

Review

The Impact of Reduced N Fertilization Rates According to the “Farm to Fork” Strategy on the Environment and Human Health

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Abstract: The use of synthetic fertilizers, including nitrogen [N] fertilizers, is an indispensable element in today’s agriculture. Through adequate fertilization, farmers have the opportunity to increase crop yields, which is essential in view of the growing population and demand for food. The European Union’s “Farm to Fork” [F2F] strategy, as part of the broader European Green Deal, aims to promote more sustainable agricultural practices by reducing chemical fertilizer use by 20% by 2030. This initiative is designed to mitigate the negative environmental impacts of excessive N application, such as soil and water contamination, greenhouse gas emissions, and biodiversity loss. In addition to addressing ecosystem concerns, this strategy also aims to reduce health risks associated with N overuse, such as the accumulation of nitrates [NO₃[−]] in crops, which can lead to the formation of carcinogenic compounds. By integrating alternative fertilization methods, the agricultural sector can work toward more resilient and environmentally friendly systems while maintaining productivity. This paper focuses on a summary of the current knowledge about the consequences of N fertilization reduction and its connection to the soil environment, crops, yields, and human health.

Keywords: agrobiotechnology; public health; nitrogen fertilization; Green Deal; environmental protection



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1. Introduction

Nitrogen [N] fertilization plays a critical role in modern agriculture, contributing to increased crop yields and the improved quality of plant production [1]. N, as a fundamental nutrient, supports plant growth; however, excessive use is associated with significant environmental and human health risks. Improper management of synthetic fertilizers, particularly N-based ones, can lead to soil and groundwater contamination, which has serious implications for ecosystem well-being and public health [1].

Excess N that is not assimilated by plants migrates from the soil to groundwater in the form of NO₃[−] [2]. This phenomenon results from processes such as leaching and irrigation, which facilitate the transfer of N to surface and groundwater, leading to their contamination [1,2]. The degradation of biodiversity and the health of aquatic ecosystems can therefore exert indirect health effects, impacting food quality and the availability of safe, natural resources. A notable consequence of water contamination is the consumption of drinking water with elevated nitrate [NO₃[−]] levels, which poses a risk of developing the so-called “blue baby syndrome” (methemoglobinemia) in infants [3]. This condition can lead to serious disturbances in oxygen transport within the body. Furthermore, prolonged dietary exposure to NO₃[−], resulting from the consumption of plant products that have been intensively fertilized with N and exhibit high concentrations of nitrogenous compounds, may promote the formation of carcinogenic nitrosamines [NOCs] [2,4]. This situation is particularly concerning, as NO₃[−] tends to accumulate most in the leafy parts of plants, including leaves and stems, due to these tissues’ high metabolic demand and NO₃[−] storage capacity [5]. Several key regulatory bodies mandate NO₃[−] testing and set maximum limits

for NO_3^- levels in specific high-risk crops, especially leafy greens, due to the health risks associated with excessive NO_3^- consumption [1,5,6]. The European Union [EU] has some of the most comprehensive regulations on NO_3^- levels in vegetables, particularly leafy greens such as spinach, lettuce, and arugula [6]. The European Commission's Regulation No 1881/2006 sets maximum allowable NO_3^- concentrations for certain fresh vegetables, with separate limits for produce grown in different conditions [i.e., open field vs. greenhouse] and seasons, as NO_3^- levels tend to be higher in winter-grown crops [5,6]. Recent studies have highlighted the risks associated with NO_3^- accumulation in vegetables, particularly leafy greens [spinach, lettuce, arugula, swiss chard], due to the intensive use of N-based fertilizers [7]. NO_3^- levels are influenced by factors like light, temperature, and fertilization methods, with leafy vegetables grown in controlled environments accumulating especially high levels [7]. Sustainable farming practices and improved environmental management can help reduce NO_3^- levels, ensuring safer consumption levels [5–7].

In addition, the EU has implemented the “Farm to Fork” [F2F] strategy as part of the European Green Deal [8,9]. This strategy aims to reduce the use of chemical fertilizers by 20% by 2030, addressing the negative impact of agriculture on the environment and human health [10,11]. A key component of this initiative is the reduction of N fertilizer applications, which, while posing challenges for farmers, could contribute to environmental quality improvement and a decrease in health threats. More particularly, the F2F strategy consists of four axes of action: (i) sustainable food production, (ii) sustainable food processing and distribution, (iii) sustainable food consumption, and (iv) food loss and waste prevention [10]. A specific goal of the first axis of the strategy is to achieve reductions of 20% in nutrient inputs and 50% in nutrient losses, including N, without deteriorating soil fertility [9–11]. Minimizing the N surplus is a key target of the F2F strategy as part of a broad effort to mitigate the impact of agriculture on both global change and environmental pollution [8–10]. However, a critical concern of adopting mitigation practices based on the reduction of N fertilization is the potential trade-off in yields [10,11]. It should be emphasized at this point that adoption of the F2F strategy is expected to result in a slight decrease in yields under current climatic conditions, but this effect is likely to be compensated in the future (mid-century) by positive effects of increased carbon dioxide [CO_2] levels [9,10].

It is also necessary to mention that before F2F, in the context of the NO_3^- environmental problem, in 1991, the Council Directive 91/676/EEC was applied, concerning the protection of waters against pollution caused by NO_3^- from agricultural sources. Under this directive, farmers across Europe have implemented numerous practices to reduce NO_3^- levels in water, particularly in designated Nitrate Vulnerable Zones [NVZs] [12]. NVZs are areas where groundwater or surface water is at a high risk of NO_3^- pollution due to agricultural runoff. In these zones, farmers must follow specific regulations to minimize NO_3^- leaching, such as restricted fertilizer application periods and mandatory buffer zones along waterways. The Nitrates Directive prohibits farmers from applying fertilizers on soils that are surface-frozen, covered with snow, or waterlogged, as fertilizers cannot be absorbed by the soil under these conditions and can easily leach into water sources. Additionally, farmers must observe designated fertilization periods and maintain minimum distances between fertilizer application areas and flowing or standing water bodies. Farmers across Europe have implemented a variety of practices to mitigate NO_3^- pollution [12]. These include the development of comprehensive nutrient management plans, supported by soil sampling and nutrient budgeting techniques. Increasingly, precision agriculture tools, such as GPS sensors, are being employed to optimize fertilizer application across different field zones, and GPS scanning data are used to establish databases that can be used to monitor soil conditions [12]. This precision helps reduce the risk of excessive NO_3^- application in specific areas, thereby minimizing environmental impact. One of the measures farmers are taking to support the idea of fertilizer reduction is the increasing use of crop rotation with legumes, which is a key agricultural practice that offers multiple environmental and economic benefits, aligning closely with the EU's F2F strategy. One of the primary benefits of including legumes in rotation systems is their ability to naturally fix N in the soil [13].

Unlike many other plants, legumes form symbiotic relationships with N-fixing bacteria, capturing atmospheric N and converting it into forms that plants can absorb and use [8,13]. This process enriches the soil and reduces the need for synthetic fertilizers, which are energy-intensive to produce and can contribute to greenhouse gas emissions [13]. By decreasing fertilizer dependency, legume rotation addresses both environmental sustainability and climate objectives central to the F2F strategy. The integration of these practices is essential for improving nutrient use efficiency and ensuring compliance with EU regulations designed to protect water quality and reduce eutrophication risks [12,13].

Consequently, it is essential to conduct extensive environmental research to better understand the impact of the moderate use of synthetic fertilizers, particularly N-based ones. Such studies should focus on developing fertilization strategies that balance the need for high yields with the need for environmental and public health protection. Fortunately, there are already tools that are able to evaluate this, such as models like STICS, which is a soil–crop model that simulates soil N (mineral and organic), water, and carbon [C] dynamics on a daily basis in bare soil and under crops according to soil characteristics, weather, crops, and crop management [14]. dos Reis Martins et al. [15] stated that yield losses in mowed grasslands (the Alpine region) due to reduced N fertilization according to F2F recommendations are offset by positive effects of rising CO₂ levels.

Concurrently, it is crucial to raise awareness among both farmers and consumers, as the quality of food production is directly linked to agricultural practices. In line with the adage “you are what you eat”, attention must be given to the entire food production chain, beginning with the initial stage of proper agricultural land preparation [16]. Education for farmers regarding sustainable fertilizer application and responsible agricultural practices should be accompanied by outreach initiatives directed at consumers to enhance their understanding of the impact of fertilization on food quality.

In this study, it was hypothesized that limitations in N fertilization could have a positive effect on soil chemistry [i.e., by reducing acidification or improving the uptake of nutrients by plants] and the biodiversity of microorganisms associated with the N cycle in the soil environment and would not, ultimately, significantly reduce yields. In addition, it was hypothesized that a reduction in fertilization could contribute to lower NO₃[−] contents in food products, with beneficial effects on human health.

2. Yielding as an Effect of Reduced N Fertilization

N is essential for plant growth as it is a key component of proteins, chlorophyll, and nucleic acids, driving processes like photosynthesis and protein synthesis [7,17]. Consequently, N directly influences the growth rate of plants, their productivity, and the quality of the yields obtained [18]. Plants that receive adequate N doses exhibit vigorous vegetative growth, characterized by lush green leaves and a strong root system, which enhances their ability to absorb water and nutrients from the soil [19]. Roots develop in response to N availability because N affects both the depth and spread of the roots [19]. When N is abundant, plants tend to develop a denser root system, which increases nutrient and water absorption [19]. This structural adaptation enables the plant to support faster growth and withstand periods of low moisture. In the soil, N typically occurs as NO₃[−] and ammonium ions [NH₄⁺], which are absorbed by roots through specialized transporters [7,19]. The transport of nitrate within the plant is facilitated by specialized NO₃[−] transporters located in root cell membranes [7]. These transporters enable the uptake and movement of NO₃[−] from the soil into root cells and, from there, throughout the plant. The two primary transporter types, NRT1 and NRT2, vary in their affinity for NO₃[−] and are activated depending on the concentration of nitrate in the soil [7,19].

Figure 1 presents the process of N absorption by the plant and its use in creating chlorophyll and the synthesis of amino acids and proteins, ultimately contributing to plant growth and photosynthesis.

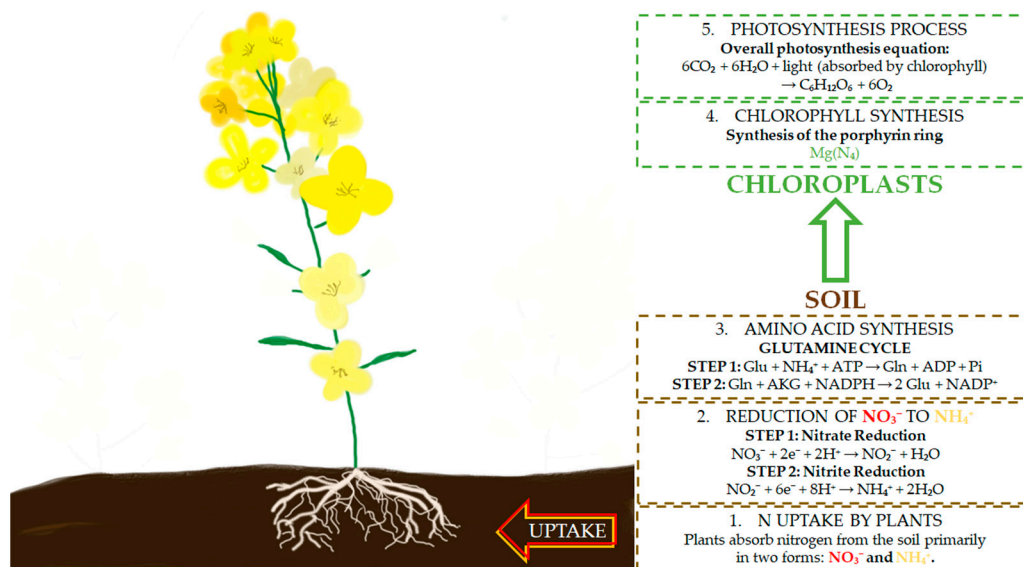


Figure 1. The role of nitrogen in photosynthesis. Gln: glutamine; Glu: glutamate; AKG: alpha-ketoglutarate (own work by Kagan K.).

