Cold response in Olive tree; A RNAseq Study.

María de la O Leyva-Pérez[1], Antonio Valverde[2], Jaime Jiménez-Ruiz [1*], Antonio Muñoz-Merida[3], Raquel Valderrama[1], Oswaldo Trelles[3], Juan Bautista Barroso[1], Jesús Mercado[2] and Francisco Luque[1]

[1]Departamento de Biología Experimental, Universidad de Jaén
[2]Departamento de Protección de Cultivos del IAS-CSIC-Córdoba
[3]Departamento de Arquitectura de Computadores, Universidad de Malaga
∗ Jaime Jiménez-Ruiz. ruiz@ujaen.es

Background

Low temperature severely affects plant growth and development. Several plant species have evolved an adaptive response, named cold acclimation. In order to study this response in olive tree (*Olea europaea*), we maintained two-months-old vegetative propagated olive trees cv. Picual at low temperature (14h at 10°C with light, 10h at 4°C at the dark) for 15 days. Control plants were maintained in field growing conditions. Cold stress symptoms were observed after the first 24h as sagging leaf and biochemical stress markers. After 5 days the plants were observed completely recovered. We extracted RNA from leaves of three control plants and the same number from 24h and 10 days cold stressed plants. We pooled RNA and made two cDNA libraries for sequencing. We used Illumina HiSeq 1000 sequencer.

Results

We obtained 129278973 101bp 35 phred quality paired-end reads (18,55Gb). We used ABySS-pe (k-mer=64) and ABySS single-end (k-mer=63) software for *de novo* ensambling in Picasso supercomputer (www.scbi.uma.es). We obtained 125969 contigs (N50=1212bp). We used DNAStar for RNAseq analysis (k-mer=63, 95% of matches). We found 7643 unigenes differentially expressed with an 8-fold-change. Among these unigenes, 3106 were found differentially expressed after 24h of cooling (2179 up-regulated and 926 down-regulated) so we considered them responsible for early cold response. From these unigenes, 904 were up-regulated and 5 repressed by cold stress at 24h but recovered to normal expression level after 10 days of cold stress so we considered them **cold stress** genes. Among the rest of early cold response unigenes, 201 kept induced and 598 kept repressed after 10 days of cold, when plants didn´t show any symptoms, so they were considered involved in **early cold acclimation response**. Other unigenes were observed differentially expressed only at 10 days so we called them **late cold acclimation** genes (42 up-regulated and 4495 down-regulated). We used Sma3 to annotate the selected unigenes. We got 53% in average of annotations from uniprot plant database. Sma3 also retrieved GO Terms and we analyzed them with Blast2GO (V.2.6.2).

Conclusions

We have found three types of response involved in the Olive tree cold stress which affect different biological process; Short term cold stress response, short term cold acclimation response and late acclimation response.