

VOLUME 11 ISSUE 2 2025

ISSN 2454 – 3055



**INTERNATIONAL
JOURNAL OF
ZOOLOGICAL
INVESTIGATIONS**

***Forum for Biological and
Environmental Sciences***

Published by Saran Publications, India



The Overlooked Aquatic Realm: A Call to Integrate Reactive Nitrogen Dynamics Across Environmental Compartments

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Received: 24th April, 2025; **Accepted:** 22nd July, 2025; **Published online:** 26th December, 2025

<https://doi.org/10.33745/ijzi.2025.v11i02.122>

Abstract: The comprehensive study on an individual reactive nitrogen (Nr) pollutant is not the foremost priority. Instead, emphasis should be placed on studying the exchange and linkages between diverse N pollutants to develop thorough insights for cascading Nr in the environment. Research that associates emissions of Nr, their transformations, and potential impact on water, soils, air, and plants with different geophysical settings like urban-rural, aquatic-marine, biosphere-troposphere-stratosphere, etc. is the present necessity. The marine and terrestrial ecosystems have been studied extensively, but a gap remains in understanding nitrogen (N) dynamics in lentic and lotic ecosystems. There is a lack of national estimates of denitrification in groundwater, as site-specific estimates were not scaled up. Interaction among wetlands, the addition of N loads, and climate dynamics are also not well understood, but considering the wetland's efficiency in removing N, assessing its potential is a matter of crucial research. Considering the area that could process anthropogenic inputs of N makes the aquatic ecosystems disproportionately important. This study undertakes a bibliometric analysis to understand the scientific landscape for Nr research, with a focus on prejudice, building on past research primarily focused on agroecosystems, plant stress, crop productivity, and their contribution to reactive nitrogen. In contrast, this study aims to draw the attention of researchers to aquatic ecosystems that act as a sink for Nr species and have been severely impacted by the effects of Nr species.

Keywords: Reactive nitrogen, Aquatic ecosystem, Nitrogen pollution, Bibliometric study, Nutrients

Citation: Dashora Milap, Kaushik Garima and Kumar Alok: The overlooked aquatic realm: A call to integrate reactive nitrogen dynamics across environmental compartments. Intern. J. Zool. Invest. 11(2): 1233-1243, 2025.

<https://doi.org/10.33745/ijzi.2025.v11i02.122>



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Introduction

The need for sustainable utilization and management of nutrients, primarily nitrogen, triggered severe concerns associated with

environmental pollution. Dinitrogen (N₂) is an amply inert gas that constitutes the largest N pool, making up ~80% of the air we breathe.

