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Factors Regulating Symbiotic Nitrogen Fixation Efficiency in Legume Crops: Mechanisms, Environmental Constraints, and Future Perspectives

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ABSTRACT: Symbiotic nitrogen fixation (SNF) is a fundamental biological process that enhances legume productivity while reducing dependence on synthetic nitrogen fertilizers. However, its efficiency varies considerably across production systems because it is regulated by complex interactions among plant genetics, rhizobial compatibility, physiological processes, environmental conditions, and agronomic management. This review synthesizes current knowledge on the mechanisms regulating SNF and critically examines the biological, environmental, and management factors that determine its effectiveness. The review integrates evidence from the literature on the molecular basis of legume–rhizobium symbiosis, physiological regulation of nitrogen fixation, the influence of soil properties, nutrient availability, climatic stresses, and agronomic practices, and evaluates emerging technologies and future research directions. The synthesis demonstrates that SNF performance is inherently context-dependent and cannot be optimized through a single intervention. Instead, successful nitrogen fixation requires coordinated management of host genotype, rhizobial inoculants, soil fertility, water availability, and crop management while accounting for local environmental conditions. The review also identifies important knowledge gaps, including limited long-term field validation, insufficient integration of multidisciplinary approaches, and challenges in translating emerging genomic, microbial, and precision agriculture technologies into practical farming systems. Overall, this review emphasizes that integrated, site-specific management combined with continued advances in biofertilizer development and multidisciplinary research is essential for maximizing SNF efficiency, reducing reliance on synthetic nitrogen fertilizers, and supporting sustainable, climate-resilient agricultural production.

KEYWORDS: Biological nitrogen fixation, Biofertilizers, Biofertilizers, Legume–rhizobium symbiosis, Nitrogen fixation efficiency, Integrated nutrient management.

INTRODUCTION

Leguminous crops (Fabaceae) are vital to global food security, nutrition, livestock production, and sustainable agriculture. More than 700 million people rely on grain legumes as an affordable protein source, especially in developing countries. Rich in protein, complex carbohydrates, dietary fiber, vitamins, minerals, and bioactive compounds, legumes support healthy diets and contribute to the United Nations Sustainable Development Goals related to zero hunger, nutrition, and sustainable agriculture (Ayilara et al., 2022). Legumes are also important forage crops, providing hay, silage, and protein-rich feed that improve livestock productivity and reduce dependence on costly protein supplements. Their integration into crop–livestock systems enhances feed quality, soil fertility, and farming system resilience (de Bruijn & Hungria, 2022). Through symbiotic nitrogen fixation, legumes enrich soil nitrogen, improve soil organic matter, stimulate microbial diversity, enhance nutrient cycling, and reduce synthetic nitrogen fertilizer requirements. In crop rotations, they also improve soil structure, increase biodiversity, suppress pests and diseases, and enhance subsequent cereal yields; a meta-analysis of over 11,000 field observations reported an average 20% yield increase in following crops (Zhao et al., 2022). Amid climate change, population growth, and declining soil fertility, legumes have become increasingly important because they simultaneously improve productivity, maintain soil health, conserve biodiversity, and reduce greenhouse gas emissions. Consequently, their integration into cropping systems is a cornerstone of climate-smart and sustainable agriculture (Kebede, 2021; Ayilara et al., 2022).

IMPORTANCE BIOLOGICAL NITROGEN FIXATION: Nitrogen (N) is the primary nutrient limiting crop productivity and is essential for amino acids, proteins, nucleic acids, chlorophyll, and other metabolic compounds. Although synthetic nitrogen fertilizers produced through the Haber–Bosch process have increased crop yields, their excessive use has caused low nitrogen use

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efficiency, environmental pollution, greenhouse gas emissions, and high economic costs (de Bruijn & Hungria, 2022). Biological nitrogen fixation (BNF) provides a sustainable alternative by enabling legumes to convert atmospheric N_2 into plant-available ammonia through symbiosis with rhizobia in root nodules. Fixed nitrogen enhances plant growth, grain protein content, soil fertility, and residual nitrogen for subsequent crops, reducing dependence on synthetic fertilizers (Kebede, 2021).

