PRIMA
PARTNERSHIP FOR RESEARCH AND INNOVATION IN THE MEDITERRANEAN AREA

AgrEcoMed
FOSTERING AGROECOLOGICAL TRANSITION

"New AGRoecological approach for soil fertility and biodiversity restoration to improve ECOnomic and social resilience of MEDiterranean farming systems"

Deliverable **D2.1**

Identification of medicinal plants suitable for agroecological cultivation in rotation with cereals

This project is part of the PRIMA Programme supported by the European Union, grant agreement PRIMA21_00018

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Acronym and abbreviations

AgrEcoMed	New AGRoecological approach for soil fertility and biodiversity restoration to improve ECOnomic and social resilience of MEDiterranean farming systems
PRIMA	Partnership for Research and Innovation in the Mediterranean Area
MEL	Monitoring, Evaluation and Learning platform
WP	Work package

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Executive summary

The AgrEcoMed project, funded under the European Union's Horizon 2020 research and innovation program PRIMA and Grant Agreement PRIMA21_00018 is a research project aimed to fill the research gaps for implementing a biodiversity-based strategy for primary crops as cereal farming systems through an Agroecological approach adapted to environments in Mediterranean countries, efficient use of natural resources, reduction of pollution, circular economy. Such a goal will be achieved through innovative approaches to support the sustainable production of staple foods in the scenario of the present and future climate changes. To support the strategic coordination and overall management structure of this project, a project execution plan is essential. This document is Deliverable 2.1 "Identification of medicinal plants suitable for agroecological cultivation in rotation with cereals". The main objective of the activities carried out was to implement scientific research activities aimed at enhancing biodiversity for primary crops, such as cereal cultivation systems, through an agroecological approach that can increase the cultivation and screening of alternative species, such as medicinal plants, to be used in large and rational crop rotations.

Keywords: PRIMA, AgrEcoMed, dissemination, medicinal plants.

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Introduction

A challenge that engages the entire agricultural scientific community is to increase the sustainability of the agri-food system through environmental and social actions that draw on an ecological approach. In order for the agroecological approach to truly be implemented and produce income, it must go beyond the purely ecological and agronomic aspect, and emphasize a strong territorial connotation that also encompasses the social, economic and cultural dimensions, as well as the politics of the agrifood system. To facilitate this new approach and consequent transition, it is necessary for all actors to be able to share a single strategy that starts from a set of sustainable practices to redesign the agroecosystem. This new agroecological strategy cannot and should not ignore the different forms of agriculture, from organic to conventional, that can interact with each other and push for the adoption of more sustainable cropping practices by farmers, especially conventional ones. In fact, the success of agroecological practice can only start from the experiences arising from organic management models and the adoption of agroecological cultivation practices developed in organic systems that are a guideline toward the “agroecological transition” of farming practice.

The principles on which the agroecological transition is based are: the enhancement of biodiversity, the efficient use of resources involved in the agrifood chain, the control of the environmental impact of agricultural systems, and the strengthening of the knowledge system and cooperation among farmers. In agricultural systems, the enhancement of biodiversity is closely linked to the planning of extensive and rational crop rotations including with alternative species such as medicinal plants and the enhancement of local genotypes and the restoration of cultivated biodiversity.

The activities carried out as part of the self-employment assignment under the AgrEcoMed project were primarily aimed at implementing scientific research activities aimed at enhancing biodiversity for primary crops, such as cereal cropping systems, through an agroecological approach that can increase the cultivation and screening of alternative species, such as medicinal plants, for use in broad and rational crop rotations.

Diversification of cropping systems is a key element in the agro-ecological transition, useful for ensuring food security while conserving and preserving natural resources. In fact, planning new crop rotations, larger than in the past, and the introduction of new crops (such as medicinal plants) is a possible agronomic innovation that can maximize the positive effects of crop rotation in terms of

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soil fertility, pathogen and pest control, plant nutrition management, and resilience to climate change.

In this context, a preventive screening of alternative officinal species that best interpret and adapt to the relevant territorial context as identified by the AgroEcoMed project is useful. The focus on these species is also due to the possibility of extracting bioherbicides and biopesticides that are well suited to the objectives of agroecology, weed control, and pests more generally.

In the AgroEcoMed project the choice of cereal crops, and in particular autumn-wheat, was dictated by the fact that they are well adapted to the semi-arid climatic conditions of the Mediterranean and are able to provide stable economic support to farmers.

In this context of change, having to pursue yield stability under sustainable agricultural production conditions, it is essential, not only, to use crop varieties that are better adapted to specific environments (greater resistance to abiotic and biotic stresses and maintain yields under adverse or low-input conditions) but also to identify crop rotations capable of preserving the production environment and healthy yields. The most promising species in this regard are officinal species.

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Officinal Plants identified for rotations with cereals

Interest in medicinal plants and their derivatives has grown steadily in recent years. From a sustainable perspective, medicinal plants have an important role to play when considered as possible new sources of molecules, of natural origin, that are highly effective and potentially useful in addressing ongoing infectious emergencies in human, veterinary, and agricultural fields. Agricultural production, according to an agro-ecological approach, cannot do without alternative molecules of natural origin that are useful in ensuring plant production free of toxic residues and defending plants from harmful organisms.

The work on the identification of medicinal species, required by WP 5 "Strengthening the knowledge system and cooperation among farmers," was based on scientific literature and the results of research and experimentation carried out in the territory of Bagnoli del Trigno (IS) experimental field of the AgrEcoMed project.

For agroecology objectives, medicinal plants to be included in crop rotations with cereal crops must perform potential herbicidal, antimicrobial, antifungal and insecticidal activities useful in organic farming. Therefore, a series of arguments on the characteristics of the soil and climate environment and the adaptive abilities of the selected species (suitability) were considered. In particular, adaptation to soil type, tolerance to environmental stresses, and ability to produce in marginal agricultural ecosystems were considered. Qualitative and functional characterization for industrial production and evaluation of potential toxic effects against broader pests were also included in this screening activity of medicinal crops.