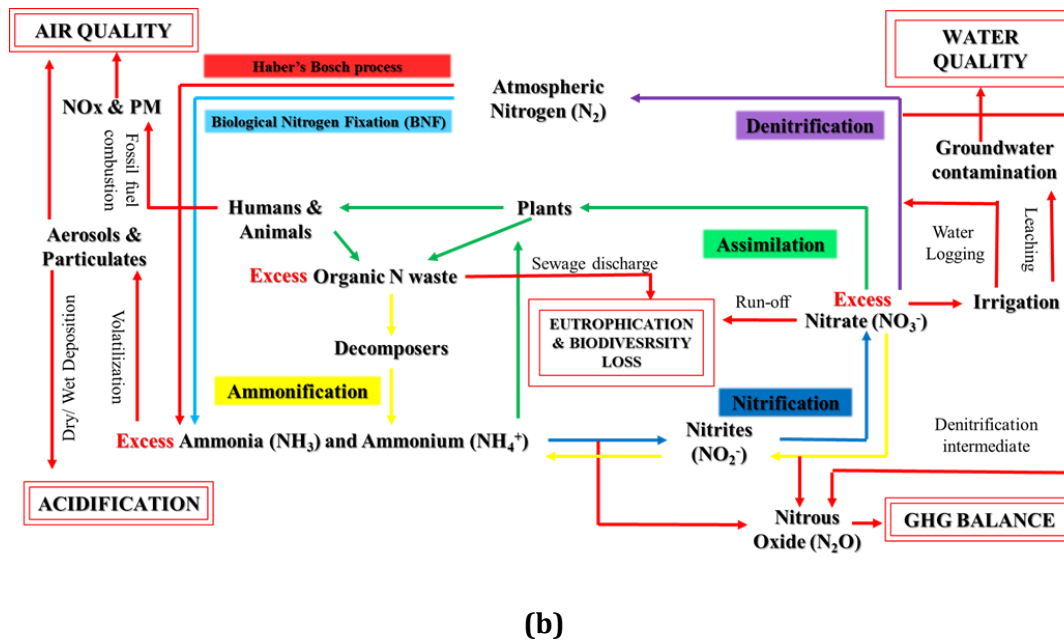
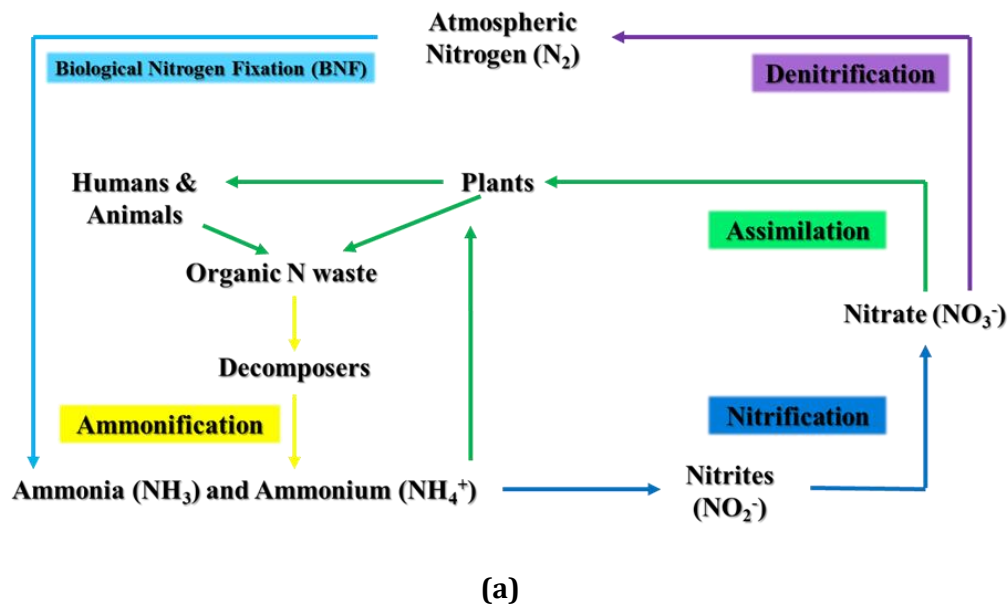


Fig. 1: Biogeochemical cycling of nitrogen (a) with ideal representations of major transformation pathways in a balanced system, and (b) addition of unintended pathways (red arrows) due to anthropogenic alterations, ensuing excessive mobile Nr species, a cascade of accelerated transformation rates resulting in five key environmental threats (red boxes).

Conversely, reactive nitrogen (Nr) broadly envelops all photochemically, biologically reactive, and radiative N species that include inorganic forms (NH_3 , NO_x ($\text{NO} + \text{NO}_2$), N_2O , NH_4^+ , NO_2^- , NO_3^-) and organic forms (nucleic acid, amines, proteins, urea, N in soil humus) (Subbarao *et al.*, 2017). The biogeochemical cycling of nitrogen seeks to

commendably balance the reactive and inert species. However, with anthropogenic perturbation in the nitrogen cycle amplified since the mid-20th century, nitrogen cycling has undergone substantial transformations within its functional framework and operational rates at different scales over the last six decades. Therefore, it

remains the most perturbed biogeochemical cycle (Abrol and Adhya 2017). These alterations engendered a cascade of unintended pathways which resulted in five critical environmental threats (Fig. 1) of reactive nitrogen (water quality, air quality, greenhouse balance, eutrophication and biodiversity loss, and soil quality) (Sutton *et al.*, 2011). As a consequence of the cascade, the excess of mobile reactive nitrogen species leaves trailing environmental impacts and eventually becomes source-independent. The complex dynamics of the ecological matrices rendered fate approximation of reactive nitrogen species quite challenging. The primary reason for these undesirable impressions resulted inadvertently within the global nitrogen cycle was anthropogenic activities like extensive combustion of fossil fuels alongside brisk urbanization, undue application of excess fertilizer in agriculturally intensive ecosystems amid low nitrogen use efficiency (NUE), boosted efflux of nitrogen to foster rapid advances, and intensive livestock production systems.

