

Review

# Alfalfa as a Biological Nitrogen Source and Biofertilizer Component in Sustainable Horticultural Production Systems

Vladimir Filipović<sup>1,\*</sup>, Elmira Saljnikov<sup>1</sup>, Snežana Dimitrijević<sup>1</sup>, Ljubica Šarčević-Todosijević<sup>2</sup>, Vera Popović<sup>3</sup>, Aleksandar Miletić<sup>4</sup>, Jelena Golijan Pantović<sup>5</sup>, Aleksandra Stanojković-Sebić<sup>6</sup> and Vladan Ugrenović<sup>6</sup>

<sup>1</sup> Institute for Multidisciplinary Research, National Institute of the Republic of Serbia, University of Belgrade, Kneza Višeslava 1, 11030 Belgrade, Serbia; esaljnikov@imsi.bg.ac.rs (E.S.); snezana.dimitrijevic@imsi.bg.ac.rs (S.D.)

<sup>2</sup> High Medical-Sanitary School of Professional Studies, Tošin Bunar 7a, 11080 Belgrade, Serbia; ljsarcevic@gmail.com

<sup>3</sup> Institute of Field and Vegetable Crops, Maksima Gorkog 30, 21000 Novi Sad, Serbia; vera.popovic@ifvcns.ns.ac.rs

<sup>4</sup> Faculty of Agriculture, University of Niš, Kosančićeva 4, 37000 Kruševac, Serbia; a.miletic78@gmail.com

<sup>5</sup> Faculty of Agriculture, University of Belgrade, Nemanjina 6, 11080 Belgrade, Serbia; golijan.j@agrif.bg.ac.rs

<sup>6</sup> Institute for Soil Science, Teodora Drajzera 7, 11000 Belgrade, Serbia; astanojkovic@yahoo.com (A.S.-S.); vladan.ugrenovic@gmail.com (V.U.)

\* Correspondence: vladimir.filipovic@imsi.bg.ac.rs; Tel.: +381-62522642

## Abstract

Alfalfa (*Medicago sativa* L.) is widely recognized as a major forage crop, yet its role as a multifunctional biological input in sustainable horticultural production remains underexplored. This review evaluates alfalfa as a biological nitrogen source, organic fertilization resource, and biofertilizer-supporting crop within vegetable, medicinal, and perennial horticultural systems. Due to its high capacity for biological nitrogen fixation, alfalfa can supply substantial amounts of plant-available nitrogen, reducing dependency on synthetic fertilizers and supporting environmentally sound nutrient management. When used as green manure, cover crop, intercrop, mulch source, compost feedstock, or processed organic fertilizer, alfalfa enhances the soil organic carbon (SOC), improves soil structure, and increases the water-holding capacity properties particularly critical in intensive horticultural production. Higher SOC levels also contribute to the improved tolerance of horticultural crops to drought and heat stress through enhanced soil moisture retention and rhizosphere buffering. Alfalfa-based organic inputs stimulate rhizosphere microbial biomass, enzymatic activity, and functional genes associated with nitrogen cycling, strengthening plant-microbe interactions that underpin biofertilizer effectiveness. Evidence from vegetable and perennial systems indicates that alfalfa-derived amendments and rotations increase soil nitrogen availability, support yield stability, and improve soil health over the long-term. In orchards and vineyards, alfalfa cover cropping contributes to carbon sequestration, erosion control, and enhanced soil biological functioning. Overall, alfalfa emerges as a strategic species for integrating organic fertilization and biofertilizer-based approaches into modern horticultural systems, supporting reduced mineral fertilizer inputs while sustaining productivity, soil health, and environmental quality.

**Keywords:** alfalfa (*Medicago sativa* L.); organic fertilization; biofertilizers; biological nitrogen fixation; green manure; soil organic carbon; soil health; nutrient cycling; sustainable horticulture; cover cropping; climate-resilient horticulture

Academic Editor: Anoop Kumar Srivastava

Received: 06 May 2026

Revised: 12 June 2026

Accepted: 16 June 2026

Published: 17 June 2026

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## 1. Introduction

In the context of accelerating climate change, soil degradation, and rising environmental pressures, horticultural production systems are increasingly required to shift from input-intensive conventional practices toward sustainable, resource-efficient models. Central to this transition is the replacement or substantial reduction in synthetic fertilizers through the use of organic fertilization strategies and biofertilizers, which enhance soil biological activity, nutrient cycling, and long-term productivity while reducing environmental externalities. As noted in the recent literature, biofertilizers have been proposed as key alternatives to synthetic inputs for sustainable crop production and for enhancing plant resilience and rhizosphere function in the face of biotic and abiotic stresses [1]. Consequently, the integration of multifunctional plant species capable of simultaneously supporting soil fertility, ecosystem services, and crop productivity has become a major focus of sustainable horticultural research.

Among such species, alfalfa (*Medicago sativa* L.) represents a particularly valuable biological resource. Although traditionally recognized as a forage crop, alfalfa possesses a suite of agronomic and ecological traits that make it highly relevant to organic fertilization and biofertilizer-based horticultural systems. As a perennial legume of the Fabaceae family, alfalfa is characterized by high biomass production, strong adaptability to diverse pedoclimatic conditions, and a pronounced capacity for biological nitrogen fixation. Globally, alfalfa accounts for approximately 30% of total legume cultivation [2], underscoring its importance as a strategic crop in sustainable agriculture.

According to the official 2023 Agricultural Census conducted by the Statistical Office of the Republic of Serbia, the national cattle population fell from 881,152 heads in 2018 to 725,408 in 2023, representing a 17.7% decrease over the five-year period [3]. Despite this decline, alfalfa retains substantial agronomic relevance beyond its traditional role as a forage crop. Through symbiosis with *Rhizobium* spp., alfalfa converts atmospheric nitrogen into plant-available forms, thereby functioning as a living biofertilizer that significantly reduces dependence on synthetic nitrogen inputs [4,5]. This characteristic is particularly valuable in horticultural cropping systems, where nitrogen availability strongly influences yield, quality, and nutrient-use efficiency. In addition, its deep and highly branched root system improves soil structure, enhances water infiltration and retention, mitigates erosion, and increases resilience to climatic extremes such as drought [6].

In recent years, alfalfa has gained renewed attention within bioeconomy and circular production models, where plant biomass is increasingly valorized as a renewable input for multiple end uses. Beyond forage, alfalfa biomass has been explored for biogas generation, plant-based protein extraction, composting, pelleting, and the production of organic fertilizers and soil amendments [7,8]. Furthermore, its capacity for phytoremediation, particularly the uptake and stabilization of heavy metals, highlights its multifunctionality in environmentally sensitive horticultural landscapes [9]. The growing scientific interest in the link between alfalfa, soil health, and regenerative agriculture is evident from the rapidly expanding body of literature, with more than 36,900 Google Scholar records, including over 17,000 published since 2022 (accessed 15 January 2026).

Current research increasingly emphasizes the role of biologically driven nutrient management, soil carbon sequestration, and water regulation as cornerstones of sustainable food production. With approximately 40% of global land already degraded and projections suggesting that up to 90% of fertile soils could be at risk by 2050 [10], the restoration of soil functionality through organic matter inputs and biologically mediated processes has become an urgent priority. According to the FAO [11], large-scale land restoration could sequester more than 50 Gt of CO<sub>2</sub> while simultaneously increasing global food production.

Within this framework, alfalfa offers a strategic contribution to sustainable horticultural production systems by acting as a source of organic nitrogen, biologically active residues, and microbial stimulation. Its ecological adaptability—thriving under annual precipitation ranging from 350 to over 1000 mm and temperatures from  $-10\text{ }^{\circ}\text{C}$  to  $28\text{ }^{\circ}\text{C}$ —combined with relatively low external input requirements, enables stable biomass yields of  $8\text{--}10\text{ t ha}^{-1}$  of dry matter in dry years and over  $15\text{ t ha}^{-1}$  under favorable conditions [6]. These attributes make alfalfa particularly suitable for integration into organic horticultural rotations, intercropping systems, and soil fertility-building phases.

Therefore, alfalfa should be viewed not only as a forage crop, but as a multifunctional component of organic fertilization and biofertilizer strategies in horticultural cultivation. This review synthesizes current knowledge on the agroecological, nutritional, and ecosystem-service functions of alfalfa, with a specific focus on its role in soil fertility enhancement, biological nutrient cycling, carbon sequestration, and resilience-building within sustainable horticultural production systems.

## 2. Alfalfa—A Green Pillar of Sustainable Agriculture and Horticulture

### 2.1. Alfalfa as a Nitrogen-Fixing Crop

Alfalfa plays a pivotal role in sustainable horticultural production systems due to its exceptional capacity for biological nitrogen fixation (BNF), positioning it as a natural biofertilizer within organic and low-input cropping frameworks (Table 1). Through a well-established symbiosis with *Rhizobium* spp., alfalfa converts atmospheric nitrogen ( $\text{N}_2$ ) into plant-available forms, substantially reducing the need for synthetic nitrogen fertilizers. This biologically mediated nitrogen input is particularly relevant for horticultural systems, where excessive mineral fertilization often leads to nitrate leaching, soil degradation, and water contamination [4]. Alfalfa may contribute to more efficient nitrogen management in horticultural systems through enhanced biological nitrogen inputs, improved nitrogen retention, and reduced nitrogen losses.

