

Metagenomic Profiling of Rhizosphere Microbiome Shifts under Integrated Biofertilizer–Organic Amendment Systems in Sustainable Agroecosystems

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ABSTRACT

Background: The rhizosphere microbiome has an essential function in some processes such as nutrient cycling, plant protection in terms of diseases and stress tolerance, and it is increasingly acknowledged as an important component of sustainable agricultural ecosystems. Integrated application of microbial bio-fertilizers with organic fertilizers (composts, vermicompost's, manure and biochar), though still not fully studied, is considered as one of the main alternatives to chemical fertilizers. The goal of this paper is to summarize metagenomic studies of changes in the rhizosphere microbiome occurring in response to the use of bio-fertilizer-organic fertilizer integrated systems by comparing the taxonomic, functional and diversity parameters of the agricultural systems used.

Literature sources: To carry out the review, a structured search was performed in Scopus, Web of Science, PubMed and Google Scholar (2015-2025) with the goal of recognizing peer-reviewed studies and reports related to shotgun and amplicon (16S rRNA/ITS) metagenomics. A total of 67 studies were downloaded based on the methodology of literature review.

Study results: The results demonstrate that integrated systems have similar effects on increasing the alpha diversity, the abundance of *Proteobacteria*, *Actinobacteria*, *Firmicutes* and *Bacteroidetes*, as well as the proliferation of functional genes responsible for nitrogen, phosphorus and carbon cycle of the crops. Bio-organic amendments are known to cause disease suppressive community restructuring although the taxa level responses are different depending on the combinations of soil type, amendment sources, and crop species as well as the experimental duration.

Research gaps: There are still several major research gaps in this field, including short study durations, lack of evidence of fungi, archaea and viruses, inconsistent metadata reporting, non-use of multi-omics approach, and little evidence of relations between the changes in microbiome and agricultural yield/production metrics.

Conclusion: Generally, the systems based on integrated biofertilizers and organic amendments lead to the formation of rhizosphere microbiomes which have a higher level of diversity and functional richness as compared to chemical fertilizers-based systems; however, further long-term experiments in laboratory conditions, including different locations, are still needed.

Keywords: Rhizosphere microbiome; metagenomics; biofertilizer; organic amendment; integrated nutrient management; soil microbial diversity; sustainable agriculture; functional gene profiling

1. Introduction

The rhizosphere, the narrow zone of soil directly influenced by root exudation and root-associated microbial activity, is among the most metabolically active interfaces in terrestrial ecosystems. It hosts dense and taxonomically diverse bacterial, archaeal and fungal assemblages that mediate nutrient acquisition, phytohormone signalling, pathogen suppression and stress buffering for the host plant ^[1, 2]. Because agricultural productivity depends heavily on these microbially mediated processes, the rhizosphere microbiome has emerged as a central target for strategies aimed at reducing dependence on synthetic agrochemicals while sustaining yield ^[3].

Two complementary management approaches have gained particular prominence: microbial biofertilizers, which introduce or stimulate plant growth-promoting rhizobacteria (PGPR), nitrogen-fixing bacteria, phosphate- and potassium-solubilizing microorganisms and arbuscular mycorrhizal fungi; and organic amendments, including farmyard manure, compost, vermicompost, green manure, crop residues and biochar, which supply carbon substrates and modify the physicochemical soil environment ^[4, 5]. When applied together as part of integrated nutrient management (INM), these inputs are hypothesized to act synergistically: organic amendments provide the carbon and nutrient substrate that sustains introduced or native beneficial microbial populations, while biofertilizer inoculants accelerate specific biochemical transformations such as nitrogen fixation, phosphate solubilization and siderophore-mediated iron acquisition ^[6, 7].

