Microsphere (2023) 2(2): 246-259
https://doi.org/10.59118/XFKD8065

Review

Elucidating the potential of nitrifying bacteria in mitigating nitrogen pollution and its industrial application

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Received: 18 November 2023 / Accepted: 16 December 2023 / Published online: 25 December 2023

Abstract: Nitrifying bacteria are specialized microorganisms that actively participate in the nitrification process, which involves the conversion of ammonia (NH_3) into nitrite (NO_2^-) and subsequently into nitrate (NO_3^-). Extensive human activities such as agriculture, thermal power generation, and automobile exhaust have led to an increase in the amounts of greenhouse gases and nitrogen pollution in soil and water. This comprehensive review highlights the importance of nitrogen-related processes for environmental sustainability and the role of nitrifying bacteria in combating nitrogen pollution. The focus of the current work is a detailed exploration of the diverse classes of nitrifying bacteria and their respective roles in environmental processes. Furthermore, the study explores practical applications of different classes of nitrifying bacteria, extending beyond the nitrification process. This review explored the potential of nitrifying bacteria in wastewater treatment, biodegradation of micropollutants, and the treatment of municipal solid waste leachate, showcasing the versatility of these bacteria in addressing broader environmental challenges.

Keywords: Ammoniacal Pollution · Nitrifying Bacteria · Wastewater Treatment · Anammox · Comammox

Introduction

Excessive nitrogen is a global issue with many hazardous impacts on the ecosystem, human health, and several problems like eutrophication. Ammonia (NH_3), nitrogen oxides (NOx), and Nitrous oxide (N_2O) are three significant nitrogen gases that are released by human activities like intensive agricultural operations such as volatilized livestock waste, nitrogen-based fertilizers, burning of fossil fuels for energy generation, and emissions from human excreta [1,2,3]. As per the United Nations Food and Agriculture Organization (FAO), in the last four decades, global crop production has doubled with an increase in synthetic Nitrogen fertilizer consumption by threefold [4]. Agriculture is responsible for nearly 81% of all global emissions of NH_3 into the atmosphere, followed by 11 % from burning biomass and 8.3 % from the energy production sectors, industries and traffic [5]. In addition to reducing the biodiversity of terrestrial and aquatic ecosystems, NH_3 pollution can also create aerosols in the atmosphere that can negatively affect human health if inhaled [6].

The nitrogen cycle comprises three processes: nitrogen fixation, nitrification, and denitrification, as shown in Figure 1. Nitrification is the degradation of organic nitrogen to an ammonia molecule called ammonification, which is subsequently oxidized to nitrite and nitrate and ultimately transformed into a dinitrogen gas molecule via denitrification [7]. Historically, microbes involved in the nitrogen cycle have been classified as “nitrogen fixers,” “nitrifiers,” or “denitrifiers” [8]. Microorganisms play a significant role in mediating the redox transformations of nitrogen, thereby influencing the concentrations of nitrogen molecules in the atmosphere, and regulating nitrogen fate in soils and marine ecosystems. Exploration of microbial communities participating in nitrogen cycling has grown in significant popularity in recent years because changes to the nitrogen balance brought on by human activities may considerably impact the health of ecosystems, biodiversity, and climate change [9].The primary aim of the current study is to critically examine the current state of information related to nitrifying bacteria, with a particular focus on their significance in industrial contexts. Additionally, the study provides insights into potential areas of future investigation to advance our understanding of this subject matter. The presence of an excessive quantity of nitrogen poses a significant environmental concern and poses a threat to ecosystems. A comprehensive evaluation of nitrifying bacteria and their counterparts would be advantageous in enhancing researchers' understanding of the complex steps involved in nitrogen removal in the environment,