N is absorbed by plants from the soil predominantly in two forms: NO_3^- and NH_4^+ . These forms become available through the processes of organic matter [OM] mineralization and soil microbial transformations, such as nitrification and denitrification [19]. Once taken up by plants, NO_3^- undergoes reduction to NH_4^+ , a necessary step for its incorporation into organic compounds [19]. The resulting NH_4^+ is utilized in amino acid synthesis through key biochemical pathways, most notably the glutamine cycle [7,19]. Amino acids like Gln and Glu derived from NH_4^+ are essential building blocks for proteins and other nitrogenous compounds crucial for plant structure and function [19]. N is also fundamental for chlorophyll synthesis [20]. Chlorophyll, the green pigment vital for capturing light energy in photosynthesis, contains N in its porphyrin ring structure: $\text{Mg}(\text{N}_4)$, where N atoms coordinate a central magnesium [Mg] ion [20]. This structure enables chlorophyll to absorb light and initiate the light-dependent reactions of photosynthesis [7,19,20]. Although N does not directly participate in the photochemical reactions of photosynthesis, it supports the process by facilitating chlorophyll production and enabling the synthesis of proteins and enzymes involved in photosynthetic metabolism [19,20].

One of the most significant effects of N fertilization is the enhancement of photosynthetic efficiency, which translates into greater biomass production and higher yields [17]. The close relationship between chlorophyll content and N content in plants is fundamental to understanding how plants capture and use light energy for growth and development. Chlorophyll, the green pigment critical for light absorption in photosynthesis, has N in its molecular structure [7,17]. This connection is especially strong because approximately 70% of a plant's leaf N is localized in chloroplasts, the structures responsible for photosynthesis and the production of chlorophyll pigments [17]. Within chloroplasts, the thylakoid membranes house the protein complexes that conduct the light-dependent reactions of photosynthesis, including photosystems [7,17]. These complexes are N-rich as they contain various proteins that require nitrogen to function. When N is plentiful, plants can produce more chlorophyll, enhancing their ability to capture light and perform photosynthesis more efficiently. This leads to increased C fixation and greater biomass production [7,17]. Furthermore, plants that receive sufficient N are capable of producing more leaves, thereby increasing their photosynthetic surface area and maximizing the conversion of solar energy into OM [17]. N also influences the processes of flowering and fruiting, which is particularly important for crop species such as cereals, vegetables, and fruits. High N availability promotes the development of seeds, grains, and fruits, thereby improving both the quality and quantity of the harvested products [17,18,21].

Figure 2 illustrates the relationship between varying N fertilizer levels and crop yields across different soil types, categorizing them into deficiency, optimal, high, and excess levels. These yield values are indicative, designed to elucidate rational N fertilization practices. Under deficient N conditions, crops display symptoms such as stunted growth and chlorosis, signaling poor health and insufficient nutrient availability. Conversely, excessive N application can lead to reduced yields despite initial growth spurts, as seen in clay soils versus sandy soils. This nuanced understanding, supported by Figure 2, underscores the importance of tailored fertilization strategies to optimize yield while minimizing environmental impact. One of the primary signs of N deficiency is stunted growth, where plants appear smaller and less vigorous compared to those receiving adequate N [22,23]. This deficiency often manifests as yellowing leaves, particularly in older foliage, a condition known as chlorosis, which impairs photosynthesis and overall plant health [23]. Ultimately, N-deficient plants produce significantly lower yields, as evidenced by the “Deficiency” section (Figure 2). Conversely, an excess of N can lead to detrimental effects on crop yield and quality. Over-fertilized plants may develop scorched leaf tips and margins due to nutrient toxicity, which is referred to as leaf burn. Additionally, excessive N can cause rapid growth, resulting in tall, weak stems that are more prone to lodging or falling over. Interestingly, while crops may initially show increased growth with higher N levels, excessive application often results in lower yields, as reflected in the “Excess” section (Figure 2).

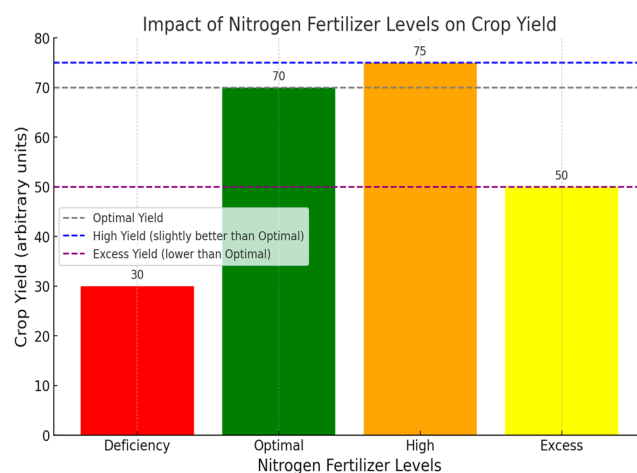


Figure 2. Relationship between nitrogen fertilizer application levels (deficient, optimal, high, excessive) and crop yields.

It is essential to note that with a balanced fertilization strategy and proper attention to the physical, chemical, and biological parameters of the soil, it is possible to maintain high yields even with reduced N applications [23,24]. This approach not only promotes sustainable agricultural practices but also enhances soil health and reduces the environmental impact associated with excessive fertilizer use [24]. Balanced N fertilization tailored to crop growth stages and specific cultivation requirements maximizes yields while minimizing the ecological footprint [23]. This is particularly important in the context of contemporary challenges related to environmental protection and sustainable agriculture [24]. However, N application must be carefully managed to avoid its excess in the soil. Excessive N fertilization can result in undesirable effects, such as excessive vegetative growth at the expense of reproductive structures, which negatively impacts fruit or grain quality [17,21]. Additionally, N surpluses can lead to nutrient losses to the environment, including leaching into groundwater, where it contributes to pollution, and emissions of nitrous oxide [N₂O], a potent greenhouse gas [19,22]. Therefore, N fertilization requires precise adjustment to the needs of the plants and soil conditions [23]. Modern agricultural techniques, such as precision farming, soil monitoring, and the use of controlled-release N fertilizers, enable the optimal utilization of this element, minimizing losses and reducing environmental impacts [21,23,24].

EU proposals suggest that a reduction in N fertilization does not necessarily lead to a significant decline in crop yields, provided that fertilization is properly balanced [23]. A key component of this strategy is the better management of available resources and tailoring fertilization to the actual needs of plants. This implies a greater emphasis on precision agriculture and advanced soil analysis methods. The optimization of N fertilization allows for the more efficient use of N, even at lower application rates, which can help prevent negative effects on crop yields [19,23]. Researchers and agronomists suggest that crops can effectively utilize lower amounts of N if fertilization is better adapted to the developmental stages of the plants and soil conditions [18]. Precise fertilizer applications in critical growth phases when the plant needs N the most can ensure that the yield potential is not compromised. In fact, such approaches may enhance soil health and reduce N losses to the environment. An important consideration is that, in many regions of Europe, excessive N fertilization is commonly practiced, with application rates often exceeding the actual needs of crops [24]. In this context, a 20% reduction in N use could benefit both the environment and farmers, lowering fertilization costs without sacrificing yield potential [22].

Wang et al. [25] evidenced that reducing N application by 20% while multiple cropping with green manure application after wheat was a suitable N fertilizer management measure for improving C mitigation in arid areas without sacrificing maize yield. Similarly, Zhang et al. [26], as a result of field experiments conducted over four consecutive seasons (2019–2023), concluded that a 20% reduction in N application increased grain yield, grain protein content, and protein yield in respect to wheat [*Triticum aestivum* L.]. They revealed that replacing 20% of the N used in conventional farming practices with organic fertilizer represents an effective strategy [26]. Also, dos Reis Martins et al. [15] examined how a 20% reduction in N fertilization according to the F2F strategy is likely to impact the yields, N₂O emissions, and N leaching of four intensively managed temperate grasslands in the Alpine region, two of them located in Switzerland and the other two in Germany. They noted that a 20% decrease in N fertilization would lead to a 5% drop in yields but also a 15% decline in N₂O emissions and a 21% decline in N leaching, mainly as NO₃⁻ [15]. Interesting research based on biogeochemical simulation was conducted by Pacofico et al. [27], which estimated the impact of reducing mineral N fertilization on the yields of maize, barley, rapeseed, and soft wheat in the EU. According to the simulation results, which examined reductions in N fertilizer application of 5%, 15%, and 25%, the largest relative yield reduction, compared to average yields from 2015 to 2018, occurred for soft wheat at all levels of fertilizer reduction. In contrast, for maize, even a 25% reduction in N application did not lead to a decline in yield. The importance of reducing and optimizing N fertilization was further highlighted by Vadillo et al. [28], who conducted a three-year field experiment on pepper cultivation at the Finca La Orden experimental farm in Spain. The applied fertilization strategy, which included doses of 0, 60, 120, and 180 kg N/ha in 2020 and 2021 and 0, 100, and 300 kg N/ha in 2022, demonstrated that N applications above 120 kg N/ha did not result in increased yield. Furthermore, higher N levels favored earlier ripening of the peppers, indicating that excessive fertilization may not only be inefficient but could also lead to earlier crop maturation without enhancing productivity. This supports the need for optimizing fertilizer use to improve both yield and environmental sustainability, which remains in agreement with the F2F target of reducing fertilizer use by 20% by 2030.

This is why balanced fertilization based on the precise monitoring of soil conditions and crop requirements can sustain high yields while reducing environmental burdens [22] and, in the long term, this could improve soil quality and yield stability.

3. Soil Features as a Consequence of Reduced Fertilization

N fertilization plays a fundamental role in shaping soil properties, particularly its structure, chemistry, and biological activity. N is an essential element within soil systems, as it drives the processes of OM mineralization, accelerates the decomposition of plant residues, and enhances microbial activity [29].

Microorganisms in the soil convert organic N into plant-available forms (Figure 3), such as NO_3^- and ammonia [NH_3], which are crucial not only for plant nutrition but also for the physical and chemical stability of the soil itself [29,30]. The conversion and enhancement of N bioavailability for plants and other organisms is facilitated by a diverse array of microorganisms through three interrelated processes: N fixation, nitrification, and denitrification [31].

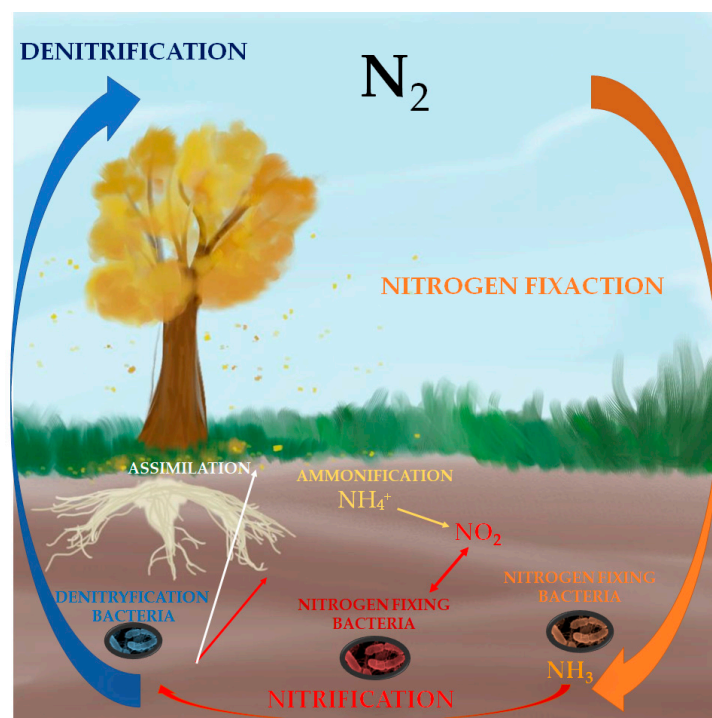


Figure 3. The N cycle in agricultural soils, highlighting processes like N fixation, nitrification, and denitrification (own work by Kagan K.).