The global scenario underscores the urgency of this research direction. Overreliance on nitrogen, phosphorus and potassium fertilizers has been linked to soil acidification, declining microbial diversity, greenhouse gas emissions and diminishing marginal returns on yield, prompting national and international bodies to promote integrated and organic-based nutrient strategies as part of climate-smart and regenerative agriculture agendas [8, 9]. At the same time, the maturation of culture-independent sequencing technologies, particularly 16S rRNA amplicon sequencing and shotgun metagenomics, has transformed the ability of researchers to resolve rhizosphere community composition and functional potential at unprecedented taxonomic and genomic resolution [10, 11]. Metagenomic approaches now permit not only description of who is present in the rhizosphere but also functional inference of what those organisms are metabolically capable of doing, including nitrogen-, phosphorus- and carbon-cycling gene abundance, biosynthetic gene clusters, and antimicrobial resistance determinants [12].

Despite an expanding body of primary research, the literature remains fragmented across crop systems, amendment types, geographic regions and sequencing platforms, and syntheses that specifically integrate biofertilizer and organic amendment effects on rhizosphere metagenomic outcomes are limited. Existing reviews tend to address either biofertilizers or organic amendments in isolation, or focus narrowly on a single crop or disease outcome, leaving open questions about the consistency, magnitude and mechanistic basis of microbiome shifts when these two input categories are deliberately combined [13, 14].

This review therefore aims to: (i) synthesize metagenomic evidence on taxonomic and functional rhizosphere microbiome shifts under integrated biofertilizer-organic amendment systems; (ii) compare findings across cropping systems, amendment sources and sequencing approaches; (iii) critically evaluate methodological strengths, weaknesses and points of disagreement across the primary literature; and (iv) identify research gaps and emerging technological trends that can guide future investigation. The scope of the review encompasses peer-reviewed metagenomic and next-generation sequencing studies published between 2015 and 2025 that examine bacterial, fungal or archaeal rhizosphere communities under biofertilizer application, organic amendment, or their combination within field, pot or

greenhouse agricultural systems; studies restricted to bulk soil without rhizosphere sampling, non-agricultural ecosystems, or purely culture-based enumeration were considered only where necessary for historical or mechanistic context.

2. Methodology of Literature Review

2.1. Literature Search Strategy

A structured search was conducted across Scopus, Web of Science Core Collection, PubMed/MEDLINE and Google Scholar for records published between January 2015 and mid-2025, supplemented by manual screening of reference lists of retrieved reviews and reports from the Food and Agriculture Organization (FAO) and national soil health mission portals. Boolean search strings combined controlled and free-text terms drawn from three concept clusters: (i) rhizosphere/soil microbiome and metagenomics; (ii) biofertilizer, plant growth-promoting rhizobacteria, and microbial inoculant; and (iii) organic amendment, compost, vermicompost, farmyard manure, biochar and integrated nutrient management. Representative search strings included: ("rhizosphere microbiome" OR "rhizosphere metagenom*") AND ("biofertiliz*" OR "PGPR" OR "microbial inoculant") AND ("organic amendment" OR "compost" OR "vermicompost" OR "biochar" OR "integrated nutrient management").

2.2. Inclusion and Exclusion Criteria

Details of the inclusion and exclusion criteria applied during screening are summarized in Table 2.

2.3. Study Selection Process

Study selection followed a PRISMA-style workflow (Figure 1). An initial pool of 1,842 records was retrieved; after removal of duplicates ($n = 546$), 1,296 unique records were screened by title and abstract. Of these, 968 were excluded for being non-English, non-peer-reviewed, or unrelated to agricultural rhizosphere systems, leaving 328 records for full-text assessment. A further 261 were excluded at full-text stage, principally for lacking metagenomic or high-throughput sequencing data, being conference abstracts or editorials, or providing insufficient methodological detail to allow comparative extraction. The final synthesis included 67 studies, spanning shotgun metagenomic, 16S rRNA amplicon, ITS amplicon and functional gene-targeted (qPCR/metagenomic) investigations.

