



NITROGEN LOSSES DUE TO NITRIFICATION: PLANT BASED REMEDIAL PROSPECTS

Kavita Arora and Alka Srivastava*

In Vitro Culture and Plant Genetics Unit, Department of Botany, University of Lucknow-226007, (U.P.) India

Received for publication: May 29, 2013; Accepted: June 06, 2013

Abstract: Nitrification of nitrogen fertilizers results into loss of nitrogen nutrient from soil along with environmental hazards, like, global warming, contamination of ground water and so on. To tackle the problem, many synthetic inhibitors of nitrification have been formulated but till now none of them has proved to be attractive option due to their cost and non-ecofriendly nature. A shift towards natural nitrification inhibitors would be beneficial, economically viable and environmentally safer option. Also, it is a matter of exhaustive research and appropriate testing before one can come out with infallible system with holistic integrated approach. To increase the potential of natural nitrification inhibitors, some neglected aspects, like, effect of genotypic variations, broader plant exploration for nitrification inhibition capacity, identification and isolation of secondary metabolites which are responsible for nitrification inhibition activity, manipulation of germplasm for superior nitrification inhibition performance require incessant persistent scientific input for transfer of information to the farmers.

Keyword: Environmental Hazard, Natural Products, Nitrification Inhibitors, Nitrogen Fertilizers, Nutrient-Loss, Nitrogen Transformation

INTRODUCTION

Nitrogen (N) a major nutrient required by plants in large amount as it serves as a constituent of many plant cell components, such as amino acids and nucleic acids. Though N is present in large quantity (78% by volume) in earth's atmosphere, still it is a key yield-limiting nutrient in crop production. Gaseous form of N cannot be utilized by majority of the organisms except cyanobacteria and legumes, which have natural ability to fix atmospheric nitrogen. Nitrogen pool in the soil is sourced from atmospheric N fixation by legumes, animal manures, soil organic matter (SOM) and N-fertilizers. Most of the plants, other than legumes, like, cereals rely on this fixed nitrogen. The modern cropping systems use external supplement of N to meet the crop nitrogen needs as N of the SOM is not sufficient for good yields^[1]. Exponential increase in population growth and scarcity of good agricultural land are the major motivating forces to increase crop production and therefore, leads towards more fertilizer consumption especially in developing countries. In India fertilizer consumption has increased to 17 million tonnes today, urea accounts for 82% of total nitrogen consumption^[2]. Nitrogen being the most essential nutrient for plant growth holds the biggest share among other nutrients/ fertilizers. Use of nitrogenous fertilizers has arisen a challenge of simultaneously improving crop productivity and nitrogen use efficiency. Indiscriminate consumption of nitrogenous fertilizers has raised certain issues pertaining to environment and energy consumption. The basic raw material for the production of nitrogenous fertilizers is NH_3 and production of NH_3 is energy and resource intensive. It accounts for 80% of the energy

consumption for nitrogenous fertilizer. Also, rapid loss of these fertilizers from soil due to urease activity and nitrification is a serious problem. To reduce the loss of fertilizer N from soil due to urease activity and nitrification, there are many synthetic compounds/ plant products known as urease inhibitors and nitrification inhibitors respectively. Synthetic compounds as nitrification inhibitor and urease inhibitor have limited success and their use is restricted more in European countries/ developed countries. In developing countries they are not so popular, due to their cost and availability. In view of present scenario, our approach should be to minimize the use of chemicals in agriculture as far as possible, as they not only are costly but lead to environment and health hazards in long run. Safer, sustainable and cheaper option is to incorporate/ focus on plant based/ natural resources.

Realizing the relevance of inhibition of nitrification for greater availability of N to the crop plants this review has been compiled to draw the attention of larger number of scientists towards this important global issue.

Plant-based remedial prospects for nitrification inhibition:

Availability of nitrogen to the crop plants through inhibition of nitrification is the current agricultural demand for minimizing the injudicious use of chemical fertilizers, which may prove toxic to the crop plants. An evaluation of the N transformation processes in the soil, the available forms of nitrogen, the reasons for its

*Corresponding Author:

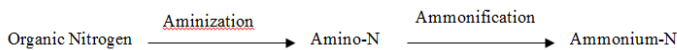
Dr. Alka Srivastava,
In Vitro Culture and Plant Genetics Unit,
Department of Botany,
University of Lucknow-226007, (U.P.) India.



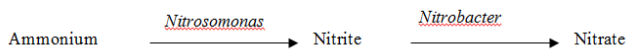
loss from the soil and the consequent environmental hazards are the matters of concern for maintaining optimum nitrogen in the soil, and all these aspects have been discussed herewith.

Nitrogen transformation processes. Nitrogen in soil exists in inorganic form such as ammonia (NH₃), ammonium (NH₄⁺), nitrate (NO₃⁻), nitrogen oxides (NO_x) and organic forms like amines, urea, proteins and nucleic acids. All forms of nitrogen are in constant state of flux and undergoing transformations. Transformation processes are:

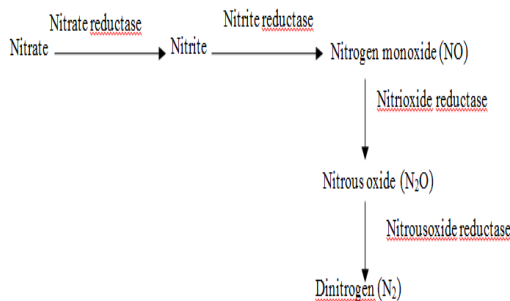
- **Mineralization:** The microbial transformation of organic nitrogen to inorganic forms is referred to as mineralization.