N fixation, the first step in the N cycle (Figure 3), involves the conversion of atmospheric nitrogen (N_2) into NH_3 by bacteria such as *Azotobacter*, making N available for plant and microbial use [32]. The NO_3^- produced during this process is subsequently available in the soil as NH_4^+ , a form that can be readily absorbed by plants, thus contributing to their nutrient uptake [33]. Following N fixation, the process of nitrification occurs. This aerobic process involves the oxidation of NH_3 to nitrite [NO_2^-] by nitrifying bacteria such as *Nitrosomonas*, followed by the further oxidation of NO_2^- to NO_3^- by *Nitrobacter*. NO_3^- is the primary form of N that plants uptake, making nitrification essential for ensuring adequate N availability in agricultural and natural ecosystems [31,32]. The final stage in the N cycle is denitrification, wherein denitrifying bacteria, such as *Pseudomonas* and *Clostridium*, convert NO_3^- back into atmospheric N_2 (Figure 3). This process is vital for regulating N levels in the soil and preventing the accumulation of excess NO_3^- , which can lead to environmental issues such as water pollution [32,33]. Together, these processes ensure the continuous cycling of N in nature, thereby allowing plants to efficiently utilize this essential element for growth and development [32]. This dynamic interplay among microorganisms not only supports plant health but also plays a crucial role in maintaining ecosystem balance and soil fertility [33]. Ren et al. [34] employed high-throughput sequencing technology to analyze the taxonomic composition of soil N-transforming microbial communities (N-fixing bacteria, NH_3 -oxidizing archaea, NH_3 -oxidizing bacteria, and denitrifying bacteria) under varying N sources and levels in grassland soils. They identified the following N-fixing bacterial community: *Azohydromonas*, *Bradyrhizobium*, and *Azospirillum*, while the NH_3 -oxidizing archaea community was mainly composed of *Candidatus Nitrosocosmicus*, *Nitrososphaera*, and *Nitrosopumilus* [34]. *Nitrosospora*, *Nitro-*

somonas, and *Nitrosovibrio* were the major genera in the NH_3 -oxidizing bacterial community, whereas *Rhodopseudomonas*, *Mesorhizobium*, and *Bradyrhizobium* were the major genera in the denitrifying bacterial community [34]. Interestingly, the application of NO_3^- -source N increased the relative abundance of *Candidatus* and *Nitrosocosmicus*, while organic-source-N application led to a significant increase in the relative abundance of *Nitrosospira* [34]. NH_4^+ - and NO_3^- source-N applications caused a decrease in the relative abundance of *Rhodopseudomonas*, while the application of organic- and NO_3^- -source N was shown to reduce the relative abundance of *Bradyrhizobium* [34].

N fertilization has a profound impact on soil structure, depending on its intensity and the balance of nutrient inputs [23]. When N is deficient, microbial activity in the soil declines, slowing the decomposition of OM. As a consequence, the formation of soil aggregates diminishes, leading to a more compacted structure with reduced water-holding capacity [WHC] [23,30]. It was proved that increased N fertilization had no significant influence on OM content because mineral fertilizer application could increase OM levels by returning crop residue to the soil; meanwhile, C mineralization led to a decrease in OM content [23]. The amount of N released during OM decomposition directly influences the amount of N consumed by soil microorganisms and the mineralization rate [30]. The effect of humidity on N dynamics in the soil environment is complex and dependent on the specific environmental conditions [30]. Increased humidity can lead to increased transpiration and microbial activity, leading to a faster rate of N uptake and processing by plants and soil microbes [30]. However, prolonged higher humidity can also result in decreased plant growth and N uptake due to water stress [30]. Over time, this depletes the soil's fertility, increasing its susceptibility to degradation. Conversely, excessive N fertilization, particularly in the form of synthetic fertilizers, can lead to soil acidification [35], especially in soils that are more sensitive to acidity fluctuations, such as sandy and loose soils.

Soil acidification leads to a reduction in microbial diversity, as most soil microorganisms, including bacteria and fungi, thrive in a pH range of 6 to 7. When the pH drops below 6, certain microbial species, particularly those responsible for N processing, exhibit decreased activity or may even become extinct [36,37]. This phenomenon disrupts critical biogeochemical processes, such as the N cycle, potentially resulting in the reduced availability of essential nutrients for plants. Additionally, acidification disrupts the soil's cation exchange complex, causing the leaching of critical elements like calcium [Ca], Mg, and potassium [K] [36]. It is also important to consider that this loss of essential cations not only affects plant growth and nutrient uptake but also further exacerbates the decline in microbial diversity, as many beneficial microbes depend on these cations for their metabolic processes [37]. The diminished activity of microorganisms accelerates the degradation of the soil structure, which can lead to a loss of the soil's capacity to retain water and nutrients, thereby increasing the risk of erosion [38]. Consequently, improper land management practices and the excessive application of N fertilizers can give rise to long-term ecological challenges, adversely affecting agricultural productivity and hindering sustainable environmental development [19,23,24]. This interplay between soil acidification and microbial health underscores the importance of adopting sustainable agricultural practices that promote soil health and maintain the balance of microbial communities essential for ecosystem function [36–38].

The mechanism by which N is assimilated and its amount in the soil environment is highly dependent on soil type (Figure 4). Sandy soils, characterized by low water retention capacity and poor structural integrity, are prone to significant N losses through leaching, particularly in the form of NO_3^- , leading to the rapid depletion of essential nutrients [39,40]. In contrast, clay soils, which exhibit greater compaction and higher cation exchange capacity [CEC], are more efficient at retaining N, with slower ion exchange processes that facilitate prolonged N availability [30]. Chernozem soils, rich in OM and microbial populations, efficiently process and stabilize N, resulting in enhanced soil structure and a more balanced nutrient profile. Heavy, poorly permeable clay soils retain N for extended periods

but are susceptible to acidification under conditions of excessive fertilization [41]. Therefore, reduced and balanced N fertilization rates are essential to prevent soil degradation and maintain long-term agricultural productivity [19,24]. Moderate fertilization practices support the natural biological and chemical equilibrium of the soil, minimizing the risks of acidification and excessive nutrient leaching [35–37].

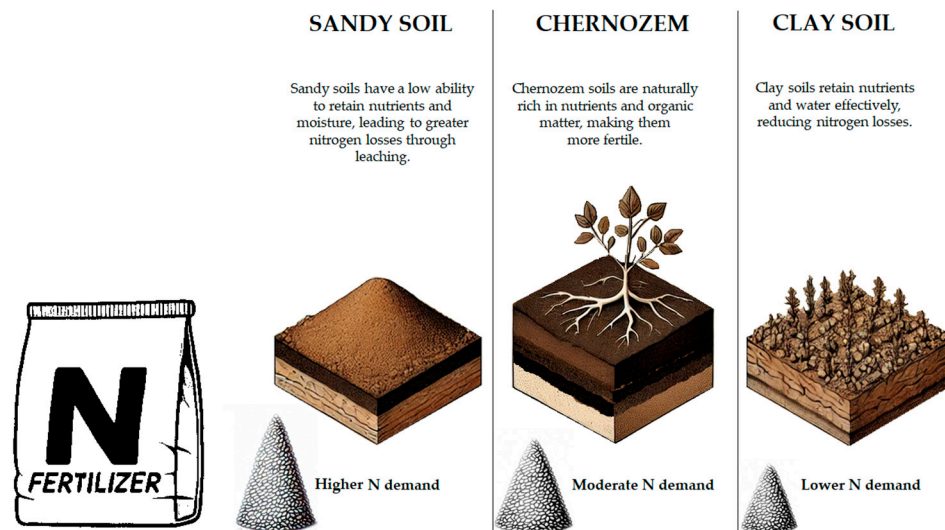


Figure 4. Nitrogen fertilization requirements and nutrient retention characteristics of different soil types: sandy, chernozem, and clay (own work by Kagan K.).

3.1. Soil Chemistry

Reduced N fertilization alters the balance of N compounds in the soil, leading to a decrease in the levels of readily available NO_3^- and NH_4^+ , which, in turn, slows down the processes of nitrification and overall nutrient cycling [33]. This can create a temporary nutrient imbalance, particularly affecting the availability of secondary nutrients like phosphorus [P] and K, as their mobilization is often driven by microbial processes that depend on sufficient N levels [42,43].

3.1.1. Soil Carbon Content

Lower N fertilization typically slows the decomposition of OM, as N plays a vital role in microbial activity, especially in the decomposition process [44,45]. However, N is often a limiting factor for microbial growth, particularly for N-dependent microbes responsible for the breakdown of organic residues. With reduced N inputs, the rate of OM mineralization decreases, allowing C compounds in OM to accumulate [45]. This can lead to an increase in soil organic carbon [SOC] over time, enhancing soil structure and fertility [46].

However, in systems with low biomass production due to N limitation, there may be less organic material returned to the soil, potentially limiting C inputs [44]. Therefore, while reduced N fertilization can enhance C sequestration by slowing decomposition, the overall C balance will depend on the interplay between organic inputs from plant residues and the rate of microbial decomposition [46–48]. In the long term, increased C retention in soil supports improved soil structure, WHC, and nutrient cycling, which are crucial for sustainable soil health.

3.1.2. Soil Aeration

The impact of reduced N fertilization on soil aeration is primarily indirect and related to changes in soil structure and microbial activity. Reduced fertilization often encourages better soil aggregation due to increased OM content [48]. OM contributes to the formation of stable soil aggregates, which improve soil porosity [49]. This enhanced porosity allows for better air circulation within the soil, leading to improved oxygen availability in the root zone [50]. Aeration enhances root growth, which can further improve oxygen availability

in the soil profile. Studies show that increased aeration supports better nutrient uptake and overall plant health by optimizing root morphology and respiration [51]. Subsurface drip irrigation, combined with reduced N levels, has been shown to improve soil oxygenation [51] and enhance root system development in crops like cucumbers, which are sensitive to hypoxia [50]. Improved aeration benefits both plant roots and aerobic microorganisms, such as those involved in the breakdown of OM and the cycling of nutrients [52].

3.1.3. Cation Exchange Capacity (CEC)

CEC, a measure of the soil's ability to retain and exchange cations such as Ca^{2+} , Mg^{2+} , K^+ , and NH_4^+ , is critical for soil fertility [23,53]. The CEC of soil is strongly influenced by the amount and quality of OM and clay minerals present. Reduced N fertilization has been shown to significantly impact soil properties, particularly through the slower rate of OM decomposition that can lead to higher OM accumulation, which directly contributes to an increase in CEC [54] and helps prevent nutrient leaching. This stabilization maintains a more consistent nutrient supply for plants by enhancing soil C content and boosting the soil's CEC. A higher CEC improves soil fertility by retaining nutrients in the root zone and making them available to plants as needed, thus contributing to more efficient nutrient cycling and supporting long-term soil health [55].