Table 2: Inclusion and Exclusion Criteria

Criterion type	Applied rule
Inclusion	Peer-reviewed articles published 2015-2025; English language; rhizosphere (not bulk soil only) sampling; use of 16S rRNA, ITS, shotgun metagenomic, or functional-gene metagenomic methods; biofertilizer, organic amendment, or integrated application as an experimental factor
Inclusion	Field, pot, or greenhouse agricultural systems (staple, vegetable, horticultural or medicinal crops)
Exclusion	Duplicate records across databases
Exclusion	Non-academic/grey web sources without peer review (except designated international/government reports used for contextual statistics)
Exclusion	Studies restricted to bulk soil, non-agricultural ecosystems, or purely culture-dependent enumeration without sequencing
Exclusion	Conference abstracts, editorials, and studies with insufficient methodological detail for comparative extraction

3. Literature Review and Thematic Analysis

3.1. Biofertilizer-Driven Taxonomic Shifts in the Rhizosphere

Multiple shotgun and amplicon metagenomic studies converge on the finding that biofertilizer application increases rhizosphere bacterial alpha diversity relative to chemical fertilization alone. In leek (*Allium ampeloprasum*), shotgun

metagenomic profiling showed that biofertilizer-treated plots harboured the most diverse microbial community among biofertilizer, chemical fertilizer and bulk soil treatments, with Bacteroidota and Verrucomicrobiota detected exclusively in the biofertilizer plot and genera such as *Dyadobacter*, *Streptomyces* and *Haliangium* uniquely enriched there [15, 16]. This pattern is broadly consistent with maize-cabbage rotation

studies, where bio-organic fertilizer regimes reshaped rhizosphere community composition and elevated the relative contribution of *Trichoderma* to yield enhancement, with crop type and fertilizer treatment identified as the dominant drivers of community variation ^[17].

Long-term field experiments applying bio-organic fertilizers have further shown that disease-suppressive outcomes are linked less to the raw abundance of an introduced biocontrol strain than to broader shifts in community composition and function; metagenomic analysis of tomato rhizosphere soils under long-term bio-organic fertilization revealed enrichment of *Sphingomonadaceae* and *Xanthomonadaceae* specifically upon pathogen invasion, associated with secondary metabolite production that suppressed *Ralstonia solanacearum* ^[18]. Comparable general disease-suppressive signatures have been reported in mustard cultivated with a bio-amendment regime, where functional annotation of metagenomic open reading frames revealed enrichment of transport- and cellular-process-related genes relative to conventionally farmed soil ^[19]. Collectively, these findings support the interpretation that biofertilizers act less as simple population additions and more as catalysts of community-level reorganization.

However, the literature also documents inconsistency in the specific taxa that respond to biofertilizer treatment, which appears strongly conditioned by crop identity, native soil microbiome composition and the biofertilizer formulation used. Consortium-based PGPR applications combining nitrogen-fixing, phosphate-solubilizing and biocontrol strains have shown crop-specific efficacy gains over single-strain inoculation in tea, chilli, mungbean and fenugreek systems, suggesting synergistic interactions among co-inoculated taxa that are not always mirrored by native community shifts ^[20]. This heterogeneity complicates efforts to generalize "core" biofertilizer-responsive taxa across cropping systems.

3.2. Organic Amendments and Microbial Diversity Indices

Organic amendments, independent of microbial inoculation, are consistently associated with elevated bacterial and fungal diversity relative to unamended or chemically fertilized soils. Comparative 16S rRNA studies of composts and vermicomposts demonstrate distinct, amendment-specific bacterial community signatures, with a substantial fraction of detected taxa corresponding to yet-uncultivated lineages ^[21, 22]. Vermicompost application, in particular, has been linked to increased abundance of antagonistic microbes and nematodes capable of suppressing soil-borne phytopathogens, alongside enrichment of plant growth-promoting metabolites such as auxins, gibberellins and cytokinins ^[23]. Compost amendment of saline-alkali soils elevated bacterial diversity dominated by *Proteobacteria*, *Firmicutes*, *Actinobacteria* and *Chloroflexi*, with redundancy analysis identifying pH, total nitrogen, organic matter and enzyme activity as the principal correlates of community structure ^[24].