The medicinal plants identified and placed in cultivation are: basil, annual wormwood, oregano, helichrysum, rosemary, wild fennel, juniper, rapeseed, coriander, and garlic. A fact sheet on botanical, morphological, cultivation and chemical peculiarities was prepared for each species. As readily apparent, there is little information available for some species due to little or no open-field cultivation and the consequent lack of objective cultivation information.

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Basil

COMMON NAME: Basil

SCIENTIFIC NAME: *Ocimum basilicum* L.

FAMILY: Lamiaceae

www.actaplantarum.org/, photo by Mauro Ottonello

www.actaplantarum.org/, photo by Franco Rossi

Basil is an herbaceous plant characterized by an erect branched stem, can reach a height of about 50 cm. It has petiolate and opposite, oval or oval-lanceolate leaves with a surface is glossy and glabrous. The flowers consist of an inflorescence formed by racemes with the flowers clustered in whorls arising above a pair of leaves. Each flower consists of a tubular calyx. The fruits are 4 achenes enclosed in the calyx, dark brown in color and oval in shape.

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The plant name “basil” comes from medieval Latin “basilicum” which originates from the Greek “basilikon” or “royal, majestic plant” which in turn originates from “basileus” or “king.” Some historians believe that it is so called because it was used in the creation of perfumes for the king.

The use of basil is lost in the mists of time. In folk tradition, basil leaves were used to stimulate lactation and as an aphrodisiac remedy. In addition, leaf infusion can be used as a mouthwash to combat bad breath. It also possesses properties:stimulant, antispasmodic, diuretic, tonic-digestive, antiseptic and anti-inflammatory.

Basil essential oil shows purifying and anti-microbial properties, also being effective as a repellent for some insects. These activities can be attributed to the presence in the oil of major compounds such as: linalool, estragole, citral, cinnamate, eugenol, methyleugenol, geranyl acetate, geraniol, 1,8-cineole, alpha-bulnesene, bergamotene, alpha-murolol, beta-caryophyllene, cadinene and alpha-terpineol.

Basil is native to the warm areas of Asia, and was introduced to Italy by the ancient Romans. Precisely because of its tropical and sub-tropical origin, it is not found wild but only cultivated.

The cultivation cycle usually begins with a sowing in a seedbed in March-April. When the seedlings are developed they can be transplanted to larger pots or to open fields in sunny locations. Basil is a demanding crop in terms of water availability and, at the same time, very sensitive to waterlogging. As with other vegetable species, basil productivity benefits from topping to promote tillering and avoid flowering too early.

Regarding nutrient requirements, basil benefits from organic or mineral (NPK) fertilization at planting and with subsequent cover applications with fast-acting nitrogenous forms (nitric and nitricoammoniacal).

Basil leaves are harvested from May to September. The flowering tops are harvested from July to September. The leaves and tops can be dried in the shade in a ventilated place and are stored in glass jars away from light.

In addition to the essential oils, the leaves and flowering tops also contain: hydroxycinnamic acid derivatives (rosmarinic acid), flavonoids (kemferol and oak heterosides, xanthomicrool, nevadensin, salvigenin), sterols and triterpenes (beta-sitosterol and oleanolic acid) and lipids (linolenic acid in the seeds).

The main compounds in the essential oil are linalool, estragole, citral, eugenol, methyleugenol, geranyl acetate, geraniol, 1,8-cineole, bergamotene, beta-caryophyllene and alpha-terpineol.

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Oregano

COMMON NAME: Oregano

SCIENTIFIC NAME: *Origanum vulgare* L.

FAMILY: Lamiaceae

www.actaplantarum.org/, photo by Franz Neidl

www.actaplantarum.org/, photo by Giorgio Faggi

Oregano is a perennial herbaceous plant with horizontally creeping rhizome from which stems grow, up to 70 cm tall, has dark green, opposite, ovate-lanceolate, spicy leaves. The flowers, pinkish or purplish, are clustered in glomerules forming corymbose inflorescences. The fruit consists of 4

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cylindrical, brown, smooth-surfaced achenes enclosed in the calyx.

Oregano is a plant native to Europe and western Asia. In Italy it is found in almost all regions, from the plains to 1,300 meters above sea level.

Oregano is a plant that prefers sunny conditions with well-exposed soils to promote flowering. It prefers loose,sandy and calcareous soils. It suffers from waterlogging. This plant can be propagated by spring sowing or by division of the trained plants during spring or after flowering.

Like other aromatics similar in size and needs, it is useful to do a plowing in the fall to a depth of about 30 cm, followed by appropriate repassing to prepare the transplant bed. Some weeding to control weeds is necessary during the first year. The crop needs relief irrigation for good rooting and during the summer period.

Regarding nutrition fertilization, it is useful to make an organic or mineral fertilizer at planting and distribute nitrogen in cover crops.

Of the oregano plant, the leaves and flowering ends are harvested at the beginning of flowering and dried in a shady and ventilated place

Next to the common oregano, very common in southern Italy and the islands is Origanum heracleoticum L., also called the southern oregano, which is very fragrant.

Oregano has numerous therapeutic properties, such as: digestive, carminative and tonic-stimulant. The leaf biomass contains: hydroxycinnamic acid derivatives (caffeic acid, rosmarinic acid, etc.) and flavonoids (luteolin heterosides, narigenin and apigenin).

The essential oil contains as main components: carvacrol, terpinene, cymene, pinene, myrcene, thymol, fellandrene, caryophyllene, elemene, linalool, germacrene and terpineol.

Oregano has interesting activities in both pest control (Lo Pinto et al,. 2022) and post-harvest management of agricultural products as a potent bactericide (Xylia et al,. 2019).

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Juniper

COMMON NAME: Juniper

SCIENTIFIC NAME: *Juniperus communis* L.

