow under anoxia and to preserve undamaged mitochondria and other organelles is not caused by the resistance of the cell organelles to oxygen deficiency, but rather by the ability of the seedling to transport organic compounds easily, even under the exclusion of oxygen, from the grain to the coleoptile where they can be utilized by glycolysis.

1. Introduction

Higher organisms, animals and plants, usually require a continuous supply of molecular oxygen from the environment for normal life activity. The maintenance of cellular fine structure of these organisms depends on aerobic metabolism. Even a short-time exclusion of molecular oxygen leads to a disorganization of the metabolism and to a degradation of the ultrastructure. Unlike other higher plants, rice coleoptiles can easily grow under conditions of total exclusion of molecular oxygen. Electron microscopic studies have shown that rice coleoptiles grown under these conditions contain all the organelles inherent to a normal aerobic cell including intact mitochondria (VARTAPETIAN et al. 1971, 1972, 1975,UEDA and TSUJI 1971, TSUJI 1972,
This remarkable resistance to anoxia of rice coleoptiles is maintained even when the seedlings suffer secondary anaerobiosis, i.e., anoxia following germination under aerobic conditions (Kursanov et al. 1973, Vartapetian et al. 1974).

Hence, rice coleoptiles may serve as a convenient model for the study of molecular and cellular mechanisms of adaptation to oxygen deficiency in higher organisms.

Using electron microscopic techniques we attempted to cast some light on the factors responsible for this high resistance to anoxia. We were particularly interested to find out whether, at the ultrastructural level, with resistance is merely a feature of the mitochondria themselves or whether it may rather be explained by physiological peculiarities of the entire plant.

We, therefore, studied the fine structural changes in the mitochondria of rice coleoptiles, leaves and roots after separating them from the seeds and placing them under conditions of anoxia. To find out the possible role of organic substances transported from the endosperm into the organs of the seedling in building up the mitochondrial tolerance to anoxia we also investigated the changes of mitochondrial ultrastructure in excised organs fed with exogeneous glucose. Intact seedlings under anaerobic conditions as well as excised coleoptiles, leaves and roots under aerobic conditions served as controls.

The results of these experiments suggest that the remarkable resistance of the mitochondria fine structure of rice coleoptiles to anoxia mainly is due to the unusual capacity of rice seedlings to transport organic substances under conditions of strict anoxia and to utilize them in anaerobic metabolism, rather than to mitochondrial peculiarities.

2. Materials and Methods

Rice seeds, Oryza sativa (Arpa-shali variety) were germinated in the dark at 25 °C for 5 days in air. The seedlings were used in the following 5 experiments:

1. Intact seedlings were placed in distilled water continuously supplied with air,
2. intact seedlings were placed in water but continuously supplied with a weak stream of N₂ (O₂ content not exceeding 0.003%),
3. excised coleoptiles, embryonic leaves and roots were placed in distilled water supplied with air,
4. excised organs were placed in water supplied with N₂,
5. excised organs were placed in 0.5 or 1% glucose solution supplied with N₂.

After 1–5 days the lower part of the coleoptile and of the leaf as well as the root tip were fixed for electron microscopy in a mixture of 2% depolymerized paraformaldehyde.

Fig. 1. Coleoptile cells; pretreatment: 5 days germination under aerobic conditions. M = mitochondrion. a Excised coleoptile, 3 days aerobic conditions in water. b Intact seedling, 5 days anoxia in water. c Excised coleoptile, 1 day anoxia in water. d Excised coleoptile, 2 days anoxia in water. e Excised coleoptile, 2 days anoxia in 0.5% glucose solution. f Excised coleoptile, 5 days anoxia in 0.5% glucose solution.