Combined biochar-vermicompost amendment of greenhouse soils increased both bacterial and fungal alpha diversity indices and elevated the abundance of *Actinomycetes*, *Acidobacteria* and beneficial fungal taxa, with organic matter content and dissolved organic carbon identified as key mediating variables ^[25]. In leafy vegetable production, 16S rRNA metagenomic profiling of produce grown with different compost types (leaf waste, municipal waste, cow dung manure and vermicompost) found consistently higher operational taxonomic unit richness under composted regimes than under

conventional fertilizer, although the ratio of beneficial to pathogenic genera varied by compost source, with cow dung manure compost showing a comparatively lower beneficial-to-pathogenic ratio than plant-derived composts ^[26]. This finding is a notable point of caution: not all organic amendments confer equivalent microbiological benefit, and manure-derived amendments in particular warrant monitoring for pathogen carry-over.

Long-duration organic amendment can also produce less favourable outcomes under some conditions. A multi-year study of heavy organic amendment in bamboo plantations found significant declines in the relative abundance of several taxa considered beneficial (including *Bradyrhizobium* and *Streptomyces*) alongside increases in acid-tolerant taxa, attributed to amendment-induced soil acidification over time ^[27]. This result illustrates that organic amendment effects are not uniformly positive and can shift toward detrimental community reconfiguration when applied at high rates over extended periods without adequate monitoring of soil pH and nutrient balance.

3.3. Functional Gene Profiling: Nitrogen, Phosphorus and Carbon Cycling

A distinguishing contribution of shotgun metagenomics, relative to amplicon sequencing, is the ability to quantify functional gene abundance directly. Multiple studies have used this capacity to characterize how integrated nutrient inputs alter nitrogen-cycling gene pools. In ginseng rhizosphere soils, moderate nitrogen addition increased yield and soil nutrient content, whereas excessive nitrogen addition reduced microbial diversity, decreased beneficial bacterial abundance, and significantly increased the abundance of denitrification and nitrification genes such as *nxrA* and *napA*, illustrating a dose-dependent, non-linear relationship between nutrient input and functional gene response ^[28]. Biochar amendment of soybean rhizosphere soils in Mollisol and alkaline soils reduced *narG* and *nosZ* gene abundance, indicating a potential mechanism for biochar-mediated mitigation of nitrous oxide emissions through modulation of denitrification gene pools rather than through nitrogen input reduction alone ^[29, 30].

Functional metagenomic profiling of lentil rhizospheres under different cultivars in a rice-fallow ecology revealed divergent nitrogen- and phosphorus-cycling gene profiles between cultivars sharing the same soil environment, with one cultivar rhizosphere significantly enriched in denitrification-associated genes relative to the other, underscoring that host genotype, not soil or amendment alone, materially shapes functional gene composition ^[31]. Similarly, functional annotation of the *Allium ampeloprasum* rhizosphere metagenome identified multifunctional genes involved in nutrient cycling and plant growth promotion that were differentially abundant between biofertilizer and chemical fertilizer plots, including RNA helicase genes (*hrpA*, *hrpB*) implicated in post-transcriptional regulation ^[32]. Integrated nutrient management field trials combining farmyard manure, wheat straw, green manure and NPK fertilizer similarly demonstrated 16S rRNA-based enrichment of *Proteobacteria*, *Actinobacteria* and *Firmicutes* associated with nutrient cycling and organic matter decomposition functions, reinforcing the pattern that combined organic-inorganic-biological inputs support a more functionally diverse microbial consortium than any single input category ^[33, 34].

3.4. Integrated Nutrient Management and Soil Health Indices

Systematic and narrative reviews of INM consistently report positive effects on yield, soil organic carbon, bulk density and microbial activity relative to sole chemical fertilizer application, while reducing the environmental burden associated with fertilizer manufacture, transport and application [35]. A long-term rice-lentil-jute cropping system study using 16S rRNA metagenomics of the V3-V4 hypervariable region found that soil test crop response-based INM increased the Shannon diversity index by 12.6% relative to control and enhanced a composite Biological Soil Health Index linked to *Azotobacter* and *Actinobacteriota* abundance, which in turn correlated significantly with rice-equivalent yield [36]. A systematic review applying PRISMA methodology to INM and soil microbes in rice systems similarly concluded that microbial abundance, diversity and activity were enhanced more consistently under INM than under sole synthetic fertilization, positioning INM as a viable strategy for reducing excessive chemical fertilizer reliance while supporting microbial consortia function [37].