Amidst sequential biotransformation reactions and several migration pathways, uncontrolled emissions of Nr have resulted in significant dry/wet depositions of atmospheric Nr species within terrestrial and aquatic ecosystems. This had consequently triggered ecosystem malfunctions, along with environmental consequences, mainly recurring climate change events, rendering this issue an imminent global concern. Atmospheric N deposition has emerged as a vital concern within the N cycle worldwide, leading to numerous issues, including nitrogen imbalances in several ecosystems. The dramatic deposition had instigated detrimental impacts, resulting in severe acidification, the disappearance of indigenous species with defunct microbial diversity, the decoupling of the stoichiometric balance of C-N-P, the unlinked confluence of surface biomass with subsurface microbial activities, the exacerbation of greenhouse gas imbalance, and hyper-eutrophication in aquatic ecosystems. Annual mean atmospheric deposition rates of reduced forms of Nr touched a mark of 180 Tg N yr⁻¹ (Xu *et al.*, 2016)

due to continual emission from anthropogenic sources. The deposited Nr causes acidification and is transformed through nitrification into nitrate (NO₃⁻), which acts as a pollutant and contaminates groundwater, causing nitrate poisoning in infants and severe health issues. Once nitrate reaches the surface water body, it could promote biodiversity loss alongside eutrophication in the aquatic ecosystem. Excess NO₃⁻ undergoes denitrification and is lost into the atmosphere. Nitrous oxide (N₂O), formed as an obligate intermediate in denitrification, is ~300 times more potent as a greenhouse gas than CO₂. Total N₂O emission increased during 2000 to 2010 from 264.16 Gg to 370.38 Gg yr⁻¹. The indirect N₂O emissions due to Nr deposition are underestimated by the Intergovernmental Panel for Climate Change (IPCC) by at least a factor of 2.

Rapid industrialization and fossil fuel combustion caused NO_x emissions, which have deteriorated air quality. Over the past decade, the NO_x emissions rate has increased by 6.5% per year. At this rate, NH₃ emissions will be surpassed by NO_x by 2055. Even the Indian per capita trends for NO_x emissions are rising (SANH and SACEP, 2022). Around 30-70% of the contribution to particulate matter (PM) by mass comes from Nr, limiting visibility and affecting respiratory health in sensitive people. Air pollutants, such as PM_{2.5}, annually claim more lives globally than the COVID-19 pandemic does in a year. Nr emissions can produce both potential cooling and warming of the climate. The positive forcings are induced through excess concentrations of greenhouse gases N₂O and O₃ in the troposphere. On the other hand, the major cooling effects are either indirect cooling caused by deposited Nr or directly through Nr aerosols. If Nr losses are moderated for any single species or pathway within the cascade, it would inadvertently promote losses in other Nr forms (e.g., moderating the leaching of NO₃⁻ would promote ammonia emissions and vice versa). Trade-offs like this for the pollutants are designated as 'pollution swapping' (Sutton *et al.*, 2011). Anthropogenic activities are a key aspect that alters the global N cycle by introducing

unregulated Nr inputs to ecosystems, which are responsible for excess Nr emissions in the form of NO (80% or more), NH₃ (70%), and N₂O (40%), globally. Societal costs of nitrogen pollution globally are around 200-2000 billion USD year⁻¹ while for Nr pollution in India, societal costs (for health, ecosystems, and climate) are around 17(8.5-34) billion USD for the year 2000 and 75(38-151) billion USD for the year 2015.

Global initiatives to address the Nr challenge

The problem of nitrogen pollution has been acknowledged for decades, even before the 1970's when the foundations were laid for environmental governance, and was not a new-fangled issue. The major issue was the fragmented focus rather than an integrated one to mitigate global nitrogen challenges. Carbon emerges as the elemental poster child of the Anthropocene, gaining recognition as both a source and solution to climate change, and ultimately, integrated management strategies are framed to address the challenge. But the impetus for the carbon challenge had shrouded another impending challenge, "The Nitrogen Challenge", unaddressed. The Anthropocene has transformed the vital resource into a major pollutant by tipping the natural balance, and we are gradually drowning in the silent sea of reactive nitrogen. The action plans and approaches to address the N challenge stalled due to a lack of a holistic approach, and in the meantime, the nitrogen waste doubled and tripled in different regions within the past mid-century. Eventually, the scientist globally addressed this as a global management problem that requires a holistic approach and integrated action plans on a global scale. The first responders were Galloway (1998) and Sutton *et al.* (2000), and the tide has turned. As a result, the Gothenburg protocol was implemented, which further paved the way for the International Nitrogen Initiative (INI), whose concept came from the first and second International Nitrogen Conferences in 1998 and 2001, respectively. An outline for the integrated effort was developed for the upcoming years.

