«VALorization of Mediterranean small-scale FARMs by cropping wild UnExploited species»

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Deliverable 2.2

Multilingual electronic handbook of technical information and best practice guides of the selected WEPs

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VALUEFARM: Valorization of Mediterranean small-scale farms by cropping wild unexploited species

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EXECUTIVE SUMMARY

Valuefarm project aims to: 1) propagate and cultivate selected WEPs species, 2) describe and evaluate agronomic performance of WEPs through laboratory-based research and farm experimentation in order to establish best practice guides of plant requirements with respect to mineral nutrition, soil and climate, environmental footprint (low GHG emissions, water and energy use), This report constitutes deliverable D2.2 – "Multilingual electronic handbook of technical information and best practice guides of the selected WEPs".

VALUEFARM IN CONTEXT

Valuefarm is an innovation action which brings together 9 partners from 8 countries:

- University of Thessaly (UTH), **Greece**
- Instituto Politécnico de Bragança (IPB), **Portugal**
- Cyprus University of Technology (CUT), **Cyprus**
- Dokuz Eylul University (DEU), **Turkey**
- Ege University (EGE), **Turkey**
- Consejo Superior de Investigaciones Científicas (CSIC), **Spain**
- Bergische Wuppertal University (BUW), **Germany**
- Greek Fresh Vegetables IKE (GFV), **Greece**
- Benha University (BU), **Egypt**
- University of Mostaganem (UM), **Algeria**

The main goal of VALUEFARM is to valorize Mediterranean small farms by introducing wild edible plants of the Mediterranean (WEPs) such as *Crithmum maritimum*, *Portulaca oleracea*, *Sonchus* sp., *Scolymus hispanicus*, and *Cichorium spinosum* as complementary crops within a competitive farming sector and a climate-changing world and cropping them in a sustainable point of view. The assessment of using WEPs in arduous conditions (drought and salinity stress) and marginal soils with low organic matter, compacted, or eroded where conventional crops cannot be cultivated will also be carried out; we will also assess the contribution of WEPs cultivation to soil properties improvement by reducing or eradicating the use of agrochemicals, and by introducing the use of a more sustainable agriculture with biostimulants, biofertilizers and biopesticides and the use of tailored composts. Finally, the selected WEPs will be assessed for their nutritional value and bioactive compounds content in order to select and propose those farming systems that increase the quality of the final product and its added value.

The key objectives of the proposal are summarized as follows: 1) to propagate and cultivate selected WEPs species, 2) to describe and evaluate agronomic performance of WEPs through laboratory-based research and farm experimentation in order to establish best practice guides of plant requirements with respect to mineral nutrition, soil and climate, environmental footprint (low GHG emissions, water and energy use), 3) to evaluate the potential of cultivating WEPs in degraded soils and assess their soil improvement properties, 4) to diversify existing farming systems from monocropping to agroecological systems rich in diversity through the incorporation of WEPs in mixed and intercropping systems and crop rotation programs combined with legumes, 5) to evaluate innovative approaches (biofertilizers, biostimulants or tailored composts that include beneficial microorganisms plant growth-promoting rhizobacteria (PGPR); plant growth-promoting fungi (PGPF); arbuscular mycorrhizal fungi (AMF)), 6) to analyze chemical composition, nutritional value and bioactive compounds content of WEPs, 7) to increase knowledge and public awareness on the nutritional value and the bioactive compounds content of WEPs, as well as on their environmental impact (resistance/tolerance to drought and salinity stress factors), 8) to create physical labs through a network of farmers for the on-farm demonstration and to implement living lab platforms for technological transfer in each zone of the project of the obtained key results, both of which will facilitate the adaptation of Mediterranean small-scale farms to the proposed farming systems.

Work package 2 aims to select WEPs to be adapted to the proposed climatic Mediterranean conditions, to provide an agronomical characterization of the selected WEPs and finally to integrate them in sustainable farming systems.

Deliverable 2.2 compiles the related technical information in a multilingual handbook, including the best practice guides for the cultivation of the selected species.

OVERVIEW OF FARMING SYSTEMS

Agriculture is facing several risks in the present; the increasing global warming that prevails throughout the world results in increasing temperatures and causes lack of water availability from crop production. Heat and drought are environmental factors that can also cause stressful conditions in plants, being water deficit the most prevalent abiotic stress responsible for major crop losses (Walters et al., 1980; Savinab and Nicolas, 1996). Also, the soil degradation caused by intensive agriculture due to irrational use of fertilizers and pesticides, is also causes a loss in soil fertility, mainly of a reduction in soil organic matter (SOM). 25% of soil occupied by agriculture is already considered degraded (Food and Agriculture Organization of the United Nations (FAO), 2022). This is why the concerns about the long-term sustainability of farming systems are increasing, especially regarding the soil quality management. Organic farming has been reported to increase soil organic matter and water holding capacity. It also increases ecological and biochemical characteristics, improves soil structure and permeability and decreases N leaching compared to soils managed exclusively by conventional farming regimes (Gomiero et al., 2011). Organic matter plays a key role in soil quality and soil ecosystems because provides substrates for decomposing microbes and increases soil microbial activity that supply mineral nutrients to plant, and therefore, increase plant growth and yield (Abiven et al., 2009). Despite that, the transition to organic management may take several years to detect the increase in SOM due to the complex nature of organic farming managements (Clark et al., 1998).