Symbiotic BNF represents one of the largest natural nitrogen inputs to agricultural ecosystems, although its efficiency varies with plant genotype, rhizobial strain, soil conditions, environmental stresses, and management practices. A quantitative synthesis highlighted this variability and emphasized the need to integrate plant genetics, microbial interactions, soil properties, and agronomic management to maximize nitrogen fixation (Palmero et al., 2022). Beyond nitrogen supply, efficient BNF improves nutrient cycling, soil biological activity, carbon sequestration, and cropping system resilience, making its optimization a priority for climate-smart agriculture (Kebede, 2021; Palmero et al., 2022). This review synthesizes the molecular, physiological, environmental, and agronomic factors regulating symbiotic nitrogen fixation efficiency and identifies key knowledge gaps and future research priorities.

PROBLEMS ASSOCIATED WITH SYNTHETIC NITROGEN FERTILIZERS: Synthetic nitrogen fertilizers, enabled by the Haber–Bosch process, have greatly increased crop productivity and global food security but have also generated major agronomic, environmental, and economic challenges that threaten agricultural sustainability (Erisman et al., 2008). Their low nitrogen use efficiency means crops recover only 30–50% of applied nitrogen, with the remainder lost through leaching, volatilization, denitrification, runoff, and microbial immobilization, reducing fertilizer efficiency while increasing production costs and environmental degradation (Raun & Johnson, 1999; Zhang et al., 2015). These nitrogen losses contaminate groundwater, accelerate eutrophication, degrade soil quality, alter microbial communities, and disrupt nutrient cycling (Galloway et al., 2008; Sutton et al., 2011; Zhang et al., 2015). Fertilizer production and use also contribute to climate change: the energy-intensive Haber–Bosch process accounts for 1–2% of global energy consumption and substantial CO_2 emissions, while fertilizer-induced N_2O emissions are a major source of greenhouse gases and stratospheric ozone depletion (Erisman et al., 2008; IPCC, 2023; Ravishankara et al., 2009). The dependence of fertilizer production on natural gas has increased costs and exposed prices to energy market fluctuations, disproportionately affecting farmers in developing countries (FAO, 2022). Consequently, improving nitrogen use efficiency and reducing reliance on synthetic fertilizers have become priorities for economically and environmentally sustainable agriculture.

WHY NITROGEN FIXATION EFFICIENCY VARIES: Although biological nitrogen fixation (BNF) is a sustainable alternative to synthetic fertilizers, its efficiency varies widely because it is regulated by interacting molecular, physiological, environmental, and agronomic factors (Kebede, 2021). At the molecular level, successful symbiosis depends on signaling between legumes and rhizobia, where root flavonoids induce bacterial *nod* genes and Nod factors that initiate infection and nodulation. Host specificity, rhizobial compatibility, and symbiosis-related gene regulation determine nodule development and nitrogenase activity (Oldroyd et al., 2011). Physiological processes, including photosynthesis, carbon allocation, oxygen regulation, and nutrient distribution, also regulate fixation because nitrogenase requires substantial ATP and reducing power (Udvardi & Poole, 2013). Environmental factors such as soil pH, phosphorus and molybdenum availability, moisture, salinity, temperature, drought, and microbial interactions further influence rhizobial survival and symbiotic performance (Kebede, 2021). Agronomic practices, including effective inoculation, integrated nutrient management, crop rotation, irrigation, conservation agriculture, and microbial inoculants, can enhance BNF, whereas excessive nitrogen fertilization suppresses nodulation (Peoples et al., 2009). Understanding these interacting controls is essential for improving nitrogen fixation and sustainable crop production.

KNOWLEDGE GAPS: Despite decades of research, important gaps remain. Most studies have addressed individual components of BNF, such as inoculants, signaling, nutrient management, or stress responses, with limited integration of molecular, physiological, microbial, environmental, and agronomic controls. Inconsistent findings caused by differences in host genotype, rhizobial strain, soil, climate, and management, together with the scarcity of long-term field studies, particularly in semi-arid and calcareous soils, hinder the development of broadly applicable strategies. Although advances in genomics, metagenomics, synthetic biology, precision agriculture, artificial intelligence, and climate-smart technologies offer new opportunities, their integration into BNF research and translation to smallholder farming remain limited. Therefore, a comprehensive multidisciplinary synthesis is needed.