In organic fertilization strategies, nitrogen-fixing crops such as alfalfa are fundamental for maintaining soil fertility and sustaining crop productivity without reliance on external chemical inputs. By continuously supplying biologically fixed nitrogen to the soil–plant system, alfalfa supports nutrient cycling, enhances microbial activity, and contributes to the stabilization of soil organic matter, key objectives in regenerative and organic horticulture [5]. Moreover, alfalfa residues and root exudates stimulate diverse microbial communities, further strengthening the soil biological functions essential for nutrient availability in subsequent horticultural crops.

Reported annual nitrogen fixation rates for alfalfa typically range from  $50$  to  $450\text{ kg N ha}^{-1}$ , with approximately  $30\text{--}80\%$  of total plant nitrogen derived from atmospheric sources, depending on cultivar, soil conditions, and management practices [5,12–14]. Under optimal agroecological conditions, nitrogen fixation values of up to  $650\text{ kg N ha}^{-1}\text{ year}^{-1}$  have been documented [15]. Consequently, mineral nitrogen fertilization is generally recommended only during the establishment phase of alfalfa stands, at modest rates of  $30\text{--}50\text{ kg N ha}^{-1}$ , primarily to support early growth before effective nodulation is achieved [13].

From a horticultural perspective, the integration of alfalfa into crop rotations, intercropping systems, or fertility-building phases offers a reliable and environmentally sound nitrogen source for high-value vegetable, fruit, and specialty crops. Numerous studies have shown that alfalfa-based rotations significantly improve nitrogen availability and nutrient cycling in subsequent crops, including cereals and horticultural species such as leafy vegetables and solanaceous crops, thereby reducing dependence on external nitrogen inputs in subsequent crops [16]. Residual nitrogen released through root

turnover and biomass decomposition can supply a substantial proportion of the nitrogen demand of the following crops, thereby functioning as an in situ organic fertilizer.

Beyond direct nitrogen inputs, alfalfa-based BNF contributes to reduced greenhouse gas emissions by lowering the demand for energy-intensive synthetic fertilizers and minimizing nitrous oxide losses from soil. This multifunctional role, combining nitrogen provision, soil biological enhancement, and environmental protection, highlights alfalfa as a cornerstone species in biofertilizer-driven horticultural production systems aligned with the goals of sustainable and climate-resilient agriculture.

**Table 1.** Annual biological fixation of nitrogen in different plant species.

Plant Species	N Fixation (kg/ha/Year)	Additional Notes/Benefits	References
<i>Leucaena leucocephala</i>	100–500	Woody legume; used in agroforestry	[17–19]
Alfalfa ( <i>Medicago sativa</i> )	50–450	Deep root system; improves fertility and microbial activity	[5,12–14]
Azolla (aquatic fern)	80–300	Symbiotic with cyanobacteria; used in rice paddies	[20,21]
Soybean ( <i>Glycine max</i> )	50–300	Efficient symbiosis with <i>Rhizobium</i> strains; used in crop rotations	[17,22,23]
<i>Sesbania sesban</i>	60–250	Fast-growing species; used for soil restoration and nitrogen fixation in marginal lands	[12,18,19]
<i>Gliricidia sepium</i>	80–250	Agroforestry species; provides shade, soil protection, and nitrogen fixation	[17–19]
Legumes (average)	50–200	Fixation depends on species, microbiota, and growing conditions	[17,23,24]
Peanut ( <i>Arachis hypogaea</i> )	50–200	Tropical legume; used for soil improvement	[17,23]
Clover ( <i>Trifolium</i> spp.)	50–200	Important pasture legume; used for grazing and hay	[18,24]
Vetch ( <i>Vicia sativa</i> )	100–200	Green manure crop; commonly used in regenerative agriculture	[5,12]
Lentil ( <i>Lens culinaris</i> )	60–120	Suitable for arid areas; stable nitrogen fixation	[17,25,26]
Pea ( <i>Pisum sativum</i> )	60–120	Suitable for temperate climates; common in organic farming	[24,25]
Common bean ( <i>Phaseolus vulgaris</i> )	30–90	Lower fixation efficiency compared to soybean	[12,25]

In addition to its direct contribution to soil nitrogen supply, alfalfa exerts a profound influence on soil microbial activity and functional diversity, which is a central mechanism underlying its biofertilizer value in sustainable horticultural systems. Its deep and highly branched root system, often extending up to 3–4 m, penetrates the subsoil layers and creates vertically connected microbial habitats, thereby improving nutrient mobilization beyond the plough layer and enhancing whole-profile soil biological functioning.

Multiple studies have demonstrated that soils under alfalfa cultivation contain up to a 30–35% higher microbial biomass compared with soils under annual arable crops, reflecting increased microbial abundance and activity associated with perennial legume systems. Alfalfa strongly stimulates rhizosphere diversity, particularly favoring beneficial and functional microbial groups, including nitrogen-fixing genera (*Rhizobium*, *Bradyrhizobium*, *Azospirillum*), as well as microorganisms involved in nitrogen mineralization and transformation processes. In crop rotation systems, alfalfa inclusion has been associated with a 20–45% increase in the activity of key nitrogen-cycling enzymes, such as urease and nitrate reductase, compared with non-legume control plots [5,13].

From the perspective of organic fertilization and biofertilizer-based horticulture, these microbial effects are particularly relevant. Enhanced nitrification efficiency coupled with reduced denitrification rates has been observed under alfalfa, resulting in improved nitrogen retention in soil and lower gaseous nitrogen losses. This biological regulation of nitrogen fluxes directly supports environmentally sound nutrient management strategies and reduces dependence on external nitrogen inputs, one of the core objectives of sustainable horticultural production systems [5,13].

Further evidence of alfalfa's role as a biological driver of soil fertility is provided by molecular studies, which report a significantly increased abundance of functional genes involved in nitrogen cycling, such as "nifH" (nitrogenase reductase), "narG" (nitrate reductase), "nirK" (nitrite reductase), and "nosZ" (nitrous oxide reductase), within the alfalfa rhizosphere. The enrichment of these genes indicates enhanced nitrogen fixation, transformation, and stabilization capacity, reinforcing alfalfa's role as a living biofertilizer platform rather than merely a nitrogen-fixing crop [13].

An additional agronomic and horticultural advantage of alfalfa lies in its strong carry-over effects on subsequent crops. The inclusion of alfalfa in cropping systems enhances biologically available nitrogen and improves nitrogen cycling, which may support the nutrient requirements of subsequent crops while reducing synthetic fertilizer demand. When incorporated into multi-year rotations, alfalfa has been shown to increase wheat yields by up to 20% and maize yields by 12–18%, depending on soil type and climatic conditions [14]. Similar benefits have been reported for horticultural crops following alfalfa-based fertility-building phases due to improved nitrogen availability, soil structure, and microbial activity.

Importantly, symbiotic nitrogen fixation in alfalfa is strongly inhibited by excessive mineral nitrogen fertilization, with little or no additional benefit observed at application rates of 75 or 150 kg N ha<sup>-1</sup> [27]. Avoiding unnecessary nitrogen inputs not only reduces the production costs, but can also lower nitrogen oxide emissions by up to 40% compared with intensively fertilized monoculture systems [14]. This highlights alfalfa's strategic value in low-input and organic horticultural systems aimed at climate mitigation, nutrient efficiency, and long-term soil health.

In this context, alfalfa does not generate 'excess' nitrogen; rather, its agronomic value lies in enhancing biologically available nitrogen, improving nitrogen retention, and reducing the need for external nitrogen inputs. These mechanisms form the biological basis for several additional agronomic, ecological, and soil-improving functions of alfalfa discussed in the following sections, including crop rotation benefits, cover cropping, intercropping performance, and carbon sequestration.

## 2.2. Alfalfa as a Protein-Rich Crop

Alfalfa is also a high-value plant protein source, with crude protein concentrations typically ranging from 18% to 25% of dry matter, positioning it among the most nutritionally significant cultivated forage crops for sustainable protein provision (Table 2).

**Table 2.** Protein content and agronomic roles of selected plant species in soil fertility.

Plant Species	Protein Content (% Dry Matter)	Role in Soil and Fertility	References
Soybean ( <i>Glycine max</i> )	35–45%	Richest plant-based protein source; nitrogen fixer; important for agroecosystems and livestock.	[4,14,28,29]
Lentil ( <i>Lens culinaris</i> )	23–28%	Rich protein source, nitrogen fixer; improves fertility; also used in human nutrition.	[13,30–33]

Mimosa ( <i>Mimosa</i> spp.)	20–26%	Agroforestry species with nitrogen-fixing ability; enhances soil fertility and biodiversity.	[13,34]
Pea ( <i>Pisum sativum</i> )	20–25%	Nitrogen fixer, commonly used in crop rotation, enhances soil microbial activity.	[4]
Alfalfa ( <i>Medicago sativa</i> )	18–25%	High protein content, nitrogen fixer, improves microbial activity and soil fertility; deep roots up to 4 m.	[12,13,35,36]
Common bean ( <i>Phaseolus vulgaris</i> )	21–24%	Used in human nutrition; improves nitrogen fertility and microbial diversity.	[37–39]
Sainfoin ( <i>Onobrychis viciifolia</i> )	18–24%	Nitrogen fixer with resilient root system; enhances fertility and protects soil.	[40,41]
Black locust ( <i>Robinia pseudoacacia</i> )	18–22%	Agroforestry species, deep roots, nitrogen fixer; used for rehabilitation of degraded land.	[5,16]
Red clover ( <i>Trifolium pratense</i> )	15–22%	Large-seeded clover, important nitrogen fixer in arable and pasture systems; boosts microbial activity.	[4,5,42]
Grapevine ( <i>Vitis vinifera</i> )	8–12%	Agroforestry species with lower protein content; important for biodiversity and soil structure.	[5,43]

In 2018, the European Parliament adopted the European Strategy for the Promotion of Protein Crops, providing a comprehensive assessment of plant-based protein production across EU member states. The strategy explicitly recommends the reintroduction and expansion of protein-rich legumes, including soybean, common bean, pea, and perennial forages such as alfalfa, clover, and sainfoin, within both arable and mixed farming systems [44]. Legumes are highlighted not only for their protein yield, but also for their ecosystem services, particularly biological nitrogen fixation, soil fertility enhancement, and reduced dependence on synthetic fertilizers, key pillars of organic fertilization and biofertilizer-based nutrient management.