Soil organic amendments like animal and green manure, inter cropping or guano have been reported to be used in agriculture for a long time and have been widely used in traditional agriculture systems until the appearance of N fertilizers during the Green Revolution in the 1950s. Nowadays, the most common organic amendments can be classified in five categories: Animal manure, municipal biosolids and sewage, green manure and crop residues, food residues and waste, waste from manufacturing processes and compost (Goss et al., 2013).

The transition to organic agriculture and the establishment of sustainable agricultural practices may enhance agricultural systems in long-term conditions, but, in already degraded soils where conventional crops are hard if not impossible to be cultivated, organic agriculture may not be sufficient to recover soil quality while maintaining economic viability for farmers (Raleigh and Urdal, 2007).

One alternative proposed to such situations is to revalorise degraded agricultural soils through the implementation of new crops/cultivars well adapted to adverse natural conditions and easy to manage. This is the case of wild edible plants (WEPs). WEPs are native plant species, able to grow in natural conditions without human intervention, traditionally used as a food source or as complementary ingredients in local recipes, or even as ´famine foods´. WEPs availability depends on the area and the environmental condition, being the cultivation of WEPs adapted to arid soils, scarcity of water and high temperatures in summer, the ones more interesting in the case of the Mediterranean basin.

Finding and successfully cultivating such WEPS might allow farmers to use their land thorough the cultivation of alternative/complementary plant species with sustainable practices, while recovering and improving the soil quality. Moreover, the valorization of WEPS may offer alternative – high value crops to the markets, due to the increasing interest over the last years for fresh, healthy and functional foods and the awareness of consumers who demand new products that enhance health combined with great gastronomic properties (Łuczaj et al., 2012; Ceccanti et al., 2018).

The main goal of this deliverable is to provide technical information regarding the temperature requirements for the sexual (seed) propagation of the species studied within the Valuefarm project e.g. *Scolymus hispanicus*, *Portulaca oleracea*, *Sonchus oleraceus*, *Cichorium spinosum* and *Crithmum maritimum*. Moreover, the best practice guides regarding the crop management requirements for the cultivation of the selected species are also provided.

PLANT DESCRIPTION

Spiny chicory (*Cichorium spinosum* L., Asteraceae) is a perennial herbaceous species commonly found in coastal and mountainous areas throughout the Mediterranean region, while it has been recently cultivated commercially as a leafy vegetable (Petropoulos et al., 2018). Its leaves are leathery-fleshy and thick and have a dark green color. They also form a spherical ground rosette, from the center of which the flowering stem of the plant grows in the spring and when it enters the reproductive phase. The flowering stem is a multi-branched inflorescence with branches that are smooth, have elongated grooves, while its upper part is spiny, blunt and leafless forming a spiny bush, hence its name "spiny chicory". During this phase (reproductive), plant height reaches 20-40 cm. It forms a taproot which can reach a depth of up to 30 cm. Its leaves are rampant and are generally (but not always) confined to the base of the shoots, 3-15 cm long, the lower ones lyreately pinnate or lanceolate, toothed with a bluntly oblong deltoid terminal lobe. Lateral lobes are usually toothed or entire. The base of the leaves is smooth with a very short petiole. The flower heads are small, with a narrow cylindrical sheath and with 5 blue flowers, which are either axillary, terminal or epiphytic and include several complete flowers, i.e. hermaphrodite, with a flowering period from June to August. After the fertilization of the flowers and the fall of their petals follows the fruiting stage of the peduncle, i.e. the creation of the syncarp, which usually includes 4-5 seeds. The seeds inside the fruits are 2.5 mm long and ovoid, with pointed apex and have a brown colour. During this period (fertilization of the flowers and beginning of fruit set) the onset of high temperatures results in the beginning of lignification of the flowering shoot, the fall and drying of the leaves, and the beginning of the lignification of the thorn.

Figure 1. Seeds of *Cichorium spinosum*.


Figure 2. Seedlings of *C. spinosum.*

Figure 3. The rosette of leaves of *C. spinosum.*

Figure 4. The formation of new leaves after harvesting the main rosette.

Figure 5. The development of the spiny inflorescence of *C. spinosum*; Left photo: the initiation of the formation of the inflorescence; Right photo: fully developed inflorescence.

Figure 6. *C. spinosum* plants in full blossom.

Figure 7. Wild *C. spinosum* plants.

Sea fennel (*Crithmum maritimum* L.) is a facultative and perennial halophyte, very common in several Mediterranean countries such as Greece, Tunisia and Spain (Jallali et al., 2012; Pereira et al., 2017; Renna & Gonnella, 2012). It is an edible and medicinal species which is commonly used in traditional dishes, while it usually grows within coastal areas, piers, rocks and sandy beaches (Renna & Gonnella, 2012). The edible parts of the species are its leaves, which can be consumed fresh as salad vegetables, or pickled, while Renna et al. (2017) have suggested the use of dried leaves for human consumption, as well as a coloring agent. Moreover, Siracusa et al. (2011) suggested the use of flower tops and stalks infusions as herbal teas, while Pereira et al. (2017) reported that all aerial parts can be an alternative source for health promoting beverages.