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OBJECTIVE AND SCOPE: This review synthesizes current knowledge on the biological mechanisms of legume–rhizobium symbiosis and critically evaluates the molecular, physiological, microbial, environmental, and agronomic factors regulating nitrogen fixation efficiency. It also examines constraints under field conditions, discusses emerging technologies and management strategies, and identifies key knowledge gaps and future research priorities to support sustainable, climate-resilient agriculture and strengthen the contribution of BNF to global food security and environmental sustainability.

BIOLOGICAL BASIS OF SYMBIOTIC NITROGEN FIXATION: Biological nitrogen fixation (BNF) is the reduction of atmospheric dinitrogen (N_2), which comprises about 78% of the atmosphere but is inaccessible to most organisms, into ammonia by prokaryotes possessing the nitrogenase enzyme complex (Galloway et al., 2004; Seefeldt et al., 2020). Although the Haber–Bosch process supports modern agriculture, BNF contributes 40–60% of fixed nitrogen entering terrestrial ecosystems, with legume–rhizobium symbiosis representing its most agronomically important form (Fowler et al., 2013). This highly specific partnership evolved through host–microbe coevolution, enabling legumes to selectively associate with compatible rhizobia (Masson Boivin & Sachs, 2018). Symbiosis begins when nitrogen-deficient legume roots release flavonoids that activate the rhizobial NodD regulator, inducing *nod* genes responsible for synthesizing Nod factors (Oldroyd, 2013; Lerouge et al., 1990). These signaling molecules are recognized by plant LysM receptor-like kinases, initiating root hair curling, infection thread formation, and nodule organogenesis (Gage, 2004). Rhizobia are released into host cells within symbiosomes, where they differentiate into nitrogen-fixing bacteroids.

Legume nodules develop as either indeterminate or determinate structures that facilitate carbon and nitrogen exchange between host and bacteroids (Ferguson et al., 2019). Nitrogenase activity is maintained by a microaerobic environment created by leghemoglobin, which regulates oxygen supply while protecting the oxygen-sensitive enzyme (Seefeldt et al., 2020). Nitrogenase, composed of Fe and MoFe proteins, catalyzes the ATP-dependent reduction of N_2 to ammonia. Fixed ammonia is assimilated through the glutamine synthetase–glutamate synthase (GS/GOGAT) pathway and transported mainly as amides in indeterminate nodules or ureides in determinate nodules, linking nitrogen fixation to whole-plant nitrogen demand and sustainable crop productivity (Udvardi & Poole, 2013). Symbiotic nitrogen fixation (SNF) is regulated by coordinated molecular, physiological, genetic, and microbial mechanisms that balance nitrogen fixation with plant demand and environmental conditions. Molecular regulation begins when host flavonoids induce rhizobial *nod* genes, producing Nod factors recognized by plant LysM receptor kinases to activate signaling pathways controlling infection thread formation and nodule development (Oldroyd, 2013). Within nodules, *nif* and *fix* genes regulate nitrogenase under microaerobic conditions, while autoregulation of nodulation (AON) mediated by CLE peptides and HAR1/NARK/SUNN receptors limits nodule number to optimize carbon investment (Dixon & Kahn, 2004; Ferguson et al., 2019). Physiological regulation links SNF to photosynthesis, carbon allocation, and oxygen homeostasis. Approximately 20–30% of photosynthate is supplied to nodules to support bacteroid respiration and ammonia assimilation, making nitrogenase highly sensitive to reduced carbon supply. Leghemoglobin and the nodule oxygen diffusion barrier maintain the microaerobic conditions required for nitrogenase activity (Udvardi & Poole, 2013; Ott et al., 2005). Host and rhizobial genotypes determine symbiotic compatibility and fixation efficiency through variation in symbiosis genes, receptor diversity, secretion systems, and effector proteins that regulate host specificity and plant defense responses (Ferguson et al., 2019; Perret et al., 2000; Wang et al., 2012). The soil microbiome also influences SNF: indigenous rhizobia compete for nodule occupancy, plant growth-promoting rhizobacteria enhance nodulation, and arbuscular mycorrhizal fungi generally improve phosphorus acquisition and fixation, although they may reduce fixation in phosphorus-rich soils (Burghardt & diCenzo, 2023; Vacheron et al., 2013; Larimer et al., 2010).

