Chapter 12
Nitrogen and Stress

Annie P. Jangam and N. Raghuram

Abstract Nitrogen is an essential macronutrient for the plants and fertilizer N-use efficiency is becoming an increasing economic and environmental concern. The nutrient stress conditions of N deficiency and N excess may get exacerbated by other abiotic stresses, which in turn are likely to be worsened by climate change. Exploring their interrelationships is being increasingly facilitated by the growing knowledge of the genome-wide N response as well as other abiotic stress responses in model plants. Nitrate and its more reduced forms are not only sources of plant N nutrition but also signals that govern their own uptake; N, C, and redox metabolism; and hormonal and other organism-wide responses. The signaling mechanisms involved in N response or response to N stress or N-use efficiency are currently far less well understood than those in other abiotic stresses. The purpose of this review is to provide an overview of the current state of knowledge on normal N response and response to N stress, as well as their interrelationships with other abiotic stresses.

Keywords Nutrient • Nitrogen • Nitrogen use efficiency (NUE) • Plants • Stress • Signaling • Integrated nutrient management • QTL • Hormones • Fertilizer management • Climate change

Nitrogen is an important macroelement for plant growth. However, plant can't use atmospheric nitrogen as such and depend on its availability in more reactive forms such as urea, nitrate, ammonium, amino acids, etc. Even legumes depend on symbiotic N-fixing bacteria to convert N_2 into ammonium ions to meet their N requirements. Therefore, the term nitrogen (N) is used in this review to represent a broad range of reactive species of N compounds. In agricultural soils, N compounds and other nutrients have to be constantly replenished as fertilizers/manures to enable repetitive cropping. As N fertilizers are expensive, N-use efficiency becomes an important determinant of crop productivity.

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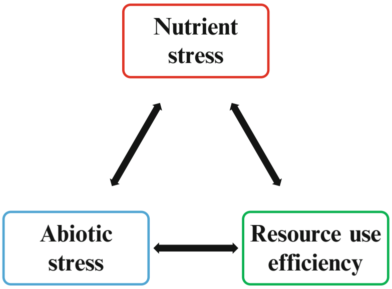


Fig. 12.1 Relationship between nutrient stress, abiotic stress, and resource-use efficiency

Since precision farming techniques to balance the plant nutrient demand with fertilizer supply are not accessible/affordable to farmers in developing countries, the actual availability of N to the crop plant varies from N-deficient state to N-excess state, depending on prevailing fertilizing practices. Both these states can cause nutrient stress to the plant. In addition, loss of reactive N species from the soil–plant system causes widespread environmental stresses, not only through N pollution of ground water and surface water bodies affecting health, biodiversity, and ecosystem services but also through air pollution and climate change (Sutton and Bleeker 2013). In fact, N_2O as a greenhouse gas is 300 times more potent than CO_2 (Galloway et al. 2008), though carbon dominates the entire climate change discussion. Climate change itself causes/exacerbates abiotic stresses to the plant and its N status (Fig. 12.1).

Nitrogen-use efficiency is therefore not only an economic problem of optimizing input costs to the farmer but also an environmental problem of preventing accumulation of reactive N species outside the agroecosystem. Accordingly, the relationship between plant, nitrogen, and stress is twofold: nutritional stress in terms of plant growth/development/productivity due to variation in N availability or climate and N-induced environmental stress (and climate change that in turn affects the plant), which can be exacerbated due to N-inefficient cultivars and/or practices. The primary focus of this book chapter is nutritional stress, but N-use efficiency is a common concern that links both nutritional and environmental aspects of reactive nitrogen.