Figure 8. Seeds of *Crithmum maritimum.*

Figure 9. Seedling of *C. maritimum* before (left photo) and after transplantation (right photo).

Figure 10. The edible part of the leaves (left photo) and the shoot (right photo) of *C. maritimum*.

Figure 11. *C. maritimum* plants in full blossom (left photo) and mature inflorescence (right photo).

Figure 12. Wild plants of *C. maritimum.*

Common purslane (*Portulaca oleracea* L.) is considered a wild edible plant distributed worldwide, and one of the three most frequently reported weeds across the world. It is mainly present in the Mediterranean basin, Asia, the Caribbean, North America, México and Australia. It is an herbaceous succulent annual plant member of the Portulacaceae family (Miyanishi and Cavers, 1980). Purslane can complete its life cycle in 2 – 4 months and has the ability to re-root after hoeing when stems remain moist (Cutney and Elmore, 1999). It can also shift to C4 metabolism (under stress condition), and therefore has a high water use efficiency, making purslane a highly competitive alternative crop in arid lands, with scarcity of water and high temperatures conditions (Yazici et al., 2007; Ren et al., 2011; Jin et al., 2015, 2016). Such characteristics has led some to consider purslane as a ´food of the future´ (Simopoulos et al., 1995). The seeds of purslane are only 0.5 mm in diameter.

Figure 13. Seeds of *Portulaca oleracea.*

Figure 14. Young plants of *P. oleracea* cultivated in field conditions.

Figure 15. Shoots (left photo) and immature seed cups (right photo) of *P. oleracea*.

Figure 16. *P. oleracea* plants at the beginning of flowering period.

Common sowthistle (*Sonchus oleraceus* L.) is an annual and/or biennial herb, 40-150 cm tall, which contains white latex in its plant parts. First it forms a ground rosette and then develops the main stem (inflorescence) of up to 1.5 m in height and with a distinct strong taproot. The taproot is fleshy, upright with many branches, especially near the soil surface. The stem below the inflorescence is simple or lightly branched and glabrous or glandular. Leaves present different morphology depending on their position on the plant (basal and lower stem leaves are smaller than middle stem leaves, while middle and upper stem leaves are extremely variable, elliptic, oblanceolate, or lanceolate being slightly spiny. Leaves are dark, glossy green in the upper side and light green in the under side.The midribs and the petioles of the leaves may contain red pigments, while red specks may be present in leaf blades. Inflorescence is shortly corymbiform or racemiform, forming a few to several capitula. Each capitula produces numerous achenes, each with a tuft of fluffy white hairs or pappus. The flower-head consists of 27-35 lance-shaped bracts, 10-13 mm long and hairy while young. Each flower-head contains 80-250 flowers which are longer than the involucre. The flowers are yellow and the ligule is about as long as the corolla tub. Achenes are brown, with dimensions of 2.5-3.75 x 0.7-1 mm, oblanceolate. The seeds of *S. oleraceus* are small (100 seed weight is approximately 0.02 g). *S. oleraceous* can produce a substantial number of seeds which can be dispersed through wind. One plant may produce 4000-6000 seeds or more of low dormancy or non-dormant seeds (Hutchinson et al., 1984; Ciocârlan, 1990). Mature seeds are brown with white ribs and slightly serrated seed coats. According to the literature, there is a great variation in plant morphology and adaptation to environmental conditions, depending on the genotype (Olivier et al., 2020).

Figure 17. Seeds of *Sonchus oleraceus.*

Figure 18. Seedling of *S. oleraceus* in a seed tray.

Figure 19. Rosette of *S. oleraceus* leaves.

Figure 20. Plant of *S. oleraceus* whith fully developed inflorescence.

Figure 21. Immature inflorescence (left photo) and open flower (capitula) of *S. oleraceus* (right photo).

Common golden thistle or **Spanish oyster thistle** (*Scolymus hispanicus* L.), is a flowering plant in the genus *Scolymus* in the family Asteraceae, native to southern and western Europe, north to northwestern France. It is a herbaceous biennial or short-lived perennial species with a thick upright stem that grows up to 80 cm tall and has spiny stems and leaves and several branches and fins. It has a deep thick root which exudes a milky, bitter sap when is cut. Its leaves are soft, lanceolate, pinnate, toothed and spiny and have long stalks. The single axillary flowerheads are bright yellow to orange-yellow, 2–3 cm diameter and consist of many flowers. Each fruit contains many elongated, small seeds (achenia), with a formation of transparent fibers at the top, to facilitate their dispersal by wind.

Figure 22. Seeds of *Scolymus hispanicus*.

Figure 23. Seedlings of *S. hispanicus* in seed trays ready for transplantation.

Figure 24. Pot (left photo) and field grown (right photo) *S. hispanicus* plants.

Figure 25. Shoot (left photo) and inflorescence (right photo) of *S. hispanicus.*

Figure 26. Roots of transplanted *S. hispanicus* plant.