3.1.4. Nutrient Content

Reduced N fertilization influences the availability of key nutrients, including O, K, Mg, and sulfur (S) [23]. Lower N inputs can decrease the availability of N in the soil, which is a primary nutrient for plant growth [18]. Without adequate N concentration, plant biomass production may decrease, leading to a reduction in organic inputs in the soil and, consequently, lower nutrient cycling rates [30,32,56].

However, N fertilization can improve the availability of other nutrients. For example, excessive N fertilization can acidify the soil (discussed further in the Section 3.1.5), reducing the availability of nutrients like Mg and K [35]. By minimizing N inputs, soil acidification is mitigated, which helps maintain a more neutral pH where these nutrients are more bioavailable [30]. Additionally, reduced N inputs may encourage the activity of N-fixing bacteria, such as *Rhizobium*, *Bradyrhizobium*, and *Azotobacter*, which can naturally supplement N levels in the soil [32].

Regarding S, reduced N fertilization may lead to lower S availability, particularly if S-containing fertilizers are also reduced [56]. S is crucial for protein synthesis in plants, and its deficiency can limit plant growth. Since S is often co-applied with N fertilizers, reducing N inputs can inadvertently decrease S levels, which would need to be managed through alternative S supplementation [57,58].

3.1.5. Soil pH

Reduced N fertilization has a noticeable impact on soil pH, primarily due to the role of N fertilizers, especially NH_4^+ -based types like ammonium sulfate or ammonium nitrate, in contributing to soil acidification [23]. This acidification occurs during the nitrification process. As this transformation takes place, hydrogen ions are released, leading to a decrease in soil pH [59]. Prolonged and excessive application of N fertilizers can significantly acidify the soil, adversely affecting nutrient availability and overall soil health [35].

By cutting back on N inputs, the acidification process slows down, allowing the soil pH to stabilize or even move toward more neutral levels. This shift brings several benefits to the soil chemistry. For instance, essential nutrients like P, Mg, Ca, and K become more available in neutral to slightly acidic soils [60]. In contrast, these nutrients can become less soluble and harder for plants to absorb in highly acidic conditions. Furthermore, the reduction of N inputs can alleviate metal toxicity, as elements like aluminum [Al^{3+}] and manganese [Mn^{2+}] ions become more soluble in acidic soils, potentially inhibiting root growth and overall plant development [61,62]. By stabilizing the soil pH, the toxicity of these metals can decrease, leading to healthier root systems.

Additionally, soil pH influences microbial activity (Table 1). Many beneficial soil microbes, including N-fixing bacteria and decomposers, thrive in neutral or slightly acidic environments [45]. By maintaining a stable pH through reduced N fertilization, a more diverse and active microbial community can be fostered, further enhancing nutrient cycling and supporting soil health [45,63].

Table 1. Soil bacterial groups and their functions depending on soil pH based on Goulding [36], Rousk et al. [64], Bautista-Cruz et al. [65], and Jankiewicz et al. [66].

Bacterial Group	Optimal pH Range	Activity
N-fixing bacteria	6.0–8.5	nitrification (pH below 6.0 stops their activity, which results in NH ₃ accumulation in the soil)
Denitrifying bacteria	6.0–8.0	denitrification (at a lower pH, denitrification is inhibited, reducing N losses from the soil)
Cellulolytic bacteria	5.5–7.5	decomposition of cellulose (lower pH reduces their activity and slows down OM breakdown)
Acidophilic bacteria	3.5–6.5	predominant in acidic soils
Ammonifying bacteria	6.0–8.0	ammonification (lower pH reduces their activity, affecting N availability)
Proteolytic bacteria	6.0–8.0	breaking down proteins and other N compounds (at a lower pH, their abundance and activity decline)
Symbiotic N-fixing bacteria	6.0–7.5	forming symbiotic relationships with leguminous plants, fixing atmospheric N (lower pH significantly reduces their ability to fix N)

3.1.6. Strategies for N Efficiency in Reduced Fertilization

Interestingly, according to Wolińska et al. [67], a 20% reduction in N fertilization, aligned with the European Union's recommendations, did not result in a decline in the chemical parameters of soils, such as pH; WHC; total C [TC]; total sulfur [TS]; easily degradable carbon [EDC]; the content of N forms [Olsen P]; total concentrations of K, Mg, and Ca; respiration activity [RA]; and amount of humus-like substances [HS], under maize cultivation. This suggests that moderate reductions in N input may not immediately impact soil chemistry, especially for certain crops like maize, which is efficient in nutrient uptake. Nevertheless, the chemical properties of agricultural soils can still be influenced by specific cultivation techniques and the type of crops grown; that is why it is crucial to implement strategies that mitigate the potential negative effects of reduced N fertilization [23]. N-fixing cover crops, like clover and alfalfa, introduce atmospheric nitrogen into the soil through symbiosis with bacteria, reducing the need for synthetic N [68]. When incorporated as green manure, they release N slowly over time as they decompose, ensuring a steady supply of N even when synthetic inputs are reduced [68,69]. Another complementary approach involves organic amendments, such as compost or manure, which enrich the soil with organic N and increase its OM content. These amendments break down gradually, releasing N and other essential nutrients at a controlled rate, improving nutrient retention and reducing the risk of leaching [53,70]. N stabilizers and inhibitors can also be applied to enhance the efficiency of the available N [71]. In tandem with improved soil management practices, such as reduced tillage and the enhancement of soil OM, the soil's ability to retain N is further strengthened. Soils with higher OM content and better structure possess greater CEC, which helps retain essential nutrients such as Ca, Mg, and K, thus preventing nutrient depletion even with less N fertilization [53,72,73].

By incorporating these strategies, it is possible to maintain an optimal balance of N and other nutrients in the soil, mitigating the negative effects of reduced N fertilization while enhancing soil fertility in a sustainable manner. These practices support a stable chemical environment in the soil, preserving its nutrient-holding capacity and ensuring long-term productivity.

3.2. Soil Biology

The biological processes within soil are closely linked to N availability, and reduced N fertilization has a direct impact on soil biological activity. Lower N inputs slow down the activity of N-dependent microbes, particularly those involved in nitrification and denitrification [32,33]. However, while there may be a decrease in overall microbial activity, a reduced N regime encourages a shift in the composition of microbial communities. Instead of favoring fast-growing, N-hungry microbes, reduced fertilization supports the proliferation of microorganisms that are better adapted to low-N environments, such as N-fixing bacteria like *Rhizobium*, *Azotobacter*, *Bradyrhizobium*, and *Frankia*; beneficial fungi like arbuscular mycorrhizal fungi (*Glomeromycota*); and decomposers like *Penicillium* and *Trichoderma* [74–76]. These organisms play a crucial role in maintaining soil health by compensating for the lower external N inputs through natural N fixation and OM decomposition [76]. In particular, symbiotic N-fixing bacteria and fungi improve nutrient cycling, enhancing the sustainability of the soil's biological system and supporting long-term fertility [31,76].

3.2.1. Enzymatic and Respiration Activities in Soil

Enzymes involved in the N cycle are particularly sensitive to reduced N fertilization because many of these processes depend on N availability in the soil. In the literature, there are various opinions regarding the role of reduced mineral fertilization in shaping soil enzymatic activity. Reduced N fertilization has a significant impact on enzymes related to the decomposition of OM and the N cycle in the soil [23,77].

Although reduced N fertilization may lead to a decrease in enzymatic activity in the short term, the soil often adapts to new conditions [32]. In the longer term, microorganisms that are better adapted to environments with low N availability may increase their activity. Such organisms include N-fixing bacteria and mycorrhizal fungi, which can support plants in assimilating N from organic and atmospheric sources, partially compensating for the reduced enzymatic activity [74–76].

One of the main enzymes involved in the decomposition of OM is cellulase, which breaks down cellulose, the primary component of plant cell walls. Under conditions of reduced N availability, the activity of microorganisms producing cellulase may decrease [63]. Microorganisms require N to build proteins and enzymes, and a lack of N limits their capacity to efficiently process OM [17].

Similarly, enzymes such as proteases, which break down proteins into amino acids, also show decreased activity under reduced N fertilization because these enzymes are directly involved in the processing of organic N [66,78].

Enzymes such as urease, responsible for breaking down urea into NH_3 , and nitrifying enzymes are critical in the N cycle [32]. Under reduced N fertilization, urease activity decreases because microorganisms have less substrate (urea) to process [78,79]. Similarly, a reduction in NH_3 levels leads to decreased nitrifying activity, limiting the conversion of N into forms more accessible to plants [35]. At the same time, the reduction in the availability of easily assimilable forms of N promotes the development of N-fixing bacteria, such as *Rhizobium*, which compensate for deficiencies by fixing atmospheric N_2 [33].

Soil dehydrogenases are enzymes critical for the respiratory processes of microorganisms, reflecting their overall metabolic activity in the soil. Their action involves catalyzing the oxidation reactions of organic compounds, which is directly related to the C cycle and the processes of OM decomposition [80]. According to Einland [81], as well as research conducted by Siwik-Ziomek and Yatsenko [82], high doses of fertilization contribute to an

increase in the activity of this group of enzymes. Under conditions of the reduced availability of this element, enzymatic activity may decrease, which should be monitored through the implementation of long-term studies. However, according to research conducted by Kagan et al. [83], no negative impact of reduced N fertilization on dehydrogenase activity was noted in soils under rapeseed and wheat cultivation.

Soil respiration, which serves as a measure of microbial activity, also changes under reduced N fertilization [84]. Microorganisms that heavily rely on N for their metabolic processes exhibit decreased RA under conditions of the low availability of this element, leading to a decline in respiration. Soil respiration, the process of releasing CO₂ by microorganisms and plant roots, is directly linked to the decomposition of OM and the C cycle [85]. Reduced N fertilization slows down the decomposition rate of organic compounds, which can hinder the C cycle in the soil [45,46].

3.2.2. Humic Acids

Humic acids [HAs] are exceptionally valuable soil components that perform numerous critical functions, including enhancing the soil's CEC and improving water retention, thereby promoting healthy plant growth [86]. Furthermore, they act as reservoirs of nutrients, supporting the activity of soil microorganisms, which is essential for biogeochemical processes. In cases of reduced N availability due to limited fertilization, the synthesis and stability of HAs may deteriorate, potentially leading to adverse effects on agricultural efficiency [87]. According to Wolińska et al. [67] and Kagan et al. [83], there was no significant negative impact of reducing N fertilization by 20% on the content of humic substances and E4/E6 ratios, which are critical indicators of the quality of OM in soil. These findings suggest that moderate reductions in N application may not only maintain but could also positively affect soil properties, contributing to the long-term sustainability of agricultural systems; however, this aspect necessitates thorough analysis.

3.3. Soil Diversity

Soil health is intrinsically linked to the complexity of its microbiome and mycobiome, both of which are critical in driving biogeochemical processes and facilitating plant growth [88]. Diverse microbial and fungal communities are fundamental to the decomposition of OM, nutrient cycling, and the suppression of soil-borne pathogens [88–90]. Greater diversity within the microbiome and mycobiome enhances soil resilience to environmental stressors, such as climate fluctuations and contamination, thereby promoting the long-term stability and functionality of soil ecosystems [88]. N is a key nutrient for microorganisms, and its availability in the soil directly influences the structure of bacterial communities, their activity, and their interactions with plants and other soil organisms. Reduced N fertilization also has significant effects on the C cycle, which is closely tied to microbial activity. N and C are interdependent in biogeochemical processes, and their ratio in the soil influences the rate of OM decomposition and C sequestration [45,91]. In addition to changes in microbial activity, N fertilization also affects the composition of microbial communities in the soil.