Despite these advantages, legumes currently occupy less than 4% of total agricultural land in the EU, reflecting a critical gap between policy objectives and on-farm adoption. Alongside biodiversity loss, agrobiodiversity erosion remains a major challenge, and the limited inclusion of legumes constrains nutrient cycling and soil regeneration within agroecosystems. Although EU plant-based protein production increased by approximately 28% over the past 15 years, the region remains heavily dependent on imports, underscoring the urgency of scaling up domestic protein crop production [45]. Recent policy and research initiatives emphasize legumes as strategic crops for reducing external protein dependency, enhancing crop diversification, and stabilizing nutrient flows in organic and sustainable systems [46].

Within this framework, alfalfa protein has emerged as a strategic bio-based resource. Advances in green biorefinery technologies have expanded its application beyond feed, enabling the extraction of high-quality leaf protein concentrates suitable for food, feed additives, and biostimulant formulations, thereby strengthening its relevance for sustainable horticultural value chains. Reflecting this growing importance, the European alfalfa protein market was valued at USD 12.62 billion in 2023 and is projected to reach USD 21.68 billion by 2031, with a compound annual growth rate (CAGR) of 9.44% between 2024 and 2031 [47]. These trends position alfalfa as a key multifunctional crop at the intersection of

organic fertilization, sustainable protein supply, and resilient horticultural production systems.

### 2.3. Alfalfa as a Pollinator Resource

Leguminous species play a dual role in sustainable horticultural systems by simultaneously contributing to soil fertility through biological nitrogen fixation and supporting pollinator communities that are essential for fruit and seed set in many horticultural crops. Within low-input and organic production systems, alfalfa represents a particularly valuable functional component, as it combines the attributes of an organic fertilization resource with a stable and abundant source of nectar and pollen for pollinators [48]. This multifunctionality aligns closely with biofertilizer-based approaches that seek to integrate ecological services into horticultural production.

Alfalfa is among the most important nectar-producing forage legumes in temperate regions (Table 3). Its flowering phase typically lasts 20–30 days and begins 10–35 days after cutting, depending on climatic and management conditions [49]. In practice, the second and third cuttings are frequently allowed to flower, extending the effective blooming period to approximately seven weeks, primarily during July and August. This period coincides with stable weather and peak pollinator activity, ensuring reliable nectar availability at a time when alternative floral resources in horticultural landscapes may be limited.

From a horticultural perspective, the inclusion of alfalfa in crop rotations, field margins, or flowering strips adjacent to vegetable, medicinal, and perennial fruit crops enhance pollination services and supports beneficial insect populations. Although the floral morphology of alfalfa may limit access for certain pollinator taxa and thus favor more specialized insect communities [50], its overall contribution to pollinator abundance and activity remains substantial. Importantly, the continuous supply of floral resources indirectly strengthens biological pest regulation, reducing the reliance on chemical plant protection products in organic and integrated horticultural systems.

Synergistic effects can be achieved by combining alfalfa with other flowering species in diversified flowering mixtures. For example, *Phacelia tanacetifolia*, which provides a 40–55-day flowering period beginning in early summer, is highly attractive to a broad spectrum of pollinators and is widely used in horticultural landscapes [51]. When integrated with alfalfa in strip or inter-row flowering systems, phacelia extends nectar availability across the growing season while simultaneously contributing organic biomass and supporting soil microbial activity. Similarly, alfalfa and yellow sweet clover (*Melilotus officinalis*) can yield up to 300–400 kg of honey ha<sup>-1</sup> while concurrently enriching soil nitrogen pools and stimulating beneficial microbial communities associated with organic fertilization processes [52,53].

As drought-tolerant legumes, alfalfa and related species ensure extended nectar provision even under water-limited conditions, a trait of increasing importance for sustainable horticulture in the context of climate change [54,55]. Additional species such as common vetch (*Vicia sativa*), which blooms for 20–30 days starting in late spring, also contribute valuable short-term nectar resources and enhance soil health through nitrogen inputs [56,57]. However, due to its longer flowering period, higher biomass production, and stronger integration with organic fertilization strategies, alfalfa remains the cornerstone species in pollinator-supportive systems designed for sustainable horticultural production.

**Table 3.** Flowering period length and honey production from selected legumes and other pollinator-friendly plants.

Plant Species	Flowering Period Length (Days)	Approximate Honey Yield (kg ha <sup>-1</sup> )	Pollinator Impact	References
Sweet clover ( <i>Melilotus officinalis</i> )	40–50	250–400	Very High	[52,53,56]
Sainfoin ( <i>Onobrychis viciifolia</i> )	30–40	200–350	High	[52,58]
Facelia ( <i>Phacelia tanacetifolia</i> )	40–50	200–300	Very High	[52,53,59]
Alfalfa ( <i>Medicago sativa</i> )	20–30	150–300	High	[56,58,60]
Buckwheat ( <i>Fagopyrum esculentum</i> )	20–30	150–250	High	[52,55,58]
Mustard ( <i>Brassica</i> spp.)	20–30	100–200	Moderate	[56,59]
Vetch ( <i>Vicia sativa</i> )	20–30	150–200	Moderate	[52,56,57]
Clover ( <i>Trifolium</i> spp.)	20–30	100–150	High	[56,59]
Soybean ( <i>Glycine max</i> )	30–50	80–150	Moderate	[50,56,58]
Cress ( <i>Lepidium sativum</i> )	10–15	50–100	Low	[52,59]

Beyond its well-recognized economic importance for apiculture, alfalfa represents a multifunctional agroecological component that significantly contributes to sustainable horticultural production systems. As a leguminous species, alfalfa plays a pivotal role in biodiversity conservation by supporting diverse and stable pollinator communities, particularly when integrated into organically managed or low-input agroecosystems. Although legume species differ in their attractiveness to specific pollinator taxa, alfalfa and related species such as yellow sweet clover (*Melilotus officinalis*) are widely acknowledged for their capacity to sustain wild pollinators and beneficial insects [59].

The ecological value of alfalfa extends beyond pollination services. Its compatibility with organic fertilization strategies and biofertilizer applications enhances soil biological activity, promotes nitrogen fixation through symbiosis with rhizobia, and improves soil structure and fertility. These functions indirectly benefit pollinators by fostering healthier, more resilient agroecosystems with increased floral resources and reduced reliance on synthetic inputs. Moreover, alfalfa stands provide refuge and foraging habitats for natural enemies of crop pests, thereby strengthening ecological balance and contributing to integrated pest management approaches aligned with sustainable horticultural practices [60].

Empirical evidence indicates that alfalfa cultivation positively influences pollinator abundance and diversity, which is critical for maintaining ecosystem resilience and long-term productivity in horticultural systems [53]. When combined with organic fertilization and biofertilizer-based soil management, alfalfa can thus serve as a strategic element within sustainable horticultural production, simultaneously enhancing soil health, pollination services, and overall agroecosystem functionality.

#### 2.4. Agro-Technical Benefits of Alfalfa Cultivation

Agronomically, alfalfa functions as a foundational biofertility-building species in sustainable horticultural production, serving as a key component of multi-year crop rotations, cover cropping systems, and intercropping designs. Building upon the biological mechanisms described in Section 2.1, alfalfa contributes to improved agro-technical performance through enhanced soil structure, erosion control, pollinator support, and improved conditions for subsequent crops. These multifunctional roles position alfalfa as a strategic crop in organic fertilization frameworks and biofertilizer-based management systems, where the emphasis is placed on internal nutrient cycling, biological nitrogen inputs, and long-term soil health rather than on external synthetic inputs [61].

Alfalfa is particularly well-adapted to erosion-prone and structurally degraded soils due to its deep and persistent taproot system, which commonly penetrates 3–5 m or more into the soil profile. This root architecture enhances soil aggregation, increases macroporosity, improves water infiltration, and stabilizes soil structure—processes that are essential for maintaining productive horticultural soils under reduced tillage and organic management regimes [62]. From a biofertilizer perspective, the extensive root system also supports a vertically stratified microbial community, facilitating nutrient mobilization and biological activity throughout the soil profile.

In organic plant production, tillage and cultivation practices shall be used that maintain or increase soil organic matter, enhance soil stability and soil biodiversity, and prevent soil compaction and soil erosion. The fertility and biological activity of the soil shall be maintained and increased: (a) by the use of multiannual crop rotation including mandatory leguminous crops (such as alfalfa) as the main or cover crop and other green manure crops; (b) in the case of greenhouses or perennial crops other than forage, by the use of short-term green manure crops and legumes as well as plant diversity; and (c) by the application of livestock manure or organic matter, preferably composted, from organic production [63].

In this regulatory and agronomic context, alfalfa represents a model crop for implementing organic fertilization strategies in horticulture, effectively bridging soil conservation, nutrient efficiency, and ecosystem-based production goals. Its multifunctionality allows it to serve simultaneously as a fertility-building phase, a biological nitrogen source, and a soil-protective cover, thereby supporting resilient and resource-efficient horticultural systems under both continental and semi-arid conditions.