Figure 27. *S. hispanicus* plants in full blossom.

PROPAGATION

Purslane (*Portulaca oleracea*)

Two genotypes of purslane were tested in the Valuefarm project; one commercial cultivar obtained from Hortus Sementi Srl. (Budrio, Italy) and one Turkish genotype. The highest germination percentage was obtained 5 days after sowing at 20 °C for the Hortus genotype (98.3%), while for the Turkish genotype the same temperature was the most beneficial although with a slower germination rate (95% after 7 days).

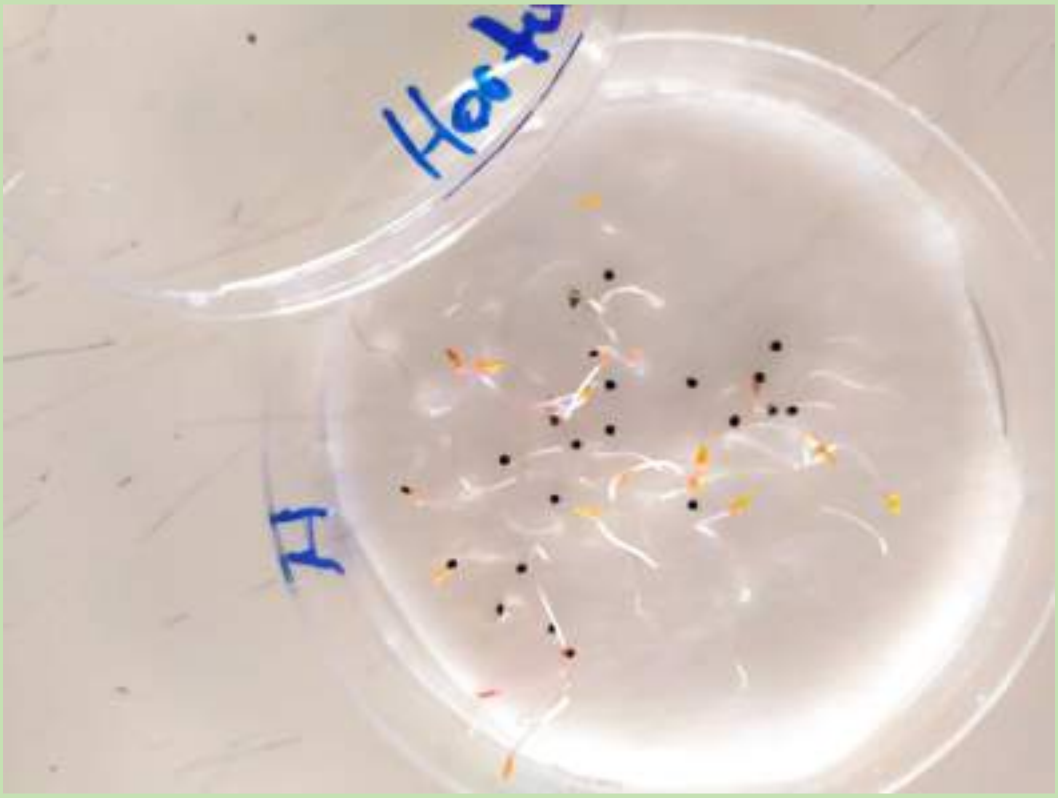

Figure 28. Germinated seeds of *P. oleracea*.

According to the literature, under suitable conditions, the germination rate can go up to 90% in 24 hours (Holm et al., 1977; Chauhan and Johnson, 2009). Purslane shows high variability on seed dormancy allowing plants to avoid the cold temperatures, while maintaining their vigour. Singh (1973) and Feng et al. (2015) collected seeds from India and China that showed dormancy but other studies did not report dormancy (Miyanishi and Cavers, 1980; Baskin and Baskin, 1987). Feng et al. (2015) tested the effect of long-term storage and obtained higher germination ratio in three year-long storage when seeds were stored at -20°C, while the storage temperature and its duration before sowing also had an effect on purslane germination; maximum germination percentage (68.4%) was obtained when seeds were kept at 45 °C for 60 days. Chauhan and Johnson (2009) found that germination of purslane is not affected by storage duration (up to 6 months) and that it is strongly stimulated by light, as only a small proportion of seeds were able to germinate in darkness, regardless of the temperature. In the presence of light, the germination rate ranged from 70% germination (25/15 °C; day/night temperature), 75% germination rate (35/25 °C; day/night temperature) and 81% germination rate (30/20 °C; day/night temperature) at different storage times (0 to 6 months) in a germination chamber, whereas in field conditions they reported seedling emergence of 17 – 20% when seeds were sown in the soil surface. Montoya-García et al., 2017 recorded similar germination rates, between 12.5 - 28.4%. Due to the need of light for germination, sowing depth is also an important factor, therefore maximum seedling emergence happens when seeds are sown at or near the soil surface and declines exponentially with increasing depth, being minimum at 1 cm of depth and 0% at 2 cm (Chauhan and Johnson, 2009; Feng et al., 2015). In contrast, Benvenuti, Macchia and Miele (2001) recorded low seedling emergence rates even at 6 cm. These differences may be be due to differences in soil structure or compaction, since the seeds are very small and the energy available for germination may not be enough to allow emergence in heavy or compacted soils.