3.3.1. Microbiome

A study conducted by Dai et al. [92] demonstrated that N fertilization results in an increased relative abundance of Proteobacteria and Actinobacteria while concurrently decreasing the abundance of Acidobacteria, aligning with the general life history strategy theory for bacteria. Acidobacteria, recognized as more fastidious oligotrophic bacteria with the capacity to catabolize recalcitrant C, are suppressed due to elevated N inputs, as noted by Fierer et al. [93]. Conversely, Proteobacteria and Actinobacteria, classified as copiotrophic organisms, exhibit a preference for nutrient-rich environments, demonstrating lower C-use efficiency compared to oligotrophic bacteria while maintaining a high growth rate [91,94]. The coexistence of oligotrophic and copiotrophic bacteria enhances microbial diversity within the soil, which is beneficial for overall soil health. The increased abundance and metabolic activity of these bacterial groups lead to the accelerated decomposition of

OM, with copiotrophic bacteria often being more effective at decomposing readily available organic substrates, thereby facilitating nutrient release. In contrast, oligotrophic bacteria are instrumental in decomposing more recalcitrant organic materials, playing a crucial role in the long-term nutrient cycling process [94,95]. Similar results were reported by Zeng et al. [91], who indicated a decline in the abundance of Acidobacteria and Alphaproteobacteria as well as Bacteroidetes and Chloroflexi. In this study, Actinobacteria and Betaproteobacteria exhibited an opposite trend, suggesting that N fertilization serves as a stimulant for their growth. Interestingly, bacterial taxa within the same group do not always respond uniformly, underscoring the importance of studying changes in microbial biodiversity under reduced fertilization regimes to maintain the stability of soil ecosystems and the resilience of microbial communities [88,91,92].

Ecologically important for the proper functioning of soil are Bacteroidota, which were found to depend on the agricultural practice and the season [96]. Furthermore, the genera *Mucilaginibacter* and *Edaphobaculum* were proven to be the most sensitive to soil chemistry over the changing seasons; thus, their reduced abundance may indicate inadequate soil chemistry parameters and, consequently, the fatigue of agricultural soils [96] in the context of over-fertilization or under-fertilization. On the other hand, the genus *Flavobacterium* was shown to be the most sensitive to agricultural practices, and its high abundance may indicate the good quality of agricultural soils [96]. Kruczyńska et al. [96] also proved that near-neutral or alkaline pH are the best conditions for Bacteroidota community presence and abundance. Additionally, these bacteria have been shown to be intolerant of high soil salinity and excessive soil moisture [96]. Efforts are currently in progress to study the dependence of Bacteroidota on the dose of reduced fertilization according to the F2F strategy.

3.3.2. Mycobiome

Fungi play a pivotal role in the decomposition of OM, nutrient cycling, and the establishment of symbiotic associations with plants. It is crucial to highlight that their function within the soil microbiota community is commensurate with that of soil bacteria [90,97]. Ascomycota, Basidiomycota, Mortierellomycota, and Mucoromycota represent the dominant fungal groups present in soil ecosystems [90,97,98]. The EU-recommended 20% reduction in N fertilization did not adversely affect the abundance of Ascomycota, Basidiomycota, and Mucoromycota in soil samples obtained from maize cultivation, as evidenced by the findings of Kruczyńska et al. [90]. Conversely, in this experiment, Mortierellomycota exhibited a markedly lower relative abundance compared to the results recorded under maximum N application. Notably, the highest abundance of this phylum was observed in soil subjected to a 40% reduction in N fertilization, suggesting that this microorganism may be particularly sensitive to N inputs. In this specific experiment, a decline in Mortierellomycota abundance was indeed noted as N levels increased.

Despite fungi being less sensitive to chemical fertilizers in comparison to other microbial groups [99], N fertilization exerts a significant impact on arbuscular mycorrhizal fungi [AMF], particularly affecting their community structure and diversity. High N levels were associated with a marked decline in AMF richness and biomass, primarily through the suppression of hyphal growth and spore germination [100]. AMF, belonging to the phylum Glomeromycota, are pivotal to plant–soil interactions, enhancing nutrient uptake, especially for N and P, through extensive hyphal networks that increase the absorptive surface area of plant roots [101,102]. These fungi also contribute to soil stability by promoting the aggregation of soil particles, thus improving water retention and structural integrity [101]. Long-term N deposition has been shown to significantly reduce AMF biomass, diversity, and spore abundance, with soil acidification induced by N inputs being a key driver of this decline [100,103]. Studies by Zhang et al. [100] revealed that AMF richness declined more dramatically after 11 years of N fertilization compared to 6 years, highlighting the cumulative impact of prolonged N enrichment. This decline in AMF diversity was further linked to shifts in fungal community composition, such as a reduction in the abundance

of Glomeraceae, which was accompanied by an increase in Diversisporaceae—a group known for producing more extensive hyphal networks and with better adaptation to acidic soils [100]. In the context of global change and intensified agricultural practices, understanding the response of soil fungal communities, particularly AMF, to N fertilization is critical for maintaining soil health and plant productivity. AMF plays a crucial role in mitigating abiotic stressors such as drought and soil pH fluctuations, thus serving as key contributors to ecosystem resilience under changing environmental conditions [101,102].

Wolińska et al. [104] evidenced that the agricultural soil mycobiome composition was dependent on both the seasons and the agricultural practices. They also found that even a 1-year break in the monoculture in favor of an intercropping mixture improved soil properties, thus contributing to greater biodiversity [104]. *Mortierella* was recommended as a potential indicator of sensitivity to long-term maize cultivation, whereas *Solicoccozyma* and *Exophiala* were proposed as indicators of resistance to long-term maize cultivation [104]. The impact of N fertilization on the soil mycobiome is an intriguing and vital topic, necessitating comprehensive, longitudinal research to fully understand its effects on fungal diversity and function. In light of increasing environmental stressors, it is important to prioritize maintaining soil stability.

4. The Effect of Reduced N Fertilization on Human Health and Food Quality

Excessive and often unbalanced N fertilization in agriculture poses significant risks to both human health and environmental sustainability. The lack of guidance on optimal N application often leads to overuse by farmers.

The presence of high levels of NO_3^- in crops, particularly leafy vegetables such as spinach, lettuce, and beets, poses potential health risks [105]. When consumed, these NO_3^- can convert into NO_2^- , which can form NOCs—carcinogenic compounds linked to an increased risk of cancers, especially in the stomach and esophagus [2,3,105,106]. Managing the levels of NO_3^- and NO_2^- in food is crucial, as prolonged exposure can elevate health threats, particularly when combined with processed meat consumption [106].

Reducing N fertilizer usage has been shown to enhance the quality and weight of food products. Babalar et al. [107] conducted studies describing the impact of N fertilization and postharvest treatments on fresh-cut celery, demonstrating that the highest loss of vegetable mass occurred with the highest level of N fertilization; however, as a result of the applied doses of fertilization, the level of vitamin C was improved with N fertilizers. Mozafar [108] also observed a decrease in the amount of vitamin C due to N fertilization among potatoes, tomatoes, and citrus fruits. Excessive N fertilization in agriculture often leads to a dilution effect, where the increased vegetative growth results in lower concentrations of vitamin C in vegetables [104]; it can therefore be suggested that the increased biomass of the plant does not correspond to a proportional increase in the minerals and vitamins that are essential for human health. Healthy soils that are not overloaded with chemicals promote more effective nutrient absorption by plants. As a result, it can be assumed that crops grown in such environments may not only be more nutritious but may also align with consumer preferences for food perceived as more natural and of higher quality.

The environmental consequences of excessive N fertilization are profound, particularly concerning groundwater quality and overall ecosystem health. When NO_3^- is not absorbed by plants, it can leach into groundwater and surface water, leading to significant contamination issues [3]. After the intake of NO_3^- , which is present in many plant-based foods and drinking water, it is absorbed in the gastrointestinal tract and undergoes conversion to NO_2^- and NOCs through a series of biochemical reactions, particularly in the mouth and stomach [109,110]. In the presence of anaerobic bacteria, NO_3^- is reduced to NO_2^- . This reaction is catalyzed by bacterial nitrate reductase, which converts NO_3^- to NO_2^- [110]. As a result of this reduction, NO_2^- is absorbed and transported to the stomach, where a highly acidic environment prevails. Under the influence of gastric acid, NO_2^- decomposes into, among other products, nitric oxide (NO), but it can also react with the amines present in food, leading to the formation of NOCs, which are carcinogenic. This process is particularly

intense when consuming processed meats, where NO_3^- and NO_2^- are added as preservatives, thus increasing the risk of NOC formation in the body [106,109]. The consumption of nitrate-laden water can lead to methemoglobinemia, thyroid dysfunction, gastrointestinal cancers, breast cancer, bladder, and neurodevelopmental disorders including neural tube defects [3,109]. Methemoglobinemia is a condition where NO_3^- is converted to NO_2^- in the body, leading to the formation of methemoglobin. This compound reduces the blood's ability to carry oxygen, which can be particularly dangerous for infants under six months of age, pregnant women, and individuals with certain genetic conditions or health issues [109,111]. The health risks associated with consuming contaminated water underscore the urgent need for the effective management of N fertilizer application.

On a global scale, a reduction in N fertilization would contribute to decreased emissions of nitrous oxide (N_2O), a potent greenhouse gas that significantly contributes to climate change. The positive impact of reduced doses of N fertilization on reducing N_2O emissions has been demonstrated by the research of Alami et al. [112] and Zhang et al. [113], among others. Limiting N_2O emissions would aid in slowing global warming and improving air quality. Additionally, sustainable agricultural practices that incorporate reduced fertilizer usage enhance soil quality and increase its capacity for C sequestration, further supporting global climate change mitigation efforts [23,24,45,76,77].

5. Conclusions

In summary, it is possible to conclude that a reduction in N fertilization according to the F2F strategy has more advantages than disadvantages both for farmers and for food consumers. It was proven that reduced N fertilization can slow the N cycle, thereby decreasing the availability of P and K—elements dependent on the microbial activity associated with N. On the other hand, lower N doses promote the accumulation of SOC, improving soil structure and water retention capacity. Increased OM content in the soil supports higher CEC, which prevents nutrient leaching and enables their long-term retention. Furthermore, reduced fertilization stabilizes soil pH, reducing acidification risks and enhancing the availability of Mg, Ca, and K. Lower acidity also limits the toxicity of heavy metals, such as Al and Mn, supporting the development of healthier root systems and beneficial microbial activity. Balanced and reduced N application can sustain productivity with fewer inputs, while the potential concern over falling yields under current climatic conditions could be mitigated in the mid-century by the positive effects of increased CO_2 levels. One should note that studies have shown that a moderate reduction in N inputs does not lead to a decrease in crop yield but to, conversely, improved N use efficiency. Additionally, setting safe NO_3^- limits and encouraging sustainable practices are extremely important and recommended for policymakers, while researchers are extremely encouraged to examine through long-term experiments how reduced N fertilization according to the F2F strategy of the EU will affect yields and N losses under global climate change conditions. Consequently, with appropriate management, reduced N doses can enhance long-term soil fertility and improve nutrient balance.

Reduced N fertilization has a complex impact on biological activity, biodiversity, crop yields, and human health. Lower N doses reduce the activity of N-dependent microorganisms, including enzymes involved in the N cycle, such as urease and protease, which may initially decrease the rate of OM decomposition. However, in the long term, reduced fertilization favors organisms better adapted to N-poor environments, such as N-fixing bacteria like *Rhizobium* and mycorrhizal fungi, which support the biogeochemical cycle and improve nutrient availability. The increase in bacterial and fungal biodiversity enhances soil resilience to environmental stresses and helps maintain ecosystem stability, benefiting soil quality and its C sequestration potential.

Less intensive N fertilization also influences plant microbiome composition, contributing to the better quality and nutritional value of crops, though it may limit their biomass. A key benefit of implementing reduced N fertilization is the lowered NO_3^- levels in plant products, which reduces health risks, such as the formation of carcinogenic NOCs, for

consumers. Moreover, reduced N input supports environmental protection by limiting groundwater pollution and reducing N₂O emissions, thus contributing to climate change mitigation and air quality improvement.