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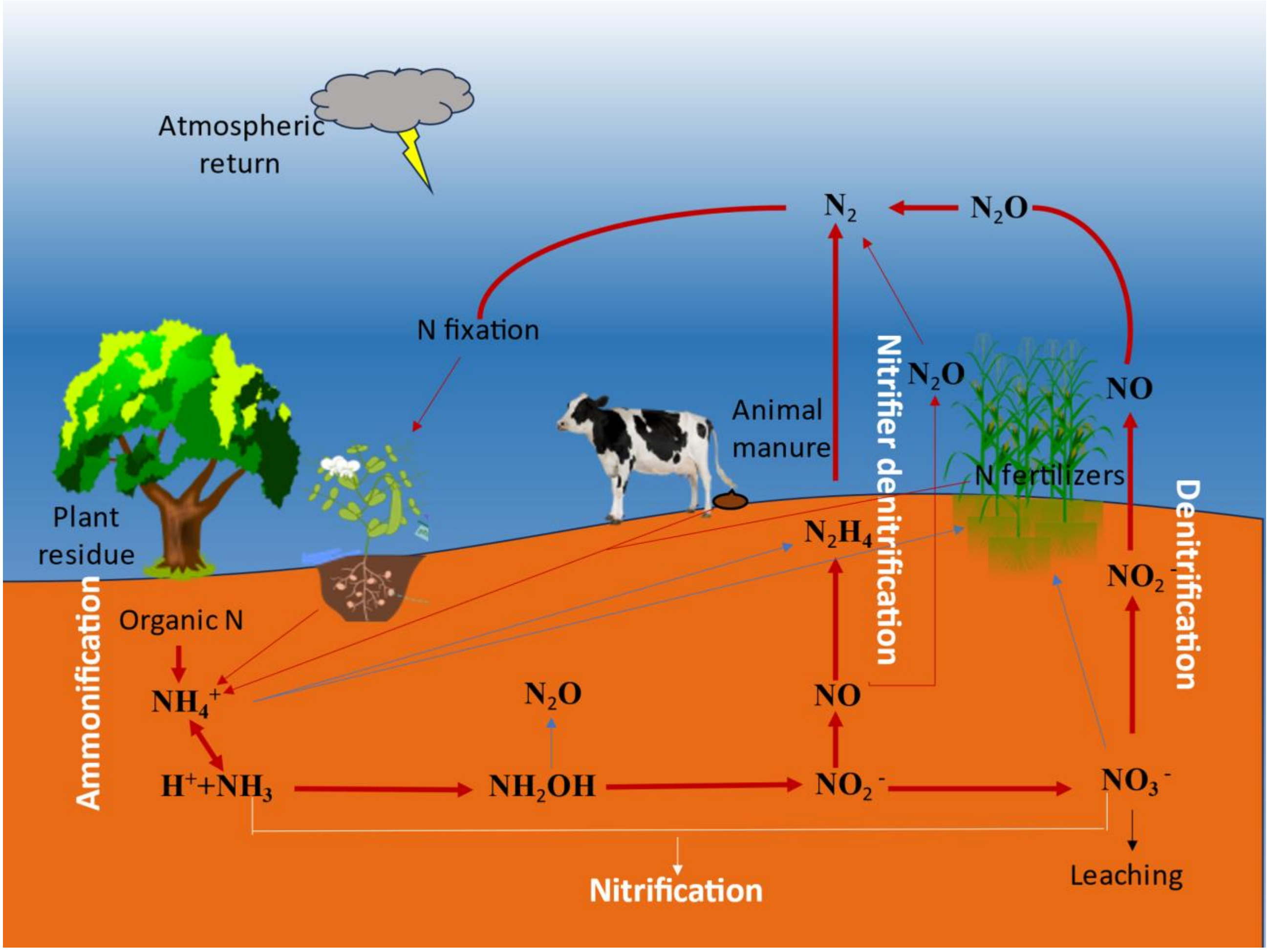

Fig 1: Schematic representation of the nitrogen cycle involving different processes of denitrification, nitrification, ammonification and nitrifiers denitrification, emphasizing significance in ecological sustainability and agriculture.

Role of nitrifying bacteria in biogeochemical recycling of nitrogen.

Nitrification is essential in the nitrogen cycle of terrestrial and aquatic environments because it converts the most reduced form of nitrogen, NH_3, to the most oxidized form, NO. Ammonia is naturally formed through the mineralization of organic matter and is also given to agricultural systems either directly as inorganic ammonium, such as ammonium nitrate or indirectly as urea and other ammonium-based fertilizers [10]. The microbial community engaged in nitrification, nitrogen fixation and denitrification are significantly influenced by the nitrogen fertilizer dosage and types because they get their energy from nitrite or oxidation of ammonia [11]. Nitrification influences nitrous oxide emission, nitrate pollution of aquatic resources, eutrophication, and agricultural areas [11,12]. Therefore, we need a greater understanding of nitrification processes to optimize the benefits of nitrogen fertilizers and reduce their detrimental effects on the environment. Sergei Winogradsky first discovered nitrifying microorganisms. Ammonia-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB) synergistically work together for nitrification. At first, ammonia is converted into nitrite by AOB, and subsequently, NOB transforms this nitrite into nitrate. Anaerobic ammonia oxidizers (anammox) and archaeal ammonia oxidizers (AOA) are also included in the groups of bacteria involved in the nitrification process. Ammonium is converted to N_2 gas by a process known as anaerobic ammonium oxidation (anammox), in which nitrite serves as an electron acceptor. Anammox is classified under *Planctomycetes phylum* and *Brocadialesorder* [5]