ENIVROMENTAL FACTORS REGULATING NITROGEN FIXATION EFFICIENCY: Environmental conditions strongly regulate SNF by affecting rhizobial survival, nodulation, and nitrogenase activity. Soil pH, texture, and organic matter determine rhizobial persistence, while phosphorus, potassium, sulfur, iron, molybdenum, and zinc are essential for nodule development and nitrogenase function (Zahran, 1999; Vance, 2001; Kaiser et al., 2005). Salinity, temperature extremes, drought, and flooding impair SNF by limiting photosynthesis, carbon supply, rhizobial growth, oxygen availability, and enzyme activity, with recovery depending on stress severity and the tolerance of both symbiotic partners (Zahran, 1999; Serraj et al., 1999; Bacanamwo & Purcell, 1999). Climate change further modifies SNF. Elevated CO_2 can increase photosynthesis, carbon allocation, and nitrogen fixation by 30–50% under favorable conditions, but these benefits are often constrained by nutrient deficiencies, drought, warming, or ozone stress, making responses highly region-specific (Rogers et al., 2009; Lam et al., 2012; Serraj et al., 1999). Improving SNF therefore requires

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integrated management, including soil pH correction, balanced nutrient supply, organic amendments, stress-tolerant legume and rhizobial genotypes, and adaptive practices such as conservation tillage and irrigation to optimize the soil–plant–microbe environment.

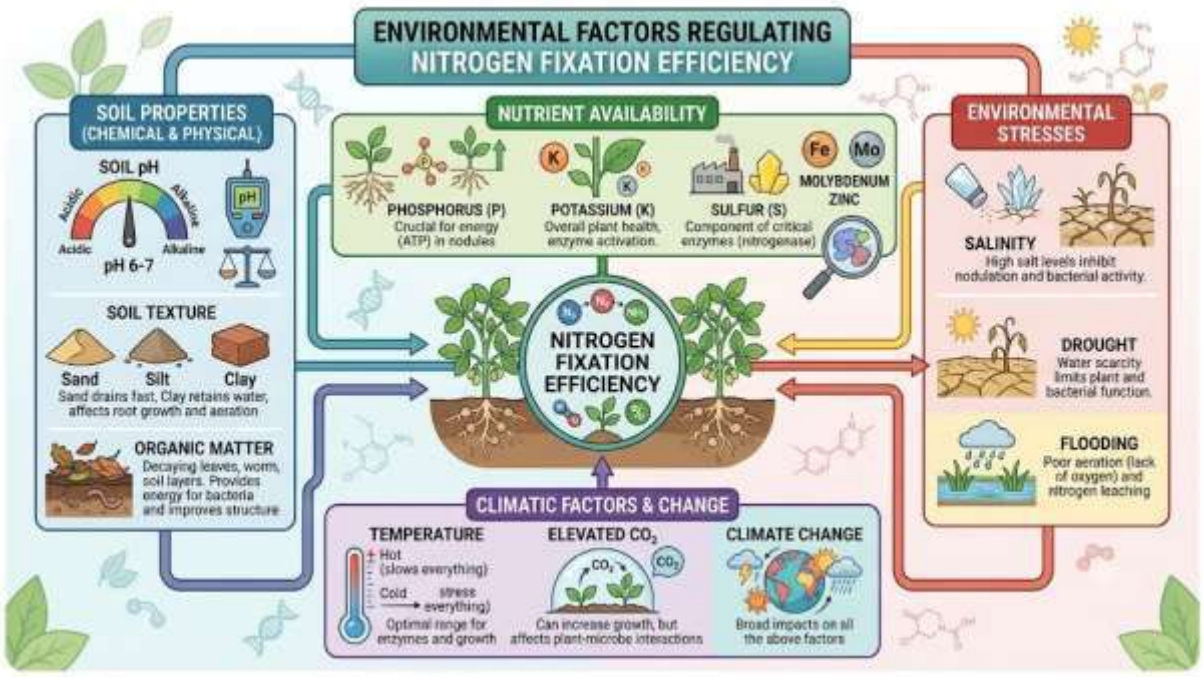

Figure 1: Environmental Factors Regulating Nitrogen Fixation Efficiency. Overview of key soil, nutrient, stress, and climatic drivers that influence biological nitrogen fixation in plants and soil microbes.