- **Nitrification:** Biological oxidation of ammonia to nitrate via nitrite.



- **Immobilization:** Uptake of NO₃⁻, NH₄⁺ from soil solution by soil microorganisms for metabolic maintenance, growth and reproduction, make it unavailable for other microorganisms and plants.
- **Denitrification:** Bacterial conversion of nitrate to nitrite and molecular nitrogen.



- **Volatilization:** Loss of nitrogen to atmosphere as NH₃ gas is called as ammonia volatilization.

Available forms of N (NH₄⁺ and NO₃⁻) are provided to plants throughout growing season by mineralization and nitrification in soil^[3], whereas immobilization, denitrification and volatilization are responsible for N losses from soil making it unavailable to plant.

Available forms of N for plant uptake. Plants generally utilize inorganic form of nitrogen, either NH₄⁺ or NO₃⁻ as their N nutrient. The metabolic energy requirement of plants for assimilation of NO₃⁻ and NH₄⁺ varies, being 20 moles and 5 moles of ATP per mole of NO₃⁻ and NH₄⁺, respectively^[4]. Also during the

assimilation of NO₃⁻, nitrous oxide is released, which is a potent green house gas. Ammonium cations are bound electrostatically to the negatively charged clay particles and the functional groups of SOM in the soil. This binding prevents leaching and thus prevents N loss, on the other hand NO₃⁻ anion is mobile and easily leached out of the root-zone as it is not electrostatically bound, like, NH₄⁺ counterpart^[5]. Nitrogen nutrition in the form of NH₄⁺ is far better for plants which consequently show increased N use efficiency because of various merits associated with it (Table 1).

Table.1: Forms of nitrogen available to plants in soil.

| Parameters | Ammonium (NH ₄ ⁺) | Nitrate (NO ₃ ⁻) |
|-------------------|---|---|
| Mobility | Less mobile, being cation held by electrostatic forces to negatively charged clay surfaces and functional groups of soil organic matter. | Highly mobile |
| Assimilation | Less metabolic energy needed to assimilate (5moles of ATP/ mole of NH ₄ ⁺) | More metabolic energy needed to assimilate (20 moles of ATP/ mole of NO ₃ ⁻) |
| Loss from soil | Loss from NH ₄ ⁺ based or NH ₄ ⁺ forming fertilizers as NH ₃ gas | Denitrification losses from NO ₃ ⁻ fertilizers or organic N fertilizers such as animal waste are generally much higher also lost via leaching |
| Gaseous emissions | Ammonia volatilization results into emission of NH ₃ into atmosphere | Nitrate assimilation by plants and denitrification causes release of nitrous oxide a potent greenhouse gas |
| Adverse effects | Eutrophication and acidification of natural ecosystems occur due to deposition of lost NH ₃ on land or in water ^[6] | Air pollution, contribute to global warming, eutrophication and pollute groundwater |

Modern cropping system leading towards losses:

To fulfill the need of mankind for food and fiber with a limited scope for horizontal geographical expansion in net cultivable land, modern agricultural systems encourage the use of N fertilizers (industrially produced inorganic nitrogen) over legumes and/or animals for N input^[7]. Adoption of modern agricultural practices receding the usual practices of crop rotation, separating the animal husbandry from agriculture, involving more irrigation and drainage of agricultural fields and practicing the use of N fertilizers. Larger inputs of N for high yield increase the chances of its losses from the field. These not only waste the resources but also cause an almost irreversible damage to environment. In the current scenario, emphasis is on increasing food production at any cost; consequently poorly educated farmers use N fertilizers in their fields, bear its cost (not economically viable) and still are not benefited by the practice. There is need to understand the N nutrient requirements of crop and it should be synchronized with the fertilizer input. The usual practice is more fertilizer more yield; therefore, N

fertilizers are applied irrespective of soil conditions, climate, etc. But the fact is about 70% of fertilizer applied in the field is lost due to nitrification and associated N losses. Since excessive and indiscriminate use of N fertilizers contaminates the environment, it has now become a global problem and a serious matter of concern.

Modes of N losses from soil:

Most of the N fertilizer applied (mostly NH_4^+ form) to the field or soil organic N is lost through nitrification at rapid rates within days or weeks. Three processes simultaneously operating in soil are responsible to a great extent for N losses in different forms. Figure 1 is an illustration showing different modes of N losses and how they are linked to each other.