FAMILY: Cupressaceae

www.actaplantarum.org/, photo by Aldo De Bastiani

www.actaplantarum.org/, photo by Anja Michelucci

Juniper is an evergreen shrub native to mountainous areas of Europe, Asia and North America. Up to 3 meters tall, it has winding, branched stems with reddish-brown bark. The leaves are needle-shaped, prickly, verticillate in threes. It is a dioecious species, with male plants with inconspicuous, yellowish flowers that appear in May-June; and female plants with three-scaled flowers that turn

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into spherical, fleshy “berries” (botanically called “galbules”), first green and blue-purple when ripe, which are completed in 2-3 years, and covered with an opaque patina, on which the three scales are visible at the apex.

Juniper berries should be harvested when ripe and then placed to dry in a shady, dry and ventilated place. The berries contain: essential oil, flavonoids, nitrogenous substances, resins and tannins. These compounds give juniper berries properties such as: soothing, anti-inflammatory, anti-rheumatic, diuretic, liver protective, choleretic and cholagogue (stimulates bile production and secretion), balsamic and expectorant, digestive and carminative.

The essential oil contains as major components: camphene, alpha-pinene, beta-phellandrene, myrcene, sabinene, limonene, linalool, humulene and germacrene D.

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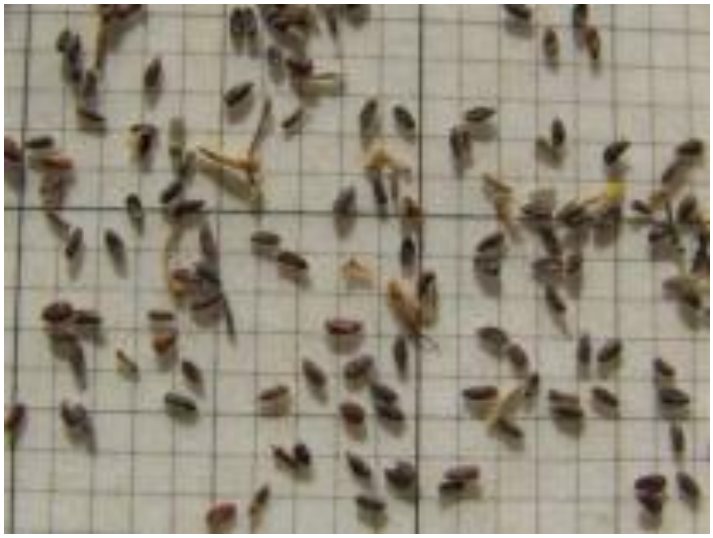
Helichrysum

COMMON NAME: Helichrysum

SCIENTIFIC NAME: *Helichrysum italicum* (Roth) Don

FAMILYA: Asteraceae

www.actaplantarum.org/, photo by Franco Rossi



www.actaplantarum.org/, photo by Franco Rossi

Helichrysum is a small shrub native to southern Europe. It grows in dry, sandy places and in uncultivated land, from flat to 800 meters above sea level; in Italy it is widespread almost everywhere.

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The suffruticose plant, can reach 50 cm in height, with silvery, linear-filiform leaves; the flower heads, bright golden in color, are united in a dense corymbose inflorescence; the fruits are shiny, white, cylindrical in shape.

Helichrysum can be sown in spring in sandy loam and, when the seedlings are sufficiently developed, transplanted into open fields or large pots placed in the sun. After harvesting, branches that are too long should be pruned back to even out the foliage and shelter the plant from the cold in the winter season. Helichrysum can also be propagated by division of heads or by tip cuttings.

It is a thermophilic plant that prefers sunny conditions with well-exposed soils to promote flowering. It shows good development and production on skeleton-rich, loose, calcareous soils. It suffers from waterlogging. Propagation can be by division of heads or by seed. Preparation of the transplant bed involves plowing in the fall 30 cm deep, followed by appropriate repassing. In cover, some weeding is useful for weed control.

Helichrysum benefits from relief irrigation at transplanting, to encourage rooting, and during the summer period. Organic or mineral fertilization with NPK should be carried out at planting, while ready-to-use nitrogen fertilizers will be used in spring.

On average, after the first less productive year, one hectare produces about 8-10 tons of fresh biomass consisting mainly of flowering tops. Harvesting is done in full bloom by removing the flowering twigs followed by drying in a cool, sunny place.

The helichrysum plant has the following therapeutic properties: sudorific, astringent, antiasthmatic and, for external use, as an antineuralgic soother. The main components of the helichrysum drug are: flavonoids, essential oil, phthalides, umbelliferone, scopoletin, esculetin, pyranoderivatives, campesterol, beta-sitosterol glucuronic acid, elipyron and alpha-pyron derivatives, sesquiterpene lactones and caffeic acid.

The essential oil contains as major components: nerol, neryl acetate, linalool and neryl-propionate. Helichrysum boasts repellent properties against harmful insects (Ninčević et al,. 2019) and herbicides (Karalija et al,. 2020), as well as fungicidal properties (Djihane et al,. 2017).

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Rosemary

COMMON NAME: Rosemary

SCIENTIFIC NAME: *Rosmarinus officinalis* L.

FAMILY: Lamiaceae

www.actaplantarum.org/, photo by Andrea Mologni

www.actaplantarum.org/, photo by Franco Rossi

Rosemary is an evergreen shrub native to Mediterranean regions. In Italy it is found throughout the territory, wild or cultivated, from the plains to 800 meters above sea level.

The characteristically aromatic, evergreen plant is compact, with prostrate or ascending, branching

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stems. The leaves are needle-shaped, opposite and turned at the margin, resinous. Flowers appear in spring, are blue-purple, pollen-rich and clustered in short axillary racemes.

Rosemary can be sown in early spring in a protected place at about 20°C. Seeds germinate after 1-2 weeks. Seedlings tend to wilt if the soil is too moist. Alternatively, propagation by cuttings (taken after flowering) or by offshoots is possible. Plants should be placed in sunny areas in acidic, well-drained soil. It grows well in pots and is sensitive to winter cold.

Preparation of the transplant bed involves fall plowing 30 cm deep, followed by proper soil refinement. Under cover, some weeding to control weeds is useful.

Rosemary benefits from relief irrigation at transplanting, to encourage rooting, and during the summer period. Organic or mineral fertilization with NPK should be carried out at planting, while ready-to-use nitrogen fertilizers will be used in spring.