Common sowthistle (*Sonchus oleraceus*)

Two genotypes of *Socnhus oleraceus* were tested; one commercial genotype obtained from Geniki Fytotechiniki S.A. (Greece) and one wild genotype type collected in Greece. In the case of wild genotype, the highest germination percentage was recorded for the temperature of 30 °C) at ten days after sowing (71.7%), while the seeds of the commercial genotype showed the highest germination percentage either at 15 °C (78.33%, 8 days after sowing) or at 25 °C (78.33%, 11 days after sowing).

Seeds of annual sowthistle were able to germinate over a broad range of temperatures (25/15, 20/12, and 15/9 °C day/night temperatures) (Chauhan et al., 2006; Manalli et al., 2018). Seed germination was favored by light; however, some germination occurred in the dark as well. More than 90% of seeds germinated at a low level of salinity (40 mM NaCl), while some seeds germinated even at 160 mM NaCl (7.5%). Seed germination was greater than 90% over a pH range of 5 to 8, but declined to 77% at pH 10. Seedling emergence was the greatest (77%) for seeds present on the soil surface but declined with sowing depth, and no seedlings emerged from a soil depth of 5 cm (Chauhan et al., 2006; Manalli et al., 2018). Although *S. oleraceous* is a major weed in the winter crops, it can flourish in the post-winter fallow phase and also in summer crops (Ali et al., 2020). Dormancy enables the species to survive any adverse environmental conditions; however, lack of primary dormancy and ability to germinate under varying environments allow *S. oleraceus* to emerge and spread by exploiting any favorable conditions (Widderick et al., 2010). Moreover, seeds of *S. oleraceus* were favoured to constant temperatures (15-22 °C) which increased germination rate compared to alternate temperatures (Masin et al., 2017), while rainfall (cumulative single or consecutive events) had also an effect on seed germination rate (Werth et al., 2017).

Spiny chicory (*Cichorium spinosum*)

One commercial genotype obtained from Geniki Fytotechiniki S.A. (Greece) was tested. Seeds showed very low germination percentage and very slow germination rate. The highest germination percentage was recorded at 25 °C, 25 days after sowing. These results indicate that spiny chicory has a low innate germination ability due to dormancy (extrnal or innate) or seed viability.

So far, no information is available regarding the temperature requirements for the germination of the species or regarding the existence of various forms of dormancy. The domesticated relative of the species (*C. intybus*) needs temperatures of 21 °C for successful germination (Bais et al., 2001), while environmental factors such as temperature, salinity, water availability, soil pH and sowing depth may also affect germination percentage (Vahabinia et al., 2019). Moreover, the growth rate of seedlings is very slow and usually at least 45 days are required before transplantation (Papafilippaki and Nikolaidis, 2020) or more (Chatzigianni et al., 2017).

Sea fennel (*Crithmum maritimum*)

Two wild genotypes of *Crithmum maritimum* were tested, one collected in Greece and another one collected in Turkey. The Greek genotype showed very low germination percentage and seeds germinated only at 15 °C (25%, 25 days after sowing), whereas at 10 °C only 3.3% of seeds germinated 22 days after sowing. On the other hand, the Turkish genotype showed better germination capacity and 93% of seeds germinated at 20 °C, 13 days after sowing. These results indicate that the genotype and the growing conditions of the mother plants may affect germination percentage of sea fennel.

Marchioni-Ortu and Bocchieri (1984), suggested the optimal conditions for seed germination of sea fennel is constant temperature of 20 °C, while Okusanya (1977) reported that alternate temperatures of 5 and 15°C, 5 and 25°C, and 15 and 25°C gave better results than constant temperatures. Strumia et al. (2020) evaluated the effect of sea water concentration in sowing medium and storage time of seeds on seed germination of sea fennel. According to the authors, seeds of *C. maritimum* showed a high germination percentage (85%) in distilled water and 100% saline water. Moreover, the same authors suggested that germination percentage decreased over time after a storage period of 4 months, from 85% to 50% (at 12 months after harvest).

Common golden thistle (*Scolymus hispanicus*)

Three wild genotypes of *Scolymus hispanicus*, collected in Greece, Spain and Turkey, were tested. Unfortunately, the seeds of the Spanish genotype did not germinate probably due to poor storage conditions or to seed viability. The Greek genotype presented the highest germination rate at 30 °C (73.3%, 16 days after sowing), while the Turkish genotype showed a higher germination percentage (87%) at 20 °C, 10 days after sowing. The resuts indicate high variability in temperature requirements for seed germination, depending on the genotype.