The metagenomic research has aided in the discovery of a new ammonia-oxidizing archaeal group that can act in a variety of conditions [13], and the metagenomic analysis has aided the isolation of ammonia-oxidizing archaea from marine and soil environments [14,15]. In recent studies, a new complete ammonia oxidation (comammox) bacterium, *Nitrospira,* that can oxidize ammonia to nitrate in one step has been isolated [16,17]. The significant microbial genes and processes involved in the transformations of nitrogen are summarized in Figure 2.

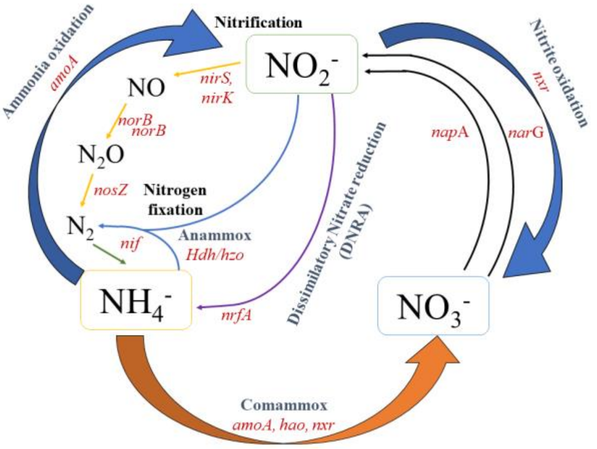

Fig.2. The schematic diagram of different microbial processes and enzymes involved in nitrogen transformations. Red italics indicate the significant enzyme-producing genes. All the processes are indicated with different individual colors. Nitrification is denoted in blue; nitrogen fixation is denoted in green; comammox reaction is represented in orange; denitrification is defined in blue; anammox is indicated in light blue; and dissimilatory nitrate reduction is represented in purple color.

Different classes of nitrifying bacteria based on their nature of oxidizing substrate.

The nitrification process relies on the combined action of ammonia-oxidizing bacteria (AOB) and ammonia-oxidizing archaea (AOA), as well as nitrite-oxidizing bacteria (NOB), anaerobic ammonia oxidation (Anammox) or solely on the functionality of complete ammonia-oxidizing bacteria (Comammox).

Ammonia Oxidizing Bacteria (AOB)

Gram-negative autotrophic ammonia oxidizers are dominant in ammonia oxidation. Due to their capacity to grow autotrophically and obtain energy from ammonia oxidation, these organisms were first classified as a single taxonomic group [18]. Most of the ammonia-oxidizing bacteria are found in the aerobic conditions. They are isolated or enriched from low-oxygen habitats like brackish water, virtually anoxic soil and sediment layers, and the subterranean parts of building materials [19].

Ammonia oxidizing bacteria are classified into proteobacteria and divided into two major groups: gamma proteobacteria and betaproteobacteria. *Nitrosomonas, Nitrosospira, Nitrosovibrio*, and *Nitrosolobus* are the four clusters that belong to the β-Proteobacteria subclass, where one cluster of *Nitrosococcus* belongs to γ-proteobacteria [19,20].

AOB derive metabolic energy through the initial conversion of ammonia or ammonium into hydroxylamine. This crucial enzymatic process is performed by an ammonia monooxygenase (AMO) enzyme. This enzyme is believed to consist of a functional *amo* operon, composed of at least three subunits expressed by the set of genes *amoA, amoB,* and *amok* [21].

Ammonia Oxidizing Archaea (AOA)