### 2.5. Alfalfa in Multi-Year Crop Rotations

The study by Ugrenović et al. [64] recommends the inclusion of a two-year alfalfa phase within a five-year crop rotation, highlighting alfalfa's strategic function as a biological fertility-building component in organic and sustainable production systems (Figure 1). In crop rotation systems, the previously described nitrogen-fixing and soil quality improving functions of alfalfa contribute to increased productivity, improved nutrient cycling, and enhanced resilience of subsequent crops.

These diversified rotations significantly promote soil microbial biodiversity and functionality, stimulating populations of key microbial groups involved in nutrient cycling, including ammonifiers, saprophytic fungi, and *Azotobacter* spp. The resulting increase in microbial activity improves nitrogen mineralization efficiency, nutrient retention, and overall soil biological stability. From a horticultural perspective, this biologically enriched soil environment creates favorable preconditions for subsequent high-value crops, reducing dependence on external organic fertilizers and enhancing the resilience and sustainability of agroecosystems over the long-term.

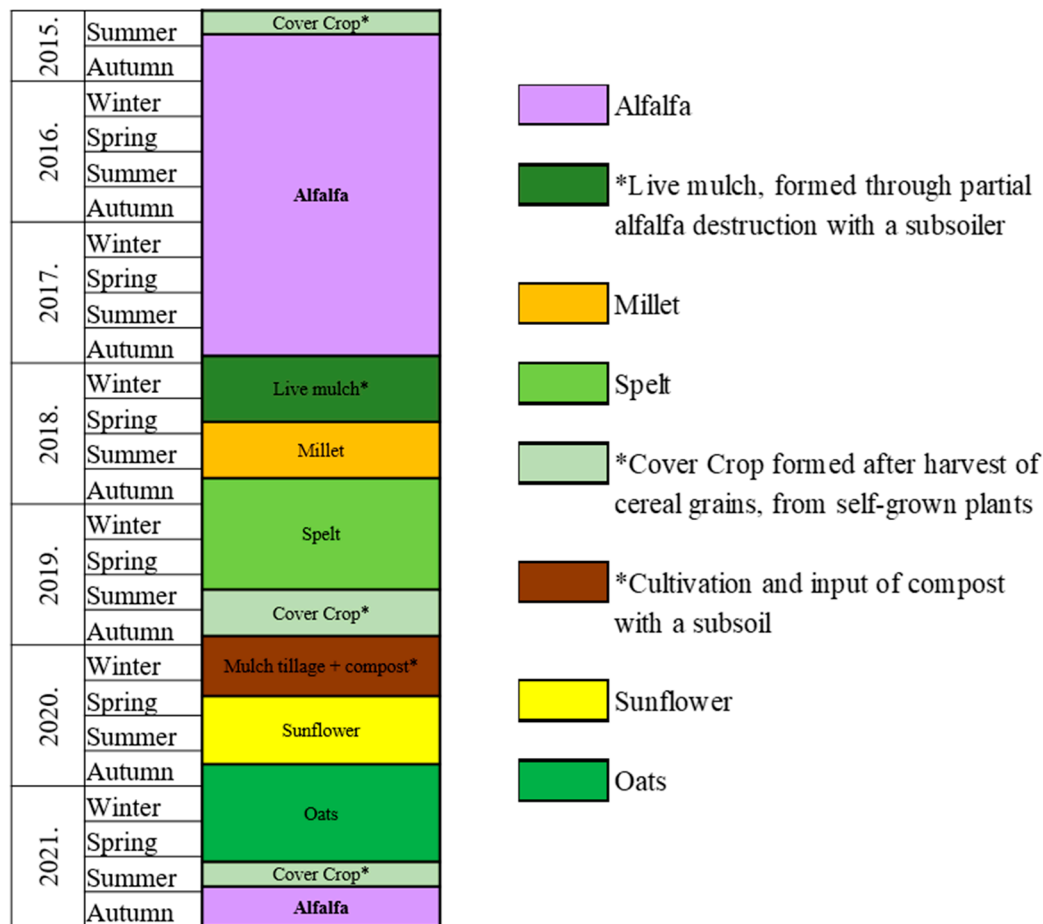


Figure 1. The model of crop rotation with the participation of alfalfa on an organic farm [64].

As a rotation crop, alfalfa plays a central role in biologically driven nutrient management, reducing nitrate leaching, interrupting pest and disease cycles, and improving both the yield and quality of subsequent crops, including cereals, oilseeds, and horticultural species. By functioning as an in-situ source of organic nitrogen, alfalfa and other grain legumes such as winter pea and vetch can supply up to 100% of the nitrogen demand of the following crop, thereby replacing or substantially reducing the need for external organic or mineral nitrogen inputs [65].

Under semi-arid and temperate continental conditions, multi-year alfalfa-based rotations have been shown to induce pronounced improvements in soil carbon and nitrogen dynamics. Field studies indicate that after four years of alfalfa cultivation, topsoil (0–5 cm) organic carbon increased by approximately 30%, accompanied by a 23.6% rise in total nitrogen content, outperforming wheat- and fallow-based systems [66,67]. These improvements are primarily driven by continuous root turnover, rhizodeposition, and the incorporation of alfalfa biomass, which together act as a biological soil conditioner and carbon-input system.

Long-term rotation trials conducted under continental to semi-arid agroecological conditions, typical of Vojvodina and the broader Pannonian Basin, further confirm alfalfa’s role in soil regeneration. On loess-derived and reclaimed soils, soil organic carbon (SOC) increases of 0.3–0.5 percentage points within 3–5 years have been reported, while multi-decadal reclamation experiments combining alfalfa with organic amendments resulted in five- to sixfold SOC accumulation, underscoring strong synergies between alfalfa-based rotations and organic fertilization strategies [68].

In addition to carbon sequestration, residual nitrogen contributions from alfalfa biomass and root turnover have been consistently quantified at 80–120 kg N ha<sup>-1</sup> available to

subsequent crops, effectively improving nitrogen use efficiency and supporting nutrient-demanding horticultural rotations [69]. These effects are most pronounced in environments receiving >450–500 mm annual precipitation and experiencing mean growing-season temperatures of 12–18 °C, conditions that are widely representative of lowland agricultural regions in Serbia.

Beyond Serbia, evidence from Mediterranean, temperate, and semi-arid production systems suggests that alfalfa-based crop rotations may provide comparable agroecological benefits, particularly through enhanced soil nitrogen availability, improved soil structure, greater resilience to drought, and reduced erosion risk. In Mediterranean and water-limited environments, alfalfa has demonstrated strong persistence and adaptive capacity due to its deep root system and efficient water-use strategies, contributing to improved resource-use efficiency stability of subsequent crops under climatic stress. Similarly, temperate cropping systems incorporating alfalfa in rotation have been associated with improved nutrient cycling, enhanced soil biological activity, and improved resource-use efficiency and long-term sustainability of production systems [70–73].

Collectively, these findings reinforce alfalfa's role as a keystone rotational crop that integrates organic fertilization, soil carbon restoration, and long-term productivity in sustainable horticultural and mixed farming systems.

From an economic perspective, although alfalfa may temporarily replace high-value horticultural crops during its establishment and multi-year persistence, its inclusion in horticultural rotations can generate substantial indirect agronomic and economic benefits through reduced fertilizer requirements, multiple annual harvests, high aboveground biomass production, relatively low-cost biological nitrogen inputs, improved soil fertility and structure, reduced pest and disease pressure, enhanced soil water retention, and improved productivity of subsequent crops. In vegetable–legume crop rotations, alfalfa may function as a fertility-building crop that sustainably enhances the agrotechnical potential of subsequent cultivated crops by improving the biological, physical, and chemical properties of soil. Therefore, the agronomic and economic value of alfalfa should be evaluated within a whole-rotation framework, considering both the direct and indirect benefits rather than through single-season crop comparisons [70,71,73,74].

#### *2.6. Alfalfa as a Cover Crop (Soil Protection and Water Relations)*

Within sustainable horticultural production systems, alfalfa functions as a multifunctional cover crop that integrates organic fertilization, soil protection, and improved water relations. Through symbiotic nitrogen fixation, alfalfa enriches soil nitrogen pools, directly supporting nutrient availability for subsequent horticultural crops while reducing dependence on synthetic fertilizers. Its perennial growth habit enables the formation of a persistent “living mulch”, which stabilizes soil structure, enhances aggregate formation, and promotes long-term soil fertility, key objectives in biofertilizer-based production systems [75].

Unlike annual cover crops, perennial legumes such as alfalfa continue to exert beneficial effects even after partial or complete termination of aboveground biomass. Nitrogen-fixing nodules remain active on the root system, supplying biologically fixed nitrogen to the soil and sustaining microbial activity associated with organic nutrient cycling. Beyond its previously discussed effects on soil fertility and biological activity, alfalfa as a cover crop particularly contributes to soil protection, water regulation, and resilience to climatic stress. Together with related perennial legumes (e.g., clovers and bird's-foot trefoil), alfalfa is among the most effective species for maintaining soil fertility and biological activity in horticultural rotations that prioritize organic inputs and ecological stability. In an apple orchard study, the use of alfalfa as a cover crop between tree rows enhanced soil nutrient status and altered fungal community structure, with alfalfa cover crops

increasing the total nitrogen (TN) and total carbon (TC) contents that are key factors regulating the soil fungal community and potentially improving soil fertility and microbial function under arid orchard conditions [76].

