











Concept Paper

LEGU-MED: Developing Biodiversity-Based Agriculture with Legume Cropping Systems in the Mediterranean Basin

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Abstract: Environmental degradation and the decrease of ecosystem service provision are currently of major concern, with current agricultural systems being a major driver. To meet our future environmental and sustainability targets a transformation of the agro-food systems and current agricultural value chain are crucial. One approach to redesign farming systems is the concept of biodiversity-based agriculture (BBA) which relies on sustainable diversification of biological components and their natural interactions in farming systems to maximize fertility, productivity, and resilience to external perturbations. Despite minimizing anthropogenic inputs, BBA is not yet able to meet all beneficial environmental objectives. BBA applied in the Mediterranean basin requires urgent innovation in approaches, methodologies, and models for small-holder traditional farming systems to ensure a stable provision of ecosystem services and better resilience to environmental stresses linked to climate change. Legumes are the backbone of the Mediterranean agro-ecosystems from ancient times, but their unique and wide biodiversity was not sufficiently valorized, especially by North-African countries. Here, we present LEGU-MED, a three-year international project funded by PRIMA initiative 2019. An international consortium was established involving five universities, 5 research institutes, and one private company from 8 countries: Italy, Germany, Spain, Algeria, Tunisia, Turkey, Lebanon,

and Croatia. The main objective of this project is to put forward an international and well-integrated plan to valorize the legume agrobiodiversity of the Mediterranean in biodiversity-based farming systems and consequently enhance agro-ecosystem functions and services in the Mediterranean basin. The successful completion of LEGU-MED will have the following impacts on Mediterranean legume-based farming systems: (1) improve water use efficiency, (2) reduce the use of anthropogenic inputs through the maintenance of soil fertility, (3) enhance pollination and improve ecological connectivity with flora and fauna, (4) protect close-by wildland ecosystems, (5) enhance other ecosystem services (e.g., pest, disease, and weed suppression), and (6) provide healthier and safer protein-rich food.

Keywords: agroecology; biodiversity; biodiversity-based agriculture; legumes; chickpea; lentil; Mediterranean; agro-ecosystems; sustainable agriculture

1. Background/Rationale

1.1. Biodiversity-Based Agriculture

A diverse and well-preserved biosphere is indispensable for human well-being to provide and maintain natural functions and ecosystem services (ES), such as regulation of water, soil and air quality, food and materials provision, and identity and cultural values conservation [1,2]. The interplay between biodiversity and ES delivery is highly shaped by societal factors, such as decisions on land-use, protection status, or governance [3–6]. The latest results on the biosphere health worldwide are alarming: a high decline of natural and seminatural ecosystems occurred during the last centuries [7], severe climatic changes and increasing human pressure lead towards more extreme conditions, higher land degradation, and potential habitat losses [8], fertilizer (nitrogen and phosphorous) reserves are well beyond critical thresholds [9], all ES but provisioning services are decreasing [10], the rate of species extinction, including wild species as well as domesticated varieties, is rising [9].

With the Agenda 2030 for Sustainable Development, the Convention on Biological Diversity (CBD), and on-going Post-2020 global biodiversity framework, international policy commits itself to a future “where all life can thrive”, people are “living in harmony with nature” (UNODA United Nations, Open-Ended Working Group OEWG-2, 2020, n. 5), and “biodiversity is valued, conserved, restored and wisely used, maintaining ecosystem services, sustaining a healthy planet and delivering benefits essential for all people” (OEWG-2, 2020, n. 9). To meet our future targets sustainable food systems and agriculture is crucial. However, currently the entire agricultural value chain “farm-to-fork” is poorly sustainable. Over-use or misuse of natural resources (water, soil, fertilizers) and pest control strategies, as well as environmentally unsuitable systems of food transformation, conservation, and transportation, make dominant agricultural systems major drivers of environmental problems [11–13]. Agriculture needs to undergo rapid transformative changes to achieve food security while maintaining resilient and healthy ecosystems [10,14–16].

In the 1990s, Hill and MacRae [17] developed a conceptual framework for a transition towards more sustainable agriculture starting with conventional-efficiency (unsustainable, shallow sustainability)—via substitution—to redesign (deep sustainability). In more recent concepts, this is also framed as “weak” and “strong” sustainability [18,19]. Since then, and mainly from the 2000s, many different terms, concepts, and approaches in the context of sustainable agriculture and ES emerged [20,21]. One of these approaches being allocated at the very end of the sustainability continuum and thus building on the redesign is biodiversity-based agriculture (BBA) [22] According to Duru et al. [23] and Therond et al. [24], BBA goes in line with ecologically intensive agriculture, eco-functional, (agro-) ecological, or sustainable intensification. At this point, we want to remark that many of these terms still lack consensus definitions [25] (find some comprehensive overviews e.g., in [21,26,27]). So BBA is still also not conclusively and officially defined.

According to Duru et al. [23] BBA is a strong ecological and eco-centric way to modernize agriculture to “promote fertility, productivity, and resilience to external perturba-