12.1 Nitrogen Nutrition and N-Use Efficiency

Nitrogen-use efficiency has been defined in many different ways by agronomists, physiologists, and others (Good et al. 2004; Pathak et al. 2008; Hirose 2011). The simplest among them is total biomass or grain yield per unit N fertilizer added (or N available in the soil). Improvement in NUE is possible to some extent by non-plant interventions such as the choice of fertilizer and method/timing of its application and other crop management practices. But only biological avenues for crop improvement are the main focus of this article, whether in relation to NUE or stress resistance. Plants show improved NUE in N-limiting conditions (Kant et al. 2011). In other words, a plant that gives the same or higher agronomic output with lesser N input is considered more N-use efficient than the plant that needs higher N input. But what constitutes yield, varies from crop to crop, such as grains in cereals or leaves/fruits/tubers in vegetables, and accordingly what constitutes NUE and how to improve it, in each case (see Chardon et al. 2012 for a recent review). This is also often true for stress resistance, if it is measured in terms of the impact of stress on yield, keeping in view the multiplicity of stresses involved. Unfortunately, many of the high-yielding and/or upmarket varieties of the green revolution era were neither designed for N-use efficiency nor stress resistance, whereas the farmers selected traditional cultivars that were more robust to such factors, even if they yielded less. This means that crop improvement strategies for NUE and stress resistance cannot be limited to the narrow germplasm of high-yielding varieties and have to include the wild/traditional varieties.

It is well known that NUE is an inherited, multigenic, quantitative trait. A study of natural variation of N uptake and metabolism in 18 accessions of Arabidopsis under high- and low-N conditions showed that while plants may vary in the way they respond to high- or low-N conditions, their NUE remained similar, indicating that NUE as a trait is exclusively genetically determined (Chardon et al. 2010, 2012). However, the multiplicity of the definitions of NUE, combined with its poor biological characterization at the phenotypic or genotypic level makes it difficult to study the impact of stress on NUE. Nevertheless, any discussion on nitrogen and stress has to be understood in terms of the impact of abiotic or nutritional stress on NUE. For example, elevated CO_2 could enhance NUE in some cases (Shimono and Bunce 2009), while heat or water stress could adversely impact NUE (Harrigan et al. 2009), as does wasteful use of N fertilizer. Understanding the various mechanisms underlying these complex interactions will equip us with the means to maintain crop productivity in a changing climate.

12.1.1 Nitrogen Uptake and Metabolism

Plant nitrogen (N) nutrition is a complex and dynamic process, as the plant has to be able to assimilate various forms/amounts of nitrogen in fluctuating micro- and macroenvironments. This can happen even in fertilized soils, depending on the soil N status, nature of the fertilizer used (organic/inorganic), the frequency of their application, and the action of nitrifying bacteria in the soil. Organic N sources (manures/urea) are broken down by nitrifying bacteria to inorganic compounds such as nitrates and ammonium salts, which are the preferred forms of N uptake for most plants, though amino acids can be used under extremely N-poor and cold conditions.

Plants uptake nitrate primarily through the high-/low-affinity transport systems (HATS/LATS) in the roots and are mostly inducible by nitrate and regulated by its downstream metabolites, hormones, stress, etc., except the constitutive HATS (Tsay et al. 2007). The signaling mechanisms involved in the nitrate regulation of NO_3^- transporter genes need to be elucidated fully, although few genes in Arabidopsis (NRT1.1, NLP7, and CIPK8) have been suggested to play a crucial role (Castaings et al. 2009). NRT1.1 is termed "transceptor" since it is both a transporter as well as a receptor for N signal (Gojon et al. 2011). There are also transporters for ammonium and urea, but their relative contribution to the overall external N acquisition by plants and their regulation are far less understood, despite the growing interest (Vert and Chory 2009; Näsholm et al. 2009; Bouguyon et al. 2012; Wang et al. 2012). Some aspects of N transport under situations of stress have been mentioned later in this chapter, but one can expect a lot more activity in this area in the coming years.