Eventually, the Scientific Committee on Problems of the Environment (SCOPE) and the International Geosphere-Biosphere Program (IGBP) agreed to sponsor the International Nitrogen Initiative (INI). Its formation was announced at the Symposium in 2003. Due to the global inadequacy of nitrogen awareness, the demand for assessing nitrogen challenges in different regions was imminent. With the support of INI and UNEP, the assessments were conducted in the various regions worldwide. The first assessment was conducted by the USA (Doering *et al.*, 2011), followed by Europe (Sutton *et al.*, 2011), India (Abrol and Adhya, 2017), and Pakistan (Aziz *et al.*, 2022).

Initiatives for Nr challenge in the Indian and south-Asian region

Indian Nitrogen brainstorming began after the INI was set up in 2003. As a result, workshops on Indian nitrogen started in 2004. With the wider consultation received from INI, the Indian Nitrogen Group (ING) was established in 2006. As nitrogen pollution was widely underestimated in the South Asian region, INI expanded to South Asia, and the South Asian N Centre was established in 2008. In 2009, the Global Partnership on Nutrient Management was launched by UNEP in South Asia and worldwide. Around 2010, the INI declaration was signed in Delhi, and with the completion of ENA 2011, the Indian Nitrogen Assessment started in 2011. In 2013, under the South Asian Co-operative Environment Program (SACEP), a study was launched to examine nutrient loading to South Asian seas. In 2014, Sustainable India Trust was established in New Delhi. Around 2016, the Indo-UK joint virtual N Centres were launched. In 2017, the Indian Nitrogen Assessment (INA) was completed in conjunction with the SACEP N resolution. A session on Nitrogen was organized on World Environment Day (2018) in Delhi. In 2019, the Colombo Declaration and the launch of the South Asian Nitrogen Hub (SANH) project came. During 2020, UNEP held its first meeting for

the nitrogen working groups, at which SANH briefed the working groups on its policy (Raghuram *et al.*, 2021). To date, the South Asian N roadmap has been strengthened, and other South Asian countries have also developed their own Nitrogen Assessments in 2022.

Importance of aquatic systems in N retention and cycling

Aquatic systems play a crucial role in nitrogen retention and cycling, which are essential for maintaining ecosystem health and mitigating the impacts of human activities on the nitrogen cycle. All reactive forms of N ultimately reach these systems (rivers, lakes, ponds, wetlands, estuaries, and oceans) and serve as global sinks for Nr species. N retention primarily occurs through immobilization, where inorganic nitrogen assimilation by benthic communities (phytoplankton and macrophytes) transforms them to organic forms, thereby buffering nitrogen transport and mitigating environmental pollution (Peng *et al.*, 2017; Orellana *et al.*, 2018). Denitrification in lakes aids in nitrogen removal, reducing local and downstream nitrogen concentrations. Wetlands, lakes, and rivers contribute to this process, with wetlands retaining the highest proportion of nitrogen due to their water discharge characteristics (Lovett *et al.*, 2009). However, the benthic community may offset some benefits by releasing ammonia, stimulating primary productivity, and increasing nitrous oxide emissions (Kelly *et al.*, 2021). These systems help reduce nitrogen pollution and maintain ecological balance, highlighting the need for effective management and conservation strategies to support their roles in nutrient cycling. Considering their importance in N transformation, further research studies are needed to address the complexity of N transformation within dynamic regimes, such as littoral zones in water bodies like lakes and ponds, as well as riparian zones in rivers and estuarine systems. We hypothesize that most of the studies, particularly in the Indian region, have primarily focused on agroecosystems, and there is a lack of

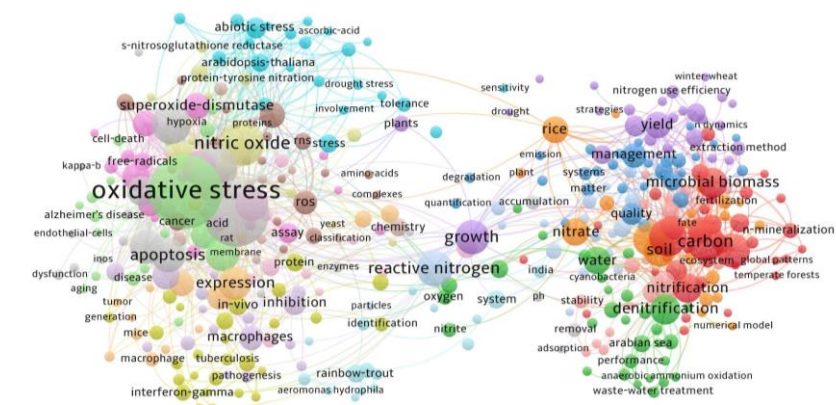
substantial estimates of N transformation rates, major processes, and controlling factors in both lentic and lotic systems. Peninsular India has 7516.6 km of coastline, followed by major rivers, inland lakes, and wetlands (freshwater and saline), yet few studies have assessed the Nr transformation in these aquatic ecosystems. Considering the above gap, we conducted a bibliometric assessment for studies on “reactive nitrogen” in the Indian region for validation.