tions" [23]. The BBA concept distinguishes between three components of agrobiodiversity: (I) planned diversity (cultivated plants/crops), (II) landscape heterogeneity (composition and configuration of surrounding habitats), (III) associated diversity (biota that immigrates from the surrounding and colonizes the field to find food or shelter) [23]. Associated diversity is strongly influenced by the planned crop diversity and landscape context as well as by field management practices [23,28–32]. To achieve high natural input services BBA follows three main principles: (I) increase plant diversity and soil cover (field level), (II) minimize soil disturbances (field level); (III) diversify the landscape matrix (farm/landscape level) [23]. For possible practices supporting these principles we refer to e.g., Gaba et al.; Nicholls and Altieri; Shackelford et al.; Wezel et al. [33–35]. As such, BBA has obvious tight connections with the emerging paradigm of agroecology [36] and we in the following use them synonymously, although BBA mainly focuses on agroecology's science and practice components [37,38] for the full definition of agroecology. Therefore, we define BBA as a farming concept with a systems perspective that aims to achieve sustainable food production by building on the fundament of (functional) biodiversity being the main driver of ES delivery by operating as an input and output component within agroecosystems at different spatial scale. Thus, BBA relies on the diversification and intensification of natural interactions between the different biophysical components of agroecosystems, leading to a stronger focus on the role of ecosystem processes and natural services provided via different functional groups [28,39] This delineates BBA and agroecology from other already established farming systems such as organic farming. Although opinions can diverge in the interpretation of their connection, mainly due to a lack of sharp definitions on the one first and the manifold of different practical implementations on the second [40]. IPBES [26] and Vanbergen et al. [22] highlight that organic farming mainly operates at the substitution stage (e.g., of synthetic by organic products) on the aforementioned sustainability continuum, whereas agroecology and diversified farming systems encompass a more holistic view and systems approach aiming at also increasing resilience and achieving transformative changes resulting in redesigning the whole food system. Moreover, Kremen et al. [27] and IPBES [26] make the point that even organic agriculture can be practiced in big monocultures on simplified farms, which does not necessarily support biodiversity enhancement also on the landscape scale. This is another difference compared to diversified farming, as this builds on the fundament of diversity increase. However, Migliorini and Wezel [41] bring into consideration, that agroecology and organic farming differ in the criterion of pesticide and fertilizer use, which is not yet officially regulated in the newly emerged approaches and thus their impact depends on how BBA/agroecology are defined in the specific cases. Nevertheless, many authors agree that indeed the two approaches share common principles and practices, such as proposed cropping practices, the view of a closed system, and a stronger ecological perspective on agroecosystems [27,36,41,42]. The High-Level Panel of Experts on Food Security and Nutrition of the Committee on World Food Security [36] for example understands agroecology as "a science, a set of practices and a social movement" which technically encompasses "the application of ecological concepts and principles to farming systems, focusing on the interactions between plants, animals, humans, and the environment, to foster sustainable agricultural development" [36], whereby the Committee allocates organic farming (as defined by official regulations) as a "related" approach, [36] which also applies agroecological practices [36].

Finally, even when implementing BBA or other agroecological farming systems, not all agronomical and ecological/sustainability goals can be met simultaneously [43–46], and thus priorities need to be set; for example, by addressing first spatial vulnerabilities, exploring the potentials to develop on-farm biodiversity, or assessing the capacity of the local farmers to operate for improving habitat at the landscape-scale collectively [47–49] The redesign of farming systems (towards BBA or similar approaches) is knowledge extensive [17]. The biophysical and socioecological context of the studied area [50] is notable not only at the field but also at the farm/landscape level, scientifically defining and proposing scale sensitive "decision spaces" and competencies [50]. It must rely on scenario and

projection techniques to illustrate the impacts of farming decisions [22,50–53]. It also needs to consider the social and cultural dimension of the agroecosystems, the reinforcement of collaborations amongst stakeholders, and the cocreation of knowledge [54,55]. Finally, as production systems are also diverse and intricate, different levels and forms of BBA (e.g., structural-functional-compositional diversity on gene-species ecosystems levels) should be considered when (re) designing sustainable agriculture [2,51,56,57]. To achieve a transition of agriculture towards sustainable alternatives, such as BBA, a holistic view and a pluralistic and interdisciplinary, multiscale research approach is needed [55].

1.2. The Importance of Legumes for BBA Applications

Following phenotypic convergence on a few traits, plant domestication was carried out, which led to few species replacing naturally available biodiversity. The Green Revolution has accentuated this phenomenon, particularly in South and Southeast Asia where cereal-based monocultures such as rice-wheat, rice-maize, and rice-rice systems replaced dozens of leguminous, oilseed and millet crops [58]. Intensive cropping systems relying on simple cropping sequences, mineral fertilizers, and chemical crop protection further reduced biodiversity and simplified landscapes [59]. Legumes were domesticated about 8000 years BC and the agronomic and food properties of legumes (with more than 7000 species, mainly wild, in the world) were known for at least 2000 years. Legumes are therefore staple food for humans since the development of agriculture, but only about two dozen vegetable legumes were domesticated. Bottlenecks during and after domestication that contributed to reducing genetic diversity in the gene pool of legumes included high-yielding monocultures, excess of nitrogen fertilizers and consequent eutrophication, and pollution caused by the use of pesticides and fertilizers entering groundwater or remaining suspended in the air, in turn reducing plant resistance to pests, diseases, and climatic conditions [60].

Despite these accompanying incongruities, legumes were successfully bred for their high nutritional composition, their ability to bind nitrogen, and other key traits [61]. Farmers know the value of legumes as atmospheric nitrogen fixers, and their tissues are rich in proteins. Legumes directly contribute to diversified landscapes, e.g., by entering into agronomical rotations, or indirectly, e.g., providing habitats and resources to various animal species [62]. Under the modern integral vision of conservation agriculture, considering the balance among the diverse factors of production, grain legumes play a significant role [63]. For example, Brazil has implemented conservation agriculture systems using soybean as a legume crop [64]. North America, Australia, and Turkey adopted conservative agriculture using grain legumes such as lentil, chickpea, and faba bean [65]. Awareness of environmental degradation has led to renewed interest in species of legumes that break pest or disease cycles typical of intensive agriculture and may be cultivated using sustainable practices without intensive applications of fertilizers and crop protection products in diversified farming systems as an alternative to modern industrial agriculture [66].

Among the additional important benefits that legumes may deliver, their role in contributing to climate change mitigation was rarely addressed. Legumes can reduce the emission of greenhouse gases such as carbon dioxide (CO₂) and nitrous oxide (NO₂) compared with agricultural systems based on mineral N fertilization, improve sequestration of carbon and nitrogen in soils, and reduce overall fossil energy inputs in the system [67]. The inclusion of legumes in rotation with cereals helps to improve system yields, enhance net carbon sequestration, and lower the carbon footprint. In a recent study, the lentil-wheat system produced the lowest carbon footprint at −552 kg CO₂ eq/ha [68]. Legume crops are well-suited to low moisture conditions due to their low protein yield-based water footprint (6.58 m³/kg pulse vs. 9.25 m³/kg cereal) [69]. The cultivation of legumes potentially reduces greenhouse gas emissions and supports biodiversity [70]. Such factors make legume crops a key for BBA. A recent study identified the economic advantages of legume-cereal rotations over cereal monocropping, including higher yields, gross margins,

and consumption [71]. Legumes also deliver ES indirectly, facilitating services such as pollination [59], and reducing dependence on a grain monoculture [60].

Legumes may contribute to addressing specific regional problems. In Europe, legumes may help to solve the double challenge of increasing plant protein production while reducing nitrogen supply [72]. In the Mediterranean, where legumes already play a prominent role in the population's diet, they may help address the need for diversified agro-ecosystems able to cope with climate change and resource scarcity (especially water). The European Union Green Deal strategy considers legume cultivation as one of the most natural and promising approaches to sustainable and competitive use of soil resources, primarily fertilizers contributing to the diversity, balance, and resilience of key European agro-ecological systems [73].