Of the rosemary plant, the leaves and flowering tops are harvested by cutting off the apical part of the twigs. If they are not used immediately, it is advisable to dry them in a shady and ventilated place.

Rosemary has the following therapeutic properties: stomachic, stimulant, aperitive, digestive, tonic-stimulant, antioxidant and antiseptic. The main components are: essential oil, polyphenols (rosmarinic acid), flavonoids and tannins.

The essential oil contains as main components: 1,8-cineole, α -pinene, β-pinene, camphor, verbenone, camphene, borneol, β-caryophyllene, p-cymene, limonene, linalool and myrcene.

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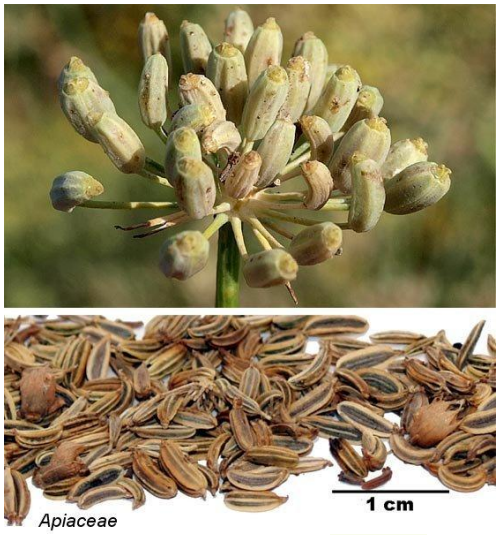
Wild fennel

COMMON NAME: Wild fennel

SCIENTIFIC NAME: *Foeniculum vulgare* Mill.

FAMILY: Ombrelliferae

www.actaplantarum.org/, photo by Erina Montoleone



Apiaceae

www.actaplantarum.org/, photo by Vito Buono

Wild fennel is a perennial herbaceous plant native to Mediterranean regions. In Italy it is especially common along coastal areas, from the plains to 1,000 meters above sea level.

Wild fennel is a rhizomatous plant with erect, branched stems up to 150 cm tall. The leaves are 3-4

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pinnate, divided into almost capillary laciniae. The yellow flowers are clustered in broad umbels. The fruit is a very aromatic achene.

Given its ecophysiological requirements, fennel is grown in open fields. It is sown on fertile soil free of waterlogging. Adult plants can also be propagated by division of heads in spring. There are particularly decorative and equally aromatic varieties.

Soil praration precedes a plowing in the fall to a depth of 40 cm, followed by repassing to refine the soil. During the first year, some weeding and topping to stimulate tillering are useful. The use of support stakes and wire to hold stems in the row before umbel formation is recommended to facilitate cultivation and harvesting. Fennelgrass is undemanding in irrigation, although some summer relief may be useful to ensure seed filling. Fertilization at planting should be done with organic fertilizers and with limited cover applications with nitrate and nitric-ammonium fertilizers. Fennel enters production as early as the first year of cultivation. Harvesting occurs in a staggered manner starting in the first decade of October with the first flowers beginning to dry and continues throughout the month. Umbrella production can reach 1, 1.5 tons per hectare.

The parts used of the plant are the leaves, for fresh consumption, and the umbels, which should be harvested when the fruits are almost ripe and dried in the shade in a dry place. When they are completely dry they can be separated from the ripe seeds.

Wild fennel has several therapeutic properties among which we can mention those: depurative, tonic-aperitive, carminative, antiseptic and antispasmodic. The flowering tops contain the following: essential oil (anethole, estragole, fencone, etc.), flavonoids (kemferol, quercetin), p-hydroxycinnamic acid, chlorogenic acid, caffeic acid, etc.

The essential oil contains as main components: anethole, estragole, fencone, limonene, camphene, α-pinene, β-pinene, α-tujene, α-phenchene, 3-carene, sabinene, α-fellandrene, myrcene, α- and β-terpinene, cis- and trans-ocimene, terpinolene and p-cymene.

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Oilseed rape

COMMON NAME: Oilseed rape

SCIENTIFIC NAME: *Brassica napus* L. var. Oleifera D.C.

FAMILY: Brassicaceae

www.actaplantarum.org/, photo by Franco Rossi

www.actaplantarum.org/, photo by Franco Rossi

Oilseed rape is an annual herbaceous plant, with taproot and erect stem 0.5 m to 1.3 m tall, much branched. The leaves, glaucous and pruinose, are simple; the lower ones are lyrate-pinnate and pedunculate, while the upper ones are sessile, oblong and partially amplexiculate. The flowers are clustered at the top of the stem; they have 4 sepals and 4 petals arranged in a cross and are yellow.

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The ovary is bicarpellar; the fruit is a siliqua containing 20-30 seeds, more or less dehiscent at maturity; they are formed by self-fertilization or through cross-fertilization. Seeds are round, red-brown to black (1,000-seed weight 3.5-4.5 grams).

The origins of rapeseed are uncertain. Today it is grown mainly in China, India, Canada and central Europe. In Italy it is found mainly in the north both as a grass forage crop in summer-autumn sowing and for grain production. The seed contains on average 45% oil, 25% protein, 5-7% fiber, 4-8% glucosinolates.

While not particularly cold-hardy, it can grow in cold environments as long as the plant reaches the 6-8 leaflet stage before winter, as it has the greatest resistance to cold at that stage. It prefers fresh, deep soils. In soils with good water retention capacity, rapeseed grows rapidly; it also grows well in rainfall-poor areas due to its greater earliness than winter cereals. It is fairly tolerant of pH, although it prefers values around 6.5; it has no particular problems with salinity. Varietally, there are winter cultivars on the market, with vernalization requirements, and cultivars that are distinguished by their toxic erucic acid and glucosinolate content.

Oilseed rape is an autumn-spring cycle plant; it improves the soil because of its abundant crop residues (roots, leaves and stems), which, if well buried, provide a good supply of organic matter. Preparing a good seedbed is important to manage the small seed size. In some dry environments and to ensure uniformity in emergence; rolling is useful.