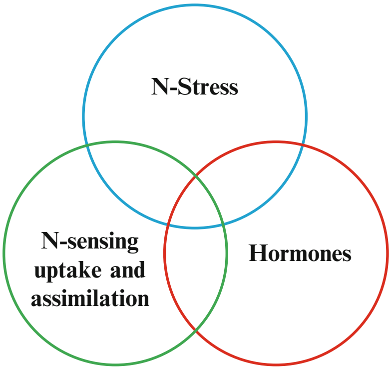
The N compounds taken up by the roots are distributed throughout the plant, where they enter the cellular nitrate assimilatory pathway. Nitrate is reduced in the cytosol by the enzyme nitrate reductase to nitrite, which is transported to the chloroplast, where it is further reduced to ammonium ions by nitrite reductase. The ammonium ions are incorporated into organic acids to form amino acids, through the glutamine synthase (GS-GOGAT cycle) and various transaminases. This is the primary N metabolic pathway in plants, but secondary N remobilization can occur during senescence, in which cytosolic isoforms of GS and GOGAT also play important roles (Xu et al. 2012). Secondary N remobilization to recycle nutrients from senescing leaves could be of critical importance for grain filling and yield in cereal crops (Kichey et al. 2007). Signaling mechanisms play a key role in N metabolic regulation to optimize the N budget under varying situations of plant N demand and supply.

12.1.2 Signaling in N Metabolic Regulation

Nitrate is not only a nutrient but also a signal for plant metabolic regulation, growth, and development (Stitt 1999). Other forms of N such as ammonium and glutamine have also been shown to have signaling roles, but nitrate remains the best-studied form of N signal, both at the local and systemic levels. Locally, it affects root growth, seed dormancy, and flowering time, but also has systemic effects on an organism-wide basis (Ho and Tsay 2010). For example, nitrate has been shown to induce genome-wide changes in gene expression in model plants such as Arabidopsis (Wang et al. 2003; Scheible et al. 2004), rice (Lian et al. 2006; Cai et al. 2012), maize (Trevisan et al. 2011), and tomato (Wang et al. 2001), involving hundreds, if not thousands, of genes. They include various nitrate transporters, enzymes of nitrate assimilation, carbon and redox metabolism, several protein kinases, cytochrome families, transcription factors, etc. The search for nitrate response elements (NREs) in the upstream sequences of a few nitrate-responsive genes has not yet produced a universally accepted consensus sequence that accounts for most, if not all nitrate-responsive genes found to date (Das et al. 2007; Konishi and Yanagisawa 2010, 2011; Pathak et al. 2011; Krapp et al. 2014). Similarly, several transcription factors are implicated in mediating nitrate response, such as ANR1, DOF1/2, LDB37/38/39, NLP6/7, and SPL9 (reviewed in Krapp et al. 2014), and further research is needed to narrow them down.

It is believed that the combinatorial action of local and systemic signaling determines the ability of a plant to adapt to fluctuating environments (Krouk et al. 2011; Alvarez et al. 2012; Huang et al. 2012). However, the mechanism of nitrate signaling continues to evade scientists for several decades. Signaling is also involved in stress response, whether due to N deficiency/excess or due to other abiotic stresses, impinging on N metabolic regulation. The pathways for stress and nutrient signaling

Fig. 12.2 Overlap between N stress, hormones, and N sensing, uptake, and assimilation



may either be separate or shared, depending on the specific stress in question, which will further define whether manipulation of one will impact the response to the other (Fig. 12.2). This is an issue of crucial agronomic relevance, which can only be addressed when the signaling mechanisms connecting N metabolic regulation and stress are better elucidated. Some developments in this regard are elaborated below.

12.2 Nitrogen and Stress

Nitrogen stress is caused by extreme fluctuations in the soil N level or due to the formation of nitroso compounds in the plant as a consequence of other stresses. The normal intracellular nitrate concentration is in the micromolar range and soil N concentration up to multi-millimolar range fall within the nutritional range (and therefore also the tolerance range) of most plants. N-limitation or N-deficiency stress occurs when soil N levels fall below the sub-millimolar range, eventually leading to N starvation. N-excess conditions require N levels to increase beyond 40 mM, though the precise threshold varies depending on the plant, duration of exposure, soil type, organic content, microbial activity, cropping practices, and climate.