Bibliometric analysis

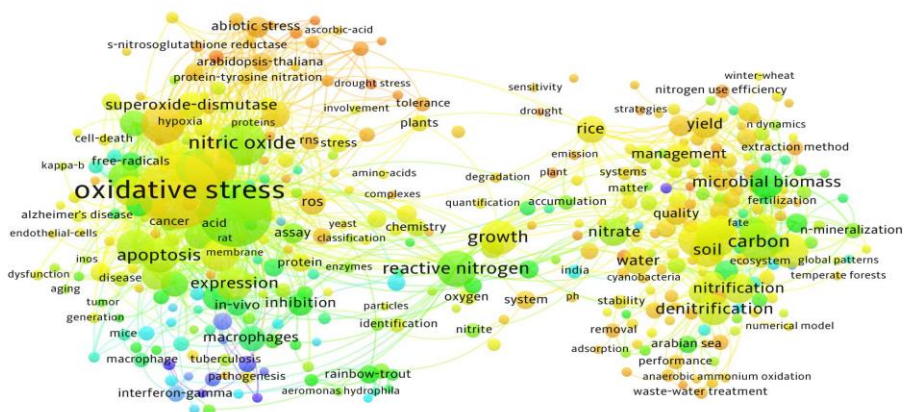
As a part of the bibliometric assessment, an advanced query search was conducted on the Web of Science (WoS) platform (Clarivate Analytics) on the research topic (reactive nitrogen), considering different variants of the term, only within the Indian region, during the period 1989 to 2025. The WoS bibliometric records were screened and exported as a query response. The bibliometric record file was further imported using VOSviewer v1.6.20 to generate co-occurrence maps. Considering the full counting method, the irrelevant keywords were omitted, and the three co-occurrence cluster maps were generated (Figs. 2 a, b, c). Additionally, analyse results feature was also used to export additional bibliometric data and visualization of the records from the WOS platform.

Bibliometric landscape for reactive nitrogen studies

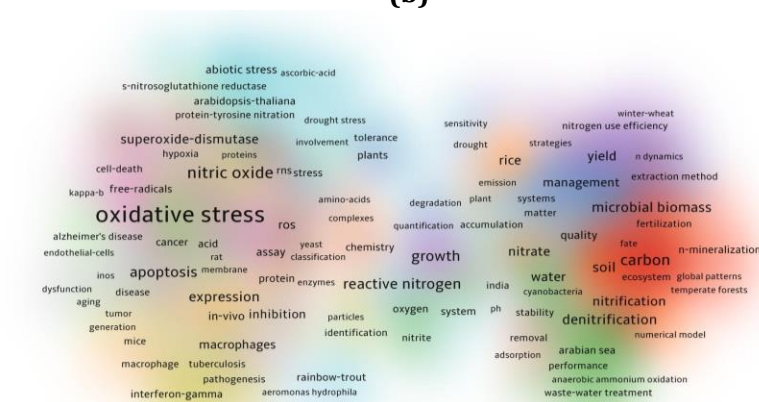
The network visualization (Fig. 2a) highlight 43 keyword clusters associated with reactive nitrogen, with major clusters on the left associated with crop stress and productivity, broadly covering aspects of agroecosystems. In contrast, clusters on the right include the N transformation process in soil water, crop yield, nutrient use efficiency (NUE), and rice management. The green cluster includes studies on aquatic systems addressing the Nr transformation process. Additionally, the overlay visualization affirms the recent contributions in plant stress studies in the past decade and the Nr transformation studies in aquatic ecosystems in the current decade (Fig. 2b). The density visualization (Fig. 2c), in the case