Because the taproot may deepen, it is necessary to avoid soil compaction, which must be managed with appropriate deep tillage.

In northern Italy, planting is done in September; in the south until November, depending also on the possibility of preparing the seedbed. The optimum density is 70-80 plants per square meter. Row spacing varies from 25 to 35 cm. Despite the fairly deep root system, oilseed rape requires fair amounts of nutrients. The elements most removed are nitrogen and phosphorus, while 90 percent of the potassium removed during the growing cycle returns to the soil with crop residues. Among the three main fertilizer elements, N is an important yield factor for oilseed rape.

The high nitrogen demand at the end of winter must be satisfied with the first application (90-120 kg of N/ha). After the start of the rise, fertilize a second time with nitrogen (50-70 kg of N/ha). The sulfur demand must be satisfied before the start of the rise (80 kg/ha). A high nitrogen fertilization leads to an increase in the sulfur deficiency. This deficiency, as well as that of boron, limits the number of pods per plant and the number of seeds per pod. Given the proximity of the rows,

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weeding is quite problematic; therefore, chemical weeding is used.

Oilseed rape harvesting is carried out when the seeds are completely brown and the pods are dry (optimal grain humidity around 12%). In Italy, productions of around 3 tons per hectare can be obtained with autumn sowing and 1.5-2.0 tons with spring sowing.

The oil always contains linoleic acid (4-10%). In the old varieties it contained up to 50% erucic acid, which is unstable and toxic. Today, varieties free of erucic acid are available: edible oil must have this characteristic, while for the non-food industry a high erucic acid content is required. In addition to the production of oilseed rape oil for food use, that for industrial purposes also has a certain importance: in this case, traditional varieties are used, as the erucic acid content is irrelevant.

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Coriander

COMMON NAME: Coriander

SCIENTIFIC NAME: *Coriandrum sativum* L.

FAMILY: Ombrelliferae

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Coriander is an annual herbaceous plant, native to the south-west Mediterranean regions. In Italy it is cultivated and rarely naturalized. It belongs to the same family as cumin, dill, fennel and parsley. Coriander has erect, branched stems, up to 50 cm tall. The leaves are alternate and 2-3 pinnate, with the lamina divided into thread-like lacinias. The leaves give off a characteristic, extremely

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unpleasant odor, which reminded the ancient Greeks of crushed bugs (the name of the plant derives from the Greek 'koris', which means bug). The inflorescences are umbels with 4-6 rays that bear small white-pink flowers. The fruit (commonly called seed) is a subglobose achene.

The cultivation technique, in some environments can be simplified and the environmental impact is minimal. The sowing period is from January to April, but it is interesting to consider that for the same variety, positive tests have been recorded with sowing in December. The technique also provides an appreciable flexibility in terms of mechanization, as it is possible to use equipment useful in the cultivation of winter cereals. On average, about 7-10 kg of seed are sown per hectare, and it varies according to the variety and the sowing period. It does not require irrigation and the harvest is carried out with any combine harvester, from mid-July to mid-August. Coriander does not require particular fertilization: for good development of the crop, a light phosphate fertilization at the base is preferable (60-70 kg/ha) and a nitrogen fertilization on top before flowering (70-80 kg/ha). As regards weeding, generally only one pre-emergence intervention is necessary.

Coriander yields generally range between 1.5 and 2.5 t/ha, depending on the variety and the more or less favorable seasonal trend. Coriander cultivation is suitable for inclusion in crop rotations in Central and Southern Italy without particular problems. As an umbellifer, it has different nutritional needs and any rebirths on subsequent wheat cultivation can be appropriately controlled.

Coriander is a melliferous species also highly appreciated by beekeepers for its prolonged white inflorescence and, as previously specified, for the absence of phytosanitary treatments on cultivation.

Therapeutic properties: antispasmodic, carminative, antiseptic, aperitif, digestive.

The essential oil contains as main components: coriander, alpha pinene, geraniol, 1,8 cineole, phellandrene, dipentene, terpinene, cymene and borneol.

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Garlic

COMMON NAME: Garlic

SCIENTIFIC NAME: *Allium sativum* L.

FAMILY: Liliaceae

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www.agraria.org

Garlic is an annual plant known and appreciated for its countless beneficial properties since the time of the Egyptians in the third millennium BC. Today it is widespread in all continents where it is also present as a perennial species with high rusticity.

The plant has cord-shaped and superficial roots, which affect the first 20-30 centimeters of soil. The leaves are basal, amplexicaul and, unlike those of the onion, do not act as reserve organs. The

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outermost one wraps around the previous one for about 10 centimeters, a value that increases as you move inward. The flower stalk is cylindrical, full, 40-80 cm long and bears at the top an umbrella-shaped inflorescence wrapped by a pointed spathe. The flowers, in variable number and carried on a long and thin peduncle, are white, pink or purple, often mixed with bulbils derived from the metamorphosis of flower buds. From fertilization a capsule originates, which rarely contains seeds. The organs used for propagation are the cloves or bulbils, which have a convex dorsal surface; which, in groups of 5-20, are inserted directly onto the stem, reduced to a small disk, called corm, which forms the bulb or head or head. The latter is wrapped in a series of metamorphosed leaves, called sterile tunics, with a protective function. The average weight of a bulb can range from a minimum of 20 grams to a maximum of over 150 g. After harvesting, the bulbils do not germinate immediately, but require a period of dormancy. To prolong this period, it is necessary to store the bulbils at a temperature of 0°C or at values above 18°C, while to shorten it, they must be stored at 5-10°C.

Garlic prefers loose soils, with good fertility, texture and structure, able to prevent water stagnation, also considering the period in which it is often grown (from autumn to early summer).

In terms of varietal choice, cultivated plants mostly belong to ecotypes rather than true varieties. Cultivated garlic is normally distinguished between "white garlic" and "red garlic". Among the red ones, we must remember the "Rosso di Sulmona" and the "Rosso Francese", while among the white ones, the "Bianco piacentino", the most widespread in Italy. Red garlic, in addition to the different color, is characterized on average by a shorter cycle of about three weeks and by larger bulbs.