Long-term integrated soil-crop management field trials have additionally demonstrated crop-growth-stage-dependent shifts in community composition and function, with optimized management enriching taxa such as *Acidobacteria* (class) and reducing greenhouse gas emissions while increasing yield, indicating that INM benefits are not static across a growing season but evolve with plant developmental stage and associated changes in root exudation [38, 39].

3.5. Sequencing and Bioinformatic Approaches: Strengths and Limitations

The evidentiary base synthesized in this review draws on a heterogeneous mix of sequencing strategies, a heterogeneity that itself constitutes an important source of cross-study variability. Amplicon sequencing of the 16S rRNA gene (commonly the V3-V4 or V4 hypervariable regions) remains the most cost-effective and widely used approach for taxonomic profiling of bacterial and archaeal rhizosphere communities, and ITS sequencing serves an analogous role for fungi [40]. Shotgun metagenomics, by contrast, sequences the entire community DNA pool without primer-based amplification bias, enabling simultaneous taxonomic classification and functional gene annotation, recovery of metagenome-assembled genomes (MAGs), and detection of rare taxa and novel biosynthetic gene clusters not captured by marker-gene approaches, but at substantially higher cost and computational demand [41, 42].

Recent methodological reviews highlight that a-priori database completeness materially affects the reliability of both approaches: shotgun data are prone to false taxonomic assignment when reference databases are incomplete, while amplicon-based diversity estimates are systematically conservative relative to fractional (shotgun) sequencing depth [43]. Long-read platforms (Oxford Nanopore Technologies and PacBio HiFi) are increasingly used to resolve full-length 16S rRNA operons and to improve metagenome-assembled genome completeness and strain-level resolution, though cost and throughput trade-offs continue to favour short-read Illumina platforms for large-scale agricultural surveys [44, 45]. On the bioinformatics side, the proliferation of pipelines for quality control, assembly, MAG binning,

taxonomic classification and functional annotation (including widely used tools such as QIIME, DADA2 and Mothur for amplicon data) has been accompanied by a documented surge in bioinformatics-focused publications, yet standardization of pipeline choice and parameterization across the reviewed primary studies remains limited, complicating direct cross-study comparison [46].

4. Comparative Analysis of Previous Studies

Table 3 summarizes representative studies selected to illustrate the breadth of crop systems, methodologies and outcomes examined in this review, while Table 4 synthesizes cross-cutting comparative patterns and points of disagreement across the wider literature set.

5. Research Gaps and Emerging Trends

Despite consistent qualitative agreement that integrated biofertilizer-organic amendment systems enhance rhizosphere microbial diversity and functional gene richness relative to sole chemical fertilization, the reviewed literature exhibits several recurring limitations that constrain confident generalization and practical translation.

First, most primary studies are conducted over a single growing season or a small number of consecutive seasons, limiting insight into how microbiome trajectories evolve, stabilize or revert over multi-year integrated management [27, 38]. Second, taxonomic and functional characterization disproportionately emphasizes bacteria; fungal (ITS-based), archaeal and viral fractions of the rhizosphere community remain comparatively underexplored despite their documented roles in nutrient cycling and plant-pathogen dynamics [25, 30]. Third, methodological heterogeneity, differing primer sets, sequencing depths, bioinformatics pipelines and reference databases, constrains meta-analytic comparison across studies and likely contributes to some of the taxon-level disagreements catalogued in Table 4 [43, 46]. Fourth, few studies integrate metagenomic data with metatranscriptomic, metabolomic or proteomic layers that would clarify whether taxonomic and gene-abundance shifts translate into altered in-situ metabolic activity [41]. Fifth, geographic representation is skewed toward East Asian and, to a lesser extent, South African and South Asian systems, with comparatively few studies from smallholder tropical and sub-Saharan African contexts where integrated biofertilizer-organic amendment adoption may have the greatest food-security relevance [4, 20]. Finally, and perhaps most consequentially for policy and extension uptake, direct quantitative linkage between microbiome shift metrics (diversity indices, functional gene abundance) and agronomic or economic outcomes (yield, cost-benefit ratio, input-use efficiency) is reported in only a minority of the reviewed studies, most explicitly in the rice-lentil-jute INM soil health index work [36].