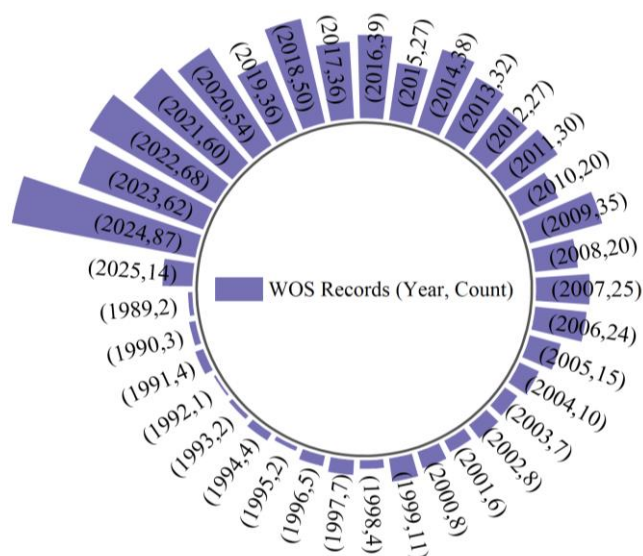
(a)



(b)



(c)



(d)

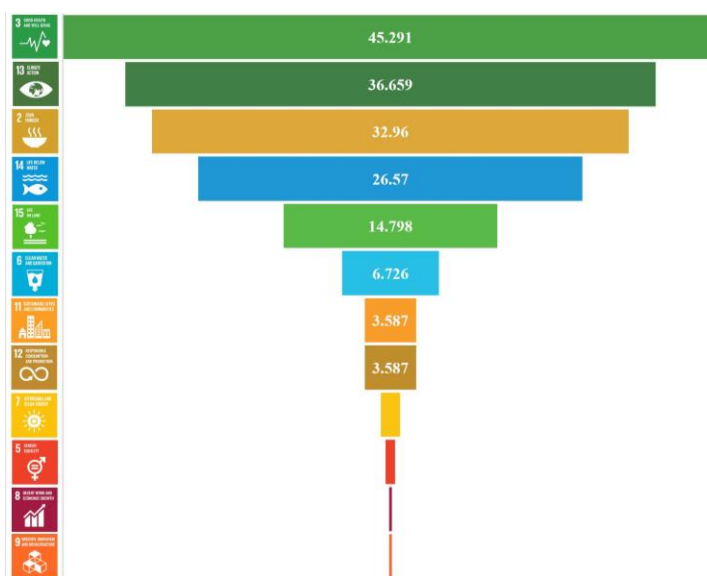
Fig. 2: Keyword (Item) clusters associated with the term reactive nitrogen as per advanced query-based search (WOS-Clarivate) depicting (a) network visualization with 23 clusters for 398 items (occurrence), at 2.5 resolution and default settings, (b) overlay visualization with color overlay (year) on item mentions in publications during the period (2000-2025), and (c) cluster density visualization representing item clusters generated through co-occurrence keyword analysis (full counting) in VOSviewer v1.6.20 and (d) year-wise counts for WOS records after advance query search (1989-2025) on the platform (Accessed on 07-04-2025).

of the aquatic ecosystems, shows that transformation processes like denitrification, nitrification, and anaerobic ammonium oxidation (ANAMOX) as major interest of the Indian researchers, but still certain transformative processes like nitrite-dependent anaerobic methane oxidation (n-DAMO), which links carbon and nitrogen cycles (Zhang *et al.*, 2024), along with assimilatory and dissimilatory nitrate reduction to ammonium (ANRA and DNRA) (Cheng *et al.*, 2023) are poorly understood and need greater attention for better understanding. Overall, the number of research studies conducted on reactive nitrogen has been increasing annually (Fig. 2 d). Still, future studies that target crucial systems to mitigate the impacts of Nr turnover into a pollutant are the present need.