According to the literature, light conditions and temperatures may affect germination percentage of *S. maculatus*. In particular, Casciaro and Damato (2011) evaluated the effect of two light conditions (dark or 8 h of light) and eight constant or alternate temperatures (10 °C, 15 °C, 20 °C, 25 °C, 30 °C, 15 °C/5 °C, 20 °C/10 °C and 25 °C/15 °C). The authors suggested that although temperature did not influence seed germination (the average germination percentage was 28%), when temperature was constant at 20 °C the germination rates T_{25}, T_{50} and T_{75} were shorter and germination values were higher. Moreover, the presence of light positively affected the germination percentage (31 vs 24%), T_{50} and germination value. Moreover, Sari and Tutar (2009) evaluated the effect of light-dark, cold storage, and selected temperatures on germination of thistle seeds obtained from two wild and one commercial genotype. The authors reported that light and cold storage improved germination percentage, while positive effects were recorded at 20 °C and 25 °C, compared to lower (15 °C) or higher temperatures (30 °C).

SEED PRIMING

The results from the above seed germination tests indicate that apart from purslane, common thistle and golden common thistle, the other two studied species, namely *Cichorium spinosum* and *C. maritimum* showed very low germination percentage and slow germination rates. Therefore, besides testing seed germination at different temperatures, seed priming of *C. maritimum*, *C. spinosum* and *S. hispanicus* was also tested in order to identify compounds that can increase the innately low germination percentage which is essential for the establishment of commercial crops.

Atia et al. (2006) suggested the presence of salt-induced dormancy in *Crithmum maritimum* seeds and tested the priming of seeds with nitrate, thiouria, water, NaCl and PEG (polyethylene glycerol) 6000. In their study, the authors reported that germination was strongly inhibited by increasing salinity, while the addition of nitrate effectively alleviated salt-induced seed dormancy under both non- and saline and conditions; and thiourea improved germination only at moderate salt concentrations. Finally, PEG 6000 delayed germination in distilled water, while priming with both water and NaCl accelerated the germination process on salt free medium. Similarly, Meot-Duros et al. (2008) suggested that L-ascorbic acid (40 or 60 mM) and ethanol (96%) significantly improved germination rate of *C. maritimum* by 10, 30 and 30%, respectively. Atia et al. (2009a) reported that salt impact was amplified by darkness, but was mitigated by nitrate supply, red light and their combination, while germination was more influenced by the light type than by the PPFD.

Moreover, Nimac et al. (2018) primed seeds of *C. maritimum* with sodium chloride (NaCl) (50, 100 and 150 mM) or distilled water (dH_2O) and reported that seeds that were primed with dH_2O and 50 mM NaCl solution showed better performance than non-primed seeds. In another study, Atia et al. (2009b) evaluated the the effects of ABA, GA3,NO^{-3}, and NH^{+4} on the germination of *C. maritimum* under NaCl-salinity (up to 200 mM NaCl) and reported that NO^{-3} and GA^3 mitigate the NaCl-induced reduction of seed germination, whereas ABA inhibited germination under optimal conditions (0 mM of NaCl). Atia et al. (2010) suggested that seed germination was negatively correlated with seed K^+, Na^+ and Cl^- content, while the sponginess of fruit coat was associated with accumulation of Na^+ and Cl^- and protected seeds from the adverse effects of these ions.

Therefore, in the context of Valuefarm we tested seed priming with NaCl (0 (dH2O), 50 and 100 mM NaCl), GA3 (10, 50 and 100 μM GA3) and ascorbic acid (50 and 100 mg of ascorbic acid) under controlled conditions (18-23 ˚C, 16 h day/8 h night), while a control treatment (untreated seeds) was also included. Seed priming has been already tested in seeds of conventional crops, however, to the best of our knowledge it is the first time that this technique is implemeneted in the seeds of most of the selected wild edible plants.

Seed priming of two *S. hispanicus* genotypes (Greek and Spanish genotype) with 100 μM of GA_3 improved germination percentage over the control treatment (77.5% and 60%, respectively) in Greek genotype, while the same treatment increased germination rate since the highest germination percentage was obtained at 3 days after germination initiation (77.5% and 35%, for the GA_3 and the control treatment respectively). However, a variable effect was recorded depending on the genotype, since the germination of the Spanish genotype was benefited from the distilled water (55%, at 7 days after seed germination initiation), although the control treatment recorded the highest overall germination percentage (70%).

In the case of *C. maritimum*, seed priming with 50 μM of GA_3 improved germination percentage over the control (30% vs 17.5%), followed by the application of 50 mM of NaCl (27.5%), although no effect on the germination rate was detected.

Finally, germination percentage of *C. spinosum* was significantly improved over the control treatment (35%) when seeds were primed with 10 μM of GA_3 (62.5%) and 100 mg/L of L-ascorbic acid (52.5%). The highest germination percentages were recorded 6 days after germination initiation.

Apart from seed priming, *in vitro* propagation has been suggested for *C. maritimum* as a means to overcome low germination percentage and slow germination rate (Grigoriadou and Maloupa, 2008). Different culture media have been used for the *in vitro* culture of other species of the family of Apiaceae, while shoot production of *C. maritimum* was significantly induced when shoot tip explants were cultured in MS medium. This particular medium seems to be the most effective for the *in vitro* cultivation of the species since it significantly increased the number of new microshoots produced / explant, as well as the shoot height (Grigoriadou and Maloupa, 2008).