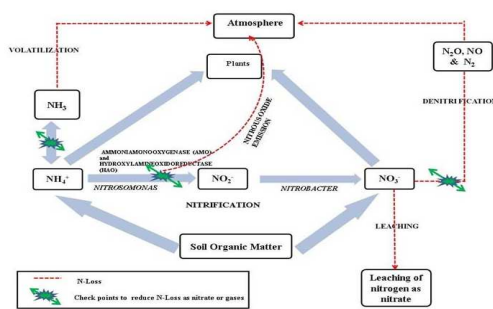


Fig. 1. Different modes of N-losses from soil

In Color Online Only

Leaching: Nitrate formed as a result of nitrification as mentioned above is highly mobile and prone to leaching, consequently not available to the plant. Since nitrification is a biological process and carried out by microbes *Nitrosomonas* sp. and *Nitrobacter* sp., therefore, rate of nitrification depends on soil factors/properties in which bacteria present; temperature, pH, organic matter, moisture, aeration and availability of NH_4^+ in soil. Conducive conditions for nitrification are a good supply of organic matter with gradual release of NH_3 , since nitrification is an aerobic process, therefore, good soil aeration is required, adequate available soil moisture nitrification rates are maximum when soil moisture is at its field capacity, a temperature between 20° and 30°C , and pH between 6.0 and 9.0. Nitrate formed leach through the soil to groundwater along with drainage or irrigation water. Concentration of NO_3^- increases in the groundwater and becomes unfit for drinking. Also, excess of nitrates lead to eutrophication.

Ammonia volatilization: Contributing factors for NH_3 volatilization are concentration of NH_4^+ , soil temperature and pH that affect the partial pressure of NH_3 in soil^[8]. Soil pH is responsible for maintaining equilibrium between NH_3 and NH_4^+ forms, as the pH changes from 6 to 7, 8 and 9, the relative concentration of NH_3 increases from 0.1 to 1, 10 and 50%,

respectively^[9], (Fig.2). Alkaline pH favours the formation of NH_3 , consequently its volatilization. Some other factors affecting NH_3 volatilization are soil texture, moisture content of the soil, pH buffering capacity, cation exchange capacity, nitrification rate and presence of plant and plant residues^[10].

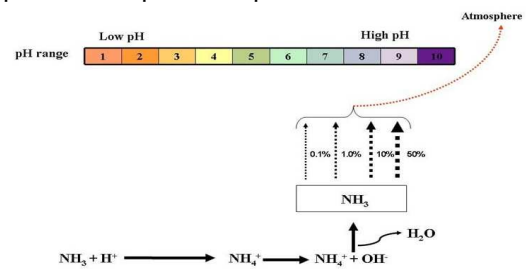


Fig. 2. Schematic diagram of equilibrium between ammonia and ammonium forms in soil with respect to pH (as per Freney et al., 1983).

In Color Online Only

Denitrification: Denitrification is the process of conversion of NO_3^- into gaseous forms such as N_2O , NO , N_2 by heterotrophic soil bacteria *Bacillus subtilis*, *Escherichia coli*, *Aspergillus flavus*, etc. Conditions that favour and support rapid microbial growth and activity viz., moist wet soils^[11] and warm temperature^[12] are responsible for denitrification. Denitrification takes place when heterotrophic bacteria under limited oxygen supply use NO_3^- as alternative electron acceptor to produce N_2O and N_2 . The N_2 : N_2O ratio increases with decreasing O_2 availability, and it decreases under high NO_3^- availability. Other factors that affect denitrification are NO_3^- concentration, pH, temperature and organic carbon^[13]. In paddy fields where alternate wetting and drying is must in its production system denitrification process is chiefly responsible for the losses and greatly affect the yield.

Environmental hazards:

Nitrification and other transformation processes not only ultimately result into N losses but also significantly contribute to environmental hazards. Excessive nitrification, denitrification and ammonia volatilization one way or other adversely affect land, water and air, viz., acidification of soil due to increased levels of NO_3^- , eutrophication of surface waters, contamination of groundwater with excessive NO_3^- and gaseous emission through denitrification result in their accumulation above permissible levels. Nitrates leached in the soil reach groundwater where it gets accumulated over the permissible limit and causes groundwater contamination. Drinking of such contaminated water causes methemoglobinemia especially in infants (blue baby syndrome). Eutrophication means nutrient enrichment, this is a process of nutrient enrichment of water bodies, which leads to rapid growth of phytoplanktons, algae and macrophytes followed by accumulation of more organic matter. Excessive input of nitrates in water

bodies through leaching results into eutrophication problems of surface waters. Two main sources of greenhouse gas emissions to the atmosphere produced as a result of soil biological processes are agricultural and forestry activities^[14]. During nitrification (oxidation of ammonium to nitrite) there is emission of nitrous oxide, which is a potent greenhouse gas and has about 300 times more global warming potential than carbon dioxide. Further, denitrification leads to emission of nitrous oxide, other oxides of nitrogen. Significant decrease (10%) in the ozone layer is predicted if the concentration of nitrous oxide in atmosphere doubles and consequently, the ultraviolet radiations reaching the surface would increase by 20%^[15]. Nitrogen monoxide being highly reactive destroys the ozone layer through an autofeedback catalytic cycle where it combines with ozone and forms NO₂ which in turn reacts with oxygen and form NO again. Further, NO can cause acid rain (by getting converted into nitric acid), which results in acidification and eutrophication of ecosystems^[16]. Production of fertilizers involves consumption of conventional energy sources. More N losses results into more incorporation of fertilizers. The use of N fertilizers appears to give short-term gains at the cost of our environment.

Approaches to reduce N losses:

Various strategies are followed to manage N loss from the field, some of them are; synchronization of fertilizer application with crop N requirements to facilitate rapid uptake, use of effective N fertilizer application methods (split or basal applications, banding of N fertilizers or broadcasting, placement of N fertilizer deep or on surface, foliar spray as and when required in case of urea). All above-mentioned approaches do help in minimizing losses of N from soil but constraints associated with them are additional labour costs and some practical difficulties, such as use of heavy machinery, availability of labour, etc.^[7]. Also, fertilizers are modified to slow down the release of nutrient so that it remain available for longer period for plant uptake (slow-released and controlled-release fertilizers, coatings) or fertilizers are supplemented with certain chemical compounds synthetic or natural which retard the nitrification process or urea hydrolysis.