Alfalfa is also highly effective in mitigating soil erosion and improving surface stability, which is particularly relevant for horticultural fields characterized by frequent soil disturbance and irrigation. Comparative studies have demonstrated that soil loss in alfalfa-covered plots was reduced by 44.3% compared with fallow systems, whereas wheat reduced soil loss by only 18.2% [66]. During peak growth (July), surface vegetation cover in alfalfa stands can reach up to 97.5%, substantially exceeding that of cereal-based systems (~82.5%), thereby minimizing runoff and enhancing rainfall infiltration. Similar results have been reported under temperate continental conditions, where alfalfa reduced soil loss by approximately 30–45% relative to fallow land and maintained surface cover above 90% during critical erosion-prone periods [77].

From a water relations perspective, alfalfa's deep and well-developed root system—commonly extending to depths of 1.5–3.0 m, and even deeper under favorable subsoil conditions—significantly improves the soil water storage and access to subsoil moisture. This trait enhances drought resilience within horticultural rotations and stabilizes production under rainfed conditions. However, biomass and forage yields remain strongly influenced by seasonal precipitation patterns, with reported rainfed yields in continental regions typically ranging from 3 to 12 t DM ha<sup>-1</sup> depending on annual climatic variability [78].

Collectively, these characteristics position alfalfa as a cornerstone cover crop for sustainable horticultural systems, particularly in erosion-prone areas and rainfed production zones in Serbia. By simultaneously providing organic nitrogen inputs, improving soil structure and hydrology, and supporting microbial-driven nutrient cycling, alfalfa exemplifies the integration of cover cropping and biofertilization strategies central to the objectives of sustainable horticultural production (Table 4).

**Table 4.** Ecosystem services provided by alfalfa when used as a cover crop.

Ecosystem Service	Mechanism	Quantitative Evidence/Relevance for Horticulture	References
Soil erosion control	Dense canopy and permanent ground cover reduce soil detachment and surface runoff	Soil loss reduced by ~30–45% compared with fallow; surface cover often >90% at peak growth	[66,77,78]
Biological nitrogen input	Symbiotic N <sub>2</sub> fixation via <i>Rhizobium</i> spp.	Residual N contribution typically 80–120 kg N ha <sup>-1</sup> to subsequent crops, reducing synthetic N demand	[12,13,65]
Improvement of soil structure	Deep taproot system and root turnover enhance aggregation and porosity	Root penetration commonly 1.5–3.0 m, improving infiltration and reducing compaction	[6,78]
Soil carbon sequestration	Continuous biomass input from roots and residues	SOC increases of 0.3–0.5 percentage points within 3–5 years under continental conditions	[13,64,66]
Water regulation	Improved infiltration and access to deep soil moisture	Reduced runoff and enhanced soil water storage, improving drought resilience in rainfed systems	[77,78]
Enhancement of soil biodiversity	Root exudates and organic inputs stimulate microbial activity	Increased abundance of beneficial microflora, including ammonifiers and free-living N-fixers	[75,79]

Weed suppression	Permanent cover and competitive growth reduce weed emergence	Lower weed pressure and reduced reliance on mechanical or chemical control [80,81]
Support to organic fertilization strategies	Living biofertilizer integrated into crop rotations	Facilitates low-input nutrient management consistent with organic and biofertilizer-based systems [61,82]

Beyond Serbia, studies from Mediterranean, temperate, and semi-arid production systems indicate that alfalfa used as a cover crop can similarly contribute to reduced soil erosion, improved soil water infiltration, enhanced moisture retention, and greater resilience to drought-related stress. In Mediterranean and water-limited environments, the deep root system of alfalfa has been associated with improved water-use efficiency and stabilization of the soil structure, thereby supporting subsequent crop performance under increasingly variable climatic conditions. Likewise, temperate agroecosystems have reported positive effects of alfalfa cover on soil aggregation, nutrient retention, and long-term soil health, particularly in diversified crop rotations [71,73,74,83].

By delivering multiple ecosystem services simultaneously, ranging from biological nitrogen input and soil carbon sequestration to erosion control and microbial stimulation, alfalfa used as a cover crop represents a cornerstone species for organic fertilization and biofertilizer-oriented horticultural systems, directly supporting the goals of sustainable input reduction and long-term soil resilience emphasized in this Special Issue.

#### Alfalfa as an Intercrop (Productivity and Resource Use Efficiency)

Intercropping systems represent a core strategy in sustainable horticultural production, particularly where organic fertilization and biofertilizer-based nutrient management are prioritized. Within such systems, the nitrogen-fixing and rhizosphere-mediated functions of alfalfa, previously described in Section 2.1, support improved resource-use efficiency productivity and resource-use efficiency. By reducing reliance on synthetic nitrogen fertilizers and enhancing soil ecological balance, alfalfa-based intercropping systems directly support the principles of low-input and organic horticulture.

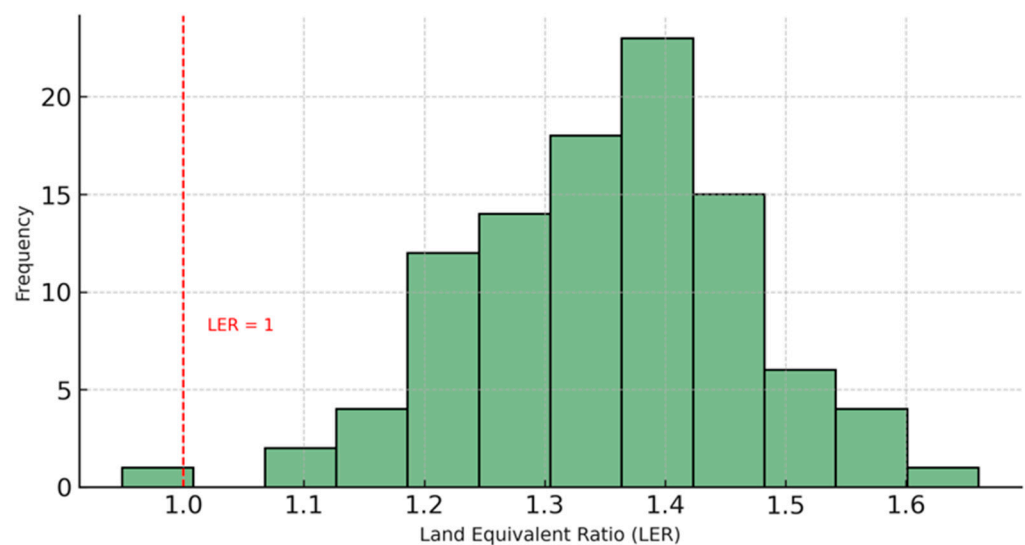
A key advantage of alfalfa as an intercrop lies in its ability to reduce interspecific competition through functional complementarity. Differences in rooting depth, nutrient acquisition strategies, and temporal resource use between alfalfa and companion crops (e.g., cereals, vegetables, or woody perennials) allow for a more efficient exploitation of soil water and nutrients, resulting in improved overall system performance [80,81,84]. In addition, root exudates and enhanced rhizosphere microbial activity associated with alfalfa promote nutrient mineralization and biological nutrient transfer, reinforcing the role of biofertilization in these systems. Intercropping alfalfa as a leguminous green manure in a citrus orchard significantly increased the soil total nitrogen by 48.7–74.7%, nitrate nitrogen by 50.0–96.7%, and available phosphorus by 44.5–45.0% compared with the monoculture. Alfalfa intercropping also enhanced soil enzyme activities such as sucrase by 63.8%, indicating improved soil nutrient cycling and biological activity in orchard systems [85].

Empirical evidence demonstrates that alfalfa intercropping significantly enhances productivity and land-use efficiency. In walnut–alfalfa intercropping systems, yield increases of 42.8% were recorded under a twice-mowed regime (MA-2), while a thrice-mowed regime (MA-3) resulted in yield gains of up to 67.5% relative to unmowed controls [86]. Across a wide range of intercrop configurations, land equivalent ratios (LERs) consistently exceeded 1.0, confirming the superior productivity of alfalfa-based intercropping systems compared with corresponding monocultures (Figure 2). Land equivalent ratio (LER), a commonly used indicator of intercropping efficiency, was used to evaluate the

productivity advantage of alfalfa-based intercrops relative to monocropping systems. In general, LER values  $> 1$  indicate a productivity advantage of intercropping, LER = 1 indicates equivalent performance, whereas LER values  $< 1$  indicate lower land-use efficiency compared with monoculture systems. Reported LER values for alfalfa–maize and related systems range from approximately 1.13 to 1.62, reflecting a 13–62% advantage in land productivity per unit area [67,69].

Beyond yield benefits, alfalfa intercropping markedly improves nitrogen availability and nutrient retention. Partial LER contributions from the legume component frequently correspond to biological nitrogen inputs exceeding  $50 \text{ kg N ha}^{-1}$ , supplied through symbiotic fixation and subsequent nitrogen transfer within the cropping system. This biological nitrogen contribution substantially reduces the external nitrogen requirements for associated horticultural crops in subsequent growing seasons, reinforcing the role of alfalfa as a living biofertilizer [68]. Management practices, particularly mowing frequency and spatial arrangement, further modulate system performance, with optimized cutting regimes increasing the total system yield by approximately 40–70% compared with less intensive management.

These benefits have been consistently observed across continental and semi-arid environments and are highly relevant for mixed and smallholder farming systems common in Serbia. When locally optimized, alfalfa-based intercropping systems offer a robust pathway to enhance productivity, nutrient cycling, and resource use efficiency while adhering to the principles of sustainable horticultural production. Collectively, these findings underscore alfalfa's multifunctional role as a key biological component in intercrop designs that integrate organic fertilization, biofertilizers, and ecological intensification.