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References

1. Walling, E.; Vaneekhaute, C. Nitrogen Fertilizers and the Environment. In *Nitrate Handbook: Environmental, Agricultural and Health Effects*, 1st ed.; CRC Press: Boca Raton, FL, USA, 2022; pp. 103–137.
2. Martínez-Dalmau, J.; Berbel, J.; Ordóñez-Fernández, R. Nitrogen Fertilization. A Review of the Risks Associated with the Inefficiency of Its Use and Policy Responses. *Sustainability* **2021**, *13*, 5625. [CrossRef]
3. de Vries, W. Impacts of nitrogen emissions on ecosystems and human health: A mini review. *Curr. Opin. Environ. Sci. Health* **2021**, *21*, 100249. [CrossRef]
4. Barabasz, W.; Albińska, D.; Jaśkowska, M.; Lipiec, J. Biological Effects of Mineral Nitrogen Fertilization on Soil Microorganisms. *Pol. J. Environ. Stud.* **2002**, *11*, 193–198.
5. Chowdhury, A.; Das, A. Nitrate Accumulation and Vegetable Quality. *Int. J. Sci. Res.* **2015**, *4*, 1668–1672.
6. EFSA Panel on Contaminants in the Food Chain (CONTAM); Schrenk, D.; Bignami, M.; Bodin, L.; Chipman, J.K.; del Mazo, J.; Grasl-Kraupp, B.; Hoogenboom, L.; Leblanc, J.C.; Nebbia, C.S.; et al. Scientific Opinion on the risk assessment of nitrate and nitrite in feed. *EFSA J.* **2020**, *18*, 6290.
7. Bian, Z.; Wang, Y.; Zhang, X.; Li, T.; Grundy, S.; Yang, Q.; Cheng, R. A Review of Environment Effects on Nitrate Accumulation in Leafy Vegetables Grown in Controlled Environments. *Foods* **2020**, *9*, 732. [CrossRef]
8. Billen, G.; Aguilera, E.; Einarsson, R.; Garnier, J.; Gingrich, S.; Grizzetti, B.; Lassaletta, L.; Le Noë, J.; Sanz-Cobena, A. Beyond the Farm to Fork Strategy: Methodology for designing a European agro-ecological future. *Sci. Total Environ.* **2024**, *908*, 168160. [CrossRef] [PubMed]
9. Montanarella, L. Soils and the European Green Deal. *Ital. J. Agron.* **2020**, *15*, 262–266. [CrossRef]
10. European Commission. A Farm to Fork Strategy for a Fair, Healthy and Environmentally-Friendly Food System. Communication from the Commission to the European Parliament, the Council, the European Economic and Social Committee and the Committee of the Regions: COM/2020/381. Available online: https://food.ec.europa.eu/horizontal-topics/farm-fork-strategy_en (accessed on 4 October 2023).
11. European Commission. Factsheet: From Farm to Fork—Our food, Our Health, Our Planet, Our Future. Available online: https://ec.europa.eu/commission/presscorner/detail/en/fs_20_908 (accessed on 4 October 2023).
12. Massarelli, C.; Losacco, D.; Tumolo, M.; Campanale, C.; Uricchio, V.F. Protection of Water Resources from Agriculture Pollution: An Integrated Methodological Approach for the Nitrates Directive 91–676-EEC Implementation. *Int. J. Environ. Res. Public Health* **2021**, *18*, 13323. [CrossRef]
13. EUR-Lex. Available online: <https://eur-lex.europa.eu/eli/dir/1991/676/2008-12-11> (accessed on 10 November 2024).
14. Constantin, J.; Le Bas, C.; Justes, E. Large-scale assessment of optimal emergence and destruction dates for cover crops to reduce nitrate leaching in temperate conditions using the STICS soil–crop model. *Eur. J. Agron.* **2015**, *69*, 75–87. [CrossRef]
15. dos Reis Martins, M.; Ammann, C.; Boos, C.; Calanca, P.; Keel, S. Reducing N fertilization in the framework of the European Farm to Fork strategy under global change: Impacts on yields, N₂O emissions and N leaching of temperate grasslands in the Alpine region. *Agric. Syst.* **2024**, *219*, 104036. [CrossRef]
16. Schebesta, H.; Bernaz, N.; Macchi, C. The European Union Farm to Fork Strategy: Sustainability and Responsible Business in the Food Supply Chain. *SSRN Electron. J.* **2020**, *15*, 420–427. [CrossRef]
17. Fathi, A. Role of nitrogen (N) in plant growth, photosynthesis pigments, and N use efficiency: A review. *Agrisost* **2022**, *28*, 1–8.
18. Leghari, S.J.; Wahocho, N.A.; Laghari, G.M.; HafeezLaghari, A.; MustafaBhabhan, G.; HussainTalpur, K.; Bhutto, T.A.; Wahocho, S.A.; Lashari, A.A. Role of nitrogen for plant growth and development: A review. *Adv. Environ. Biol.* **2016**, *10*, 209–218.
19. Anas, M.; Liao, F.; Verma, K.K.; Sarwar, M.A.; Mahmood, A.; Chen, Z.L.; Li, Q.; Zeng, X.P.; Liu, Y.; Li, Y.R. Fate of nitrogen in agriculture and environment: Agronomic, eco-physiological and molecular approaches to improve nitrogen use efficiency. *Biol. Res.* **2020**, *53*, 47. [CrossRef]

20. Martins, T.; Barros, A.N.; Rosa, E.; Antunes, L. Enhancing Health Benefits through Chlorophylls and Chlorophyll-Rich Agro-Food: A Comprehensive Review. *Molecules* **2023**, *28*, 5344. [[CrossRef](#)]
21. Niemiec, M.; Chowaniak, M.; Zuzek, D.; Komorowska, M.; Mamurovich, G.S.; Gafurovich, K.K.; Usmanov, N.; Kamilova, D.; Rahmonova, J.; Rashidov, N. Effect of nitrogen fertilization on yield of grapes and fertilization efficiency in Gissar Valley of the Republic of Tajikistan. *J. Elem.* **2021**, *26*, 19–31.
22. Sutton, M.A.; Howard, C.M.; Kanter, D.R.; David, R.; Lassaletta, L.; Möring, A.; Raghuram, N.; Read, N. The nitrogen decade: Mobilizing global action on nitrogen to 2030 and beyond. *One Earth* **2021**, *4*, 10–14. [[CrossRef](#)]
23. Sun, J.; Li, W.; Li, C.; Chang, W.; Zhang, S.; Zeng, Y.; Zeng, C.; Peng, M. Effect of Different Rates of Nitrogen Fertilization on Crop Yield, Soil Properties and Leaf Physiological Attributes in Banana Under Subtropical Regions of China. *Front. Plant Sci.* **2020**, *11*, 613760. [[CrossRef](#)]
24. Penuelas, J.; Coello, F.; Sardans, J. A Better Use of Fertilizers is Needed for Global Food Security and Environmental Sustainability. *Agric. Food Secur.* **2023**, *12*, 5. [[CrossRef](#)]
25. Wang, P.; Yu, A.; Li, Y.; Wang, Y.; Lyu, H.; Wang, F.; Shang, Y.; Yang, X.; Chai, Q. Reducing nitrogen application by 20% under the condition of multiple cropping using green manure after wheat harvesting can mitigate carbon emission without sacrificing maize yield in arid areas. *Field Crops Res.* **2024**, *306*, 109232. [[CrossRef](#)]
26. Zhang, J.; Li, S.; Jiang, P.; Wang, R.; Guo, J.; Xiao, H.; Wu, J.; Shaaban, M.; Li, Y.; Huang, M. Organic fertilizer substituting 20% chemical N increases wheat productivity and soil fertility but reduces soil nitrate-N residue in drought-prone regions. *Front. Plant Sci.* **2024**, *15*, 1379485. [[CrossRef](#)] [[PubMed](#)]
27. Pacifico, F.; Ronchetti, G.; Dentener, F.; Velde, M.; van den Berg, M.; Lugato, E. Quantifying the impact of an abrupt reduction in mineral nitrogen fertilization on crop yield in the European Union. *Sci. Total Environ.* **2024**, *954*, 176692. [[CrossRef](#)] [[PubMed](#)]
28. Vadillo, J.; Campillo Torres, C.; González, V.; Prieto, M. Assessing Nitrogen Fertilization in Processing Pepper: Critical Nitrogen Curve, Yield Response, and Crop Development. *Horticulturae* **2024**, *10*, 1141. [[CrossRef](#)]
29. Moran, K.K.; Six, J.; Horwath, W.R.; Chris, v.K. Role of mineral-nitrogen in residue decomposition and stable soil organic matter formation. *Soil Sci. Soc. Am. J.* **2005**, *69*, 1730–1736. [[CrossRef](#)]
30. Sardar, M.F.; Younas, F.; Farooqi, Z.U.R.; Li, Y. Soil nitrogen dynamics in the natural forest ecosystem: A review. *Front. For. Glob. Chang.* **2023**, *6*, 1144930. [[CrossRef](#)]
31. Pajares, S.; Bohannan, B.J.M. Ecology of Nitrogen Fixing, Nitrifying, and Denitrifying Microorganisms in Tropical Forest Soils. *Front. Microbiol.* **2016**, *7*, 1045. [[CrossRef](#)]
32. Grzyb, A.; Wolna-Maruwka, A.; Niewiadomska, A. The Significance of Microbial Transformation of Nitrogen Compounds in the Light of Integrated Crop Management. *Agronomy* **2021**, *11*, 1415. [[CrossRef](#)]
33. Takai, K. The Nitrogen Cycle: A Large, Fast, and Mystifying Cycle. *Microbes Environ.* **2019**, *34*, 223–225. [[CrossRef](#)]
34. Ren, B.; Ma, X.; Li, D.; Bai, L.; Li, J.; Yu, J.; Meng, M.; Li, H. Nitrogen-cycling microbial communities respond differently to nitrogen addition under two contrasting grassland soil types. *Front. Microbiol.* **2024**, *15*, 1290248. [[CrossRef](#)]
35. Zamanian, K.; Taghizadeh-Mehrjardi, R.; Tao, J.; Fan, L.; Raza, S.; Guggenberger, G.; Kuzyakov, Y. Acidification of European croplands by nitrogen fertilization: Consequences for carbonate losses, and soil health. *Sci. Total Environ.* **2024**, *924*, 171631. [[CrossRef](#)] [[PubMed](#)]
36. Goulding, K.W.T. Soil acidification and the importance of liming agricultural soils with particular reference to the United Kingdom. *Soil Use Manag.* **2016**, *32*, 390–399. [[CrossRef](#)] [[PubMed](#)]
37. Wang, J.; Zhang, Z.; Liang, F.; Che, Z.; Wen, Y.; Zhang, M.; Jin, W.; Dong, Z.; Song, H. Decreased soil pH weakens the positive rhizosphere effect on denitrification capacity. *Pedosphere* **2024**, *34*, 905–915. [[CrossRef](#)]
38. Chenu, C.; Cosentino, D. Microbial regulation of soil structural dynamics. In *The Architecture and Biology of Soils: Life in Inner Space*; CABI Publishing: Wallingford, UK, 2011; pp. 37–70.
39. Zotarelli, L.; Scholberg, J.M.; Dukes, M.D.; Muñoz-Carpena, R. Monitoring of Nitrate Leaching in Sandy Soils. *J. Environ. Qual.* **2007**, *36*, 953–962. [[CrossRef](#)] [[PubMed](#)]
40. Köhler, K.; Duynisveld, W.H.M.; Böttcher, J. Nitrogen fertilization and nitrate leaching into groundwater on arable sandy soils. *J. Plant Nutr. Soil Sci.* **2006**, *169*, 185–195. [[CrossRef](#)]
41. Salm, C.; Toorn, A.; Chardon, W.J.; Koopmans, G.F. Water and nutrients transport on a heavy clay soil in a fluvial plain in the Netherlands. *J. Environ. Qual.* **2012**, *41*, 229–241. [[CrossRef](#)]
42. Poelau, C.; Herrmann, A.M.; Kätterer, T. Opposing effects of nitrogen and phosphorus on soil microbial metabolism and the implications for soil carbon storage. *Soil Biol. Biochem.* **2016**, *100*, 83–91. [[CrossRef](#)]
43. Fageria, N.K.; Baligar, V.C. Enhancing Nitrogen Use Efficiency in Crop Plants. *Adv. Agron.* **2005**, *88*, 97–185.
44. Hobbie, S.E. Nitrogen effects on decomposition: A five-year experiment in eight temperate sites. *Ecology* **2008**, *89*, 2633–2644. [[CrossRef](#)]
45. Averill, C.; Waring, B. Nitrogen limitation of decomposition and decay: How can it occur? *Glob Chang. Biol.* **2018**, *24*, 1417–1427. [[CrossRef](#)]
46. Li, C.; Aluko, O.O.; Li, J.; Liu, H. The responses of soil organic carbon and total nitrogen to chemical nitrogen fertilizers reduction based on a meta-analysis. *Sci. Rep.* **2022**, *12*, 16326.
47. Gaiser, T.; Stahr, K. Soil organic carbon, soil formation and soil fertility. In *Ecosystem Services and Carbon Sequestration in the Biosphere*; Lal, R., Lorenz, K., Hüttl, R.F., Schneider, B.U., Braun, J., Eds.; Springer: Dordrecht, The Netherlands, 2013; pp. 407–417.

