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STUDY OF GENOMICS AND PROTEOMIC RESPONSE TO ABIOTIC STRESS IN TOBACCO CELL LINE

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Plant growth and productivity depend on the interaction of genotype with various external factors. Every time that there is a drastic change of environment factors a stress occurs. The principal goal of this research is to study the characterization of cellular response, analysis of protein expression and DNA epigenetic modifications changes during abiotic stresses.

Nicotiana tabacum L. cell cultures have been used and Heat shock has been used as model system of abiotic stress.

The work has been performed on cellular lines of tobacco (TBY-2) subjected to different timing of abiotic stress (Heat-Shock at 35°C).

The Southern Blot analysis have been used to verify changes of methylation pattern in cellular lines when subjected to different timing of stress temperature. We digested DNAs by using some isoschizomeric enzymes, differently sensitive to the pattern of methylation (HpaII/MspI; MboI/Sau3AI). Afterward, they have been hybridized using, as radioactive probes, regions of repeated DNA (rDNA 5S, rDNA 18S, Tto1 retrotrasposon, repeat BamHI DNA etc)(Wade, 2004).

Moreover, in order to assess the establish of oxidative stress and damage (determination of production of reactive oxygen and nitrogen species and protein oxidation and lipid peroxidation), study level and redox state of ascorbate, glutathione, pyridine nucleotides and level of SH protein group oxidation have been carry out.

These analyses quickly allowed us acquiring useful details about the variation of the methylative state of the DNA in the cells during the time course.

This kind of experimental approach allows us to quickly and clearly highlight both the effects of applied stress on the genome methylation state and the proteomics response induced.

The analyses conducted so far revealed both there is a clear genome hypomethylation and an increase in total protein, besides we found a drastically change of proteomic dowry, probably due to the increased expression of heat-shock proteins.