1.3. Lentil and Chickpea: An Overview of Improved Varieties and Landraces Cultivation

The continued development of legume varieties is essential to meet new environmental challenges due to climate change improve production and quality traits [61]. Compared to that of the closest wild relatives and forage legumes, domestication and plant breeding altered many traits in modern cultivars. For example, farmer selection aimed to increase the seed size, nonshattering pods, and nondormancy as main characters for legumes [74].

On the other hand, different phenotypic traits may be required to cope with the present, and future challenges posed by climate change and sustainable land use, and native ecotypes may provide higher fitness and production stability under specific regional conditions. For this reason, it is important to safeguard genetic resources as a unique source of diversification for breeders.

Advances in the genetic knowledge of legume model species such as *Medicago truncatula* and *Lotus japonicus* [75,76] led to remarkable progress in understanding the genetic structure of legumes. Genome-wide association studies conducted on natural accessions of these model legumes recently allowed the identification of candidate genes responsible for specific phenotypic traits [77,78].

In addition to cereals and other legumes, lentil and chickpea are among the founder crops of agriculture in the Mediterranean and Near East areas. These two species spread progressively from east to west Mediterranean basin, in both northern and southern sides of the basin up to the Nile valley and Ethiopia during the diffusion of primeval agriculture [79,80]. Archaeological remains dating back to the Neolithic age, testify to their cultivation in several locations of Turkey, Balkan Peninsula, southern Italy, and Iberian regions [81,82]. Moreover, the uninterrupted cultivation that lasted for millennia is testified by their mentions in many classical writers' scripts of different ages and civilizations [81,82]. The cultivation environments dissimilar for climatic conditions and soil composition, together with the selective pressure operated over time by local farmers gave rise to the selection of a myriad of landraces. Each one of these landraces was well adapted to particular microclimatic and edaphic conditions and was able to satisfy the agronomic, nutritional, and aesthetic preferences of the community growing it. Although the concept of landrace has evolved in the last decades [83], it is undoubted that for millennia, lentil and chickpea cultivation was based on plant material obtained by unconscious human selection. Genetic diversity among and within the landraces is recognizable by the differences in seed traits (size, shape, coat, and cotyledon color) and plant morphological features.

The selection of improved varieties, characterized by higher yield and resistance to biotic and abiotic stress, started in the last century, has significantly affected the material under cultivation. Breeding efforts in the past developed ~3700 years improved varieties that are grown in diverse agroecosystems across the world [84]. These numbers are a fraction of landraces and crop wild relatives that used to exist in natural habitats.

Farmers progressively replaced the landraces with improved varieties able to assure higher incomes. For this reason, lentil and chickpea experienced significant genetic erosion in a short time [85]. An unknown number of landraces was irremediably lost before starting actions aimed to safeguard this precious material through the systematic germplasm

collection and storage of the acquired samples in national and international gene banks (ex-situ conservation). The safeguard of autochthonous plant genetic resources is an important goal because the narrow genetic base of modern cultivars is a limiting factor to the release of new varieties through future breeding programs.

On the other hand, the diffusion of improved varieties has certainly contributed to the increase of the world production of lentil and chickpea allowing wider access for poor people to cheap proteins with a good nutritional value. The global lentil production passed from 2,565,138 tons in 1990 to 5,734,201 tons in 2019. In the same period, the world chickpea production has more than doubled, from 6,786,780 to 14,246,295 tons (source FAO). In the last century, the cultivated genotypes were replaced and the principal countries of production of both legumes have changed. Although lentil was domesticated in the Fertile Crescent [81], Canada is the principal producer and exporter worldwide (2,166,900 tons in 2019, source FAO) though lentil was introduced in recent times in this country.

Similarly, India has the worldwide leadership of chickpea production (9,937,990 tons in 2019, source FAO), though this species was domesticated in a small area close to the Syrian border [86]. In contrast with Canada, Indian production is mainly sold in local markets. Figures 1 and 2 show the lentil and chickpea production trend in Mediterranean macro-areas during the last 30 years. It is easy to observe that Turkey is constantly the principal producer of both species in this timeframe. The production-related to the cluster of countries located in the northern and southern side of the Mediterranean Sea, even if with small variations, has constantly remained inferior to that of Turkey. However, the whole production of chickpea and lentil in the Mediterranean basin constantly decreases year by year (see Figures 1 and 2). Although the FAO data does not give information about the cultivated genotype, only a very low fraction of the production is attributable to the landraces in each country. This descends from the requests of both national legislation and seed trade companies that promote the commercialization of genetically homogeneous material, a condition that the improved varieties can satisfy but not the landraces.

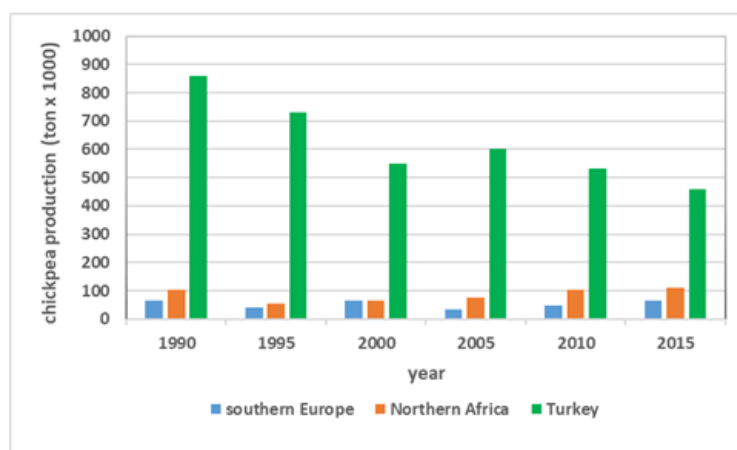


Figure 1. Chickpea production from 1990 to 2015 (FAOSTAT). Northern African countries: Algeria, Egypt, Libya, Morocco, Tunisia. Southern European countries: Croatia, France, Greece, Italy, North Macedonia, Portugal, Spain.