Use of slow-release and controlled-release (SCR) fertilizers:

Slow- and controlled release fertilizers for eg., urea-formaldehyde based fertilizer, sulfur-coated urea and polymer-coated/encapsulated products ensures the availability of nutrients to plants for longer duration as compared to conventional fertilizers, as they are designed to slow down the nutrient availability^[17]. The chemical and physical properties of such fertilizers are so managed that N is slowly

released for uptake by plants, and is thus available for longer duration^[18,16]. Besides this, conventional fertilizers can be converted into SCR fertilizers by encapsulating them with water-insoluble, semi-permeable or impermeable with pores which regulate water entry and rate of dissolution and nutrient release^[19]. Higher N recovery in mint-wheat-rice cropping system has been reported when urea coated with mint oil by-products are used^[20]. Coating with urease inhibitor can improve the bioavailability of N, resulting in increased dry matter yield and N uptake^[21]. However, use of these fertilizers is beset with certain problems such as, high cost compared to conventional fertilizers. Polymer coated urea is 4-8 times expensive than normal urea, therefore their use in agriculture is limited^[22]. Besides economic viability, there are other limitations which restrict the use of these fertilizers, like, requirement of special handling skills, storage facilities and lack of research-based recommendations on their effectiveness and management under different environment and cropping systems^[23].

Application of nitrification inhibitors and/or urease inhibitors:

Nitrification inhibitors and urease inhibitors are variously known as magic bullets or tools. Nitrification inhibitors are the chemicals that slow down, delay or restrict the nitrification process by retarding the metabolism of soil nitrifiers, so that fertilizer N can be utilized by the plants before it is lost from the soil as nitrate^[23]. There are number of compounds known as nitrification inhibitors, many of them are patented also. Some examples of synthetic nitrification inhibitors are 2-chloro-6 (trichloromethyl) pyridine (nitrapyrin or N-serve), DMPP (3, 4-dimethylpyrazole phosphate), Dicyandiamide (DCD), sulfathiazole, 2-amino-4-chloro-6-methyl pyrimidine, 2-mercaptobenzothiazole, thiourea, etc. but only a few of them have been thoroughly evaluated under field conditions eg. Nitrapyrin, DCD and DMPP^[24, 25, 26, 27, 16]. But use of synthetic nitrification inhibitor is not successful to much extent. They are expensive for broadscale application, effects are not long-term, availability is a constraint, unstable, as nitrapyrin hydrolyzed into 6-chloropicnic acid and lost by volatilization^[28, 29]. The ill-effects of nitrapyrin on plants was reported as leaf chlorosis in cow peas, and interveinal chlorosis in chick peas^[30]. Reports suggests that as soil temperature increases half-life of DCD decreases^[31]. Moreover there is rarely any report on the long-term impacts of synthetic nitrification inhibitors on non-target flora and fauna, microbial diversity, soil microbial community composition.

Urease inhibitors are the chemical compounds that reduce the rate at which urea hydrolyzed and converted to NH₄⁺ by inhibiting the activity of urease enzyme responsible for the hydrolysis of urea^[17]. Volatile losses of NH₃ which occur primarily at the soil

surface can be reduced by reducing the rate of urea hydrolysis. Examples are hydroxyurea, hydroquinone, thiourea, nBTPT (N-(n-butyl) thiophosphoric triamide; trade name Agrotain), PPD (phenylphosphorodiamidate). A number of chemical compounds have been tested as urease inhibitor but their toxic effects were poorly documented [32,33,34]. Also, like synthetic nitrification inhibitors, synthetic urease inhibitors are also not cost-effective for large-scale applications and also do not remain stable eg. PPD is effective but known to decompose rapidly [33]. We need to make sure that unsystematic continued use of these chemicals will have no adverse effects on soil and water quality and human and animal health in long-term. High costs of synthetic compounds as nitrification inhibitors/ urease inhibitors and urge to minimize the use of chemicals in agriculture system has prompted scientists to find out /identify the alternative options. Numerous earlier investigations pointed out that some of the substances produced by higher plants are antagonistic to soil bacteria. In the same direction work was oriented towards the substances /inhibitors for nitrifying bacteria. These findings have become landmarks for today's research in the area of nitrification/ urease inhibitors. Initial research was solely based on natural resources for nitrification inhibitors, i.e., exploration of plant resources having the potentiality to inhibit nitrification.