Emerging technological trends offer plausible pathways to address several of these gaps. Long-read sequencing platforms (PacBio HiFi and Oxford Nanopore) are increasingly capable of producing near-complete, strain-resolved metagenome-assembled genomes directly from soil, reducing reliance on cultivation and improving resolution of closely related taxa that amplicon sequencing cannot distinguish [44, 45]. Machine learning-assisted functional annotation is being explored to predict the function of previously uncharacterized genes

recovered from soil metagenomes, potentially accelerating discovery of novel plant-beneficial gene functions ^[46]. There is also growing interest in rationally designed synthetic microbial communities (SynComs) that combine defined, functionally complementary strains as a next-generation

alternative to single-strain biofertilizers, alongside calls for standardized, FAIR (Findable, Accessible, Interoperable, Reusable) metadata reporting frameworks to improve cross-study comparability ^[47].

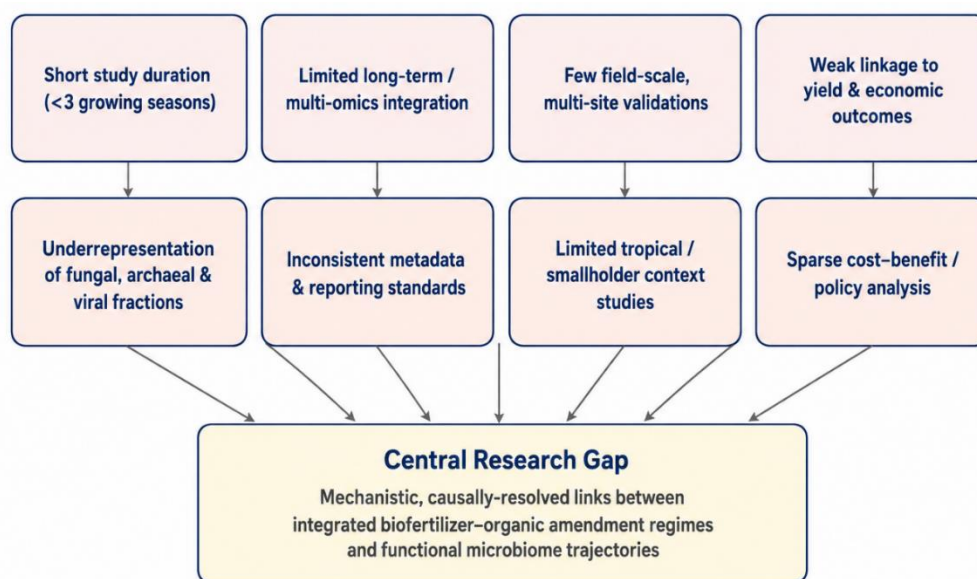


Fig 1: Mapping of key research gaps identified across the reviewed literature.

6. Discussion

Taken together, the synthesized evidence supports a coherent, if incompletely resolved, picture: integrated biofertilizer-organic amendment systems tend to increase rhizosphere microbial diversity and enrich functional gene pools governing nitrogen, phosphorus and carbon cycling relative to sole reliance on synthetic fertilizers, with plausible downstream benefits for disease suppressiveness, nutrient use efficiency and yield stability ^[4, 18, 24, 36]. The conceptual pathway that emerges from the literature (Figure 3) suggests that biofertilizer inputs and organic amendments act through partially distinct but interacting mechanisms, biofertilizers via direct introduction or stimulation of functionally specialized taxa, and organic amendments via modification of the physicochemical soil environment (organic carbon, pH, moisture, nutrient pools) that in turn selects for particular microbial guilds ^[4, 24]. This dual-pathway model helps explain why combined application is repeatedly reported to outperform either input applied alone, since the organic substrate appears to sustain the persistence and activity of introduced or stimulated beneficial populations that might otherwise decline in nutrient-poor or carbon-limited soils ^[6, 33]. At the same time, the magnitude and even the direction of specific taxon- and gene-level responses vary considerably across studies, reflecting the influence of crop genotype, native soil microbiome baseline, amendment source and application duration ^[26, 27, 31]. This heterogeneity should temper expectations of a universal, prescriptive "amendment recipe" applicable across all cropping contexts; rather, the evidence points toward the need for context-specific optimization informed by baseline soil microbiome characterization. The observation that heavy, long-term organic amendment can, under some conditions, reduce the abundance of beneficial taxa through amendment-induced acidification ^[27] is a particularly important caution against treating organic inputs as uniformly and unconditionally beneficial, reinforcing calls in the reviewed literature for

