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Isolation of gibberellins producing fungi from the root of plants

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Gibberellins(GAs) are a large family of isoprenoid plant hormones, that control many aspects of plant development, including seed germination, shoot elongation, flower formation and development, fruit-setting, seed development, sex determination, and chlorophyll content. 238 fungi were isolated from the 25 plant roots, and the production of GAs was spectrophotometrically examined. As a result sixty nine fungi were shown to produce GAs. Bioassay of culture broth from sixty nine GAs-producing fungi was carried out with waito-c rice. Sixty nine fungi with GAs-producing activity were incubated for seven days in 40 ml of Czapek's liquid medium at 25°C and 120 rpm, and the culture broth of fungi were treated on the 2-leaf rice sprout. Fifteen of these have plant growth promoting activity and the amount of each GAs in the medium was measured by Gas chromatography-mass spectrometer (GC-MS). The fifteen fungi were shown activity of gibberellin by GC-MS. Ten of these fungi were also identified by genetic analysis of the nucleotide sequences in the internal transcribed spacer region of the ribosomal DNA.

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Contribution of Antigenicity by GAPDH, a Fish Pathogen *Edwardsiella tarda*
Surface Antigen in the Immune Response

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Edwardsiella tarda is a Gram-negative bacteria in the family of *Enterobacteriaceae* and causes a systemic fish disease called edwardsiellosis. It infects a wide-range of commercial fish. In a recent study (Kenji *et al.*, *Vaccine* **22**, 2004, 3411-3418), the 37 kDa protein from *E. tarda*, which is conserved in various serotype, was suggested to have an antigenicity. Therefore, on the basis of the N-terminal sequence of 37 kDa protein, the gene was amplified by PCR was analyzed by nucleotide sequencing. The gene encoding 37 kDa protein was identified as a *gapA* encoding a GAPDH (glyceraldehyde-3-phosphate dehydrogenase). It shows antigenicity and is on progress in research for its practical use as a vaccine candidate. We cured plasmids having antibiotic markers and inactivate *gapA* gene for analysis whether the gene play a role in pathogenesis, we constructed a suicide plasmid containing a kanamycin resistant gene cassette in *gapA* gene, pBP428. The next we will construct a *gapA* mutant by an allelic exchange. With the employment of several protein purification approaches, the GAPDH was purified for immunogenic characteristics. GAPDH-specific polyclonal antibody is produced in a rabbit and an immuno-reactive protein band is detected in western blot analysis. In progress, to inject GAPDH protein that vaccine candidate for flounder, we expect that this protein imply a possibility towards generating high-titer antibody responses during infection of *E. tarda*.