Although garlic is a perennial plant, it behaves like an annual if cultivated. Plowing is carried out in a trough at a depth of 25-30 cm, followed by harrowing or milling. Although garlic does not require deep tillage, it is considered a renewal crop. In rotation, garlic usually follows wheat, and must not be preceded by onion, carrot and vegetable crops.

The largest and healthiest bulbils should be planted in autumn in mild climates and in early spring in cold and humid areas. The bulbils can be planted manually, in which case they are placed with the tip facing upwards, at a depth of about 4-5 cm, at a distance of 10-15 cm on the row and 30-50 cm between rows. However, mechanical "seeding" is increasingly used. The transplant must be preceded, even by a few days, by the separation of the bulbils from the disk, which can be manual or mechanical.

Garlic fertilization is exclusively mineral; organic fertilization can cause serious problems and must

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be done in the previous crop. With a production of 10 t/ha of bulbs, the crop removes about 100 kg of nitrogen, 28 kg of phosphorus (phosphoric anhydride), 130 kg of potassium (potash), 25 kg of calcium and 18 kg of magnesium. Phosphorus and potassium must be added during soil preparation, while nitrogen must be distributed as a top dressing, in two applications. Garlic generally does not require irrigation because the normal amount of rainfall is sufficient to satisfy the plant's needs. It only needs irrigation in the event of prolonged drought. Having a very superficial root system, any weeding must be done very carefully; for this reason, weed control is increasingly done using herbicides. Flower stems must be cut when they are still in bud to prevent the plant from using the reserves contained in the bulb for flowering.

Harvesting can be done either manually or mechanically, and is done by uprooting the plants and leaving them to dry on the ground for about a week. The bulbs are then cleaned of the outermost, dirty, broken or blackened tunics, the roots are cut and braids of 20-30 bulbs are made, or the leaves are cut and the bulbs are placed in airy containers. The yield of garlic is around 8-12 tons per hectare of bulbs. They must be stored in a cool and ventilated place in boxes or hung braided in the typical “reste”. The best conservation is obtained in warehouses at 0°C and with 70% relative humidity (up to 6-7 months). Garlic can be marketed both fresh and semi-dry or dry.

Garlic has therapeutic properties, such as: anthelmintic (against ascarids and pinworms), hypotensive, expectorant, digestive, carminative, antiseptic, hypoglycemic. From garlic, it is also possible to extract essential oil (60 g from 100 kg of bulbs), yellow-brown in color, containing sulfur compounds (derivatives of allicin) with bactericidal properties. In addition, the essential oil contains: allyl bisulfide, allipropyl, allicin, garlicin (with antibiotic action), allin (glucoside), vitamins A, B, C, sugars, phytosterols, lipids, mucilages.

Garlic, among its countless properties, has that of being used as a biopesticide (Dusi et al,. 2022) and insecticide, as also evidenced by popular uses (Dougoud et al,. 2019). Its use in food preservation against food pathogens is also relevant (Polito et al,. 2022).

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Experimental activities

Experimental activities were carried out in two different environments, Genzano di Lucania in Basilicata and Bagnoli del Trigno in Molise. In the Lucanian one, the activities focused on annual wormwood in rotation with durum wheat and leguminous plants. While, in the Molise one, species whose biomasses and extracts (essential oil), were cultured and made available to the other project partners (UNIBAS) under WP 2 "Screening of alternative species as medicinal plants and enhancement of the natural biodiversity of ancient grains."

The experimental site of Genzano di Lucania

The Lucanian experimental site is located in a hilly environment, from the soil-climate point of view, in the countryside of Genzano di Lucania (PZ) (average altitude 400 m above sea level). The experimental site has good fertility considering together: soil texture, organic matter content, nutrient content and climate. The Lucanian area in which the experimental field is located is on the eastern slope of the Apennine watershed and has a Mediterranean climate typical of the inland lands of central-southern Italy, with higher rainfall in winter. Rapeseed, coriander and annual wormwood were tested in rotation with durum wheat at this site. The field activities were shared with the University of Basilicata (UNIBAS) unit, which investigated aspects of rapeseed and coriander.

Annual wormwood (*Artemisia annua* L.) seedlings were produced in seedbeds and were selected and certified. Prior to fertilizer application, soil samples (0-30 cm) were taken from each block and analyzed according to standard procedures (SISS, 2000). The soil texture was loose and the soil was found to be rich in skeleton. The content of organic matter and total N is typical for the area. The soil profile is uniform overall and contains good amounts of available P and exchangeable K. The soil had a very low level of active CaCO3 and the pH was neutral on average; salinity was low.

After conventional plowing (35 cm depth), the equivalent of 80 kg P ha^{-1}, 120 kg K2O ha^{-1} and 90 kg N ha^{-1} were applied, on average. Doses were calculated considering crop removals, soil balance, spatial and temporal variability and vegetative state of the crop.

Seedlings were transplanted in late spring, followed by relief irrigation, in rows 100 cm apart. Two planting densities per square meter of soil were compared (2.5 and 5 plants per square meter). These parameters are a compromise between the limited literature data, expected biomass quality

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and growing environment. The field was surrounded by a buffer strip to allow uniform growing conditions.

The experimental site of Bagnoli del Trigno

The Molise experimental site is located in a hilly environment, from the soil-climatic point of view, in the countryside of Bagnoli del Trigno (IS) (altitude 600 m above sea level). The experimental site has high fertility considering together: soil texture, organic matter content, nutrient content and climate. The Molise area where the experimental field is located is situated on the eastern slope of the Apennine watershed and has a Mediterranean climate typical of the inland lands of central-southern Italy, with higher precipitation in winter and spring, and a cool climate.