48. Wang, J.; Sun, X.; Du, L.; Sun, W.; Wang, X.; Gaafar, A.R.; Zhang, P.; Cai, T.; Liu, T.; Jia, Z.; et al. Appropriate fertilization increases carbon and nitrogen sequestration and economic benefit for straw-incorporated upland farming. *Geoderma* **2024**, *441*, 116743. [[CrossRef](#)]
49. Kay, B.D.; VandenBygaart, A.J. Conservation tillage and depth stratification of porosity and soil organic matter. *Soil Tillage Res.* **2002**, *66*, 107–118. [[CrossRef](#)]
50. Jin, C.; Lei, H.; Chen, J.; Xiao, Z.; Leghari, S.J.; Yuan, T.; Pan, H. Effect of Soil Aeration and Root Morphology on Yield under Aerated Irrigation. *Agronomy* **2023**, *13*, 369. [[CrossRef](#)]
51. Heuberger, H.; Livet, J.; Schnitzler, W. Effect of soil aeration on nitrogen availability and growth of selected vegetables—preliminary results. *Acta Hort.* **2001**, *563*, 147–154. [[CrossRef](#)]
52. Qian, Z.; Zhuang, S.; Gao, J.; Tang, L.; Harindintwali, J.D.; Wang, F. Aeration increases soil bacterial diversity and nutrient transformation under mulching-induced hypoxic conditions. *Sci. Total Environ.* **2022**, *817*, 153017. [[CrossRef](#)]
53. Fawole, F.O.; Ayodele, J.; Adeoye, G.O.; Adeyemi, F.O. Soil Nitrogen Contents as Affected by Composts Enriched with Organic Nitrogen Sources. *J. Exp. Agric. Int.* **2019**, *34*, 1–11. [[CrossRef](#)]
54. Ramos, F.; Dores, E.; Weber, O.; Beber, C.; Campelo, J.; Maia, J. Soil organic matter doubles the cation exchange capacity of tropical soil under no-till farming in Brazil. *J. Sci. Food Agric.* **2018**, *98*, 8881. [[CrossRef](#)]
55. Ćirić, V.; Prekop, N.; Šeremešić, C.; Vojnov, B.; Pejic, B.; Radovanović, D.; Marinkovic, D. The implication of cation exchange capacity (CEC) assessment for soil quality management and improvement. *Agric. For.* **2023**, *69*, 113–133.
56. Dawar, K.; Khan, A.A.; Jahangir, M.M.R.; Mian, I.A.; Khan, B.; Ahmad, B.; Fahad, S.; Moustafa, M.; Al-Shehri, M.; Mubashir, M.; et al. Effect of Nitrogen in Combination with Different Levels of Sulfur on Wheat Growth and Yield. *ACS Omega* **2022**, *8*, 279–288. [[CrossRef](#)]
57. Wang, F.; Liu, H.; Yao, H.; Zhang, B.; Li, Y.; Jin, S.; Cao, H. Reducing Application of Nitrogen Fertilizer Increases Soil Bacterial Diversity and Drives Co-Occurrence Networks. *Microorganisms* **2024**, *12*, 1434. [[CrossRef](#)] [[PubMed](#)]
58. Zörb, C.; Ludewig, U.; Hawkesford, M.J. Perspective on Wheat Yield and Quality with Reduced Nitrogen Supply. *Trends Plant Sci.* **2018**, *23*, 1029–1037. [[CrossRef](#)] [[PubMed](#)]
59. Wang, Z.; Tao, T.; Wang, H.; Chen, J.; Small, G.E.; Johnson, D.; Chen, J.; Zhang, Y.; Zhu, Q.; Zhang, S.; et al. Forms of nitrogen inputs regulate the intensity of soil acidification. *Glob. Chang. Biol.* **2023**, *29*, 4044–4055. [[CrossRef](#)]
60. Ferrarezi, R.S.; Lin, X.; Gonzalez Neira, A.C.; Tabay Zambon, F.; Hu, H.; Wang, X.; Huang, J.H.; Fan, G. Substrate pH Influences the Nutrient Absorption and Rhizosphere Microbiome of Huanglongbing-Affected Grapefruit Plants. *Front. Plant Sci.* **2022**, *13*, 856937. [[CrossRef](#)]
61. Li, K.W.; Lu, H.; Nkoh, J.N.; Hong, Z.N.; Xu, R.K. Aluminum mobilization as influenced by soil organic matter during soil and mineral acidification: A constant pH study. *Geoderma* **2022**, *418*, 115853. [[CrossRef](#)]
62. Fernando, D.R.; Lynch, J.P. Manganese phytotoxicity: New light on an old problem. *Ann. Bot.* **2015**, *116*, 313–319. [[CrossRef](#)] [[PubMed](#)]
63. Zhang, L.; Zhao, Z.; Jiang, B.; Baoyin, B.; Cui, Z.; Wang, H.; Li, Q.; Cui, J. Effects of Long-Term Application of Nitrogen Fertilizer on Soil Acidification and Biological Properties in China: A Meta-Analysis. *Microorganisms* **2024**, *12*, 1683. [[CrossRef](#)]
64. Rousk, J.; Bååth, E.; Brookes, P.C.; Lauber, C.L.; Lozupone, C.; Caporaso, J.G.; Knight, R.; Fierer, N. Soil bacterial and fungal communities across a pH gradient in an arable soil. *ISME J.* **2010**, *4*, 1340–1351. [[CrossRef](#)]
65. Bautista-Cruz, A.; Aquino-Bolaños, T.; Hernández-Canseco, J.; Quiñones-Aguilar, E.E. Cellulolytic Aerobic Bacteria Isolated from Agricultural and Forest Soils: An Overview. *Biology* **2024**, *13*, 102. [[CrossRef](#)]
66. Jankiewicz, U.; Kilszczyk, A.; Russel, S. Charakterystyka właściwości proteolitycznych dwóch wybranych szczepów bakterii z rzędu Myxococcales. *Woda-Sr. Obsz. Wiej.* **2012**, *12*, 53–62. (In Polish)
67. Wolińska, A.; Banach, A.; Kruczyńska, A.; Sochaczewska, A.; Goraj, W.; Górski, A.; Podlewski, J.; Słomczewski, A.; Kuźniar, A. Effect of Reduced Nitrogen Fertilization on the Chemical and Biological Traits of Soils under Maize Crops. *Agronomy* **2023**, *13*, 2913. [[CrossRef](#)]
68. Scavo, A.; Fontanazza, S.; Restuccia, A.; Pesce, G.R.; Abbate, C.; Mauromicale, G. The role of cover crops in improving soil fertility and plant nutritional status in temperate climates. A review. *Agron. Sustain. Dev.* **2022**, *42*, 93. [[CrossRef](#)]
69. Lauren, B.C.; Mooshammer, M.; Bowles, T.; Jin, V.; Schmer, M.; Thompson, B.; Grandy, S. Complex crop rotations improve organic nitrogen cycling. *Soil Biol. Biochem.* **2022**, *177*, 108911.
70. Uddin, S.; Islam, M.R.; Jahangir, M.M.R.; Rahman, M.M.; Hassan, S.; Hassan, M.M.; Abo-Shosha, A.A.; Ahmed, A.F.; Rahman, M.M. Nitrogen Release in Soils Amended with Different Organic and Inorganic Fertilizers Under Contrasting Moisture Regimes: A Laboratory Incubation Study. *Agronomy* **2021**, *11*, 2163. [[CrossRef](#)]
71. Benckiser, G.; Schartel, T.; Weiske, A. Control of NO₃⁻ and N₂O emissions in agroecosystems: A review. *Agron. Sustain. Dev.* **2015**, *35*, 1059–1074. [[CrossRef](#)]
72. Caravaca, F.; Lax, A.; Albaladejo, J. Organic matter, nutrient contents and cation exchange capacity in fine fractions from semiarid calcareous soils. *Geoderma* **1999**, *93*, 161–176. [[CrossRef](#)]
73. Angelova, V.; Akova, V.; Artinova, N.S.; Ivanov, K. The effect of organic amendments on soil chemical characteristics. *Bulg. J. Agric. Sci.* **2013**, *19*, 958–971.
74. Yuan, X.; Zhang, J.; Chang, F.; Wang, X.; Zhang, X.; Luan, H.; Qi, G.; Guo, S. Effects of nitrogen reduction combined with bio-organic fertilizer on soil bacterial community diversity of red raspberry orchard. *PLoS ONE* **2023**, *18*, e0283718. [[CrossRef](#)]