Accomplishments and present stakes for Nr studies

The various bibliometric categories on the WoS

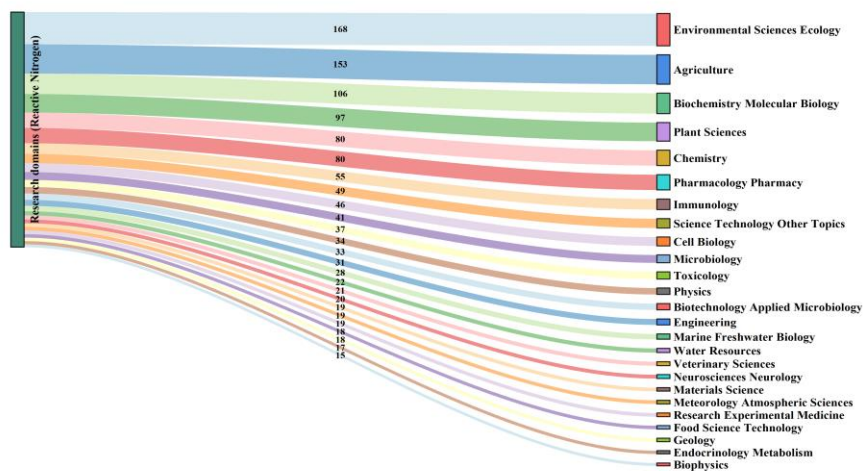
platform provide sufficient information about significant contributions related to the query topic. The Sustainable Development Goals (SDGs) category pinpoints the research studies' alignment with the targeted SDGs. Our query results highlight that SDG goals 3, 13, and 2 were the top 3 SDGs addressed by researchers in India associated with reactive nitrogen (Fig. 3a), followed by goals 14, 15, and 6, which need to be prioritized. Within the category of citation topics meso (Fig. 3b), Soil Science, Molecular and Cell Biology-Pharmacology, Crop Science were the prime citation topics. Looking at the research areas under the reactive nitrogen domain (Fig. 3c), water resources are relatively low, and the majorly focused topics were Agriculture, Biochemistry, Molecular Biology, and Plant Sciences, which broadly fall under agroecosystem studies. These findings confirm our hypothesis that reactive nitrogen studies are more biased at



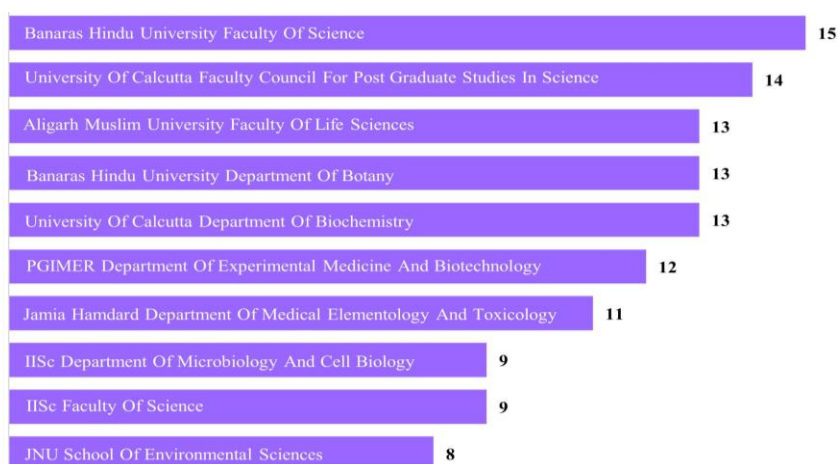
(a)



(b)



(c)



(d)

Fig. 3: Bibliometric categories from WOS platform for Nr topic (a) pyramid plot representing (%) contributions of Nr records aligning with the different SDGs, (b) treemap representing citation topics associated with Nr studies, (c) sankey plot representing research domains undertaking Nr studies, and (d) top ten institutions undertaking the Nr research studies (Accessed on 07-04-2025).

domains like agriculture, crop production, and plant productivity in India, which has undoubtedly turned India into a Golden Bird, but the ensuing impacts of non-judicious use of nutrients have turned these nutrients into pollutants, particularly the three species of Nr (NO_2 , NH_3 , N_2O) had been categorized as Triple trouble (Raghuram *et al.*, 2021). Nr species ultimately reaches the aquatic ecosystem, so studies for assessment of N transformation within the distinct aquatic ecosystem should be undertaken to mitigate the Nr associated impacts early.

Significance of aquatic systems studies for Nr transformation

The comprehensive study on an individual Nr pollutant is not the foremost priority. Instead, emphasis should be placed on studying the exchange and linkages between diverse N pollutants to develop thorough insights for cascading Nr in the environment (Rudek *et al.*, 2017). Research that associates emissions of Nr, their transformations, and potential impacts on water, soils, air, and plants with different geophysical settings, such as urban-rural, aquatic-marine, and biosphere-troposphere-stratosphere, is a present necessity. The marine and terrestrial ecosystems have been studied extensively, but a