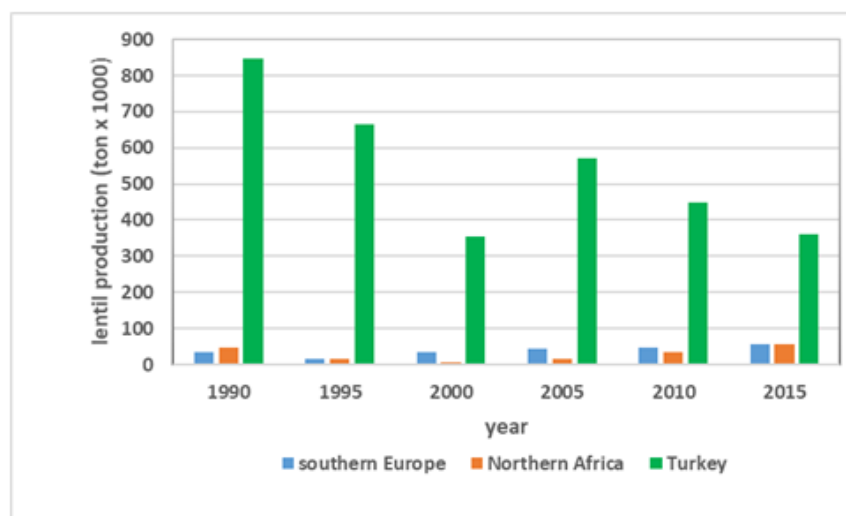


Figure 2. Lentil production from 1990 to 2015 (FAOSTAT). Northern African countries: Algeria, Egypt, Libya, Morocco, Tunisia. Southern European countries: Croatia, France, Greece, Italy, North Macedonia, Portugal, Spain.

The increased attention towards preserving agrobiodiversity, the threats that agriculture faces as a consequence of climate change and the increasing request, especially by European consumers, of traditional foods perceived as healthier renewed the attention towards landraces. Many papers, published in the last two decades, evidenced that the improved cultivars did not completely replace the landraces everywhere. Several authors documented the on-farm survival of lentil and chickpea landraces in Italy [87,88], Spain [89,90], Turkey [91], and Morocco [92]. Generally, these landraces are cultivated on small plots located in the traditional area devoted to their cultivation. Self-consumption is the prevalent form of harvest used. Occasionally it is sold in local markets without any label. This persistent cultivation has allowed the survival of this precious germplasm and testifies the strong link between each landrace with its territory and inhabitants. They are part of the agrarian landscape of a region, are used in traditional dishes, and sometimes can be related to the cultural patrimony of one community. Finally, legumes are among the milestones of the Mediterranean diet recently declared Immaterial Patrimony by UNESCO [93].

In 1992, the European Community introduced the three following quality marks: Protected Designations of Origin (PDOs), Protected Geographical Indications (PGIs), and Traditional Specialities Guaranteed (TSGs) to identify, to support the production, and to regulate the commercialization of the traditional products of each European country (EC Reg. no. 2081/92 and no. 2082/92). Among the vegetable products, the attribution of these marks has recognized the relevance of some prestigious landraces. The lentil and chickpea that have already obtained one European recognition are listed in Table 1. Of course, they are a very little fraction of the rich patrimony of agrobiodiversity associated with European lentil and chickpea germplasm. This suggests that further actions should be taken at local, national, and international levels to support landraces survival. The safeguard of agrobiodiversity is central for the resilience of agriculture and the perpetuation of landraces cultivation allows their coevolution with the ecosystems of belonging.

Table 1. List of lentil and chickpea landraces that obtained one European quality mark.

Species	Landrace Name	Country of Origin	Type and Code of EU Mark
Lentil	Lenticchia di Castelluccio di Norcia	Italy	PGI-IT-1557
	Lenticchia di Altamura	Italy	PGI-IT-02204
	Lenticchia di Onano	Italy	PGI-IT-02651
	Lenteja de La Armuña	Spain	PGI-ES-0102
	Lenteja de Tierra de Campos	Spain	PGI-ES-0313
Chickpea	Garbanzo de Fuentesauco	Spain	PGI-ES-0264
	Garbanzo de Escacena	Spain	PGI-ES-0945

1.4. Varietal Mixture and Composite Cross Population

Farmers and policy makers recently started to get interested in the role played by intra-specific diversity in agricultural systems: an initial sign of recognition and support was the Council directive 66/402/EEC1 of March 2014, which temporarily allowed the marketing of heterogeneous material for the first time in Europe. After that, the new organic regulation (EU) 2018/848 (Article 3, 18) officially introduced heterogeneous material within the categories of authorized plant reproductive material, confirming its crucial role in organic and low input farming, due to the better adaptability and to the capacity to adjust to climate change [94].

Intraspecific biodiversity can be achieved through the breeding for (i) multigenomic mixtures (or dynamic populations), (ii) multiparental populations (or composite cross populations), and (iii) farmer's selections [94,95]. Multigenomic mixtures (i) consist of a certain number of cultivars or landraces grown together on the same field for a few generations until the mixture evolves and adapts to local conditions. Multiparental populations (ii) result from the intercrossing of several cultivars, then evolved in a specific environment. Farmer selections (iii) are local landraces selected by farmers during the time in specific areas, exhibiting a very high genetic variability [94].

Brumlop et al. [96] assert that genetic diversity within crop species is a powerful means to increase resilience in the face of increasing environmental variability. Above all, because it allows a great expression of phenotypic plasticity in response to the environment with positive outcomes for the crops, such as yield stability [97] or adaptability [98]. Several authors agreed on the capacity of cultivar mixtures to buffer biotic and abiotic stress [99–101], especially in organic and low-input farming, through compensation, complementarity, and competition processes [102].

The specific breeding programs to obtain such material (called Evolutionary Breeding) are getting more and more popular with regards to cereals: many multiparental wheat populations were successfully developed in Europe [95] and proved high local adaptability and yield stability, especially if compared to pure lines [103].

However, some examples are also available for other crops: Wolfe [104] reported higher resistance to fungal disease of rice mixture than pure strains, with an experiment that ended with the abandonment of fungal treatments in the region. Among the hypothesized reasons is the immunization process among mixed plants, resulting in the early activation of the plant's disease-resistance mechanisms after the attack of ineffective pathogens (e.g., adapted to other components of the mixture). Another mechanism could be the competition among individual pathogens' genotypes well adapted to specific varieties, and those, less specialized, that thrive on different combinations of varieties [105]. The increase in the complexity of the pathogen population may also slow its adaptation to the mixture [106].

