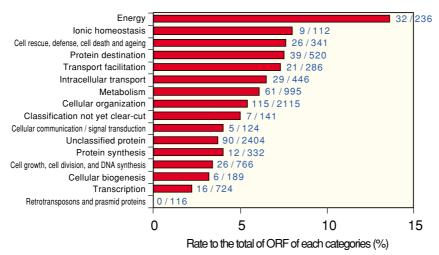
## Screening of Genes that Respond to Freeze-Thawing Stress using DNA Chip

The International Patent Organism Depositary (IPOD) is authorized by the Commissioner of the Japanese Patent Office as the sole depositary for the organization patent in Japan. Deposited organisms are preserved in optimum conditions such as freeze-drying, cryopreservation and subculture. However, there may be a possible deficit in biological activities and transformed characters in the deposited organisms. Therefore, we should understand the damage caused by preservation and develop new preservation methods. It may be possible that the characteristics of organisms may change by the present preservation method. This problem has discussed by organism-related inventors. Therefore, it is quite important to maintain the precious property in this depositary for clarifying details of the damage and establishing an efficient preservation method.

Freeze-thawing stress is one of the main stresses that can be caused during preservation. In order to analyze the effect by freeze-thawing stress, we have carried out DNA microarrays containing approximately 6,000

open reading frames (ORFs) from Saccharomyces cerevisiae. The yeast cells were cultivated until the logarithmic phase, and the cells were frozen at -80 °C. These cells were thawed and cultivated for 15 min, 30 min, and 60 min. From these cells, we isolated mRNA and compared them with those obtained from the cells without freeze-thawing stress. Consequently, 282 ORFs showed more than a twofold increase in expression levels at 60 min, 15 and 30 min. When the induced genes at 60 min were classified according to functional categories of MIPS (http://mips.gsf.de/), it was suggested that the number of genes concerning the function of "energy" were induced. Many other induced genes may belong to the categories on "ionic homeostasis", "cell rescue, defense, cell death and aging", and "protein destination" (Fig). These results suggest that the expression of these genes may be induced for the proteinic restoration and decomposition caused by freeze-thawing. These elevated expression levels may also be related to their energy. These genes can be a good marker for searching the best freeze-thawing condition and developing new preservation methods.

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Classification of induced genes