AOA represent one of the most abundant and widespread categories of microorganisms across the Earth's ecosystems [22]. Ammonia oxidizing archaea (AOA) are found in many habitats, including soil, marine and freshwater environments, and waste-water treatment plants. AOA is categorized as chemolithoautotrophs, which obtain energy through ammonia oxidation and utilize carbon dioxide to produce biomass [23]. AOA play a crucial role in the initial and controlling phase of nitrification, where they convert ammonia (NH_3) into nitrite (NO_2^-), producing energy for their autotrophic growth. Consequently, they significantly contribute to the worldwide nitrogen and carbon cycles [24]. Ammonia oxidizing archaea (AOA) are categorized into five primary clusters according to the phylogenetic analysis of the amoA gene, i.e., is responsible for encoding the functional subunit of the ammonia monooxygenase (AMO) enzyme, crucial for the starting step of ammonia oxidation [23]. The ammonia-oxidizing archaea (AOA) community within the phylum *Thaumarchaeota*is a diverse, widely distributed, and ecologically significant set of microorganisms in various ecosystems [25]. Based on the revised taxonomic proposal, the former *Thaumarchaeota* are now reclassified as the class *Nitrososphaeria* within the phylum *Crenarchaeota*, and all identified AOA belong to the order *Nitrososphaerales* [24]. The primary clusters are generally found in Group `1, known as the marine group, and Group 1.1b, referred to as the soil group" [23,26]. AOA is categorized into five clusters: *Nitrosopumilus, Nitrososphaera, Nitrosotalea, Nitrososphaera,* and *Nitrosocaldus*[27].

Nitrite oxidizing bacteria

Winogradsky isolated chemolithoautotrophic bacteria that use ammonia and nitrite as electron donors and energy sources. He reported that the Two distinct bacterial classes, ammonia-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB), play roles in this nitrification process. NOB is challenging to grow in the laboratory. However, Winogradsky successfully isolated nitrite-oxidizing bacteria by introducing a selective culture enriched with nitrite as the substrate, inoculating it with soil, and conducting consecutive subcultures in the identical medium. This method effectively eliminated ammonia oxidizers, and the nitrite oxidizer was obtained by plating on silica gel plates., which could not oxidize ammonia [28]. Nitrite oxidizing bacteria were considered obligate chemolithoautotrophs, accounting for a remarkable 88% of the fixed nitrogen in the oceans. They convert nitrite to nitrate to counteract nitrogen loss, which is utilized by many microbes and plants [29]. Nitrite is highly toxic to living organisms and inhibits bacterial growth by targeting heme-copper oxidase [30]. NOB activity tends to be unstable in waste-water treatment plants (WWTPs) associated with industrial systems. If nitrite from these treatment plants leaks into natural waters, tremendous ecological damage will occur [31]. *Nitrospinae* and *Nitrococcus* stand out as the primary marine nitrite-oxidizing bacteria (NOB), constituting as much as 9% of the microbial community abundances in oxygen minimum zones (OMZs), deep-sea waters, and sediments.[32]. *Nitrospira* is divided into six phylogenetic sublineages ubiquitously. *Nitrospira* has been found to carry out nitrite oxidation in moderately thermophilic habitats such as geothermal hot springs with a temperature range of 60–65°C, thus indicating an unexpected diversity of heat-adapted *Nitrospira* [33].Nitrite oxidoreductase is a crucial enzyme in NOB. The *nxrA* and *B* genes are functional and phylogenetic markers for detecting and identifying uncultured NOB.[34].

Anammox

Anammox bacteria, discovered in the 1990s, have been observed in diverse environments, including waste-water treatment facilities, lakes, marine subtoxic regions, and coastal sediment. These bacteria play a fundamental duty in the nitrogen cycle and are identified as a crucial global source of dinitrogen gas production. The average diameter of anammox bacteria, which are coccoid in shape and are anaerobic chemolithoautotrophs, falls within the range of 800 to 1,100 nanometers [35]. The anammox process represents a one-step mechanism wherein ammonia is transformed into nitrogen in the presence of nitrite. Bacteria prevalent in anaerobic NH_4^+ oxidation are widely distributed in various environmental conditions and are classified into the Planctomycetes phylum. Nineteen anammox species have been identified and distributed in five genera [36]. Five recognized genera of anammox bacteria include *Candidatus Kuenenia, C. Anammoxoglobus, C. Brocadia, C. Scalindua and C. Jettenia*. [35]. Under anoxic conditions, the anammox reaction converts ammonium and nitrite into N_2 gas [37].

$$NH_4^+ + NO_2^- \rightarrow N_2 + 2H_2O.$$

Comammox

Different groups of microbes carry out the ammonia and nitrite oxidation process. a microbe can perform complete nitrification [17]. Therefore, complete ammonia oxidizers and nitrifiers named comammox (complete ammonia oxidizer) microbes were discovered, with a relatively low growth rate but higher growth yields than other ammonia oxidizers. Comammox is categorized within the genus *Nitrospira* and performs complete ammonia oxidation using genes involved in ammonia oxidation (hydroxylamine dehydrogenase e[*hao*] and ammonia monooxygenase[*amo*]) and reduction of nitrite (nitrite oxidoreductase [*nxr*]). therefore, directly convertingNH_4^+ to NO_2^-[38,39]. The reaction involved in comammox is as follows:

$$NH_4^+ + 2O_2 \rightarrow NO_2^- + H_2O + 2H^+$$

Unlike chemolithoautotrophic nitrite oxidizers, comammox *Nitrospora* can utilize both ammonia and nitrite as energy sources [39].Heterotrophic nitrification involves the conversion of ammonia into nitrite and nitrate, a process carried out by diverse chemoorganotrophic microorganisms [40,41]. Also, various heterotrophic nitrifiers can perform aerobic denitrification by catalyzing the nitrate reduction into nitrite in the presence of oxygen nitrous oxide production results from microbial denitrification, contributing to greenhouse gas emissions [40]. Denitrifiers can catalyze nitrous oxide reduction to nitrogen using nitrous oxide reductase (*nosZ*) [42]. Earlier, the *nitrospora* was assumed as NOB, but the discovery of comammox evidenced that it can encode for both ammonia and nitrite metabolism as it contains both dehydrogenase and oxygenase gene clusters mediating the complete oxidation of ammonia to nitrate [39,43,44]. Limited comammox-like *Nitrospira* genomes have been reported to date, which include *Nitrospira sp. strain Ga0074138, C. Nitrospiranitrificans, C. Nitrospirainopinata, and C. Nitrospiranitrosa* [39,45,5]. It has been reported that the comammox yields higher energy ($G = -349$ kJ·mol/L NH_3) in comparison to traditional nitrifiers [39] . Since the availability of ammonia has been acknowledged as a critical element determining the niche distinction between AOA and AOB, it is anticipated that ammonia, rather than ammonium, is the direct substrate for AMO.

These different classes of bacteria have adapted to use nitrogen in various ways. Table 1 enlists some of the nitrifying bacteria with their genes, enzyme and the reactions during nitrogen metabolism

Table 1. Various genes, enzymes, and reactions catalysed by nitrifying bacteria for nitrogen metabolism.

S. no.	**Nitrifying bacteria**	**Modus Operandi**	**Genes**	**Enzyme encoded**	**Reaction catalysed**	**Reference**
1	*Nitrosomonas spp., Nitrosospira briensis, Nitrosovibrio tenuis, Nitrosolobus multiformis, Nitrosococcus spp.,*	Ammonia oxidizing bacteria	amoA	Ammonia monooxygenase A	$NH_4^+ + O_2 + H^+ + 2e^- \rightarrow NH_2OH + H_2O$	[15, 18, 27, 36, 39, 47, 48, 50, 51, 52, 53, 54, 55]
			hao	Hydroxylamine oxidoreductase	$NH_2OH \rightarrow NO + 3H^+ + 3e^-$	
2	*Nitrospira spp.*	Nitrite Oxidizing Bacteria	amtB	Ammonium transporter	$NH_3 \rightarrow NH_4^+$	[34, 36, 46, 48, 49, 50, 51, 52, 53, 54, 55]
			glnA	Glutamine synthetase	Glutamate + ATP + NH_3 → Glutamine + ADP + phosphate	
			nirA	Cytoplasmic ferredoxin-dependent nitrite reductase	$NO_2^- \rightarrow NO$	
			nrfAH	Periplasmic cytochrome c nitrite reductase	$NO_2^- \rightarrow NO$	
			nxrABC	Nitrite oxidoreductase α, β & γ subunit	$NO_2^- + H_2O \rightarrow NO_3^- + 2H^+ + 2e^-$	
			ONR	Octaheme cytochrome c nitrite reductase	$NO_2^- + 6e^- + 8H^+ \rightarrow NH_4^+ + 2H_2O$	
3	*Nitrospira sp.* ND1	Nitrite Oxidizing Bacteria	fdhABC	Formate dehydrogenase	$CH_2O_2 \rightarrow CO_2^+ + H^+ + 2e^-$	[36, 51]
6	Candiatus *Scalindua spp.,*	Anaerobic ammonia	hzh	Hydrazine hydrolase	$NO + H_2O + NH4^+ + O_2 + H^+ + 2e^- \rightarrow NH_2 OH + H_2O$	[36, 56]