AGRONOMIC FACTORS REGULATING NITROGEN FIXAITON: Agronomic management strongly influences symbiotic nitrogen fixation (SNF), beginning with effective rhizobial inoculation. Where native rhizobia are absent or ineffective, elite stress-tolerant strains can substantially improve nodulation and yield (Deaker et al., 2004). The effectiveness of seed, liquid, or granular inoculants depends on environmental conditions, strain quality, and proper handling, while high temperatures, pesticides, and competition from indigenous rhizobia can reduce nodule occupancy. Selecting appropriate inoculation methods is therefore essential (Deaker et al., 2004). Nutrient management also regulates SNF. Organic amendments improve soil structure, water retention, microbial activity, and nutrient availability, whereas balanced mineral fertilization corrects nutrient deficiencies supporting nodule function (Giller, 2001). However, excessive nitrate or ammonium suppresses nodulation and accelerates nodule senescence (Streeter, 1988). Limited starter nitrogen (20–40 kg N ha^{-1}) may enhance early growth without inhibiting fixation if applied below suppressive levels (Salvagiotti et al., 2008). Integrated nutrient management therefore optimizes nutrient supply while avoiding excessive mineral nitrogen.

Cropping systems further influence SNF. Legume rotations improve soil fertility, exploit residual nitrogen, and reduce pests and diseases, while conservation agriculture enhances soil structure, microbial diversity, and rhizobial persistence through minimum tillage, residue retention, and crop diversification (Peoples et al., 2009; Jensen et al., 2012).

Irrigation and precision agriculture improve SNF by maintaining adequate soil moisture, preventing drought and waterlogging stress, and enabling site-specific application of inoculants and nutrients. Variable-rate technologies guided by soil and crop sensors optimize phosphorus and micronutrient placement while avoiding excess nitrogen, thereby enhancing nitrogen fixation efficiency (Serraj et al., 1999; Bacanamwo & Purcell, 1999; Gebbers & Adamchuk, 2010).

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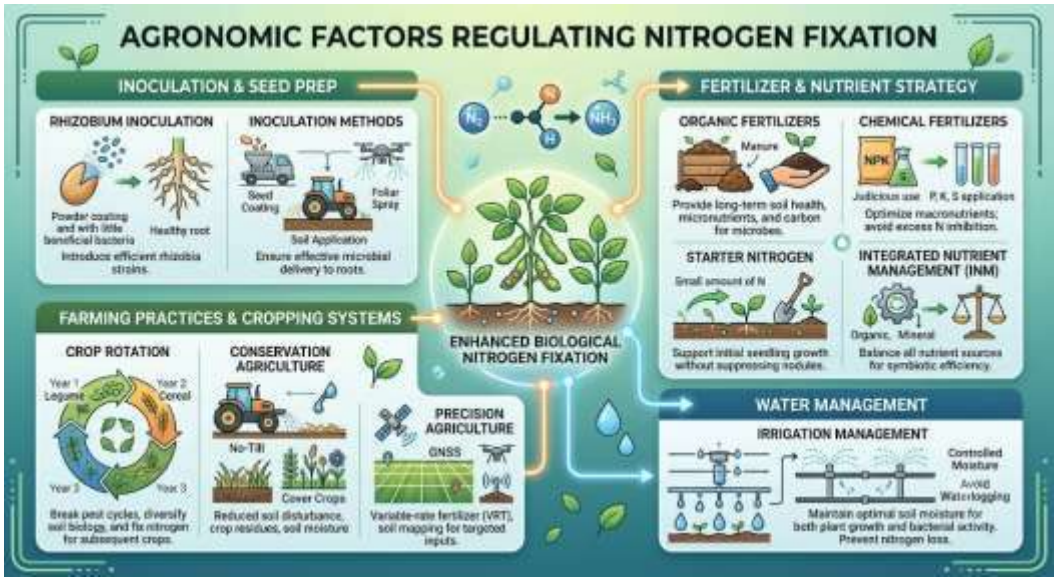

Figure 2. Overview of agronomic factors regulating biological nitrogen fixation, including seed inoculation, cropping systems, nutrient strategies, and water management.