gap remains in understanding nitrogen (N) dynamics in lentic and lotic ecosystems. There is a lack of national estimates of denitrification in groundwater, as site-specific estimates were not scaled up. Interaction among wetlands, the addition of N loads, and climate dynamics is also not well understood, but given the wetland's efficiency in removing N, assessing its potential is a matter of crucial research. Considering the area that can process anthropogenic inputs of N makes aquatic ecosystems disproportionately important (Galloway *et al.*, 2003). Nr inflow into lentic ecosystems predominantly through direct runoff through the surface, subsurface underground flow channels, contiguous exchange of groundwater, and dry deposition of atmospheric N in the form of ammonium (NH_4^+), nitrate (NO_3^-), or dissolved organic nitrogen (DON). NH_4^+ might undergo nitrifier-assisted transformation under aerobic conditions (with few exceptions) into nitrate (NO_3^-), nitric oxide (NO), a precursor for the formation of tropospheric ozone, and nitrous oxide (N_2O), a potent greenhouse gas (Baron *et al.*, 2012). Denitrification occurs in anoxic zones with sufficient available nitrate, which is transformed into nitrous oxide as an obligatory intermediate and ultimately into molecular nitrogen. Lentic ecosystems are crucially vital hotspots of

denitrification, which on average, have per unit area denitrification rates ten times higher than the soils (Seitzinger *et al.*, 2006). Both natural and artificial wetlands have high N storage capacity, either in soils or biomass, and denitrification is the major mode of removal (Seitzinger, 1988). Small lakes with an area < 50 km² are considered important as they remove (84% of N) and retain more N (1.1 Tg N yr⁻¹) than larger lakes (0.59 Tg N yr⁻¹) that are > 50 km² in area. Multiple factors could affect this, such as a greater number of smaller lakes than larger ones (Downing *et al.*, 2008). Nitrous oxide emission is proportional to the surface area. In larger lakes, it can cause eutrophication and hypoxic conditions, indirectly favoring denitrification and promoting nitrous oxide yield during denitrification. It is also unclear whether the environmental conditions or biotic communities have a major role in N dynamics within lentic ecosystems. It is reasonably obscure to deduce the microbial community's response to a moderate surge in surface water temperature. Deduction of the mechanisms by which microbial processes and aquatic stoichiometry are altered is a current area of research. It is highly ambiguous to assess the amount of N-rich buried organic matter within the reservoir or aquatic ecosystems and its dependence on the trophic state and land use. There is a lack of understanding of how the spatial variation of nitrification and denitrification within the lentic ecosystem contributes to the N dynamics of the lentic ecosystem. Often, lentic N cycling studies overlook the lake's stratification and the importance of the littoral zone, as well as its potential as a hotspot for nitrification and denitrification processes (Bruesewitz *et al.*, 2012). As evident from the above points, lentic ecosystems are a crucial component of N cycling, but there is a lack of a complete understanding of the different N transformation processes in lentic ecosystems.

Conclusion

Ultimately, the transition from viewing reactive nitrogen solely as a nutrient to recognizing it as a cascading environmental pollutant represents the

paramount challenge for contemporary Indian research. Scientific efforts must evolve beyond site-specific agricultural assessments to encompass integrated "source-to-sink" dynamics that link terrestrial runoff with the health of aquatic and atmospheric systems. Addressing these obscured microbial mechanisms and spatial variabilities within wetlands and small lakes is not merely a bibliometric necessity but a fundamental requirement for developing the mitigation strategies needed to safeguard national water resources and biodiversity by the 2030 deadline. The present need is a shift from isolated pollutant monitoring to a holistic understanding of N_r linkages across urban-rural and terrestrial-aquatic gradients. To achieve sustainable nitrogen management in India, policy interventions must transition from a yield-centric agricultural paradigm to a multi-sectoral Integrated Nitrogen Management (INM) framework. Ultimately, inter-departmental synergy between the ministries of Agriculture, Environment, and Water Resources is essential to mitigate the impacts of nitrogen turnover into pollutants.

Ethical Statement

No animal or microbes have been used or sacrificed for this study, hence Ethical Approval not required.

Author Contributions

Dashora Milap: methodology, data analysis, writing original draft; *Kaushik Garima*: formal analysis of manuscript, review and editing, validation; *Kumar Alok*: conceptualization, supervision, validation, editing and finalizing the manuscript. All authors have read and agreed to the published version of the manuscript.

Acknowledgements

Dashora Milap would like to thank the Department of Environmental Science, Central University of Rajasthan, Ajmer, and the Department of Environmental Studies, University of Delhi, for providing the facilities necessary for compiling this work.

Conflict of Interest

The authors declare no conflicts of interest.

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