	Candidatus *Jettenia asiatica*	oxidizing bacteria				
			hzs	Hydrazine synthase	$NH_4 + NO + 2H^+ + 3e^- \rightarrow N_2H_4 + 2H_2O$	
8	Candidatus *Kuenenia stuttgartiensis,*	Anaerobic ammonia oxidizing bacteria	hzo	Hydrazine oxidoreductase	$NH_2OH + H_2O \rightarrow NO_2^- + 5H^+ + 4e^-$	[36, 56]
9	*Nitrospira inopinnata*	Anaerobic ammonia oxidizing bacteria	narG	Nitrate reductase	$NO_3^- + 2H^+ + 2e^- \rightarrow NO_2^- + H_2O$	36, 51, 52
			hzo	Hydrazine oxidoreductase	$NH_2OH + H_2O \rightarrow NO_2^- + 5H^+ + 4e^-$	
			nosZ	Nitrous oxide reductase	$N_2O + H_2O \rightarrow N_2 + H_2O$	
			pmoA	Methane monooxygenase	$CH_4 + O_2 + NAD(P)H + H^+ \rightarrow CH_3\,OH + NAD(P)^+ + H_2O$	
11	Candidatus *Anammoxoglobus spp., Nitrospira marina* 295	Ammonia Oxidizing bacteria	nirK	Copper-dependent NO-forming nitrite reductase	$NO_2^- \rightarrow NO$	36, 37, 48
			amoA	Ammonia monooxygenase A	$NH_4^+ + O_2 + H^+ + 2e^- \rightarrow NH_2OH + H_2O$	
			hao	Hydroxylamine oxidoreductase	$NH_2OH \rightarrow NO + 3H^+ + 3e^-$	
12	*Nitrosomonas europaea*	Anaerobic ammonia oxidizing bacteria	norB	Nitric oxide reductase	$NO_2^- + 5H^+ + 4e^- \rightarrow NO_3^- + 2H^+ + 2e^-$	[36, 46]

Applications of nitrifying bacteria resolving ammoniacal pollution in various industries and treatment facilities.

Applications of Ammonia Oxidizing Bacteria

Waste-water treatment

The broad substrate specificity exhibited by AOB, coupled with their capacity to break down pharmaceutical molecules, can significantly enhance the removal of pharmaceuticals. The degradation process strongly correlates with the measured nitrification rate in the context of waste-water treatment. The presence of pharmaceutical residues in aquatic ecosystems and their possible negative impacts on the environment and human health are of concern. Waste-water treatment is recognized as an essential path for introducing pharmaceuticals into the environment. The enhanced removal of pharmaceutical chemicals through AOB has been extensively observed in diverse waste-water treatment methods [55]. In recent decades, the emergence of micropollutants in the aquatic environment has become a significant concern. Micropollutants encompass a range of substances, such as medications, personal care products, hormones, detergents, and disinfectants. These substances have the potential to cause risks to the ecosystem [57]. The occurrence of pharmaceutical compounds has been extensively documented in wastewater, surface water, and groundwater, exhibiting varied concentrations ranging from a few nanograms per liter to several hundred micrograms per liter [58,59,60]. The enzyme ammonia monooxygenase (AMO), which lacks specificity, has demonstrated the ability to convert aliphatic and aromatic chemicals [61,62].

Biodegradation of micropollutants

The study revealed that the degradation of pharmaceuticals by AOB follows a metabolic pathway. Ammonia is the primary substrate and energy source for developing microorganisms and creating enzymes in this pathway [63]. AOB are paramount in the nitrification process as they facilitate the conversion of NH4+ to NO2−. The mentioned transformation holds great importance as it facilitates the breakdown of various aromatic compounds via metabolic biodegradation. The capability in issue is assumed to be attributed to the non-specific enzyme AMO found in AOB [64,65]. AOB has demonstrated efficient degradation capabilities towards hydrocarbons, phenol, and other aromatic chemicals, as reported [61,65].

Application of Ammonia oxidizing Archaea

Waste-water treatment

The use of AOA within activated sludge processes can yield significant knowledge regarding the distinct contributions made by AOA in the context of nitrogen removal within waste-water treatment plants (WWTPs). Using oxidizers and tetraether lipids as diagnostic indicators for *Crenarchaeota* is a common practice in scientific investigations, as illustrated by [4]. AOA have the potential for carrying out both nitrification and denitrification processes in environments with restricted aeration and low levels of dissolved oxygen [66]. These environments are often typified by alternating aerobic and anaerobic conditions. AOA exhibits a significant potential for ammonia oxidation and is abundant in different wastewater treatment systems. As reported, AOA was initially identified in the activated sludge of 9 WWTPs in the United States [1,67].