Again, barley multiparental populations were studied since 1991, when Soliman et al. [107] noticed their higher adaptability and stability than commercial cultivars. Commercial cultivars indeed showed high deviation from regression due to their ability to perform well in limited geographical areas, while population adaptation was proven by (1) yield increase over a generation, (2) lower sensitivity to foliar diseases, and (3) lodging.

Similar studies were conducted on maize and oat [108], attesting that heterogeneous material generally shows a smaller or absent genotype-environment interaction compared to pure lines. Nevertheless, the literature lacks food legumes, especially lentils and chickpeas, which miss a scientific observation of multigenomic mixtures and multiparental populations.

1.5. Market Trends of BBA Approach

The adoption of a BBA approach, and the already discussed benefits in environmental terms are also important at an economic level and indicators show that in the future an increasing component of agriculture will use all or part of solutions related to this system. Performing a market analysis of the BBA approach is a complex exercise that would require massive data collection. Moreover, the definition of practices related to the BBA approach is often nuanced and heavily determined by the agronomic contexts of reference. Some authors have attempted to determine the value of ES [109,110] and certainly, a BBA approach has among its prerogatives the maintenance of high ecosystem functionality. However, although scientifically the economic assessment of ES and payment for ecosystem services (PES) schemes are more and more intensively studied and the assessment of the value of a given ES is still little perceived by markets, politicians, and citizens, while the growth of a given sector in the real economy is more easily understood and closer to the sensitive experience of people. Since there is no sector in the market related to the BBA approach, it was decided here to select some benchmark segments as indicators of the economic development of certain approaches. Therefore, the data below do not aim to be complete market analysis, but instead, highlight market trends related to a BBA approach.

The push towards the adoption of more environmental-friendly production strategies is very strong, as evidenced by numerous actions at a political level, including the Paris COP 21 climate agreements (https://ec.europa.eu/clima/policies/international/negotiations/paris_en (accessed on 29 December 2021)) or the European Green Deal (https://ec.europa.eu/info/strategy/priorities-2019-2024/european-green-deal_en (accessed on 29 December 2021)); but also social, as evidenced by the emergence of movements such as Fridays For Future (<https://fridaysforfuture.org/> (accessed on 29 December 2021)). The BBA approach is not yet defined at the regulatory level. The practices associated with this management system are certainly close to those of organic agriculture, even if the limits of these two management models do not fully coincide, and probably organic farming can represent a step before the adoption of a BBA approach (as mentioned in Section 1.1). As previously mentioned, the analysis of the development of organic farming provides us with the first trends within a more environmentally conscious way of farming.

Organically managed land in 2019 amounted to 1.4% globally [111] for a total of roughly 70 million ha. However, the rate of conversion to organically managed land is increasing, and it is estimated that by 2026 the area managed under this practice may exceed 100 million ha, and the trend does not appear to be waning in the coming decades [111]. On an economic level, organic food products (food and drinks) had a market value of US\$97 billion in 2017, and again the trend is considered to be upward [111]. The growth of organic farming is closely linked to the development and use of innovative products no longer based on traditional chemistry but with important biological and microbiological components. To this end, it is possible to identify the new class of plant biostimulants as elements of a transition towards a BBA approach. Undoubtedly, soils managed with BBA approaches will, in the long term, see their need for inputs of any kind reduced. However, in the current scenario, where most agricultural soils were depleted in many of their

components, the use of (micro)-biological inputs, such as biostimulants, is a crucial step in halting the loss of soil fertility and can reintroduce classes of microorganisms important for re-establish a proper soil community.

In this context, biostimulants are considered as those products that fall strictly within the category defined by Regulation (EU) 2019/1009, such as amino acids, organic acids, seaweed extracts, plant extracts, mycorrhizal fungi, and plant growth-promoting rhizobacteria (PGPR). The Compound Annual Growth Rate (CAGR) of this class of products, for the period 2020–2025, is estimated at an annual rate of 11.56%, with the class of beneficial microorganisms showing even higher growth rates of 14.98% over the period under review. Growth in monetary terms is expected to take the global value of this market from US\$ 2000 million in 2019 to US\$ 3943 million in 2025 [112]. Apart from biostimulants, microorganism-based bioinsecticides are also showing steady growth rates at the market level. At the European level, the bioinsecticides market was worth US\$ 479 million in 2019 and is expected to be worth US\$ 1171.4 million in 2025, at a CAGR of 16% [113]. These numbers may appear poor when compared to those of conventional agriculture tools, where the fertilizer market alone has reached a value of US\$ 83.5 billion in 2020 [114] and the pesticide market of US\$ 84.5 billion in 2019 [115], however, a strong emerging interest on biostimulants and biocontrol agents is increasing. To this end, it is important to notice that the growth of the biostimulants market is higher than that of organic agriculture, indicating that more and more conventional farmers are relying on such innovative class of products [112], putting the focus on soil health as a foundational element of profitable agriculture. The use of biological products such as microbial-based biostimulants and bioinsecticides can be interpreted as one of the signs of interest in BBA approaches. The market trends of these products highlight that the attention towards productions that tend to preserve and enrich the biological components of the soil is high and, in the future, they will play an important role in the global market. The good aspect related to the diffusion in the use of these products is that their economic value is not limited only to the market price. In fact, the use of certain products, in view of a BBA, provides positive externalities related to the regeneration and conservation of ES, thus promoting positive feedback able to lead to the achievement of sustainable intensification of agriculture and going against the constant loss of ES caused also by conventional farming practices [109,116], in addition to their socioeconomic and environmental value in the diversification of production systems for other crops (e.g., cereals), food legumes have considerable value. Indeed, in countries of the global South, where galloping demography and natural resources are drying up under climate change, food legumes are the primary source of protein. Unfortunately, under the combined effect of the drought and salinity of the soils, their production has dropped drastically, which increased demand and thus their prices.

The 1986 oil shock, the reduction in the foreign exchange reserve, and the transition to a market economy in recent years did not help the situation and considerably penalized agriculture, including that of food legumes. Still, they are little cultivated despite their agronomic benefits, albeit the development of their culture is limited by the high instability of yields in the face of biotic and abiotic constraints such as water stress due to the instability of rainfall, salt stress resulting mainly from the rainfall deficit and evaporation during high temperatures summer and the deficiency of mineral elements in particular phosphorus [111].

In addition to the biotic and abiotic factors limiting their production other elements also disrupt their rehabilitation, in particular the absence of a suitable genotype selection, the nonexistence of a seed bank, the loss of suitable genetic resources, the high cost of chemical inputs, ignorance of new cultivation technologies and noncompliance by operators with suitable agricultural routes [111].