CULTIVATION PRACTICES

Spiny chicory (*Cichorium spinosum*)

Spiny chicory is a wild halophyte, commonly found in coastal areas, which is used for its edible tender leaves. Plants in the wild are perennial and develop new shoots and rosettes of leaves every autumn. The growth cycle is completed at the end of spring or in the summer through the formation of the spiny inflorescence which has the form of a small spiny bush.

For commercial cultivation, seeds can be sown in the autumn or early in the spring depending on the climate conditions and frost incidences. Seeds can be sown directly in the field or in seed trays and then transplanted in rows in the field at distances of 30 cm within the row and 50 cm between the rows (approximately 65000 plants/ha). If direct sowing is applied, it is better to mill fruit prior to sowing in order to separate the seeds from the fruit and facilitate seed germination. Transplantation is the best option since it ensures uniform development of plants by selecting the best developed seedlings, is lowers the amounts of seeds required and minimizes the gaps in the field due to germination failure. It can be cultivated as an annual or as a perennial plant.

Crop management includes soil preparation prior to sowing which must be done in a depth of 1-2 cm. Nutrient management requires a base dressing with a complex fertilizer (12-12-17 or 14-7-14, N-P-K). High amounts of nitrogen should be avoided to reduce the risk of increased nitrates content in the final product. Moreover, nitrogen form (nitrate or ammonium nitrogen) and their corresponding ratio may affect the chemical composition and the yield of the final product. Organic cultivation is also possible due to low nutrients requirements of the species. Irrigation should be performed with a drip irrigation system on a regular basis, depending on the growing period and the climate conditions.

Several harvests can be carried out throughout the growth cycle of the species by cutting rosettes of leaves with a sharp knife at the upper part of hypocotyl without disturbing the auxiliary buds at the apex of the meristem. Successive harvests are continued starting 3-4 months after sowing and until plants enter the reproductive stage and the inflorescence is developed in the center of the rosette. After that point, leaves are not edible because are withered and become less tender.

Apart from soil cultivation, hydroponic cropping systems have been also suggested with higher yields due to optimal growth conditions which result in faster growth and more harvests throughout the growth cycle. Greenhouse soil or pot cultivation is also an option for out of season production which can improve the product availability throughout the year and cover the increased market needs for healthy and functional food products.

Yield of fresh edible leaves can be as high as 20 tons/ha or higher, depending on the climate conditions and the number of harvests. In pot cultivation, the yield can reach 60 ton/ha when 3 harvests are applied.

Purslane (*Portulaca olearacea*)

Purslane is an annual herb with prostrate growth, which can be found as a weed in commercial crops throughout the world. It is commonly used for its edible fleshy stems and leaves which are considered the richest plant source of omega-3 (α-linolenic acid) fatty acids. It has a short growth cycle which is completed in the summer or early in the autumn, depending on the climate. In some areas it is cultivated as a minor or orphan crop and distributed in local markets, however its commercial cultivation is not wide-spread so far since it is mostly treated as a troublesome weed. Commercial cultivation is essential for covering marketing needs, while it ensures the safety of the final product since wild plants are usually collected in commercial fields where pesticide residuals may be present.

Propagation is carried out with seeds, although tissue culture and propagation with cuttings can be also applied. Considering the short cycle of the species, it can be cultivated either twice within the same growing period when climate is mild (no late frosts in the spring and early frosts in the autumn), starting with sowing early in the spring and performing a second sowing in the mid-summer; or apply 1-2 successive harvests by cutting the marketable stems and allow the rest of the plant to regrow. For this purpose, commercial cultivars are available with plants having an erect growth habit which facilitates successive harvests or even mechanization of harvesting.

Crop management includes soil preparation prior to sowing which must be done in a depth of 0.5-1 cm in equidistant rows at 30 cm. Therefore, soil must be plowed moldboard to a depth of 20-25 cm followed by two rotary-harrowings. Irrigation with a sprinkler irrigation system is essential after sowing to keep soil surface moist and allow seed germination and seedling emergence (5-7 days after sowing). However, after seedling emergence irrigation should be performed with a drip irrigation system on a regular basis, since the species is susceptible to white rust and wetting of foliage could increase the disease incidence.

The plant has no high nutrient requirements. Base dressing should include the application of manure (up to 2-3 ton/ha) and nitrogen in the form of urea (40 kg/ha) or a complete fertilizer (10-10-10 N-P-K) at 100 kg/ha. Otherwise, fertigation at regular intervals with a nutrient solution that contains N-P-K in a ratio of 3:1:1 and up to 6:1:1 can be applied.

Harvesting is performed by cutting stems just above soil surface with a sharp knife before flower initiation (30-45 days after sowing, depending on the conditions and the genotype) or at the stage of 14-16 true leaves. Multiple harvests can be applied.

Hydroponic cultivation can be also applied allowing higher yields, better availability of the product throughout the year and cleaner products which are ready to market, especially when prostrate genotypes are cultivated.

Yield of fresh biomass ranges from 15 to 30 tons/ha, while maximum yields of 50 tons/ha have been also obtained.

Sea fennel (*Crithmum maritimum*)

Sea fennel is another perennial halophyte of the Mediterranean basin which is commonly used for its tender edible leaves or for the essential oils of its seeds and aerial plant parts. Ground seeds can be also used as salt substitute.