75. Ding, Y.; Jin, Y.; He, K.; Yi, Z.; Tan, L.; Liu, L.; Tang, M.; Du, A.; Fang, Y.; Zhao, H. Low Nitrogen Fertilization Alter Rhizosphere Microorganism Community and Improve Sweetpotato Yield in a Nitrogen-Deficient Rocky Soil. *Front. Microbiol.* **2020**, *11*, 678. [[CrossRef](#)]
76. Abd-Alla, M.H.; Al-Amri, S.M.; El-Enany, A.-W.E. Enhancing Rhizobium–Legume Symbiosis and Reducing Nitrogen Fertilizer Use Are Potential Options for Mitigating Climate Change. *Agriculture* **2023**, *13*, 2092. [[CrossRef](#)]
77. Daunoras, J.; Kačergius, A.; Gudiukaitė, R. Role of Soil Microbiota Enzymes in Soil Health and Activity Changes Depending on Climate Change and the Type of Soil Ecosystem. *Biology* **2024**, *13*, 85. [[CrossRef](#)] [[PubMed](#)]
78. Mencil, J.; Mocek-Plóćiniak, A.; Kryszak, A. Soil Microbial Community and Enzymatic Activity of Grasslands under Different Use Practices: A Review. *Agronomy* **2022**, *12*, 1136. [[CrossRef](#)]
79. Korsakov, K.; Stepanov, A.; Pozdnyakov, L.; Yakimenko, O. Humate-Coated Urea as a Tool to Decrease Nitrogen Losses in Soil. *Agronomy* **2023**, *13*, 1958. [[CrossRef](#)]
80. Wolińska, A.; Stępniewska, Z. Dehydrogenase Activity in the Soil Environment. In *Dehydrogenases*; Canuto, R.A., Ed.; InTech Open: London, UK, 2012; Volume 10, pp. 183–210.
81. Einland, F. The effect of manure and NPK fertilizers on the soil microorganisms in a Danish long term field experiment. *J. Plant Soil Sci.* **1980**, *84*, 447–454.
82. Siwik-Ziomek, A.; Yatsenko, D. Total sulfur and its fractions content and enzymatic activity of luvisols. *Infrastrukt. Ekol. Teren. Wiew.* **2023**, *18*, 154–168.
83. Kagan, K.; Goraj, W.; Kuźniar, A.; Kruczyńska, A.; Sochaczewska, A.; Słomczewski, A.; Wolińska, A. Exploring the Synergy between Humic Acid Substances, Dehydrogenase Activity and Soil Fertility. *Agronomy* **2024**, *14*, 1031. [[CrossRef](#)]
84. Liu, Q.; Wang, R.; Li, R.; Hu, Y.; Guo, S. Temperature Sensitivity of Soil Respiration to Nitrogen Fertilization: Varying Effects between Growing and Non-Growing Seasons. *PLoS ONE* **2016**, *11*, e0168599. [[CrossRef](#)]
85. Darenova, E.; Holub, P.; Bednařík, A.; Klem, K. Responses of soil CO₂ efflux and microbial activity to water deficit under conventional and adaptation technology. *Soil Tillage Res.* **2023**, *234*, 105856. [[CrossRef](#)]
86. Vikram, N.; Sagar, A.; Gangwar, C.; Husain, R.; Kewat, R.N. Properties of humic acid substances and their effect in soil quality and plant health. In *Humus and Humic Substances-Recent Advances*; Makan, A., Ed.; IntechOpen: London, UK, 2022; p. 105803.
87. Martins, T.; Saab, S.; Milori, D.; Brinatti, A.; Rosa, J.; Cassaro, F.A.M.; Pires, L. Soil organic matter humification under different tillage managements evaluated by Laser Induced Fluorescence (LIF) and C/N ratio. *Soil Tillage Res.* **2011**, *111*, 231–235. [[CrossRef](#)]
88. Zhang, J.; van der Heijden, M.G.A.; Zhang, F.; Bender, S.F. Soil biodiversity and crop diversification are vital components of healthy soils and agricultural sustainability. *Front. Agric. Sci. Eng.* **2020**, *7*, 236–242. [[CrossRef](#)]
89. Singh, B. Are Nitrogen Fertilizers Deleterious to Soil Health? *Agronomy* **2018**, *8*, 48. [[CrossRef](#)]
90. Kruczyńska, A.; Kuźniar, A.; Banach, A.; Jurczyk, S.; Podlewski, J.; Słomczewski, A.; Marzec-Grządziel, A.; Sochaczewska, A.; Gałazka, A.; Wolińska, A. Changes in the mycobiome structure in response to reduced nitrogen fertilization in two cropping systems of maize. *Sci. Total Environ.* **2023**, *904*, 166343. [[CrossRef](#)] [[PubMed](#)]
91. Zeng, J.; Liu, X.; Song, L.; Lin, X.; Zhang, H.; Shen, C.; Chu, H. Nitrogen fertilization directly affects soil bacterial diversity and indirectly affects bacterial community composition. *Soil Biol. Biochem.* **2016**, *92*, 41–49. [[CrossRef](#)]
92. Dai, Z.; Su, W.; Chen, H.; Barberán, A.; Zhao, H.; Yu, M.; Yu, L.; Brookes, P.C.; Schadt, C.W.; Chang, S.X.; et al. Long-term nitrogen fertilization decreases bacterial diversity and favors the growth of *Actinobacteria* and *Proteobacteria* in agro-ecosystems across the globe. *Glob. Chang. Biol.* **2018**, *24*, 3452–3461. [[CrossRef](#)]
93. Fierer, N.; Bradford, M.A.; Jackson, R.B. Toward an ecological classification of soil bacteria. *Ecology* **2007**, *88*, 1354–1364. [[CrossRef](#)]
94. Stone, B.W.G.; Dijkstra, P.; Finley, B.K.; Fitzpatrick, R.; Foley, M.M.; Hayer, M.; Hofmockel, K.S.; Koch, B.J.; Li, J.; Liu, X.J.A.; et al. Life history strategies among soil bacteria—dichotomy for few, continuum for many. *ISME J.* **2023**, *17*, 611–619. [[CrossRef](#)]
95. Li, Y.; Wang, Z.B.; Zhang, X.Y.; Dang, Y.R.; Sun, L.L.; Zhang, W.P.; Fu, H.H.; Yang, G.P.; Wang, M.; McMinn, A.; et al. Experimental evidence for long-term coexistence of copiotrophic and oligotrophic bacteria in pelagic surface seawater. *Environ. Microbiol.* **2021**, *23*, 1162–1173. [[CrossRef](#)]
96. Kruczyńska, A.; Kuźniar, A.; Podlewski, J.; Słomczewski, A.; Grządziel, J.; Marzec-Grządziel, A.; Gałazka, A.; Wolińska, A. Bacteroidota structure in the face of varying agricultural practices as an important indicator of soil quality—A culture independent approach. *Agric. Ecosyst. Environ.* **2023**, *342*, 108252. [[CrossRef](#)]
97. Ullah, S.; Ai, C.; Ding, W.; Jiang, R.; Zhao, S.; Zhang, J.; Zhou, W.; Hou, Y.; He, P. The response of soil fungal diversity and community composition to long-term fertilization. *Appl. Soil Ecol.* **2019**, *140*, 35–41. [[CrossRef](#)]
98. Tedersoo, L.; Bahram, M.; Pöhlme, S.; Kõljalg, U.; Yorou, N.S.; Wijesundera, R.; Ruiz, L.V.; Vasco-Palacios, A.M.; Thu, P.Q.; Suija, A.; et al. Fungal biogeography: Global diversity and geography of soil fungi. *Science* **2014**, *346*, 1256688. [[CrossRef](#)]
99. Ullah, S.; Ai, C.; Huang, S.; Zhang, J.; Jia, L.; Ma, J.; Zhou, W.; He, P. The responses of extracellular enzyme activities and microbial community composition under nitrogen addition in an upland soil. *PLoS ONE* **2019**, *14*, e0223026. [[CrossRef](#)] [[PubMed](#)]
100. Zhang, C.; Xiang, X.; Yang, T.; Liu, X.; Ma, Y.; Kaoping, Z.; Liu, X.; Chu, H. Nitrogen fertilization reduces plant diversity by changing the diversity and stability of arbuscular mycorrhizal fungal community in a temperate steppe community. *Sci. Total Environ.* **2024**, *918*, 170775. [[CrossRef](#)] [[PubMed](#)]
101. Wahab, A.; Muhammad, M.; Munir, A.; Abdi, G.; Zaman, W.; Ayaz, A.; Khizar, C.; Reddy, S.P.P. Role of Arbuscular Mycorrhizal Fungi in Regulating Growth, Enhancing Productivity, and Potentially Influencing Ecosystems under Abiotic and Biotic Stresses. *Plants* **2023**, *12*, 3102. [[CrossRef](#)] [[PubMed](#)]

102. Avio, L.; Castaldini, M.; Fabiani, A.; Bedini, S.; Sbrana, C.; Turrini, A. Giovannetti, Impact of nitrogen fertilization and soil tillage on arbuscular mycorrhizal fungal communities in a Mediterranean agroecosystem. *Soil Biol. Biochem.* **2013**, *67*, 285–294. [[CrossRef](#)]
103. Pan, S.; Wang, Y.; Qiu, Y.P.; Chen, D.M.; Zhang, L.; Ye, C.L.; Guo, H.; Zhu, W.X.; Chen, A.Q.; Xu, G.H.; et al. Nitrogen-induced acidification, not N-nutrient, dominates suppressive N effects on arbuscular mycorrhizal fungi. *Glob. Chang. Biol.* **2020**, *26*, 6568–6580. [[CrossRef](#)]
104. Wolińska, A.; Podlewski, J.; Słomczewski, A.; Grządziel, J.; Gałazka, A.; Kuźniar, A. Fungal indicators of sensitivity and resistance to long-term maize monoculture: A culture-independent approach. *Front. Microbiol.* **2022**, *12*, 799378. [[CrossRef](#)] [[PubMed](#)]
105. Brkić, D.; Bošnjir, J.; Bevardi, M.; Bošković, A.G.; Miloš, S.; Lasić, D.; Krivohlavek, A.; Racz, A.; Mojsović-Čuić, A.; Trstenjak, N.U. Nitrate in leafy green vegetables and estimated intake. *Afr. J. Tradit. Complement. Altern. Med.* **2017**, *14*, 31–41. [[CrossRef](#)]
106. Keszei, A.P.; Goldbohm, R.A.; Schouten, L.J.; Jakszyn, P.; van den Brandt, P.A. Dietary N-nitroso compounds, endogenous nitrosation, and the risk of esophageal and gastric cancer subtypes in the Netherlands Cohort Study. *Am. J. Clin. Nutr.* **2013**, *97*, 135–146. [[CrossRef](#)]
107. Babalar, M.; Daneshvar, H.; Díaz-Pérez, J.C.; Nambeesan, S.; Tabrizi, L.; Delshad, M. Effects of organic and chemical nitrogen fertilization and postharvest treatments on the visual and nutritional quality of fresh-cut celery (*Apium graveolens* L.) during storage. *Food Sci. Nutr.* **2022**, *11*, 320–333. [[CrossRef](#)]
108. Mozafar, A. Nitrogen fertilizers and the amount of vitamins in plants: A review. *J. Plant Nutr.* **1993**, *16*, 2479–2506. [[CrossRef](#)]
109. Ward, M.H.; Jones, R.R.; Brender, J.D.; de Kok, T.M.; Weyer, P.J.; Nolan, B.T.; Villanueva, C.M.; van Breda, S.G. Drinking Water Nitrate and Human Health: An Updated Review. *Int. J. Environ. Res. Public Health* **2018**, *15*, 1557. [[CrossRef](#)] [[PubMed](#)]
110. Milton-Laskibar, I.; Martínez, J.A.; Portillo, M.P. Current Knowledge on Beetroot Bioactive Compounds: Role of Nitrate and Betalains in Health and Disease. *Foods* **2021**, *10*, 1314. [[CrossRef](#)] [[PubMed](#)]
111. Wagh, V.M.; Panaskar, D.B.; Mukate, S.V.; Aamalawar, M.L.; Laxman Sahu, U. Nitrate associated health risks from groundwater of Kadava River Basin Nashik, Maharashtra, India. *Hum. Ecol. Risk Assess* **2019**, *26*, 654–672. [[CrossRef](#)]
112. Alami, M.J.; Fang, X.; Zhong, D.; Zhou, W.; Gao, B.; Huang, W.; Cui, S. Impacts of High-Frequency Chicken Manure Biochar Application on N₂O and CH₄ Emissions from Vegetable Field in Subtropical China. *Agronomy* **2024**, *14*, 926. [[CrossRef](#)]
113. Zhang, F.; Ma, X.; Gao, X.; Cao, H.; Liu, F.; Wang, J.; Guo, G.; Liang, T.; Wang, Y.; Chen, X.; et al. Innovative nitrogen management strategy reduced N₂O emission while maintaining high pepper yield in subtropical condition. *Agric. Ecosyst. Environ.* **2023**, *354*, 108565. [[CrossRef](#)]

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