## Summary

In recent times, we are witnessing to a growing demand of craft beers with original flavours and to an increasing interest for the hop functional compounds. Together with the widening of hop growing area, also the research world is more and more focusing on hop, not only on its propagation, but also on its breeding; other than confering resistance to pests and diseases, breeding programs aim to identify hop genotypes with valuable aroma profiles for craft beer field, and rich in bioactives compounds, with anticabacterial and antiviral properties for herbal and pharmaceutics industry.

However, the process of hop domestication, cultivation and breeding has led to a decreasing of hop intraspecific agrobiodiversity, making these goals more challenging.

In this context, in order to overcome these limitations, it is necessary to valorise the existing hop biodiversity, but also to renew the varietal panorama. Even though the increasing interest in this species, very few are the research going on aiming at valorizing and improving hop, especially through biotechnologies.

Based on these considerations, the general objective of my PhD thesis project was to deep knowledge on hop, following two main research lines, agrobiodiversity valorisation and agrobiodiversity enrichment.

In the framework of **agrobiodiversity valorisation**, in this PhD thesis, the adaptability of International cultivars to the Italian climate was investigated, by comparing hop cone characteristics of cultivar Cascade, grown in different areas of Italy, with those grown in the main producing areas of the United States of America (Oregon and Michigan), Germany (Tettnang region) and Slovenia (Carniola region). Obtained results showed significant differences in bitter acids and xanthohumol content, among the analyzed samples, underlining how geography and climatic conditions may affect hop secondary metabolism.

Further to a recurrent demand for hop plants carrying peculiar features, the depletion of germplasm has been attracting the International attention. In this context, a biotechnological approach to propagate hop plants and to preserve their germplasm has been described. Specifically, organogenesis was induced from petioles of hop, cultivar Gianni, confirming the possibility of using this kind of hop explants to obtain a great number of plantlets, in limited space and, independently of the season.

Moreover, to overcome the problem of somaclonal variation that may occur during regeneration and the issues connected to conventional germplasm storage methods, for the first time, in our

knowledge, hop microcutting encapsulation technology was tested. The high conversion recorded proved the suitability of hop microcuttings to encapsulation and opened new insights to investigate other applications of this technology for hop germplasm propagation and storage.

In the context of **agrobiodiversity enrichment**, the influence of several pre-treatments (timing of chemical scarification, cold stratification, imbibition in water and in gibberellic solution) on *in vitro* hop (cv. Columbus) seed germination was evaluated. The study allowed to set up an efficient protocol for the obtainment of a high number of seedlings, in a relatively short time and out of the natural season, resorting to chemical scarification with sulphuric acid and to the use of gibberellic acid, both in the imbibition solution and in the culture medium. Moreover, by resorting to sex-linked molecular markers it was possible to precocious individuate the seedling gender.

All in the interest of enriching the variability within which will be possible to implement the selection of new genetic combinations, two studies have been carried out to induce new variability, investigating the gamatoclonal and the somaclonal variation. Gametoclonal variation offers the opportunity to discover the natural variability present in plant gametes and to exploit this genetic variability, in order to create new genetic combinations. Since in hop, there is a complete lack of information on gametic embryogenesis, a preliminary study on structure and biology of the hop male flower, microspore and pollen development has been carried out. This study has been carried out in collaboration with the Comav, Polytechnic University of Valencia. The obtained results provided a deep description of male flower, with a peculiar attention to development of microspore and pollen grains. Moreover, it was possible to establish a correlation between microspore/pollen developmental stages and flower bud/anther size, making easier and faster the selection of buds containing microspores at the most appropriate developmental stage in order to obtain androgenesis.

Since, some aspects of the *in vitro* culture environment can led to the occurrence of genetic and epigenetic change, indirect organogenesis was induced from leaves of hop cultivar Gianni, testing the effect of the type and the concentration of several growth regulators in the culture medium and the time in which the explants were maintained in culture on the insorgence of somaclonal variation. Plants obtained from leaves culture have been subjected to cytofluorimetric analysis and to molecular analysis which revealed that 16.8% of regenerants were mutated, some of which were tetraploids.