To increase their production and meet the demand of the food legume market, several solutions can be proposed on the research and development level and on the organizational level. Concerning the research plan, the urgency is to select resistant genotypes capable of maintaining satisfactory production in the extreme climates that prevail in these regions,

while allowing the market to be stabilized and satisfied. The selection of symbiotic microorganisms associated with these legumes is also an inevitable part of increasing the yields of these crops, mainly nitrogen fixers, PGPR, and mycorrhizal fungi. The choice of the appropriate symbiotic partner (s) allows these legumes to grow on poor soils while reducing expensive and potentially polluting synthetic fertilizers [66].

We conclude that the organizational aspect, the creation of an extension body, the link between the producers, and the marketing structures for the food legume sector, remain inactive in most Mediterranean countries.

2. Collaborative Research Project LEGU-MED: Legumes in Biodiversity-Based Farming Systems in Mediterranean Basin

To meet the above-mentioned issues, research gaps, and desirable targets for a valorization of legumes in biodiversity-based farming systems of the Mediterranean basin, we are currently developing this three-year project, which began on 1st November 2020. LEGU-MED is funded by PRIMA Foundation (Partnership for Research and Innovation in Mediterranean area) within Topic 2.2.2: RIA Use and management of biodiversity as a major lever of sustainability in farming systems. We established an international consortium made of 5 universities, 5 research institutes, and 1 private company from 8 countries: Italy, Germany, Spain, Algeria, Tunisia, Turkey, Lebanon, and Croatia (Figure 3).

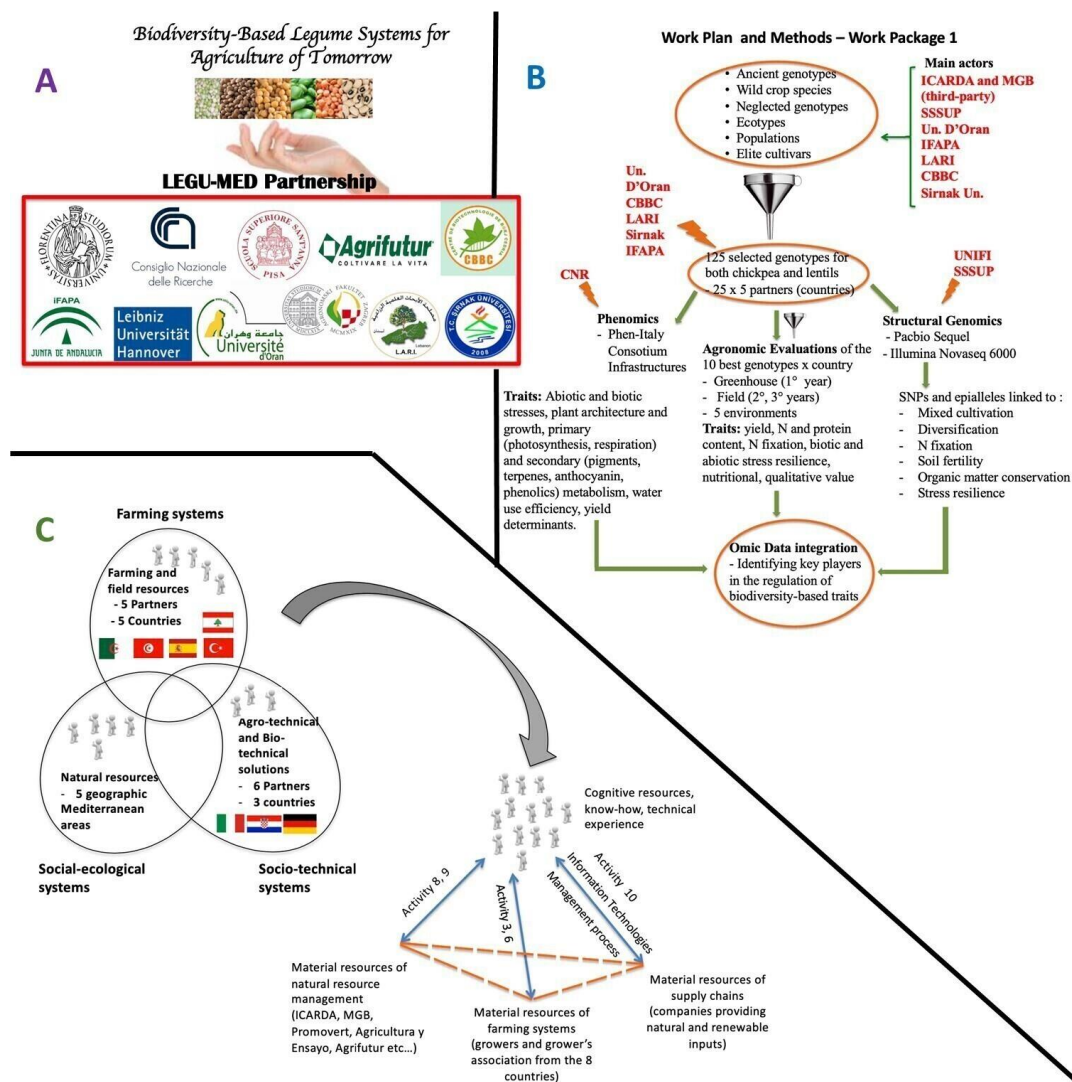


Figure 3. (A) International partnership of Legu-med proposal. (B) Structure of work package

1 composed of genomic, phenomic, and agronomic characterization of selected local and under-investigated autochthonous chickpea and lentil germplasm. (C) Integrated framework to design biodiversity-based farming systems at local level in Mediterranean basin.

The main objective of this project is to put forward an international and well-integrated plan to valorize, restore and manage the legume agrobiodiversity (including neglected genotypes and wild crop relatives) of the Mediterranean in biodiversity-based farming systems and consequently enhance agro-ecosystem functions and services in the Mediterranean basin. The proposal will use lentils and chickpeas as models for all grain legumes.