CONSTRAINTS REDUCING NITROGEN FICATION EFFICIENCY: Symbiotic nitrogen fixation (SNF) is constrained by interacting biological, soil, climatic, agronomic, socioeconomic, and policy factors. Biological limitations include the scarcity of effective rhizobia, competition from inefficient native strains, and pests and diseases affecting nodules (Graham & Vance, 2003). Soil acidity, alkalinity, salinity, nutrient deficiencies—particularly phosphorus, molybdenum, and iron—and coarse-textured soils reduce rhizobial survival and nitrogenase activity, while drought, extreme temperatures, waterlogging, and erratic rainfall disrupt the carbon–water–oxygen balance required for fixation (Zahran, 1999; Serraj et al., 1999).

Agronomic constraints include excessive nitrogen fertilization, poor-quality or improperly applied inoculants, inadequate nutrient management, continuous monocropping, and intensive tillage, all of which reduce nodulation and soil microbial health (Giller, 2001). Adoption of SNF-enhancing practices is further limited by restricted access to quality inoculants, credit, extension services, improved cultivars, and farmer awareness, particularly in developing countries (Chianu et al., 2011). Policy barriers, including fertilizer subsidies favoring synthetic nitrogen, limited investment in research and extension, and insufficient incentives for the environmental benefits of SNF, further hinder widespread adoption (Graham & Vance, 2003; Chianu et al., 2011). Addressing these constraints requires integrated advances in microbiology, breeding, agronomy, farmer support, and policy.

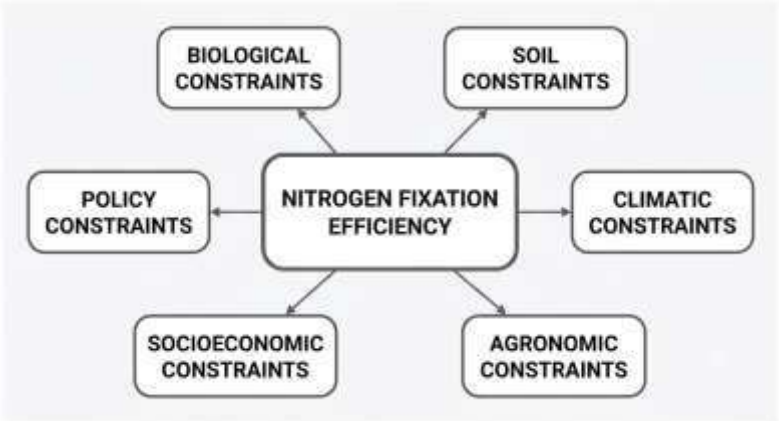

Figure 3: Conceptual framework illustrating the primary biological, soil, climatic, agronomic, socioeconomic, and policy constraints influencing nitrogen fixation efficiency.

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EMERGING TECHNOLOGIES AND FURTRER INNOVATIONS: Emerging technologies are transforming biofertilizers from single-strain inoculants to multi-strain microbial consortia that combine rhizobia with plant growth-promoting rhizobacteria and mycorrhizal fungi to improve nutrient acquisition, stress tolerance, and nodulation (Santos et al., 2019). Advanced seed coatings and nano-biofertilizers enhance microbial survival, delivery, and controlled release under field conditions (Rocha et al., 2019; Raliya et al., 2018). Genomics, metagenomics, synthetic biology, and CRISPR-based genome editing are accelerating the identification and engineering of elite rhizobial strains with improved competitiveness, stress tolerance, host specificity, and nitrogen fixation efficiency (Geddes et al., 2015; Haskett et al., 2021). Artificial intelligence and precision agriculture further optimize inoculant selection and site-specific application using genomic, soil, and climatic data, while climate-smart biofertilizers employ stress-tolerant strains capable of maintaining nitrogenase activity under drought, salinity, and heat stress (Haskett et al., 2021; Wolfert et al., 2017; Gebbers & Adamchuk, 2010; Vurukonda et al., 2016). Together, these innovations provide data-driven strategies for enhancing SNF and sustainable agricultural intensification.