Earlier workers hypothesized that grasses excrete bacteriostatic substances [35,36]. Stiven [37] has given an experimental proof that roots of *Trachypogon plumorus* a climax grass sp. on the Transvaal Highveld contain a water-soluble substance which is bacteriostatic for some bacteria. Roux [38], showed nitrogen sensitivity of some grasses, whereas Rice [39] focused on to seed plants for their inhibitory activity on nitrifiers and Munro [40] moved forward used grass root extracts and analyzed its effect on nitrifiers. Since then many plants have been listed as having potential for nitrification inhibition activity, such as *Pongamia glabra* [41], *Azadirachta indica* [41,42], *Mentha spicata* [43] and *Artemisia annua* [44,43], etc., (Table 2). The work of Patra et al. [59] centered their study on assessing the activity of essential oils present in plants. Similarly, the litter of trees like, Mahogany, Teak, Jackfruit, Mango, and Breadfruit has potential for inhibiting nitrification process due to their tannin content, which has been shown to increase nitrogen fertilizer efficiency for soybean [60]. In this direction, the natural potential of plants is exploited to inhibit nitrification or urease activity by some workers. Merits of natural nitrification inhibitors/urease inhibitors are presented in table 3.

Table.2: Plant parts/products as natural nitrification inhibitors.

| Plant | Common name | Plant part/ products | References |
|--|----------------------------------|---|------------------|
| <i>Abies balsamea</i> | Balsam fir | Foliar leachates, leaf extracts, bud extracts | [45] |
| <i>Anacardium occidentale</i> | Cashew | Cashew shell powder | [46] |
| <i>Artemisia annua</i> Linn. | Sweet Wormwood | Leaves | [44] |
| <i>Azadirachta indica</i> | Neem | Seed kernel powder, seed cake, leaves, seed oil | [47, 48, 49, 42] |
| <i>Bassia latifolia</i> or <i>Madhuca indica</i> | Mahua | Seed cake, seed oil | [50, 48, 51] |
| <i>Camellia sinensis</i> | Tea | Tea fluff | [52] |
| <i>Chrysanthemum cinerariaefolium</i> | Pyrethrum | Flower dust | [53] |
| <i>Citrullus colocynthis</i> (Linn.) Schrad. | Bitter apple | Seed cake | [54] |
| <i>Curcuma longa</i> | Turmeric | Turmeric Powder | [46] |
| <i>Jatropha curcus</i> | Ratanjot/ Physic-nut/ Purgingnut | Seed oil | [48] |
| <i>Linum usitatissimum</i> | Linseed/Flax | Seed oil | [55] |
| <i>Liriodendron tulipifera</i> | Tulip tree | Leaf leachates | [56] |
| <i>Mentha arvensis</i> L. | Mint | Distillation waste (mentha-spent) | [53] |
| <i>Mentha spicata</i> Linn. | Spearmint | Dried plant material (Spearmint oil) | [43] |
| <i>Pongamia pinnata</i> | Karanja | Seed, bark, leaves | [57, 58] |
| <i>Populus balsamifera</i> | Balsam poplar | Foliar leachates, leaf extracts, bud extracts | [45] |
| <i>Quercus rubra</i> | Red Oak | Leaf leachates | [56] |
| <i>Ricinus communis</i> | Castor | Seed oil | [48] |

Table.3: Natural vs Synthetic nitrification inhibitors/ urease inhibitors.

| Parameters | Natural | Synthetic |
|------------------------|--------------------------|---|
| Cost | Cost-effective | Expensive |
| Availability | No problem | Not frequently available |
| Effect | Long-term | Short-term/ less effective |
| Performance | Consistent | Non-consistent |
| Stability | Stable | Unstable |
| Side-effects/Biosafety | None | Thorough research required |
| Regulatory control | Not of much significance | No regulatory controls on its use in agriculture |
| Government input | Lack of Government push | Lack of Government push and absence of any usage legislation as yet |
| Benefit to cost ratio | High | Low |
| Ecological impact | Biodegradable | Largely non-biodegradable |

Scope for improvement:

Efficient natural nitrification inhibitors can be produced, if focus of research shifts towards the

manipulation of their active ingredients. Active principles present in various parts of plants are responsible for nitrification inhibitory properties, which vary from genotype to genotype especially in case of heterozygous plants/ trees. For example, in case of neem, active ingredients (meliacins) present in most of its parts are reported to be responsible for nitrification inhibition^[47,43], these active ingredients vary from plant to plant because of high heterozygosity of neem. To get the maximum benefit in such case, mass propagation of elite tree can be done by *in vitro* cloning methods^[61], which can yield progenies with almost same level of active ingredients. Pertaining to genotypic variation, extent of variation between different genotypes of *Oryza sativa* for nitrification inhibition activity in their root exudates has been reported^[62]. Also, genotypic differences in nitrification inhibition for sorghum root exudates and tissue extracts has earlier been seen^[63]. Genetic interventions for improving the nitrification inhibition capacity of plant products used as supplements appears possible and also shows promise of reducing nitrogen loss from the soil. Selection of better performing genotypes, testing of large number of local plants (plant exploration studies) and manipulation of the existing germplasm for better retention of N in the soil appears to be a must for improving the fertility of the soil. Genetic strategies, like, selection and intermating, hybridization between appropriate varieties, induced mutation and careful evaluation of the variability generated can lead to the production of varieties with high levels of nitrification inhibition activity. Identification of genes imparting nitrification inhibition ability and their judicious incorporation in otherwise elite germplasm can help in making superior natural supplements of fertilizers. Therefore, potential plant exploration should be followed by its genotypic selection and genetic improvement. Further, aspects to be focused upon are identification and enhancement of active principles responsible for nitrification inhibition activity for their commercial exploitation. The biological nitrification inhibition (BNI) ability of the roots of various plants seems to depend on the wide range of biomolecules, like, phenolics, alkaloids, etc. that are released in the rhizosphere^[64]. Hairy root cultures and callus cultures may be used to enhance the level of valuable biomolecules and further their up scaling can be done in bioreactors. These are some of the neglected aspects of plant based nitrification inhibitors which seek attention.