2.1. Goals

The project is structured in four work packages (WPs) and 10 activities aiming at obtaining the following three major objectives (Obj) (Figure 4):

Objectives-Activities	Countries	Institutions	Key Persons
Objective 1: Deeply characterize biodiversity of important legumes in Mediterranean basin			
Work Package 1: Agronomic, phenotypic and molecular characterization of lentil and chickpea biodiversity in Mediterranean basin			
Activity 1: Recovery, selection and collection of germplasm	MOROCCO ITALY ITALY TURKEY CROATIA TUNISIA SPAIN ALGERIA	ICARDA CNR UNIFI SU-AF ZAGREB UNIV. CBBC-INRAT IFAPA UNIV. D'ORAN	Shiv Kumar Agrawal Giuseppe Vendramin Federico Martinelli Derya Yucel Sanja Sikora Mariem Bouhadida Francisco Perez-Montano Abdelkader Bekki
Activity 2: Genomic and phenomic characterizations of the selected germplasm	ITALY ITALY ITALY	UNIFI CNR SSSUP	Federico Martinelli Francesco Loreto Mario Enrico Pe'
Activity 3: Agronomic screening and evaluation of germplasm in different geographic areas	TURKEY ALGERIA TUNISIA LEBANON	SU-AF UN. D'ORAN CBBC-INRAT LARI	Derya Yucel Abdelkader Bekki Mariem Bouhadida Rania El Nabbout
Objective 2: Enhance use and management of agro-biodiversity to enhance ecosystem services and farming system's sustainability in Mediterranean basin			
Work Package 2: Models and approaches for a biodiversity-based agriculture in legume-based farming systems			
Activity 4: Development of new models, tools and approaches for the exploitation of legumes in biodiversity-based agriculture	GERMANY ITALY SPAIN	UN. HANNOVER SSSUP IFAPA	Christina Von Haaren Paolo Barberi Dulce Rodriguez-Navarro
Activity 5: Agronomic approaches to enhance ecosystem services of legume-based farming systems	GERMANY SPAIN ALGERIA TUNISIA LEBANON	UN. HANNOVER IFAPA UN. D'ORAN CBBC LARI	Christina Von Haaren Dulce Rodriguez-Navarro Abdelkader Bekki Darine Trabelsi Rania El Nabbout
Work Package 3: Agro-technical and biotechnical solutions to improve management of functional agro-biodiversity in legume-based farming systems			
Activity 6: Developing new legume-rhizobia combinations with enhanced SNF	ITALY CROATIA SPAIN TUNISIA ITALY	UNIFI UN.ZAGREB IFAPA CBBC AGRIFUTUR	Alessio Mengoni Sanja Sikora Francisco Perez-Montano Ridha Mhamdi Roberto Kron Morelli
Activity 7: Agro-technical and bio-technical novelties to increase sustainability of farming systems	GERMANY SPAIN ALGERIA TUNISIA LEBANON	UN. HANNOVER IFAPA UN. D'ORAN CBBC LARI	Christina Von Haaren Dulce Rodriguez-Navarro Abdelkader Bekki Ridha Mhamdi Rania El Nabbout
Objective 3: Evaluation of trade-offs proposed measures with a cost/benefit analysis performed by stakeholder's			
Work Package 4: Socio-economic evaluations of proposed measures, information management, outcome disseminations and exploitation			
Activity 8: Socio-cultural assessment of the proposed biodiversity-based agriculture measures	GERMANY MOROCCO ITALY SPAIN	UN.HANNOVER ICARDA UNIFI IFAPA	Miguel Cebrian-Piqueras Shiv Kumar Agrawal Federico Martinelli Dulce Rodriguez-Navarro
Activity 9: Socio-economic evaluation of the proposed measures through stakeholder's involvement	MOROCCO TUNISIA ITALY MOROCCO	ICARDA CBBC AGRIFUTUR ICARDA	Aladdin Hamwiah Ridha Mhamdi Roberto Kron Morelli Shiv Kumar Agrawal
Activity 10: Outreach, stakeholder updates, training and deliverables dissemination	ALL COUNTRIES	ALL PARTNERS	All key persons

Figure 4. Objectives, activities, countries, and institutions of Legu-med proposal.

- (1) Deeply characterize the biodiversity of key legume species in the Mediterranean basin
- (2) Enhance use and management of agro-biodiversity to improve the provision of legume-based ecosystem services and farming system sustainability in the Mediterranean basin
- (3) Evaluate trade-offs of proposed measures, with cost/benefit analysis performed by stakeholders.

2.2. Research Project Structure

In the first WP, three activities will select chickpea and lentil germplasm and perform an agronomic, phenomic, and molecular characterization of these autochthonous and under-investigated plant materials (Figure 3). At least 125 genotypes for each legume (chickpea and lentil) for agronomic evaluation. These genotypes will be represented by wild relatives, local populations, landraces, ICARDA elite cultivars, neglected genotypes, under-investigated Mediterranean local genotypes from the eight countries with high breeding potential. We will perform a genomic characterization of at least 125 genotypes per legume using next-generation sequencing technology. The GWA (genome-wide association) study will make use of mixed linear models accounting for population structure such as FarmCPU [117].

GWA will result in the prioritization of molecular markers accelerating breeding for traits that will enhance the sustainability of legume-based farming systems. High-precision phenotyping will be performed on the ten best performing genotypes identified by the agronomic screening. These data will be matched with genomic data and GWA outcomes to validate the SNP identification and deliver potential markers usable for future breeding activities. The agronomic characterization in greenhouse and field conditions will include at least the following traits: drought resistance, nutritional qualitative parameters (protein, anti-nutritional, and nutraceutical compounds), biological nitrogen fixation (BNF), nutritional requirements, biomass production, other key plant growth measurements and plant functional traits important for BBA (e.g., resistance/tolerance to pests and diseases, weed suppression, tolerance of weed competition).

In WP2, models and approaches will be developed for BBA in legume-based farming systems. Within one activity, we will develop new spatially concrete models (based on multivariate statistics and geographical information system tools) allowing for a simple assessment of agrobiodiversity and different ecosystem services (provisioning, regulating, cultural, and supporting) at patch and farm level. The models will be developed and calibrated in collaboration with other research activities within LEGU-MED and will include a broad range of different typical biophysical conditions in the Mediterranean area, local farmers' know-how as well as socio-cultural and socioeconomic assessments. In the other WP2 activity (Table 1), these models will be applied in selected farm fields and landscapes from the participant collaborators from the regions under several scenarios of land-use intensity, as well as climate- and site conditions.

In WP3, agrotechnical and biotechnical solutions will be designed and tested to improve the management of functional agrobiodiversity in legume-based farming systems. Screening for persistent and stress-tolerant rhizobia strains which are at the same time highly efficient and compatible with host plant variety is of great importance for improving sustainable agricultural production without adverse impact on soil and environment. Collection of rhizobial strains nodulating chickpea and lentil will be prepared and biodiversity within rhizobial field populations will be estimated for the capacity to trigger better N₂-fixation performances. Therefore, rhizobial selection programs based on biodiversity studies can greatly contribute to the more successful utilization of BNF in BBA agriculture and achieve greater benefits from improved inoculants.