**Figure 2.** Distribution of land equivalent ratio (LER) values demonstrating productivity and resource-use efficiency advantages in alfalfa-based intercropping systems.

### 2.7. Alfalfa as a Nutritional Source of Minerals, Vitamins, and Chlorophyll

Alfalfa is notably rich in essential nutrients, including vitamins (A, B1, B2, B4, C, D, E, K, PP), minerals (K, P, Ca, Mg, Fe, Zn, Na), xanthophylls, saponins, amino acids, and organic acids such as malic, malonic, and citric acid [87]. Table 5 provides a comparative overview of key vitamins and minerals in alfalfa and several other commonly cultivated plant species.

**Table 5.** Key vitamins and minerals in alfalfa and selected cultivated plants.

Plant Species	Dominant Vitamins	Dominant Minerals	References
Alfalfa	Vitamin K (531 mcg/100 g), Vitamin A (4500 IU/100 g), Vitamin C (100 mg/100 g)	Calcium (250 mg/100 g), Magnesium (80 mg/100 g), Iron (6 mg/100 g)	[88–90]
Soybean	Vitamin K (25 mcg/100 g), Vitamin B6 (0.2 mg/100 g), Vitamin C (6 mg/100 g)	Calcium (277 mg/100 g), Iron (15 mg/100 g), Magnesium (280 mg/100 g)	[88,91,92]
Clover	Vitamin K (120 mcg/100 g), Vitamin A (800 IU/100 g), Vitamin C (30 mg/100 g)	Calcium (160 mg/100 g), Magnesium (80 mg/100 g), Iron (5 mg/100 g)	[88,89]
Pea	Vitamin K (24 mcg/100 g), Vitamin C (40 mg/100 g), Vitamin B1 (0.3 mg/100 g)	Calcium (25 mg/100 g), Magnesium (50 mg/100 g), Iron (2.5 mg/100 g)	[88,93,94]
Black bean	Vitamin K (6 mcg/100 g), Vitamin C (5 mg/100 g), Vitamin A (20 IU/100 g)	Calcium (50 mg/100 g), Iron (2.5 mg/100 g), Magnesium (100 mg/100 g)	[88,95]
Maize (Corn)	Vitamin B1 (0.3 mg/100 g), Vitamin C (6 mg/100 g), Vitamin A (187 IU/100 g)	Calcium (10 mg/100 g), Magnesium (37 mg/100 g), Iron (0.5 mg/100 g)	[88,95]
Spinach	Vitamin K (483 mcg/100 g), Vitamin A (9376 IU/100 g), Vitamin C (28 mg/100 g)	Calcium (99 mg/100 g), Magnesium (79 mg/100 g), Iron (2.7 mg/100 g)	[88,93]
Swiss chard	Vitamin K (830 mcg/100 g), Vitamin A (4000 IU/100 g), Vitamin C (30 mg/100 g)	Calcium (102 mg/100 g), Magnesium (80 mg/100 g), Iron (1.5 mg/100 g)	[88,89]
Pumpkin	Vitamin A (5000 IU/100 g), Vitamin C (9 mg/100 g), Vitamin B6 (0.1 mg/100 g)	Calcium (20 mg/100 g), Magnesium (37 mg/100 g), Iron (1.2 mg/100 g)	[88,93,95]

In addition to these essential vitamins and minerals, alfalfa is especially valued for its high chlorophyll content, which has been widely utilized for decades as a health-promoting dietary supplement (e.g., in powdered green supplements or capsules). In addition to the agronomic functions described previously, alfalfa also represents an important nutritional resource of chlorophyll content (Table 6).

**Table 6.** Chlorophyll content and common applications of selected plant species.

Plant Species	Chlorophyll Content (mg/g Fresh Biomass)	Common Applications and Purposes	References
Green Tea ( <i>Camellia sinensis</i> )	3.0–3.5	Used in food and beverage industries; rich in antioxidants	[96,97]
Kale ( <i>Brassica oleracea</i> )	2.5–3.0	Vitamin-rich vegetable used in human diets	[98,99]
Spirulina *	1.5–3.0 (dry matter)	Dietary supplement; rich in protein and chlorophyll	[100,101]
Alfalfa ( <i>Medicago sativa</i> )	2.0–2.5	Used as livestock feed, dietary supplement, in the food industry, biostimulant, soil enhancer	[102,103]
Soybean ( <i>Glycine max</i> )	1.5–2.2	Source of soybean meal, oil, and animal feed	[104,105]
Green Bean ( <i>Phaseolus vulgaris</i> )	1.0–1.5	Protein- and nutrient-rich vegetable; consumed fresh or cooked	[106,107]
Wheat ( <i>Triticum aestivum</i> )	0.8–1.1	Staple cereal; flour and animal feed production	[108,109]

\* Cyanobacteria belonging to the genus *Arthrospira*.

As shown in Table 6, alfalfa is among the plant species with the highest chlorophyll content, which justifies its cultivation for chlorophyll extraction and processing. Chlorophyll derived from alfalfa possesses notable antioxidant and immunomodulatory properties, making it useful as a dietary supplement in both human and veterinary nutrition [9]. Medical studies suggest that it may aid in managing skin disorders, body odor, and even certain types of cancer. Chlorophyll from alfalfa is generally considered safe for human consumption for those interested in its potential health benefits.

Furthermore, the chlorophyll content in alfalfa and its hybrid *Medicago × varia* Martyn can be significantly enhanced through the application of biostimulants such as humic acids and amino acids. Studies have shown that these treatments lead to increased chlorophyll concentrations, thereby improving photosynthetic efficiency and boosting fresh biomass yield [102].

Given its rich chlorophyll content and associated bioactivities, alfalfa has potential as a functional dietary ingredient in the prevention and management of metabolic disorders—particularly metabolic syndrome, which comprises a cluster of interconnected conditions such as obesity, hypertension, dyslipidemia, and impaired glucose regulation [110].

### 2.8. Alfalfa as Organic Fertilizer and Soil Enhancer

Building upon the soil-enhancing mechanisms previously described, alfalfa biomass and processed materials can function as valuable organic amendments that improve nutrient availability, soil organic matter, and biological activity, which are central principles of biofertilizer-based horticultural systems. This positions alfalfa not only as an organic fertilizer source, but also as a biological soil activator that enhances the effectiveness of microbial biofertilizers by improving habitat conditions, carbon supply, and root–microbe interactions. Alfalfa is also recognized for its capacity to serve as a raw material in the production of nitrogen-rich organic fertilizers containing essential macro- and micronutrients. Due to its high potassium uptake, the aboveground biomass of alfalfa is particularly rich in potassium [82], an essential nutrient involved in various physiological processes related to plant growth, development, and resilience.

Moreover, alfalfa is increasingly used as green manure (sideration). When incorporated into the soil at the appropriate growth stage, typically 6 to 8 months after sowing, depending on agroecological conditions, it contributes to improved soil structure, nutrient content, and reduced environmental footprint. The plants are mowed, and the green biomass is shallowly incorporated into the soil, serving as a natural fertilizer [111]. To maintain effectiveness, the alfalfa should not be allowed to overgrow, as excessive biomass may hinder soil incorporation.

Green manuring with alfalfa also suppresses weeds and enhances soil biodiversity by supplying organic matter and habitat for soil microorganisms, fungi, and beneficial invertebrates [79].

Alfalfa biomass can further be used to produce high-quality compost, contributing to sustainable nutrient supply in organic farming systems. Compost made from the first and third swaths of alfalfa, as well as residues from the second swath used for seed production, achieved optimal values of total nitrogen (5.04%), organic matter (42.56%), C/N ratio, pH, moisture content, and electrical conductivity [64]. The study recommends a two-year alfalfa cycle within a five-year rotation, with fields fertilized using on-farm compost during non-alfalfa years (Figure 1). Microbiological analysis including total microflora, ammonifiers, fungi, and *Azotobacter* species, demonstrates alfalfa's positive impact on soil microbial biodiversity and activity. Furthermore, organic management practices that boost SOC and microbial biomass carbon by ~18–30% demonstrate how biologically

driven systems typical of horticulture outperform many conventional cereal rotations in sustaining soil health [112].