Biodegradation of micropollutants

AOA exhibited a much higher efficiency than AOB in the biotransformation of mianserin and ranitidine. Additionally, AOA has shown the capability to degrade lincomycin by hydroxylation, S-oxidation, and demethylation processes. *Nitrosospha eragargensis*an has demonstrated the ability to biotransform various micropollutants, including tertiary amines, mainserin, and ranitidine. Notably, the biotransformation rates of these micropollutants by *Nitrososphaera gargensis* during ammonia oxidation surpass those observed in AOB under equivalent conditions [63,68].

Application of Nitrite Oxidizing Bacteria

Biosensor and water treatment

Early detection of nitrite, with consideration for both timeliness and sensitivity, holds substantial importance in securing human health and addressing the issue of water pollution. Nevertheless, it is worth noting that a significant limitation of several biosensors lies in their ability to assess the overall toxicity of water solely, hence lacking specificity in detecting pollutants. Consequently, there are few electroactive biofilm sensors explicitly designed to identify pollutants. The utilization of biofilms generated by bacteria exhibiting specialized functionalities can enhance the selectivity of nitrite detection through the utilization of biosensors. The biosensor's selectivity in detecting nitrite was demonstrated by identifying NOB and nitrite oxidase enzymes within the electrode biofilm. The biosensor demonstrated satisfactory performance in wetland and river environments, with a relative standard deviation of less than 14.8% in detecting nitrite at low concentrations. Additionally, the biosensor provided insights into the existence of nitrification. During catalytic nitrification, NOB facilitates nitrite oxidation to nitrate via the enzyme nitrite oxidoreductase. This enzymatic reaction exhibits a greater degree of specificity towards detecting nitrite. In microbial ecology, it has been observed that NOB serves as a natural host for nitrite oxidoreductase and can undergo self-regeneration [31].NOB are crucial for facilitating the biological nitrogen removal in contemporary WWTPs facilities [13]. *Nitrotoga* is the primary bacteria accountable for nitrite oxidation in extensive WWTPs. The bacterium *Candidatus Nitrotoga* was identified in wastewater treatment plants operating within a temperature range of 7 to 16 degrees Celsius, as documented by [17]. *Candidatus Nitrotoga* potentially has a distinct temperature preference, emphasizing the significance of operating temperature when choosing NOB communities for wastewater treatment procedures [35].

Application of anammox

Roles of anammox in water treatment

Anammox has a significant interest as a feasible alternative for nitrogen removal by utilizing the organic content in urban waste-water. Compared to conventional nitrification-denitrification processes, the anammox method offers significant potential for sustainable waste-water treatment. This is primarily owing to its reduced demand for dissolved oxygen, lower requirement for organic carbon, and decreased creation of extra sludge and greenhouse gas emissions. The anammox process exhibits broad applicability across many technological systems, encompassing both single- and two-stage configurations, which may be implemented in side-stream and mainstream waste-water treatment [69]. For example, step-feed anoxic/oxic (A/O) process through integrated partial nitrification-denitrification/anammox (PN-PD/A) processes was applied for the removal of N from municipal wastewater having low C/N, which resulted in N removal with 85.6 % efficacy at C/N of 2.8 [69]. Advanced techniques, such as the combination of denitrifying anaerobic methane oxidation (DAMO) with anaerobic ammonium oxidation (Anammox), as well as the utilization of biofilm technology, have been devised to eliminate pollutants throughout the process of wastewater treatment effectively [39]. In their study, suggested that the combined approach of DAMO-Anammox can convert ammonium, nitrite, nitrate, and methane into harmless nitrogen and carbon dioxide. This process has a fascinating energy-positive capability [34].

Biodegradation of micropollutants

The mitigation of micropollutants is a significant concern in the context of water reuse. The study reports a significant removal of over 80% of some pharmaceutical and personal care products (PPCPs) using the primary removal mechanism of biodegradation in a single-stage anammox process. The substances primarily encompass estradiol, estrone, ethinylestradiol, ibuprofen, naproxen, bisphenol A, and celestite. For the removal of PPCPs, the 'ELiminación Autótrofa de Nitrógeno' (ELAN®) rector was operated [70].

Municipal solid waste leachate treatments

The anammox method has attracted considerable attention for treating leachate from municipal solid waste (MSW) due to its advantageous characteristics, such as reduced oxygen consumption, carbon source requirements, and sludge production. In the realm of MSW leachate treatment, the anammox technique demonstrates versatility due to its potential for integration with partial nitrification, denitrification, fermentation, and methane oxidation processes. The application of Anammox offers a promising biotechnological strategy for efficiently removing nitrogen from leachate derived from MSW [71]. In a recent study, a two-stage PN-A system was operated in the Zentraldeponie Emscherbruch landfill leachate treatment plant (Herten, Germany) to treat the landfill leachate, which resulted in the removal of ammonia and COD by 93.7% and 92.1%, respectively [72]. Another study revealed that 70.0–80.0% and 60.0 ± 5.0% of ammonia and COD were efficiently removed from landfill leachate through the one-stage PNA process in the New Taipei landfill leachate treatment plant [73].