Future prospects:

An integrated approach is warranted to deal with the issue of nitrogen losses from soil. First and foremost need is to explore more and more plants with nitrification and /urease inhibition ability/ activity, as has been done in case of *Brachiaria humidicola*. This

should then be followed by their improvement at genetic level, biochemical level and finally working on an effective market product as an outcome. Since farmers have to bear the cost of everything right from seed to fertilizer, pesticides etc. therefore, best quality, cheaper product as nitrification and/ urease inhibitors which is at the same time eco-friendly are to be provided and recommended to the farmers. Best management practices are to be developed not only centralizing higher yields but also other related facts as nutrient use efficiency, environmental conditions and farmers' interest.

CONCLUSIONS

High N input from artificial source (N fertilizers), moving away from multiple cropping system in modern agricultural practice are chiefly responsible for N losses and environmental pollution. Our approach should be to increase the yields by using best management practices and improving nutrient use efficiency rather than through applications of higher fertilizer. Conventional (multiple cropping, crop rotation, use of animal manure etc.) and non-conventional approaches (use of N fertilizers and their modified forms, nitrification inhibitors and urease inhibitors) go hand in hand and minimize the indiscriminate use of N fertilizers as far as far possible, as their excessive use is not environment friendly and require lot of energy for their production. Natural/ plant-based nitrification inhibitors will go a long way as far as increase in yield, economic-viability and safety of environment is concerned and to achieve this we need to tap the diverse treasure of potential plants. The remedy not only lies in scientific solutions but along with that government push and implementation of regulatory measures are also required, so that whatever research is done at laboratory level should ultimately reach the farmer.

ACKNOWLEDGMENT

The authors thank the Council of Scientific and Industrial Research (CSIR), India, for financial assistance.

REFERENCES

1. Evenson RE, Gollin D, Assessing the impact of the green revolution, 1960 to 2000. *Science*, 2003, 300, 758-762.
2. Nautiyal CS, Biofertilizer and biopesticide for developing world: A boon for sustainable agriculture. *Environews Newslett*, 2011, 17, 1-2.
3. Huber DM, Wareen HL, Nelson DW, Tsai CY, Nitrification inhibitors-new tools for food production. *BioScience*, 1977, 27, 523-529.
4. Salsac L, Chaillou S, Morot-Gaudry J, Lesaint C, Nitrate and ammonium nutrition in plants. *Plant Physiol Biochem*, 1987, 25, 805-812.