The combined presence of nitrogen (N) and potassium (K) makes alfalfa a valuable organic input, not only in the form of green manure and compost as previously discussed, but increasingly also as processed fertilizer products such as pellets and briquettes [61]. The combined effect of reducing mineral fertilization and adding biofertilizers on broccoli yield, soil microbial abundance, and activity demonstrated that biofertilizer application can maintain or improve crop yield and soil quality while lowering dependence on mineral inputs in intensive horticultural systems [113]. Owing to its dual capacity to fix atmospheric nitrogen and accumulate potassium, alfalfa-derived biofertilizers (e.g., compost, pellets = Figure 3) offer promising alternatives to synthetic N and K fertilizers in sustainable agricultural systems. Although alfalfa pellets are traditionally and often more profitably used in animal nutrition, their application in horticultural systems may provide important agronomic and ecological benefits beyond direct market value. In regions with limited or absent livestock production, alfalfa biomass and pelletized materials may assume a stronger bio-agrotechnical role by improving soil fertility, stimulating microbial activity, reducing dependence on external fertilizer inputs, and contributing to long-term soil sustainability. Therefore, the economic suitability of alfalfa pellets depends strongly on regional production context and intended end use [70,114,115]. Compared with animal manures and composts, alfalfa-derived inputs offer a more predictable nitrogen release pattern due to their relatively narrow C:N ratio, lower risk of salinity, and lower pathogen load [116]. These properties make alfalfa-based fertilizers particularly suitable for intensive horticultural systems, including greenhouse vegetable production and young orchards, where nutrient synchronization and soil biological balance are critical [117]. In organic greenhouse tomato production, the application of legume-derived green manure including alfalfa pellets increased the plant available soil nitrogen and significantly enhanced yield compared to the unfertilized control. In horticultural systems, alfalfa-derived composts and pellets have shown particular value in vegetable production (e.g., tomato, pepper, lettuce), orchards, and perennial horticultural crops, where nutrient release synchronization and soil biological stimulation are critical. Specifically, in orchards where legumes were intercropped as green manure, the inclusion of alfalfa increased the content of soil available nitrogen by over 90% compared with control plots without legume green manure, indicating the potential of alfalfa to function as a biofertilizer–nitrogen source in organic horticultural systems [118]. Specifically, alfalfa pellet treatments at 330 g m<sup>-2</sup> and 660 g m<sup>-2</sup> increased the total fruit yield of tomatoes by approximately 19% and 33%, respectively, relative to the control, and promoted higher leaf nitrogen concentrations, demonstrating the effectiveness of legume-based organic fertilization in horticultural systems [119]. Organic fertilizers improve nitrogen availability, nutrient retention, and soil fertility in horticultural systems, helping meet the high nitrogen demand of intensive vegetable and perennial crops. Improved nitrogen synchronization from alfalfa-derived amendments is particularly important for horticultural crops characterized by shallow root systems and high nitrogen uptake rates, such as lettuce, spinach, tomato, and pepper, where insufficient N availability rapidly reduces yield and quality [120,121]. For instance, legume-derived amendments significantly increased the soil available N and tomato yields by up to ~33%, and similar organic amendments boosted growth and N uptake in leafy vegetables such as Chinese cabbage by enhancing N mineralization and availability [122,123]. In greenhouse lettuce (*Lactuca sativa* L.) production, the application of organic fertilizers supplying 90 kg N ha<sup>-1</sup> significantly enhanced the shoot fresh and dry weight compared to unfertilized controls, supporting the role of legume-derived or plant-based organic fertilization approaches for soil fertility and crop performance in short-cycle horticultural crops [124].



**Figure 3.** Pelleted alfalfa biomass used as an organic fertilizer and carrier matrix for biofertilizer application in sustainable horticultural production systems (photo: V. Filipović 18 December 2025).

This is particularly relevant in the context of EU policy goals. The “Farm to Fork Strategy for a fair, healthy and environmentally-friendly food system” [COM (2020) 381] and the “Biodiversity Strategy for 2030—Bringing nature back into our lives” [COM (2020) 380] both aim to reduce overall pesticide use and associated risks by 50% and cut synthetic fertilizer use by at least 20% by 2030 [125,126]. These targets emphasize the need for alternative plant nutrition and soil enhancement strategies, of which alfalfa-based organic fertilizers represent a viable solution.

Although comprehensive global data regarding the commercial production and use of alfalfa-based fertilizers (e.g., pelleted, briquetted, composted, or extract-based products) remain limited, emerging evidence suggests growing interest in alfalfa-derived amendments within sustainable and low-input agricultural systems. Rather than representing a fully quantified global market trend, current observations primarily indicate region-specific adoption driven by increasing interest in organic soil amendments, reduced dependence on synthetic fertilizers, and environmentally oriented farming practices. In this context, alfalfa-based products should be considered a promising but still insufficiently documented component of the broader transition toward sustainable nutrient management, warranting further agronomic and economic investigation [70,114,115,127,128].

### 2.9. Alfalfa as a Carbon Sequestration Crop

Alfalfa, when integrated into crop rotations with annual species, helps reduce nitrate leaching and enhance soil organic carbon (SOC) sequestration. Due to its efficient nitrogen uptake, alfalfa can significantly lower groundwater nitrate contamination when used in rotation, without negatively affecting subsequent crops. In general, legumes in rotations, especially alfalfa, reduce the need for mineral and organic nitrogen fertilizers, which account for approximately 25% of direct agricultural greenhouse gas (GHG) emissions in the EU [129,130].

Incorporating legumes into long-term rotations positively impacts soil properties and reduces herbicide requirements, as alfalfa and clover suppress weed growth, benefiting subsequent crops. Although most data originate from arable rotations, the soil-regenerating effects of alfalfa are particularly relevant for horticultural soils, which are typically more intensively managed, frequently tilled, and more prone to structural degradation. However, expanding crop rotations can improve agroecosystem stability, biodiversity, and yield. Product quality also improves (e.g., higher protein content, reduced mycotoxins), pest pressure is reduced by breaking pest life cycles, and pollinator and predator populations increase. Nevertheless, legumes are also susceptible to specific pests, such as rodents [131].

Unfortunately, the lack of awareness about sound agronomic practices limits the adoption of systems aligned with the EU Green Deal. Strategies such as long, diverse rotations, intercropping, and permanent soil cover can disrupt pest and weed cycles, potentially rendering synthetic pesticides obsolete [132]. In addition to the biological and agroecological functions discussed previously, alfalfa contributes to long-term carbon sequestration through sustained biomass production, root turnover, and reduced reliance on synthetic nitrogen fertilizers. Legume production systems are often more variable and less productive than cereals or oilseeds, leading to a yield gap. A Canadian study found that cereal-legume rotations reduced the net GHG emissions by 17–35% compared to monocultures due to enhanced N availability from mineralized organic residues. In Europe, legume integration into cereal rotations has improved plant-based protein production while reducing the environmental impacts [14].

Soil carbon plays a critical role in the carbon cycle, plant development, and climate regulation. Carbon sequestration refers to the process of absorbing CO<sub>2</sub> from the atmosphere and storing it as carbon in soil organic matter. In horticultural production systems, enhanced soil organic carbon (SOC) levels are directly associated with improved soil structure, increased aggregate stability, higher water-holding capacity, and improved nutrient buffering capacity. Soil organic carbon significantly enhances the soil water-holding capacity and aggregate stability, in some soils increasing the available water capacity by ~1.5–2.5 mm per 30 cm soil profile per 1% SOC increase [133]. These soil functions are particularly critical for high-value vegetable and perennial horticultural crops, where yield stability, root development, and nutrient availability strongly depend on soil physical and biochemical quality. Because horticultural crops often have shallow and highly active root systems, improvements in topsoil SOC exert a disproportionately stronger effect on their performance compared with deep-rooted field crops. Long-term management practices such as intercropping with native grasses significantly increased the soil organic carbon (SOC) and total nitrogen in orchard systems, with the SOC content rising by 118.3–184.2% compared to conventional tillage, indicating that soil biological enhancement through organic and cover cropping practices can profoundly improve the soil nutrient status in horticultural production systems [134].

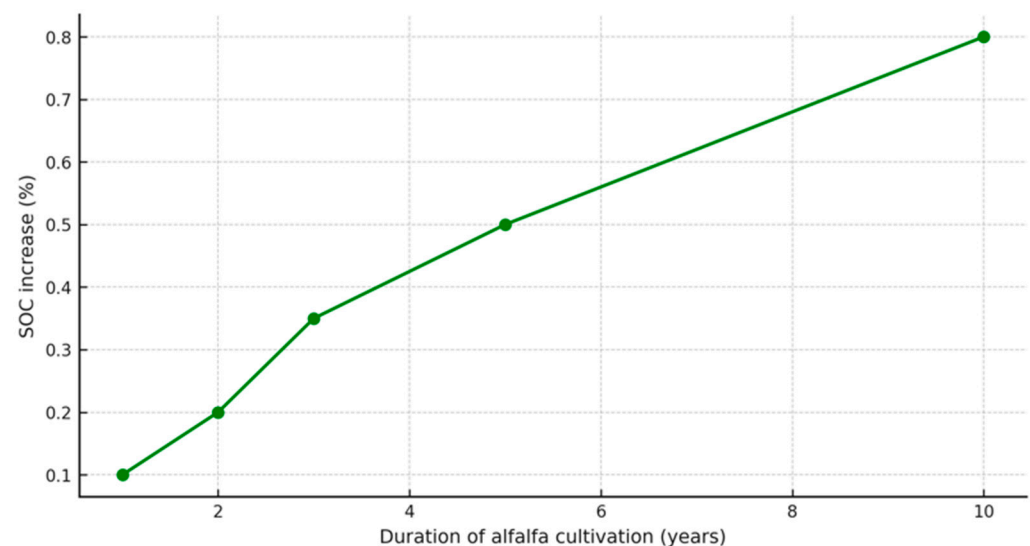
Conversion to intensive vegetable cultivation significantly increases the soil organic carbon (SOC), with the SOC levels rising on average by ~58% after conversion from regular field crops, indicating that horticultural systems inherently accumulate more SOC as a response to organic amendments and intensive management than most cereal-dominated

systems [135]. Numerous studies have demonstrated that SOC-enriching practices, including the use of organic amendments and legume-based rotations, significantly improve soil porosity, reduce bulk density, and enhance plant-available water and nutrient retention in intensive horticultural systems [133,136]. In vegetable production systems, higher SOC levels significantly improve crop tolerance to abiotic stresses such as drought and heat by enhancing soil moisture retention, nutrient buffering, and rhizosphere biological activity, leading to improved physiological performance and yield stability under stress conditions [137]. One of the most effective ways to reduce atmospheric CO<sub>2</sub> is through photosynthesis. Wide rotations that include alfalfa, combined with proper residue management, enhance SOC sequestration, particularly on low-fertility soils [138]. According to Saljnikov & Ugrenović [139], alfalfa residues with a low C:N ratio (7–11:1) decompose faster than cereal residues with wider ratios, contributing to improved organic matter dynamics.