SYNTHESIS AND DISCUSSION

The literature reviewed across the biological, regulatory, environmental, agronomic, and technological dimensions of symbiotic nitrogen fixation reveals a robust consensus on several core principles. There is broad agreement that the molecular dialogue between flavonoids and rhizobial NodD proteins, leading to Nod factor perception by plant LysM-domain receptors, is the conserved mechanism initiating symbiosis (Oldroyd, 2013). Similarly, the indispensability of the nitrogenase enzyme, its oxygen sensitivity, and the protective role of leghemoglobin within a precisely controlled microaerobic nodule environment are universally acknowledged (Ott et al., 2005; Seefeldt et al., 2020). Studies consistently demonstrate that the carbon cost of fixation—often 20–30 % of net photosynthate—ties nitrogenase activity tightly to photosynthetic rate, and that autoregulation of nodulation (AON) via CLAVATA3/ESR-related peptides prevents excessive nodule formation (Ferguson et al., 2019; Udvardi & Poole, 2013). It is also undisputed that phosphorus is the most frequently limiting nutrient after nitrogen, because of the high ATP requirement of nitrogenase, and that excess mineral nitrogen, whether from fertilizer or soil mineralization, systemically suppresses nodulation and accelerates nodule senescence (Vance, 2001; Streeter, 1988). In the agronomic realm, the value of crop rotation with legumes, the potential of conservation agriculture to stabilize the soil habitat for rhizobia, and the necessity of inoculation when compatible indigenous strains are absent are all strongly supported (Peoples et al., 2009; Deaker et al., 2004).

Despite these areas of consensus, contradictory findings emerge, particularly when controlled-environment results are extrapolated to the field. The response of SNF to elevated atmospheric CO_2 exemplifies this tension: free-air CO_2 enrichment studies frequently report a 30–50 % stimulation of fixation, driven by increased carbon supply (Lam et al., 2012), yet this benefit often disappears when legumes are simultaneously subjected to drought, low phosphorus, or high ozone, leading to minimal or even negative net gains (Rogers et al., 2009). Similarly, the practice of applying a small dose of starter nitrogen remains contentious; while some experiments show that 20–40 kg N ha^{-1} can boost early growth without permanently inhibiting nodules, others report that even low nitrate concentrations down-regulate nitrogenase gene expression, causing an irreversible yield penalty (Salvagiotti et al., 2008). The role of arbuscular mycorrhizal fungi likewise shows context-dependency: co-inoculation with AMF often enhances nodulation and phosphorus uptake under low-P conditions, but in phosphorus-rich soils the fungi may compete for carbon, temporarily reducing nitrogen fixation (Larimer et al., 2010). Even the superiority of multi-strain consortia over single-strain inoculants is not universal, as strain compatibility and niche overlap can lead to antagonism rather than synergism (Santos et al., 2019).

These discrepancies arise from the pronounced influence of soil properties, climatic regimes, host-symbiont genotypes, and experimental methodologies. Soil pH and texture modulate the severity of nutrient deficiencies and the survival of introduced rhizobia, so results from a fertile loam cannot be directly applied to an acid sandy soil (Zahran, 1999). Climatic factors, particularly the timing and intensity of drought or flooding, interact with plant developmental stage; a moderate mid-season water deficit may trigger reversible feedback inhibition via ureide accumulation in soybean, whereas a severe terminal drought causes irreversible nodule damage and carbon starvation (Serraj et al., 1999). Legume species differ fundamentally in nodule morphology—indeterminate nodules of alfalfa possess a persistent meristem and export amides, while determinate nodules of common bean export ureides—leading to divergent physiological responses to the same stress. Rhizobial strain identity further complicates comparisons because effectiveness in nitrogen fixation, competitiveness for nodule occupancy, and tolerance to environmental extremes vary markedly even within a single species (Burghardt & diCenzo, 2023). Finally, experimental methods such as pot versus field trials,

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the use of sterilised versus native soil, and the duration of observation introduce systematic biases: short-term pot studies frequently overestimate inoculation benefits because they exclude the competitive indigenous microbiota present in agricultural fields (Deaker et al., 2004).

