Sink or swim: submergence tolerance and survival strategies in Rorippa and Arabidopsis

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SUMMARY

Like humans, plants also struggle and cannot survive in limiting oxygen levels although they are the major suppliers of earth's oxygen. Plants experience low oxygen levels in many occasions one of which is flooding. Interestingly, both plants and humans have similar solutions to deal with the challenging environment under water. Humans came up with many ways to explore the extraordinary underwater life; one of the easiest and cheapest ways being breathing through a snorkel. Similarly, plants can also grow stems or leaves that behave as snorkels to transport gases from the surface of the water to the lower parts attached to soils. However, these plant snorkels are not as cheap as their human counterparts; they need energy to grow. If the waters are deep and all the energy is consumed before the plant snorkels can reach the surface, mortality is inevitable. Nevertheless, if the snorkel can reach the surface on time, the plants can survive for a longer time.

Another strategy that human free divers use to dive deeper or stay under water for a longer time is through a meditative state in which they maintain their metabolism at the minimum. A free diver, Tom Sietas recently entered the Guinness World Records by holding his breath under water for more than twenty-two minutes, needless to mention the importance of meditation in his achievement. Likewise some plants also decrease their metabolic processes when submerged, trying to consume as little as possible. This is achieved by an inhibition of growth and preparing to underwater life by means of several acclimations leading to down-regulation of energy consuming processes.

These are only a few strategies for coping with excess water stress for plants and there is still a great amount of natural variation to be explored that accumulated throughout the evolutionary history. Understanding the mechanisms behind the evolution of these traits will enlighten our views in submergence tolerance of plants. Considering the predictions of increasing frequencies of floods in a changing climate, these advances will help us to enhance crop resistance to floods and/or sustain natural populations in flood-prone areas. This thesis explores natural variation in both intra- and interspecific levels by investigating physiological, genetic and evolutionary aspects of submergence tolerance by using species of three distinct genera: *Rorippa*, *Arabidopsis* and *Rumex*.

In Chapter 2 of this thesis, we investigated extreme submergence tolerance in two yellow cress species; *Rorippa amphibia* and *Rorippa sylvestris*. *Rorippa amphibia* elongates its stem like a snorkel, when submerged. However, if the snorkel cannot reach the surface on time, since its energy is consumed for growth, the plant dies. On the other hand, the meditating species *Rorippa sylvestris* can survive for a longer time since the energy is not consumed

for growth. The environments they inhabit can explain the strategies these species have. The elongating *Rorippa amphibia* usually occurs on sites flooded with shallower waters in which the snorkel can reach the surface before the plant is exhausted. On the other hand *Rorippa sylvestris* inhabits sites with shorter periodic deep floods in which growth might constitute a cost but no gain since reaching the surface is less likely.

The sensing mechanisms of low oxygen and the onset of these strategies are still not entirely known. Nevertheless, recently some gene families were shown to have a key role in both sensing of low oxygen and regulation of these strategies. These genes are of importance since understanding how they regulate these valuable traits might lead to a deeper understanding of how nature shapes plant distributions and eventually help crop improvements for flood-prone agricultural fields. Accordingly, in the third chapter, we studied potential regulators of these two strategies in the *Rorippa* and *Rumex* species (*Rumex palustris* and *Rumex acetosa*) from two distinct lineages. We did not observe a pattern in regulation of these genes that would indicate a parallel evolution of regulation of submergence tolerance strategies. However, we identified candidate oxygen sensors that might be involved in enhancing submergence tolerance in both *Rorippa* and *Rumex*.

All plants are equal but fortunately in this case, some are more equal than others. *Arabidopsis thaliana*, with its great range of molecular and genetic resources is more valuable than any other plant in the laboratories. By using this model plant, in chapter 4, we identified genomic regions that explain the variation in submergence tolerance between two accessions collected from different sites. By using this information, we were able to select candidate genes that increase submergence survival in this species. In the final research chapter (5), we used modern gene sequencing techniques to narrow down this region even more, and additionally we revealed the important gene groups regulated under submergence stress in both of these accessions.

Our results indicate that exploring natural variation will aid submergence research in order to identify different means of coping with underwater life and to understand how these traits are regulated. The results presented in this thesis constitute a promising background for identifying genes with fundamental roles in submergence stress in *Rorippa*, *Arabidopsis* and *Rumex*.