5. Amberger A, Efficient management of nitrogen fertilization in modern cropping systems, Optimization of plant nutrition, (eds) Fragoso MAC, van Beusichem ML, Kluwer Academic Publishers, Dordrecht, 1993.
6. Sommer SG, Hutchings NJ, Ammonia emission from field applied manure and its reduction—invited paper. Eur J Agron, 2001, 15, 1-15.
7. Dinnes DL, Karlen DL, Jaynes DB, Kaspar TC, Hatfield JL, Colvin TS, et al. Nitrogen management strategies to reduce nitrate leaching in tile-drained Midwestern soils. Agronomy Journal, 2002, 94, 153-171.
8. Singh J, Bolan NS, Saggar S, Zaman M, The role of inhibitors in controlling the bioavailability and losses of nitrogen, Chemicals bioavailability in terrestrial environment (eds.) Naidu R, Bolan NS, Megharaj M, Juhasz A, Gupta S, Clothier B, et al., Elsevier, Amsterdam, The Netherlands, 2008, p. 329-362.
9. Freney JR, Simpson JR, Denmead, OT, Volatilization of ammonia, Gaseous loss of nitrogen from plant-soil systems, (eds.) Freney JR, Simpson JR, M. Martinus Nijhoff/Dr. W. Junk Publishers, The Hague, 1983, p. 1-32.
10. Freney JR, Black, AS, Importance of ammonia volatilization as a loss process, Advances in Nitrogen Cycling in Agricultural Ecosystems, (ed.) Wilson JR, C.A.B International, Wallingford, 1988, p. 156-173.
11. Linn, DM, Doran JW, Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and nontilled soils. Soil Sci Soc Am J, 1984, 48, 1267-1272.
12. Sahrawat KL, Keeney DR, Nitrous oxide emissions from soils. Adv Soil Sci, 1986, 4, 103-148.
13. Peoples MB, Freney JR, Mosier AR, Minimizing gaseous losses of nitrogen, Nitrogen fertilization in the environment, (ed.) Bacon PE, Marcel Dekker, Inc. New York, 1995, p. 565-602.
14. Muñoz C, Paulino L, Monreal C, Zagal E, Greenhouse gas (CO₂ and N₂O) emissions from soils: A review. Chilean J Agri Res, 2010, 70, 485-497.
15. Crutzen PJ, Ehhalt DH, Effects of nitrogen fertilizers and combustion on the stratospheric ozone layer. Ambio, 1977, 6, 112-116.
16. Subbarao GV, Ito O, Sahrawat KL, Berry WL, Nakahara K, Ishikawa T, et al., Scope and strategies for regulation of nitrification in agricultural systems—challenges and opportunities. Crit Rev Plant Sci, 2006, 25, 303-335.
17. Trenkel ME, Improving fertilizer use efficiency—controlled release and stabilized fertilizers in agriculture. International Fertilizer Industry Association (IFA), Paris, France, 1997.
18. Freney JR, Strategies to reduce gaseous emissions of nitrogen from irrigated agriculture. Nutr Cycling Agroecosyst, 1997, 48, 155-160.
19. Fujita T, Yamashita Y, Yoshida S, Yamahira K, Can. Pat. 1295849, Chisso Corp, Japan, 1992.
20. Kiran U, Patra DD, Augmenting the yield and urea-N utilization efficiency in wheat (*Triticum aestivum*) through use of natural essential oils and dicyandiamide coated urea in light textured soils of Central Indian sub-tropical conditions. Agric Ecosyst Environ, 2002, 50, 64-69.
21. Junejo N, Khanif MY, Hanfi MM, Wan Yunus WMZ, Dharejo KA, Role of inhibitors and biodegradable material in mitigation of nitrogen losses from fertilized lands. Afr J Biotechnol, 2011, 10, 3504-3514.
22. Detrick J, RLC membrane encapsulated fertilizer technology can deliver high yield value benefits for agriculture, Proceedings of Great Plains Soil Fertility Conference, (ed.) Havlin J, Denver, Colorado, 1996.
23. Motavalli PP, Goynne KW, Udawatta RP, The environmental impacts of enhanced efficiency nitrogen fertilizers. Crop Manage, 2008, Available at <http://www.plantmanagementnetwork.org/pub/cm/symposium/enhanced/impacts/>
24. Goring CAI, Control of nitrification of ammonium fertilizers and urea by 2-chloro-6-(trichloromethyl)-pyridine. Soil Sci, 1962, 93, 211-218.
25. Weiske A, Benckiser G, Herbert T, Ottow JCG, Influence of the nitrification inhibitor 3, 4-Dimethylpyrazole phosphate (DMPP) in comparison to dicyandiamide (DCD) on nitrous oxide emissions, carbon dioxide fluxes and methane oxidation during 3 years of repeated application in field experiments. Biol Fertil Soils, 2001, 34, 109-117.
26. Zerulla W, Barth T, Dressel J, Erhardt K, Locquenghien KHV, Pasda G, et al., 3, 4-Dimethylpyrazole phosphate (DMPP)—a new nitrification inhibitor for agriculture and horticulture. Biol Fertil Soils, 2001, 34, 79-84.
27. Di HJ, Cameron KC, The use of a nitrification inhibitor, dicyanamide (DCD) to decrease nitrate leaching and nitrous oxide emissions in simulated grazed and irrigated grassland. Soil Use Manage, 2002, 18, 395-403.
28. Hoeft RG, Current status of nitrification inhibitor use in U.S. agriculture, Nitrogen in crop production, (ed.) Hauck RD, American Society of Agronomy, Madison, 1984, p. 561-570.
29. Slangen JHG, Kerkhoff P, Nitrification inhibitors in agriculture and horticulture: A literature review. Fert Res, 1984, 5, 1-76.
30. Maftoun M, Yasrebi J, Darbeheshti M, Comparative phytotoxicity of nitrapyrin and ATC to several leguminous species. Plant Soil, 1981, 63, 303-306.
31. Di HJ, Cameron KC, Effects of temperature and application rate of a nitrification inhibitor, dicyandiamide (DCD), on nitrification rate and microbial biomass in a grazed pasture soil. Aust J Soil Res, 2004, 42, 927-932.
32. Krogmeier MJ, McCarty GW, Bremner JM, Potential phytotoxicity associated with the use of soil urease inhibitors. Proc Natl Acad Sci U.S.A., 1989, 86, 1110-1112.
33. Bremner JM, Recent research on problems in the use of urea as a nitrogen fertilizer. Fert Res, 1995, 42, 321-329.
34. Watson CJ, Urease activity and inhibition—principles and practice, Proceedings of International Fertiliser Society, (ed.) Watson CJ, U.K., 2000, p. 1-40.
35. Lyon, TL, Bizzell JA, Wilson BD, Depressive influence of certain higher plants on the accumulation of nitrates in the soil. J Am Soc Agron, 1923, 15, 457-466.
36. Theron JJ, The influence of plants on the mineralization of nitrogen and the maintenance of organic matter in the soil. J Agric Sci, 1951, 41, 289-296.