Propagation is carried out with seeds; however, apart from sexual propagation with seeds, *in vitro* propagation with cuttings is also suggested allowing the production of high numbers of seedlings with uniform development and characteristics identical to the mother plant. In the case of sowing, the low germination rates and the slow seedling growth necessitate the sowing of seeds in seed trays and the transplantation of seedlings in the field. Plants develop new shoots at the beginning of spring and as soon as the temperatures rise, while the growth cycle ends in the autumn with the fruit maturation and withering of the plants. Sowing should be performed early in the spring in the autumn, unless seeds are sown in seed trays in heated nurseries for seedling production, when sowing should start 2-3 months earlier due to slow growth rates. Plant is cultivated as a perennial species.

Crop management includes soil preparation prior to sowing or seedling transplantation. Irrigation should be performed with a drip irrigation system on a regular basis; however, water requirements are considered very low, although water availability increases the fresh biomass yield. For nutrients management, the application of a complex fertilizer (11-15-15, N-P-K) at 500 kg/ha or liquid fertilizers (25 kg of N/ha) can be applied.

Plants are arranged in rows with a distance of 30 cm within the row and 60-80 cm between the rows, which allows the unhampered growth of plants (plant density of 41000-55000 plants/ha). Apart from soil cultivation, plants can be grown in pots under controlled environments in various growth substrates (e.g. peat, perlite, vermiculite etc.) or in hydroponic systems where plant density may increase up to 230 plants/m^2.

Plants intended for the production of fresh leaves are harvested before the reproduction phase, while for other purposes such as the essential production plants should be harvested when seeds are mature. Multiple harvests of leaves can be performed throughout the growing period.

Yield of harvested fresh total biomass for soil cultivation ranges between 10-13 tons/ha (for a single harvest), while for pot cultivation under greenhouse conditions and multiple harvests fresh yield can reach 55 tons/ha, depending on the fertilization regime.

Common thistle (*Sonchus oleraceus*)

Common thistle is a widespread weed which can be used for its edible leaves, seeds and flowers. Propagation is carried out with seeds which are sown in a depth of 0.5-1 cm, while sowing is carried out early in the spring or in the autumn. Plant density should be around 100000-110000 plants/ha (plant distances of 30 cm x 30 cm). Commercial cultivation is essential for covering marketing needs, while it ensures the safety of the final product since wild plants are usually collected in commercial fields where pesticide residuals may be present. It can be grown throught the year due to the broad range of temperatures that seeds can germinate, while soil moisture is also an important factor for seed germination. Hydroponic cultivation is also suggested for higher yields and better availability of fresh leaves throughout the project.

So far there are no specific guidelines for the cultivation of the species. Valuefarm results showed that the application of fertilizers with a nutrient solution that contained nutrients in a ratio of 3:1:1 or 6:1:1 of N-P-K was beneficial for fresh biomass yield. A base dressing with 60 kg/ha of N and 30 kg/h of P_2O_5 is also suggested. Drip irrigation is suggested for covering water requirements of the species, although rain fed cropping is also applicable with proper selection of the time of sowing.

Harvest should be performed before the formation of the inflorescence by cutting the rosette of leaves with a sharp knife.

Total yield of fresh biomass can be higher than 13-15 tons/ha, depending on the growing season and the fertilization regime.

Common golden thistle (*Scolymus hispanicus*)

Common golden thistle is a prickly perennial herb with a wide distribution in the Mediterannean basin, which can be found in uncultivated agricultural fields, weed areas, roadsides etc.

Propagation is carried out with seeds which can be sown directly in the field or with transplantation of seedlings. However, when plants are grown for the production of leaves and roots, transplantation should be avoided since it results in malformed roots. Sowing should take place early in the spring or in the autumn, depending on the climate conditions. Plants can be cultivated as annual or perennial. Plant density should be around 65000 plants/ha (plant distances of 30 cm x 50 cm) under field conditions or 40000 plants/ha for hydroponic cultivation.

Crop management includes fertilization with a complex fertilizer (20-20-20, N-P-K) or fertigation with a nuutrient solution that contains nutrients in a amounts of 300 mg/L (N-P-K). Drip or sprinkler irrigation is suggested for covering water requirements of the species, although rain fed cropping is also applicable with proper selection of the time of sowing. Apart from open field cultivation, hydroponic systems are also suggested using various substrates such as perlite or coir.

Harvesting is performed before the formation of the inflorescence, while a single cutting is implemented in each growing period (in perennial plants). Apart from the leaves, roots are also edible and can be harvested at the end of the growing period increasing the added value of the crop with a dual purpose cultivation (leaves and roots).

Total yield of fresh aerial biomass of 6-7.5 tons/ha can be achieved in pot cultivation, depending on the growing period and the fertilization regime, while root biomass yield ranged between 4.7 and 6.5 tons/ha. Similarly, high biomass yields (6-7.8 ton/ha and 8.9-14.5 tons/ha, for leaves and roots, respectively) were also achieved under field conditions, depending on the irrigation schedule.

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