Current limitations in the science and practice of SNF management therefore reflect this complexity. Mechanistic understanding of how microbial consortia interact in the rhizosphere and how plants integrate multiple simultaneous stresses—drought, heat, and nutrient deficiency—remains incomplete (Haskett et al., 2021). Predictive tools that translate genomic and environmental data into reliable field-scale recommendations for biofertilizer formulation are still in their infancy, and the regulatory pathways for genetically edited rhizobia or synthetic consortia are underdeveloped in many countries. On the socioeconomic side, the gap between research-validated technologies and smallholder adoption persists because of inadequate extension services, insecure land tenure, and policy environments that subsidise synthetic nitrogen rather than ecological services (Chianu et al., 2011; Graham & Vance, 2003).

The practical implications of this synthesis are clear: no single management practice can optimise SNF across all contexts. Site-specific integrated nutrient management that combines liming, balanced mineral nutrition, and organic amendments is essential to create a soil environment that is hospitable to rhizobia and mycorrhizal partners (Giller, 2001). Inoculant selection must be matched not only to the host legume genotype but also to the anticipated abiotic stresses, using locally adapted or climate-smart strains and, where appropriate, carefully designed consortia (Vurukonda et al., 2016). Advanced seed coating and nano-formulation technologies can improve the survival and precision delivery of these microbes (Rocha et al., 2019). Equally, agronomic decisions—from crop rotation and conservation tillage to irrigation scheduling and variable-rate technology in precision agriculture—must be orchestrated to stabilise the soil-plant-microbe system. Finally, realising the full potential of biological nitrogen fixation requires policy incentives that reward farmers for reducing reliance on synthetic nitrogen, funding for long-term, multi-location field trials that capture the genotype-by-environment-by-management interactions, and continued investment in the genomics and synthetic biology tools that will underpin the next generation of biofertilizers.

KNOWLEDGE GAPS AND RESEARCH PRIORITIES

Significant knowledge gaps remain in understanding how microbial consortia—including rhizobia, mycorrhizal fungi, and plant growth-promoting rhizobacteria—interact under multiple stresses and how host plants regulate ineffective symbionts. Long-term, multi-location field trials examining genotype × environment × management interactions are limited, and standardized protocols for evaluating biofertilizer efficacy are lacking. Most mechanistic studies are based on temperate systems, leaving tropical smallholder agroecosystems with phosphorus deficiency and erratic rainfall underrepresented (Chianu et al., 2011). Climate-related uncertainties include the combined effects of heat and drought on nitrogenase activity and nodule senescence, and whether elevated CO_2 can mitigate these impacts. Translating laboratory innovations, such as nano-biofertilizers and CRISPR-engineered strains, into field applications is constrained by weak regulatory frameworks and limited extension services (Haskett et al., 2021). Future research should prioritize: (1) multi-strain consortia performance under field conditions; (2) genetic markers predicting symbiotic efficiency across soils and climates; (3) precision inoculation using real-time soil and climate data; and (4) policy frameworks promoting biological nitrogen fixation over synthetic fertilizer dependence.

CONCLUSION

Symbiotic nitrogen fixation efficiency is determined by the coordinated interaction of molecular signaling, host–microbe compatibility, plant physiology, environmental conditions, and agronomic management rather than by any single factor. This synthesis highlights that the effectiveness of biological nitrogen fixation is inherently context-dependent, explaining why similar practices can produce contrasting outcomes across different soils, climates, and cropping systems. Consequently, improving nitrogen fixation requires integrated, site-specific strategies that combine appropriate rhizobial inoculation, balanced nutrient management, sound soil and water management, and adaptation to local environmental conditions. Advancing these integrated approaches, together with continued innovation in biofertilizer technologies and multidisciplinary research, will be essential to enhance legume productivity, reduce reliance on synthetic nitrogen fertilizers, and strengthen the contribution of biological nitrogen fixation to sustainable and climate-resilient agriculture.

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