Application of COMAMMOX

Role of Comammox in water treatment

In specific waste-water treatment facilities, there was a significant increase in activity abundance, reaching up to 24-fold higher values. The potential involvement of comammox bacteria in the waste-water treatment is significant. The copolymerization process can be enhanced by comammox and anammox bacteria in a hypoxic environment [17,74,75].

Biodegradation of micropollutants

Comammox bacteria have demonstrated the ability to efficiently degrade specific micropollutants, presenting potential opportunities for using

Comammox in WTPs and WWTPs. As an example, the research revealed that *Nitrospira inopinata* exhibits a significant proficiency in the degradation of several compounds, such as asulam, fenhexamid, mianserin, fantidine, and carbendazim, in the presence of ammonia [76]. The strong indication of Comammox bacteria's potential to degrade a wide array of organic nitrogen compounds, apart from free NH_3, is supported by active genes involving urea, amines, and cyanates. Nitrifying bacteria are known to have a significant impact on the degradation of a range of micropollutants, including ibuprofen, naproxen, mianserin, ranitidine, sulfamethoxazole, erythromycin, roxithromycin, fluoxetine, and trichloroethane, in both WTPs and WWTPs [77].

Biotransformation of micropollutants

Comammox bacteria exhibit a significant propensity to participate in co-metabolic biotransformation reactions while simultaneously oxidizing NH3, leading to the conversion of diverse micropollutants. This encompasses the transformation of several substances, such as artificial sweeteners, including acesulfame, aspartame, cyclamate, saccharin, sucralose, sulfamethoxazole and 2-chlorophenol [76]. The study examined the biotransformation of artificial sweeteners, such as acesulfame, aspartame, cyclamate, saccharin, and sucralose, by nitrifying activated sludge and found that all the compounds were degraded. Additionally, it was also observed that the metabolic biodegradation of target artificial sweeteners was directly proportional to the nitrification rate, resulting in a linear relationship between nitrification rate and co-metabolic biodegradation rate. Hence, for the biodegradation of artificial sweeteners, ammonia oxidizers played a significant role [76].

Conclusions

The advent of extensive agriculture has posed ammoniacal pollution as a global nuisance, which has adverse implications for human health, an imbalanced global nitrogen cycle, and increased greenhouse gas emissions. There is a significant role nitrifying bacteria in regulating the nitrogen cycle and reducing greenhouse gas emissions by converting the excess ammonia into nitrites and nitrates, which are readily utilized by plants, preventing nitrogen loss as ammoniacal emissions. Furthermore, this chapter has discussed the different types of nitrifying bacteria and their industrial applications in waste-water treatment, biosensors, and the degradation and biotransformation of micropollutants. Studying these groups of bacteria further would be beneficial to understand their role in ameliorating global ammoniacal pollution.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Author(s) contributions

PS: Conceptualization, investigation, writing original draft and compiling the manuscript. KKP: investigation, writing original draft and compiling the manuscript. AL: Conceptualization, figure illustration, writing-original draft and compiling the manuscript. SS: Data collection, investigation, and writing-original draft. NM: Data collection, investigation, and writing-original draft. SKS: Data collection, investigation, and writing-original draft. RP: Data collection and writing-original draft. RK: Conceptualization, visualization, funding acquisition, finalizing the manuscript.

Acknowledgements

AL is thankful to UGC, Govt. of India for the 'Research Fellowship' Grant (NTA Ref. No.: 200510046070/ UGC-NET). SKS is thankful to the Department of Science and Technology for the 'Research Fellowship' (DST/INSPIRE/03/2022/001242/IF210305). The authors acknowledge the financial support from the NMHS project of MoEF&CC (sanction no. NMHS2022-23/MG11/01/282), DST-TDT project no.DST/TDT/WM/2019/43 and Council of Scientific and Industrial Research (CSIR) Fast Track Commercialization and Fast Track Translational project (Fourth Tranche of E3OW) MLP-0182 and MLP0183 to work on the aspect of waste management. The manuscript represents CSIR-IHBT communication no 5475.

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