Understanding conifer responses to environmental changes

Conifers are a group of ancient plants that include tree species of great ecological and economical relevance. Inside this group, we can found trees such as pines, cedars, firs, spruces or cypresses. Coniferous forests dominate vast regions in the northern hemisphere being the main source of wood for industrial purposes. Conifers comprised species that grow in different climates and altitudes with wide geographical distributions and long lifespans. As example, a bristlecone pine is the longest-lived individual on Earth with around 5,000 years. These characteristics highlight their great capacity of acclimation and adaptation to different environmental conditions. Thus, during their long lives conifers must respond to seasonal environmental changes, adapt to unpredictable environmental stresses and co-ordinate their adaptive adjustments with internal developmental programs. Among conifers, the maritime pine (*Pinus pinaster*) is a species with great economic and environmental value that is widely distributed in the Mediterranean southwestern region, dominating forests in France, Portugal and Spain. This pine has high tolerance to environmental stresses such as drought; hence it is distributed in widely varying environments at altitudes ranging from sea level to 2000 m. For that reason, maritime pine is a good model to study the plant responses to the environmental changes.

Fig. 1. (A) Climatic data at the harvesting location during 2012. (B) Needle whorls from an adult maritime pine. The emergence of each whorl was in consecutive years from 2009 to 2012. (C) Hierarchical clustering of samples based on their gene expression profiles.

To understand the way in which these responses occur we conducted a systems-based approach to develop further studies on the relationship between maritime pine and the environment. In this work, we examined the effect of environmental parameters such as temperature and rain on metabolites (the intermediary compounds of metabolism as sugars or amino acids) and gene expression. We analysed 39 soluble metabolites and 8,000 genes in needles grouped by age from 25-year-old maritime pine trees growing in a forest at 1245 m altitude in the South of Spain, under natural conditions, during the course of a year (Fig. 1.). The analysis of the results was performed through statistical methods using bioinformatics resources. With this approach, we were able to correlate the obtained data with the environmental and developmental factors.

Fig. 2. Gene ontology terms for biological processes founds in de gene clusters related to needle development and secondary metabolism. Red squares correspond to biological processes with higher number of related genes with them inside the group.

The results indicated that an important portion of metabolites was strongly affected by inherent acclimation to winter (notably sucrose and other sugars) and needle age (e.g., betaine and methionine). In contrast, gene expression profiles were dependent on environmental variables mainly temperature (notably, the genes involved in photosynthesis and winter acclimation). The employed analyses let us to group the data by their profiles. In the case of the gene expression, we identified 14 different gene clusters, most of them correlated with the environmental conditions. However, two gene clusters were related to the needle development and were mainly composed by genes encoding enzymes of the secondary metabolism pathways (Fig. 2.). These metabolic routes are the responsible of the synthesis of phenylpropanoids, flavonoids and terpenoids which are metabolites involved in wood formation or defense against stress and pathogens. Inside these groups, we also found several genes that are responsible of the secondary metabolism regulation. Thus, these results provided valuable information on the function of the gene clusters and their co-regulation.

In summary, our results suggest that environmental changes module the gene expression for fine regulation of metabolism and metabolites during development. Thus, the adaptive responses would influence the development programme through the maintenance of a metabolic balance. In addition, the results support the existence of alternative groups of genes involved in the same metabolic pathways that contribute to the essential flexibility in responding to varying environmental and/or developmental conditions. The system-based analysis presented here provides a general vision of the seasonal regulation of maritime pine growth and opens new perspectives for understanding the complex regulatory mechanisms underlying adaptive responses in conifers.

Rafael A. Cañas and Francisco M. Cánovas
Departamento de Biología Molecular y Bioquímica, Facultad de Ciencias,
Universidad de Málaga, Spain

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