Proceedings of the Joint Meeting AGI-SIBV-SIGA
Assisi, Italy – 19/22 September, 2011
ISBN **978-88-904570-2-9**

Oral Communication Abstract – 5C.02

GREEN MICROALGAE AND PLANTS EXPRESSING FUNGAL LACCASES USEFUL FOR ENVIROMENTAL AND INDUSTRIAL APPLICATIONS

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Laccase, tobacco, green microalgae, bioremediation

Industrial activities release a wide range of toxic chemicals into the biosphere. Among such chemicals, phenolic compounds represent a major risk for human health and biodiversity mainly due to their relative persistence in the environment. In the Mediterranean area the olive oil industry accounts for about 95% of the entirely world olive oil production. Olive oil mill wastewaters are characterised by low pH and a high content of mono- and polyaromatic compounds that exert microbial and phytotoxic activity. White-rot fungi have been proved to secrete a large array of ligninolytic enzymes which are highly efficient in degrading aromatic compounds, such as lignin peroxidases, manganese peroxidase and laccases. The latter enzymes, have been proved to be useful for industrial applications, such as biobleaching of pulp, organic synthesis and fruit juice process. Our final goal is to enhance the laccase production in green organisms and to evaluate their ability to secrete laccases into the environment.

A biotechnological approach to enhance the efficiency of xenobiotic removal from green organisms is to overexpress genes involved in metabolism, uptake or transport of specific organic pollutants. However, a useful approach is to secrete laccases in the soil or water. The laccase cDNA of the *pox*A1b gene from *Pleurotus ostreatus*, carrying a signal peptide sequence for enzyme secretion and driven by the CaMV 35S promoter, was cloned into a plant expression vector. Nuclear genetic transformation was carried out by co-cultivation of *Agrobacterium tumefaciens* with tobacco cv Samsun NN leaves and cells of five different microalgae accessions belonging to the genera *Chlamydomonas, Chlorella* and *Ankistrodesmus*. Transgenic plants and microalgae were able to express and secrete the recombinant laccase in the root exudates and the culture medium, respectively. In comparison to untransformed controls, the ability to reduce phenol content in OMW solution was enhanced up to 2.8-fold in transgenic tobacco lines and by up to about 40% in two microalgae accessions. The present work provides new evidence for metabolic improvement of green organisms through the transgenic approach useful for environmental and industrial applications.