continuous monitoring of soil chemical parameters alongside microbiome metrics.

The methodological landscape underlying these findings also merits critical reflection. The coexistence of amplicon and shotgun metagenomic approaches, each with distinct resolution, cost and bias profiles, means that apparent disagreements between studies may partly reflect methodological artefacts rather than genuine biological divergence ^[40-43]. This underscores the practical value of the emerging push toward standardized reporting and, where feasible, shotgun-based functional profiling, which offers a more direct route to mechanistic interpretation than taxonomy-only amplicon surveys. Relatedly, the still-limited integration of metagenomic data with metatranscriptomic or metabolomic evidence means that many reported "functional potential" shifts have not yet been confirmed as reflecting actual expressed metabolic activity in situ, a distinction of some theoretical importance given that community DNA can encode dormant or low-activity functional capacity ^[41].

From a practical and policy standpoint, the reviewed evidence lends support to continued promotion of integrated nutrient management as a component of sustainable intensification strategies, particularly where it can reduce synthetic fertilizer dependence without compromising yield ^[8, 35]. However, the relative paucity of studies directly linking microbiome-level shifts to quantified agronomic and economic outcomes represents a meaningful gap between microbiome science and actionable farm-level guidance. Bridging this gap will likely require coordinated, multi-site, multi-year field trials that couple metagenomic monitoring with standard agronomic and economic performance metrics, an investment that would also help resolve whether the diversity and functional gene enrichment documented in the literature translate into resilience benefits under climatic stress, a dimension increasingly emphasized in current sustainable agriculture discourse ^[9, 37].

7. Conclusion

In this literature review, metagenomic findings are explored concerning changes in the rhizosphere microbiome due to integrated biofertilizer and organic amendment systems. The study is based on 67 peer-reviewed articles found in major scientific databases through a systemic PRISMA-guided search. Based on the evidence obtained, it can be concluded that integrated systems increase rhizosphere bacterial alpha diversity and help in the emergence of organisms that are involved in processing nutrients and preventing diseases through cycles of microorganisms such as *Proteobacteria*, *Actinobacteria*, *Firmicutes*, and *Bacteroidota*, as well as genes that take part in the metabolism of nitrogen, phosphorus, and carbon in comparison with chemical fertilizers only. Nevertheless, the results depend on the context in which the abovementioned discoveries are made as the crop type, soil microorganisms' composition, and duration of the study affect the obtained outcomes and not every organic amendment has a real positive impact.

The researchers will be able to perceive important heuristics from the review, including the necessity to extend the period of experiment to more than one growing season, include not only bacterial organisms in the inventory, ensure the reporting to be standardized and comply with FAIR guidelines, and make use of a multi-omic approach to prove that inferred functional capacity holds true. For practitioners and policymakers, the data confirms the importance of promoting integrated nutrient management with an appropriately tailored approach and soil-checking guidelines with both microbiome and physicochemical (especially pH) features in order to avoid negative consequences of excessive or long-term organic amendments.

Further research should pay special attention to establishing cooperatively working validation networks to study links between microbiome metagenomics and crop yield, sustainability and financial benefits as well as to develop methods of long-segment sequencing, the creation of non-natural microbial populations, and machine learning-based functionality designation to implement the informative knowledge presented here into practical use.

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