37. Stiven G, Production of antibiotic substances by the roots of a grass (*Trachypogon plumosus* (H.B.K.) Nees.) and of *Pentanisia variabilis* (E. May) Harv. (Rubiaceae). *Nature*, 1952, 170, 712-713.
38. Roux ER, The nitrogen sensitivity of *Eragrostis curvula* and *Trachypogon plumosus* in relation to grassland succession. *S Afr J Sci*, 1954, 50, 173-176.
39. Rice EL, Inhibition of nitrogen-fixing and nitrifying bacteria by seed plants. *Ecology*, 1964, 45, 824-837.
40. Munro PE, Inhibition of nitrite-oxidizers by roots of grass. *J Appl Ecol*, 1966, 3, 227-229.
41. Sahrawat KL, Comparative evaluation of Karanj and extracts of Karanja (*Pongamia glabra* Vent.) and neem (*Azadirachta indica* L.) seeds for retardation of nitrification of urea in soil. *J Indian Soc Soil Sci*, 1982, 30, 156-159.
42. Mohanty S, Chhonkar AK, Patra PK, Neem (*Azadirachta indica*) seed kernel powder retards urease and nitrification activities in different soils at contrasting moisture and temperature regimes. *Bioresour Technol*, 2008, 99, 894-899.
43. Kiran U, Patra DD, Medicinal and aromatic plant materials as nitrification inhibitors for augmenting yield and nitrogen uptake of Japanese mint (*Mentha arvensis* L. Var. Piperascens). *Bioresour Technol*, 2003, 86, 267-276.
44. Patra DD, Kiran U, Kumar S, Urease and nitrification inhibitors from natural source: *Artemisia annua*. *J Indian Soc Soil Sci*, 2002, 50, 508-510.
45. Thibault JR, Fortin JA, Smirnoff WA, *In vitro* allelopathic inhibition of nitrification by balsam poplar and balsam fir. *Am J Bot*, 1982, 69, 676-679.
46. Geethalakshmi V, Lourduraj CA, Maragatham N, Nitrification retardation property of some plant products and their effect on N uptake and nitrogen use efficiency in cotton. *Indian J Agric Res*, 1998, 32, 271-277.
47. Sahrawat KL, Parmar BS, Alcohol extract of "neem" seed as nitrification inhibitor. *J Indian Soc Soil Sci*, 1975, 23, 131-134.
48. Prasad B, Prasad R, Prasad J, Evaluation of nitrification retardation property of non-edible oils and their influence on yield and N uptake by wheat in calcareous soil. *J Indian Soc Soil Sci*, 1986, 34, 281-285.
49. Santhi SR, Palaniappan Sp, Effect of neem leaf (*Azadirachta indica* L.) on growth and yield of low land Rice. *J Agron Crop Sci*, 1986, 157, 114-117.
50. Muthuswamy P, Raju GSN, Krishnamoorthy KK, Mineralization of urea coated with nitrification inhibitors. *J Indian Soc Soil Sci*, 1975, 23, 332-335.
51. Mago GS, Totawat KL, Efficacy of coating materials for urea applied under saline water irrigation. I. Mineralization of urea-N. *Ann Arid Zone*, 1989, 28, 79-87.
52. Krishnapillai S, Inhibition of nitrification by waste tea (tea fluff). *Plant Soil*, 1979, 51, 563-569.
53. Ram M, Patra DD, Subramanyam K, Singh DV, Nitrification inhibitory properties in mentha-spent and pyrethrum flowers. *J Indian Soc Soil Sci*, 1993, 41, 176-177.
54. Jain JM, Narayanasamy G, Sarkar MC, Datta MN, An evaluation of nitrification retardation property of *Citrullus colosynthis* cake and its influence on yield and N uptake by wheat. *J Indian Soc Soil Sci*, 1980, 28, 480-484.
55. Suri VK, Datta BK, Linseed oil coated urea and ammelide as slow release nitrogenous fertilizers. *J Indian Soc Soil Sci*, 1995, 43, 615-618.
56. Strauss E A, The effects of organic carbon and nitrogen availability on nitrification rates in stream sediments. Ph.D. dissertation, 2000, University of Notre Dame, South Bend, Indiana.
57. Sahrawat KL, Parmar BS, Mukerjee SK, Note on the nitrification-inhibitors in the seeds, bark and leaves of *Pongamia glabra* Vent. *Indian J Agric Sci*, 1974, 44, 415-418.
58. Saharawat KL, Karanj (*Pongamia glabra* Vent.) as source of nitrification inhibitors. *Fert News*, 1981, 26, 29-33.
59. Patra DD, Kiran U, Pande P, Urease and nitrification retardation properties in natural essential oils and their by-products. *Commun Soil Sci Plant Anal*, 2006, 37, 1663-1673.
60. Purnomo D, Suryono, Sulistyio TD, Budiastuti S, Supriyadi, Potential of varies trees litter containing tannin on agroforestry system as nitrification inhibitor for Increasing nitrogen fertilizer efficiency for soybean. *J Agri Sci and Tech*, 2012, B 2, 198-203.
61. Arora K, Sharma M, Srivastava J, Ranade SA, Sharma AK, Rapid *in vitro* cloning of a 40-year-old tree of *Azadirachta indica* A. Juss. (Neem) employing nodal stem segments. *Agroforest Syst*, 2010, 78, 53-63.
62. Tanaka JP, Nardi P, Wissuwa M, Nitrification inhibition activity, a novel trait in root exudates of rice. *AOB Plants*, 2010, Available at <http://aobpla.oxfordjournals.org/content/2010/plq014.full.pdf+html>.
63. Alsaadawi IS, Al-Uquili JK, Alrubeaa AJ, Al-Hadithy SM, Allelopathic suppression of weed and nitrification by selected cultivars of *Sorghum bicolor* (L.) Moench. *J Chem Ecol*, 1986, 12, 209-219.
64. Subbarao GV, Kishii M, Nakahara K, Ishikawa T, Ban T, Tsujimoto H, et al., Biological nitrification inhibition (BNI)-Is there potential for genetic interventions in the Triticeae? *Breed Sci*, 2009, 59, 529-545.

Source of support: Council of Scientific and Industrial Research (CSIR), India

Conflict of interest: None Declared