The species grown are basil (*Ocimum basilicum* L.), oregano (*Origanum vulgare* L.), helichrysum (*Helichrysum italicum* (Roth) Don), rosemary (*Rosmarinus officinalis* L.), wild fennel (*Foeniculum vulgare* Mill.), juniper (*Juniperus communis* L.), and *garlic* (Allium sativum L.), to which mint (*Mentha x piperita* L.) was added during the course of the work and for some specific needs. The seedlings from seedbeds used are selected and certified.

Before fertilizer application, soil samples (0-30 cm) were taken from each block and analyzed according to standard procedures (SISS, 2000). The soil texture was medium sandy-clayey. The content of organic matter and total N was good. The soil profile was uniform overall and contained good amounts of available P and exchangeable K. The soil had a very low level of active CaCO3 and the pH was medium alkaline; salinity was low.

After conventional plowing (35 cm depth), the equivalent of 90 kg P ha^{-1}, 80 kg K2O ha^{-1} and 100 kg N ha^{-1} were applied, on average. Doses were calculated considering crop removals, soil balance, spatial and temporal variability, and vegetative state of the crop.

Seedlings were transplanted in early spring, followed by rescue irrigation, in rows 75 cm apart. The planting densities for the different species per square meter of soil are: basil (20), mint (10), oregano (10), helichrysum (10), rosemary (5), wild fennel (10), juniper (0.25), and garlic (25). These parameters are a compromise between little literature data, expected biomass quality and cultivation environment. The field was surrounded by a buffer strip to allow uniform growth conditions. The appropriately treated biomass produced from mint, rosemary, annual wormwood and coriander were transferred to the University of Basilicata unit for biocontrol activities.

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Essential oil extraction

Also in Bagnoli del Trigno, the essential oil subsequently delivered to UNIBAS (mint, annual wormwood and rosemary oils) was extracted.

To isolate the essential oils, each sample (1000 g in three replicates) was extracted by hydrodistillation for 3 h, using a Clevenger-type apparatus as recommended by the European Pharmacopoeia. The essential oils produced were transferred to the unit of the University of Basilicata for biocontrol activities.

Annual wormwood essential oils were obtained with different yields for the two densities (5 plants m^{-2} 0.117±0.011% - 2.5 plants m^{-2} 0.157±0.01%) on a fresh mass and were yellowish in color (5 plants m^{-2} intense and 2.5 plants m^{-2} light).

Helichrysum essential oils were obtained with average yields of 0.301±0.015% and were yellowish in color.

Wild fennel essential oils were obtained with average yields of 0.621±0.013% and is deep yellowish in color.

Mint essential oils were obtained with average yields of 0.411±0.009% and is yellowish in color.

Oregano essential oils were obtained with average yields of 0.309±0.01% and is yellowish in color.

Basil essential oils were obtained with average yields of 0.204±0.008% and is yellowish in color.

Juniper essential oils were obtained with average yields of 0.315±0.014% and is light greenish in

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color.

Garlic essential oils were obtained with average yields of 0.323±0.016% and is light greenish in color.

Rosemary essential oils were obtained with average yields of 0.601±0.009% and is light yellowish in color.

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Annual wormwood: Literature and experimentation

Medicinal plants and particularly some of their derivatives, such as essential oils, are widely used in the food (Maisanaba et al., 2017), herbal (Bajer et al., 2017) and pharmaceutical (Adhavan et al., 2017) industries. Recently, their use in agriculture (Romano et al., 2024) and post-harvest (Allagui et al., 2024) to defend plants and food from pests is also common. The scientific research behind the dissemination of essential oils in these production areas, however, often produces contradictory results depending on the chemotype of plant used. These inconsistencies can be attributed to qualitative differences related to genetics (Ben Farhat et al., 2019; Schmiderer et al., 2013), environment (Russo et al., 2013; Delfine et al., 2017), and the cultivation practice adopted (Formisano, et al., 2015). In particular, an important role on essential oil yield and composition is played by planting density, which has the potential to induce natural variations in plant development and, therefore, in its primary and secondary metabolism (Tibaldi et al., 2022; Saki et al., 2019; Tuttolomondo et al., 2016).

Annual wormwood (Asteraceae), native to temperate Asia, has become endemic in a large number of countries (Mojarab-Mahboubkar et l., 2023). Artemisia annua is a medicinal plant that has been much studied because of its biological properties. The EO of annual wormwood is known for its different modes of action against pests (Mojarab-Mahboubkar et al., 2022; Zhang et al., 2014).

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Annual wormwood is an annual herbaceous plant that in temperate climates can reach 2 meters in height. The plant can assume a monocaulescent habit (with alternate branches), or a bushy habit (branching at the base). The leaves are deeply septate, ranging from 2.5 to 5 cm in length and covered with trichomes. The inflorescence (capitula) is about 2 to 3 mm in diameter, is usually

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yellowish in color and is enclosed by numerous bracts. The capitula has numerous bisexual (hermaphrodite) flowers in the center, surrounded by female (pistillate) flowers. All floral organs also contain trichomes (filamentous cells).

The active substance that characterizes this plant is artemisinin and is more present in the inflorescences (% S.S., 4 to 11 times higher) than in the leaves. This substance is mainly present in the glandular trichomes. The fruit is a very small achene and the seeds are very small in size (about 10,000 per gram).

Annual wormwood is an aromatic plant of Eastern European and Asian origin. Today it is widespread in various parts of the globe characterized by temperate climate. In Italy it is found throughout most of the country and behaves as a ruderal.

Historically, its use became particularly well-known and renowned in China, where annual wormwood was indicated as an anti-hemorrhoidal and anti-malarial remedy, and for the treatment of dermatitis.

More recently, it is also known to be used as a bio-fumigant against insects in stored foodstuffs. In general, according to folk medicine, annual wormwood has the following medicinal properties: antibacterial, antiseptic, carminative, digestive and febrifuge.

Annual wormwood prefers loose, south-facing soils, but it is also adapted in clay soils. The plant is undemanding of nutrients. In our latitudes the species has a spring-summer growth cycle, and flowering occurs in late summer. The plant is moderately resistant to low temperatures and totally desiccates with the first frosts. The limiting factor for growth is photoperiodism. In fact, when the day is about 12 hours long the plant induces flowering and slows down the growing season.

