**Título:** ESTUDIO DEL EFECTO DE LA INTERACCIÓN ENTRE AUMENTO DE CO2, TEMPERATURA Y SIMBIOSIS CON DIFERENTES CEPAS DE SINORHIZOBIUM MELILOTI EN LA FOTOSÍNTESIS, FIJACIÓN DE N2 Y CALIDAD DE LA ALFALFA (MEDICAGO SATIVA L. CV. ARAGÓN)

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**Resumen:** Effect of elevated CO2, high temperature and symbiosis with different Sinorhizobium meliloti strains, in photosynthesis, N2 fixation and alfalfa (Medicago sativa L. cv. Aragón) quality.

According to the predictions of the IPCC, at the end of the present century, CO2 concentration may be around 700 µmol mol-1 and the resulting mean global warming may lead to a temperature increase of 4°C. Increasing atmospheric CO2 concentration is expected to enhance plant photosynthesis and yield. Nevertheless, after long-term exposure, plants acclimate and show a reduction in photosynthetic activity (down-regulation), which may cause reductions in potential yield. Some authors suggest that down-regulation is related to nutrient availability and more specifically to an insufficient plant C sink strength caused by limited N supply. Alfalfa (Medicago sativa L.) is an important forage crop for ecological and economical reasons, it establishes a symbiotic relationship with a N2-fixing bacterium (Sinorhizobium meliloti) providing an extra source of N for the plant. N2-fixing legumes often show a larger stimulation of growth and photosynthetic rate at elevated atmospheric CO2 than species without this capability. Therefore, the first objective of the PhD thesis was to test if nodule activity was affected by elevated CO2 and if this activity was sufficient to prevent down-regulation in strictly N2-fixing conditions. The second objective was to study the effect of elevated CO2, high temperature and symbiosis with different Sinorhizobium meliloti strains on photosynthesis, N2 fixation and forage quality of alfalfa plants grown in two different seasons (summer and
In exclusively N2 fixing plants exposed to elevated CO2, N supply by the nodules was insufficient to cover plant N demand and avoid photosynthetic down-regulation. It was due to reductions in nodule metabolism, N2 fixation and N transport to the plant. However, it was concluded, that the addition of high doses of external N, enabled the avoidance of photosynthetic down-regulation.

Other finding of the PhD thesis was that alfalfa yield was dependent on the S. meliloti strain and that the efficiency of the different strains varies according to growth season. Being the plants inoculated with 102F78 strain the most productive at the higher summer temperature, and 102F34 inoculated plants in autumn, because of mild temperatures. Plants inoculated with 102F78 showed similar shoot yield than 102F34 ones, but higher crude protein content at elevated CO2 and temperature. In this climate change conditions, 102F78 inoculated plants produced more quality forage. However, the higher digestibility of plants inoculated with 102F34 strain under any CO2 or temperature conditions, makes them more adequate for growing under climate change scenario conditions. Plants grown at elevated CO2, presented down-regulation in both seasons, especially when combined with high temperatures in summer. The most adequate parameters for detecting acclimation in order of sensibility were Agrowth, rubisco in-vitro activity, rubisco content, A400, Vcmax, A700 and rubisco gene expression. The absence of correlation between photosynthesis and rubisco content with rubisco gene expression was consequence of a lag between both parameters resulting on gene expression lowest sensibility. The genomic and proteomic studies concluded that small subunit of rubisco was the limiting subunit of photosynthesis under ambient and elevated CO2 conditions. The fall of this subunit under elevated CO2 during all the day, was the cause of photosynthetic down-regulation, especially at noon.