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Examination of *Trichoderma* strains isolated from the rhizosphere of vegetables for the purposes of developing environment-friendly in field technologies

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Organic farming is becoming nowadays more and more important in the agriculture. Organic farmlands are exposed to dangerous xenobiotics through distinct pollution drift effects such as wind-driven, pesticide-containing dusts and xenobiotic-containing rains. In order to achieve organic farming, there is a need for the development of new techniques which allow the bioremediation of lands previously used in common, intensive agricultural practice. Organic agriculture also faces the problem of pests including the damage caused by plant pathogenic fungi, therefore the implementation of biological control as a possible, environment-friendly solution is also of increasing importance.

Trichoderma strains were isolated from vegetable rhizosphere samples on dichloran-Rose Bengal medium. After purification of genomic DNA, the PCR amplification of the internal transcribed spacer (ITS1-5.8S rDNA-ITS2) region and its sequence analysis were used for the identification of the isolates at the species level. Altogether, 45 *Trichoderma* isolates were identified from the examined samples. The detected *Trichoderma* species were *T. asperellum, T. atroviride, T. citrinoviride, T. gamsii, T. hamatum, T. harzianum, T. koningiopsis/T. ovalisporum, T. longibrachiatum/H. orientalis, T. pleuroticola* and *T. virens*.

In vitro antagonism of selected isolates was examined in dual culture tests and the Biocontrol Index (BCI) values were determined for the particular isolates. Certain *T. asperellum, T. virens* and *T. atroviride* isolates proved to possess good *in vitro* antagonistic activities against plant pathogenic *Fusarium solani, F. oxysporum, Phoma cucurbitacearum, Alternaria alternata, Botrytis cinerea, B. pseudocinerea* and *Rhizoctonia solani* strains.

Fungicide susceptibilities were measured by the microdilution method and the Minimum Inhibitory Concentration (MIC) values were recorded. Ten fungicides were tested in the concentration range of 512 µg/ml to 1 µg/ml. Strain *T. asperellum* SZMC 20866 showed resistance to 9 fungicides and was sensitive only to Maneb (MIC: 256 µg/ml). The *T. atroviride* strain SZMC 20781 showed similar fungicide resistance properties to those of *T. asperellum* SZMC 20866. MIC values of *T. harzianum* SZMC 20770 were 256, 512, 32, 64, 512 and 128 µg/ml for Cyproconazole, Fenarimol, Imazalil, Maneb, Penconazole and Thiram, respectively. The strain most sensitive to the tested fungicides was *T. virens* SZMC 20779.

The effect of temperature on growth in a range of 5 – 40 °C was also examined, and the water activity (a_w 0.997 – 0.922) and pH (2.2 – 8.0) dependence determined in the case of the isolated *Trichoderma* strains. Temperature values of 20-30 °C were optimal for the growth of *Trichoderma* strains, while none of the strains were able to grow at 5 °C. The examined strains were able to grow in a wide range of pH from 2.2 to 8.0, the maximal growth was observed under acidic conditions at pH 4.0. The highest tested a_w value (0.997) seemed to be optimal for the growth of all strains. Only limited growth was observed at 0.945 in the case of only three examined strains.

The results of the recent study suggest that the rhizosphere of vegetables may be a rich source of potential biocontrol agents for environment-friendly, organic agricultural production. We identified 3 *Trichoderma* strains which seem to be very promising for the development of microbial products with multiple beneficial effects for the purposes of organic farming.

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Physiological and molecular analysis of salt stress-induced PCD in tomato

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As saline soils and waters are common around the world, salinity is one of the major abiotic stress which largely limits plant growth and productivity. The ability of plants to tolerate salt stress is determined by multiple biochemical pathways; the most important is that the plant facilitates retention and/or acquisition of water, protects chloroplast functions, and maintains ion homeostasis. Severe salinity induces programmed cell death (PCD) in plants takes place in eukaryotic cells of different origin. One typical hallmark of PCD in plants is an increase in the process of protein degradation which is initiated by reactive oxygen species (ROS) and nitric oxid (NO) and involves the action of proteolytic enzymes. ROS and NO generation is one of the earliest response of plant cells under abiotic stresses. Protein degradation is probably the most important degradation process that occurs during PCD. The total protein content of tomato leaf gradually decreased with increasing concentration of NaCl. This decrease in protein content might be due to the increasing activity of cysteine- and serine proteases. For this reason, many of the genes up-regulated during PCD are proteases. The four major classes of proteases: cysteine, serine, aspartic acid and metalloproteases, exist in plant cells. Genes that encode proteases are activated by different ways. Expression of these genes that encode cysteine proteases has been shown to induced by environmental stress such as salinity. We studied different genes, for instance *MCA1, CYP, CP,* which encode various types of proteases participating in plant PCD. In addition, inhibitors encoding genes (*PI2* and *LTC*)