Seeding is done in seedbeds because of the very small seed size. Seedlings at the 2-4 leaf stage and about 10 cm tall are transplanted in the open field using common transplanters. There is no optimal density for industrial cultivation at present because few agronomic trials have been conducted on this plant. Based on the few data available, it seems that a density of about 6 plants per square meter is the right compromise for plant development and active substance (artemisinin) accumulation. Nitrogen does not seem to affect the productivity of annual wormwood, which instead benefits from modest inputs of phosphorus and potassium. Irrigation also positively affects productivity but no crop coefficients are available for calculating irrigation requirements. Weed control (lacking registered herbicides) is done mechanically, by means of appropriate weeding.

The low content of artemisinin (max 0.5 percent) in the genotypes of annual wormwood species

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grown in the recent past in Europe and America has been the main limiting factor for the spread of the crop over large areas. In recent years, genetic improvement has led to a marked increase in artemisinin in the aerial parts of the plant. This result, combined with abundant dry biomass production (20-25 t/ha) could make the crop economically viable.

As with most aromatic plants, the maximum content of active substances (including artemisinin) coincides with the flowering stage. This is the appropriate time for harvesting, which is done by mowing the plants. These are then placed to dry in the shade in a covered and ventilated room. To date, there are no machines suitable for mechanized harvesting. It would be useful to be able to harvest only the leaves or apical tops of the plant, leaving the conspicuous woody part in the field.

The main compounds found in the essential oil are selinene, 1,8 cineole, camphor, verbenol, artemisia ketone, borneol and trans caryophyllene.

Annual wormwood is counted among the species that can control weeds at different stages of growth (Pannacci et al,. 2015; Benvenuti et al,. 2017; Pannacci et al,. 2020).

The main compounds detected in the EOs of Artemisia annua include camphor, 1,8-cineole, and mugwort ketones, but the chemical composition varies according to geographic area, harvest period, and method of extraction and cultivation of the plant (Marinas et al., 2015). In addition, the scientific importance of the Artemisia plant derives from the presence of biologically active substances, such as sesquiterpenes and terpenoids (mainly monoterpenes in OEs) (Trendafilova et al., 2021). These phytochemical classes are recognized for a wide variety of interesting biological activities (Ur Rashid et al., 2019).

Annual wormwood is cultivated, not only for the production of essential oil, but also for the production of interesting and promising biomass for other uses including agricultural. Cultivation of annual wormwood, with appropriate cultural management, could represent a valuable opportunity to increase agro-environmental sustainability and diversification of typical Mediterranean crops, as well as to help reduce the use of synthetic molecules for pest control such as fungi, bacteria and weeds.

Considering the potential economic importance of this crop, the activities summarized in this report aim to produce essential oil and quality green biomass useful for agricultural uses from a sustainable perspective. Therefore, the influence of planting density on the main agronomic and herbicidal traits (plant biomass production and weed seed germination suppression performance), as well as

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on the yield and composition of essential oil useful for other investigative activities under the AgrEcoMed project was evaluated.

The results of the artemisia trial carried out in rotation with cereals in the Genzano di Lucania experimental field as part of the AgrEcoMed project are reported below.

The field activity was carried out in the experimental site of Genzano di Lucania (PZ), previously described. After transplanting, annual wormwood seedlings were grown following environmentally friendly protocols and fed by meeting the physiological needs of the crop at different phenological stages. During the balsamic period, after sampling representative plats for essential oil extraction, the plants were chopped and buried by plowing. In the following autumn, durum wheat (cv Tirex) was sown in order to highlight the effects of biomass burial on plant development and durum wheat grain production.

Durum wheat was sown both on the two plots where annual wormwood was buried at the two different densities, and in one plot, where annual wormwood had not been buried and kept bare fallow (control treatment). In total, therefore, three plots of about 1,000 square meters each were made.

The durum wheat crop had good emergence and plant development despite poor rainfall. Water shortage, unfortunately, showed its negative effects at the end of the rising when stunted growth was followed by poor earing. This condition negatively affected grain yield. Surveys carried out at this stage and for the short period when the crop was not adversely affected by water shortage, showed a higher vegetative luxuriance, by about 30 %, in treatments where annual wormwood had been buried than in the control. Accompanying this statement are data collected, in collaboration with the University of Basilicata, from a drone that calculated the NDVI (Normalized Difference Vegetation Index), which is the vegetation index that describes the level of vigor of the crop.

Other evidence is that related to good weed containment (allelopathy) compared to control. By analyzing the presence of weeds and in particular the presence of some botanical families, herbicidal activity, or more properly, containment of weed development, was shown to be about 40 percent compared to control.

Regarding the presence of segetal weed species, the differences found were significant between control (bare fallow) and the D20 (5 plants square meter) and D40 (2.5 plants square meter) treatments.

For the species richness present in the three theses under comparison, the values per plot vary from

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a maximum of 18 in the control to a minimum of 10 in the D40 thesis. In particular, evaluating species abundance shows that the D20 and D40 treatments show no significant differences between them, while both treatments show lower values than the control.

The allelopathic effect was found to be different among the three treatments compared. The measurements then focused on those species that exceeded a threshold of potential damage to crop yield. Specifically, in surveying the harmful species present, and relative abundance, a low number (about five) were found in the D20 and D40 treatments (Malva sylvestris, Polygonum aviculare, Amaranthus retroflexus, Chenopodium album and Cota altissima). In the control treatment, however, there are more than 15 diffrential species: Amaranthus ssp., Chenopodium album, Convolvulus arvensis, Galium aparine, Heliotropium europaeum, Lactuca sativa, Papaver rhoeas ssp., Picris hieracioides, Sinapis arvensis, Sonchus arvensis. Poppy, bindweed and amaranthus the most representative species.

From the first preliminary data available regarding the presence of the weed flora in a durum wheat crop, it is evident that precession with annual wormwood, on the whole, has an excellent effect in controlling the development of segetal weed vegetation, with average reductions of more than 50 percent.

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