Atmospheric carbon sequestration has emerged as a promising strategy to offset GHG emissions and mitigate climate change. Enhancing SOC also restores degraded soils [140]. However, no global studies to date have quantified the effect of converting land to alfalfa cultivation on SOC levels. Nevertheless, due to its deep root system (up to 3 m), alfalfa has considerable carbon sequestration potential. Long-term cultivation can increase SOC by 0.3–0.5% over five years, contributing to climate stability [6,10]. The cultivation of perennial legumes such as alfalfa significantly contributes to the accumulation of soil organic carbon, particularly when grown continuously over multiple years. Numerous studies confirm that extended alfalfa cultivation leads to a progressive increase in soil organic carbon, especially after the fifth year (Table 7 and Figure 4).

**Table 7.** Duration of alfalfa cultivation and increase in soil organic carbon content.

Duration of Cultivation (Years)	Increase in Soil Organic Carbon (%)	References
1–2	0.1–0.2	[141]
3–5	0.3–0.5	[142]
>5	>0.5	[143]



**Figure 4.** The increase in soil organic carbon (SOC) content over time.

In combination with practices such as reduced tillage and intercropping, this effect is further enhanced. Due to its deep root system, alfalfa improves the soil carbon retention, thereby reducing greenhouse gas emissions. Long-term cultivation of alfalfa, especially in crop rotation systems, increases the soil organic matter content, enhancing both carbon

sequestration and soil quality. These practices not only lower the carbon footprint of agricultural systems, but also improve soil fertility, making it more suitable for subsequent crops.

### 3. Additional Considerations for Horticultural Systems

In perennial horticultural systems such as orchards and vineyards, the integration of alfalfa as a cover crop or intercrop presents specific management challenges. Long-term ground cover may increase the habitat suitability for rodents (e.g., voles and mice), which can cause bark damage to fruit trees, particularly during winter periods. This risk is more pronounced in young orchards and high-density plantings, where trunk protection and root systems are more vulnerable. Furthermore, the perennial nature of alfalfa may compete with fruit trees for water and nutrients under rainfed conditions if mowing regimes and spatial arrangements are not properly optimized. These constraints highlight the necessity of adaptive management strategies, including regulated mowing, strip or alley cropping designs, and integrated pest control, to balance ecosystem services with orchard productivity.

In vegetable production systems, additional limitations arise from short cropping cycles, intensive soil disturbance, and the need for precise crop termination timing. While alfalfa-derived inputs (green manure, compost, pellets) offer clear benefits for soil fertility and microbial activity, the direct use of standing alfalfa as a cover crop may be constrained by the narrow planting windows typical for high-value vegetables. Improper or delayed termination can temporarily immobilize nutrients or interfere with seedbed preparation, particularly in early spring plantings. Therefore, in horticultural rotations with short-season vegetables, alfalfa is most effectively utilized as a preceding crop, off-season green manure, or processed organic fertilizer rather than as a continuous living cover.

Overall, these system-specific constraints do not diminish alfalfa's relevance for sustainable horticulture but instead emphasize that its benefits are highly management-dependent. When alfalfa integration is tailored to the biological, temporal, and structural characteristics of orchards and vegetable systems, potential risks can be mitigated, allowing its multifunctional contributions to organic fertilization, soil health, and agroecosystem resilience to be fully realized. Such system-specific constraints further emphasize that alfalfa functions most effectively in horticulture when managed as part of an integrated organic fertilization strategy rather than as a stand-alone cover crop.

The synthesis of benefits and constraints indicates that when appropriately managed, alfalfa consistently delivers net agronomic, ecological, and environmental gains, positioning it as a strategically important crop for sustainable, regenerative, and climate-resilient agricultural systems rather than a short-term production input (Table 8).

**Table 8.** Agronomic, ecological, and horticultural implications of alfalfa-based organic fertilization systems.

Aspect	Key Benefits	Potential Constraints/Risks	Management Implications
Nitrogen fixation	Biological nitrogen fixation of 50–450 kg N ha <sup>-1</sup> year <sup>-1</sup> ; reduced dependence on synthetic fertilizers; improved N availability for subsequent crops	Reduced fixation under excessive mineral N fertilization or poor nodulation	Optimize inoculation, avoid excessive N inputs, integrate into rotations
Soil fertility and structure	Increased soil organic carbon (+0.3–0.5% over 3–5 years); improved aggregation, porosity, and microbial activity	Lower performance on acidic, compacted, or poorly drained soils	Soil pH correction (liming), drainage improvement, proper site selection

Carbon sequestration	High belowground biomass and deep rooting (up to 3–5 m); enhanced long-term SOC accumulation	Carbon gains require multi-year persistence and proper residue management	Long-term rotations, reduced tillage, residue retention
Water relations and erosion control	Deep root system enhances water infiltration and drought resilience; soil erosion reduced by 30–45% compared to fallow	Yield variability under prolonged drought in rainfed systems	Cultivar selection, adaptive cutting regimes, moisture-conserving practices
Crop rotation and system productivity	Yield increases of subsequent crops (e.g., +10–20% for cereals); break in pest and disease cycles	Multi-year stand may limit short-term cropping flexibility	Strategic rotation planning aligned with farm objectives
Intercropping and land-use efficiency	Land equivalent ratios (LERs) > 1.0 (up to 1.62); improved nutrient and resource use efficiency	Requires careful spatial and temporal management	Optimized row spacing, mowing regimes, crop combinations
Biodiversity and pollinator support	Extended flowering period; high nectar value (150–300 kg honey ha <sup>-1</sup> ); support for beneficial insects	Floral morphology may favor specific pollinator groups	Use in diversified flowering mixtures and agroecological designs
Nutritional and bioeconomic value	High protein (18–25% DM), rich in minerals, vitamins, chlorophyll; biomass usable for feed, fertilizers, and bio-based products	Market and processing infrastructure may be locally limited	Development of local value chains and circular bioeconomy
Pests and diseases	Generally resilient in diversified systems; contributes to reduced pesticide use	Susceptible to specific pests, diseases, and rodents in long-term monocultures	Integrated pest management, rotation diversity
Economic considerations	Long-term soil improvement and reduced input costs	Higher establishment costs; delayed economic returns	Long-term planning, policy support, incentives for sustainable practices

#### 4. Conclusions

Alfalfa should be viewed not only as a forage crop, but as a strategic biological input for sustainable horticultural production systems. This review highlights that alfalfa-based practices directly support the core principles of organic fertilization and biofertilizer-driven nutrient management emphasized in this Special Issue. Through its high biological nitrogen fixation capacity, substantial biomass production, and favorable residue quality, alfalfa functions as a natural nitrogen biofertilizer, reducing reliance on synthetic N fertilizers in vegetable, fruit, and medicinal crop systems. Alfalfa-derived inputs including green manure, compost, mulches, and processed organic fertilizers enhance soil nitrogen availability while simultaneously stimulating rhizosphere microbial activity, improving nutrient cycling and nutrient use efficiency in horticultural crops with high nutrient demands. In addition, alfalfa-based organic fertilization significantly contributes to soil organic carbon (SOC) enrichment, which is particularly important in horticultural systems characterized by shallow root zones, intensive soil disturbance, and high irrigation requirements. Improved SOC enhances soil structure, water-holding capacity, and nutrient buffering, thereby increasing crop resilience to abiotic stresses such as drought and heat, key challenges in modern horticulture. Beyond nutrient supply, alfalfa integration through rotations, cover cropping, or intercrops supports soil biological fertility, carbon sequestration, erosion control, and biodiversity, reinforcing the ecological sustainability of horticultural production. These multifunctional benefits make alfalfa especially valuable in low-input, organic, and climate-resilient horticultural systems. Overall, the

evidence positions alfalfa as a cornerstone species for organic fertilization strategies and biofertilizer-based intensification in horticultural cultivation. Its integration into vegetable, orchard, vineyard, and medicinal plant systems provides a practical pathway to improve soil health, enhance biological nutrient cycling, and maintain productivity while reducing dependence on synthetic inputs. These functions are especially relevant for intensive vegetable and perennial horticultural systems, where soil degradation, high nutrient demand, and increasing climate stress necessitate biologically based fertilization strategies.

**Author Contributions:** Conceptualization, V.F., A.M. and V.U.; methodology, V.F. and V.U.; software, V.F.; validation, V.F., E.S., V.P., L.Š.-T., A.M. and V.U.; formal analysis, V.F. and S.D.; investigation, L.Š.-T., J.G.P. and V.P.; resources, V.F., S.D., L.Š.-T. and V.U.; data curation, V.F., V.P. and J.G.P.; writing—original draft preparation, V.F.; writing—review and editing, V.F. and E.S.; visualization, V.F.; supervision, V.F., E.S., A.S.-S. and V.U.; project administration, E.S. and A.S.-S.; funding acquisition, V.F. and A.S.-S. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grants numbers 451-03-33/2026-03/200053, 451-03-33/2026-03/200032, 451-03-34/2026-03/200383, 451-03-34/2026-03/200116, and 451-03-33/2026-03/200011. The APC was funded by 451-03-33/2026-03/200053.

**Data Availability Statement:** The raw data supporting the conclusions of this article will be made available by the authors on request.

**Conflicts of Interest:** The authors declare no conflicts of interest.

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