In synergy with the introduction of selected legumes in diversified cropping systems based on improved crop rotations and intercropping/living mulches, we will optimize management practices to enhance the provision of ecosystem services. In particular, given the recognized importance of conservation agriculture in Mediterranean environments,

reduced or no-tillage and continuous soil cover will be promoted to increase soil biological activity, fertility organic carbon stocks, water retention and on-field biodiversity.

In WP4, the activities consist of a well-defined plan of socioeconomic evaluations of proposed measures, information management, outcome dissemination, and exploitation. Social perceptions, acceptance, knowledge and demand, and cultural values (e.g., aesthetic and recreational value, context-based traditions) of components of BBA will be explored throughout the project, from the selection of genotypes to landscape arrangements. Besides, a detailed cost/benefit analysis will be performed to determine the trade-offs of the proposed measures tested in the previous WPs.

It is expected that WP4 will promote the facilitation of the transfer of proposed systems (e.g., cropping systems, biological fertilizers, and pesticides) to growers and other stakeholders as an eco-friendly system to boost plant productivity and their nutritional quality. The results will contribute to the generation of new models and strategies for knowledge cocreation, social learning, and identification of knowledge systems synergies.

2.3. Expected Results

In WP1, we expect to obtain: (1) an extensive molecular characterization of the germplasm collections (10K–20K SNP markers); (2) a description of the relatedness within germplasm collections, allowing prioritization of allele pools untapped by breeding; (3) an identification of Quantitative Trait Locus responsible for traits of agronomic relevance and improved understanding of the molecular mechanisms underlying agronomic performance. Some genotypes are expected to be found with higher values in comparison with current cultivars for these following desirable traits: productivity in drought conditions, N-fixation capability, key nutritional components.

In WP2, we will develop and test cropping systems based on legumes using two strategies: (1) integrating genetic diversity by matching the use of improved genotypes with improved crop rotations/intercropping, (2) increasing habitat diversity by including 5% of arable land to semi-natural areas aimed at increasing functional biodiversity (e.g., hedges, field margins, fallows). The enhancement of most of these services will enable farming systems to depend less on marketed inputs such as mineral fertilizers, pesticides, and irrigation water and will increase the sustainability and resilience capacity against expected and unexpected climate change scenarios. An improvement of actual provision of ES is expected using novel biodiversity-based cropping systems, optimizing the use and agronomic management of local legume germplasm. In WP3, methods to maintain high provisioning ES and non-marketable “environmental services” such as cultural values and biodiversity will also be developed. Improvements on management practices and seminatural habitats will be obtained to further scale up the provision of multiple ES by improved cropping systems based on higher genetic and species diversity. We expect to identify at least five to ten rhizobial strains with higher nitrogen fixation capacity than current strains specific for chickpea and lentil. The competitiveness of rhizobia strains will be tested in controlled conditions reproducing semiarid, partially desalinated, and acidic soils. The evaluation of growth capacity in semiarid and acidic soils represents a crucial and very promising tool for the competitiveness of selected rhizobia strains in those specific environments [118,119]. This approach is of extreme importance as an introduction of an elite rhizobia population with more efficient N-fixation might be hindered, due to competition with indigenous bacteria and genetic instability of the inoculating strains [120,121].

In WP4 the following results will be expected: (1) the development of new socioeconomic indicators for enhanced sustainability and resilience of tested farming systems based on BBA with focus on several sociocultural and economic domains, improvement of local markets and circular and solidarity economy, diversification, and variety of income sources for supporting a transition to BBA [122]; (2) innovations developed and used by multiactors indicating enhanced sustainability and resilience of at least two to three tested BBA farming systems, in comparison with those currently used; (3) cost/benefit analysis showed that at least two to three agrotechnological proposed solutions provide

improvements in ecosystem services compared to currently used ones; (4) indicators (participant's surveys) showing high interest of stakeholders to the project events (conferences, technical workshops, training sessions); (5) publications in high-impact scientific journals or trade magazines.

2.4. Impacts

Our ambition is to address the three key BBA challenges: valorization, restoration, and management. WP1 will ultimately valorize the wide biodiversity available in the Mediterranean basin selecting varieties, ecotypes, populations, local genotypes, and wild crop relatives adapted to different geographic environments. Restoration will be addressed by WP2 and WP3. Activities will develop agro-technologies and biotechnologies (e.g., the use of PGPR) to restore soil fertility and agriculture sustainability over the entire project lifespan and beyond. WP1 will: (1) identify and deliver new (under-investigated) plant genotypes; (2) identify new genes, molecular markers, molecular mechanisms of expression of key traits; (3) identify phenotypic variability of key traits for high-throughput selection of climate-ready plants; (4) characterize BBI encoding genes in chickpea, and (5) identify trait-trait correlations and tailor models capturing yield and yield components. WP2 will deliver new cropping systems to implement BBA. WP3 will: (1) identify and exploit a useful combination of new rhizobia elite populations and best legume germplasms; (2) identify and exploit useful plant-plant and plant-microorganism-animal (tritrophic) interactions; (3) conserve soil fertility and water resources; (4) improve nutrient recycling and use efficiency, and (5) lead to better environmental stress management, (6) optimize the provision of ecosystem services by integrating genetic, environmental, management and socio-economics aspects and priorities. Management will be addressed by WP4 which will have the following impacts: (1) creating a national task-force composed of 6–10 multiactors for each country with the aim not only to test solutions in multiactors fields but cocreate innovations and work strictly together in their development from the beginning to the need of the project; (2) demonstrating that the proposed changes of farming systems have a positive socioeconomic impact on Mediterranean agriculture; (3) creating an outreach program to train stakeholder communities to become multipliers of project innovations, and (4) disseminating the results of the project with an intense organization of meetings and publications.

In conclusion, LEGU-MED will establish a framework for the sustainable use of legumes in BBA as well as in other agroecological farming systems. Focusing on chickpea and lentil our aim is to test a multidisciplinary and integrative approach for BBA agriculture which can be applied for all kinds of legumes and similar renovation crops.

To be an example of well-integrated and coordinated research work performed in a participatory way with all food chain actors and in a way as modern agricultural research is performed in a multiactor approach as “living labs”.

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