Plants respond in many different ways to changes in N provision (Krouk et al. 2010; Kraiser et al. 2011; Kant et al. 2011). Their responsiveness to N availability depends on both genotype and the interaction of genotype with N fertilization level (Gallais and Hirel 2004; Chardon et al. 2010). They can adjust their molecular machinery in accordance with N nutritional status or abiotic stress, often rapidly and sometimes indefinitely (Daniel-Vedele et al. 2010). For example, plants respond to N starvation or deficiency by changes such as increase in the root to shoot ratio by enhancing lateral root growth or suppressing shoot growth or early senescence of leaves (Marschner 1995). Characterization of the machinery responsible for N homeostasis in stress helps to identify appropriate sites of intervention for crop improvement. This machinery includes, but is not limited to, the affinity-based N transport systems such as LATS and HATS, which can be reprogrammed to achieve

N homeostasis at the local or whole plant level. Several studies have shown that the root uptake capacities for nitrate, ammonia, and urea are strongly downregulated under conditions of N excess and upregulated during N starvation or N limitation (Tsay et al. 2007; Nacry et al. 2013). However, going beyond transporters, it seems that the limiting steps in plant N metabolism are different under high and low N levels (Coque and Gallais 2006). Moreover, while studies that report "high" N often do not make clear distinction between N sufficiency and N excess, studies that report "low" N either deal with N limitation or N starvation but not both, making it necessary to discuss them in their own separate context.

12.2.1 Nitrogen Starvation/Limitation

Nitrogen-starved plants show poor development with shunted growth; chlorosis; reduced photosynthesis; poor yield; poor pigmentation due to carbohydrate accumulation, anthocyanin induction, and phenylpropanoid biosynthesis. Studies in which nitrate was supplied to nitrate-starved plants like Arabidopsis and rice (Wang et al. 2000, 2003; Scheible et al. 2004; Lian et al. 2006; Cai et al. 2012) showed the involvement of genes from N/C metabolism, redox metabolism, hormonal response, etc. Many gene families such as cytochrome, protein kinases, and hormone/nutrient transporter were differentially regulated by nitrate in both rice and Arabidopsis (Cai et al. 2012). Various N transporters such as NRT2.1 and NRT2.2 for nitrate; AMT1.1, AMT1.2, and AMT1.3 for ammonia (Tsay et al. 2007); and DUR3 for urea (Kojima et al. 2007) were differentially regulated by N source/availability/concentration. The expression of GLN and GDH genes (Masclaux-Daubresse et al. 2005) were also altered during N starvation. The possible role of NLP7 as a key regulator in N-starved conditions has been suggested recently (Marchive et al. 2013). Studies in Arabidopsis and maize have shown that chronic N limitation elicits a genome-wide response and the genes involved are far more differentially regulated than genes supplied with sufficient N (Bi et al. 2007; Wu et al. 2011).

In the low concentration range such as 1 μM, high-affinity transport systems (HATS) are able to scavenge ions from the soil. During N starvation or limitation, NRT1.1 represses lateral root in Arabidopsis by remobilizing auxin, mimicking the role of an auxin transporter (Krouk et al. 2010). Low concentrations of ammonia have also been known to strongly regulate nitrate transport systems. NRT1.1 mutant studies have shown that a protein kinase CIPK23 phosphorylates NRT1.1 during nitrate limitation, thereby influencing primary nitrate response (Ho et al. 2009). The rate of N uptake in roots is determined by ionic concentration (Tsay et al. 2007), which is influenced by various stress conditions (Segonzac et al. 2007). Recently, it was reported that in maize, N-deficiency stress resembled the response of plants to a number of other biotic and abiotic stresses, in terms of transcript, protein, and metabolite accumulation (Amiour et al. 2012). In rice, two proteins, fibrillin and hairpin-binding protein, have been identified previously as N-deficiency stress-responsive proteins (Song et al. 2010; Amiour et al. 2012).

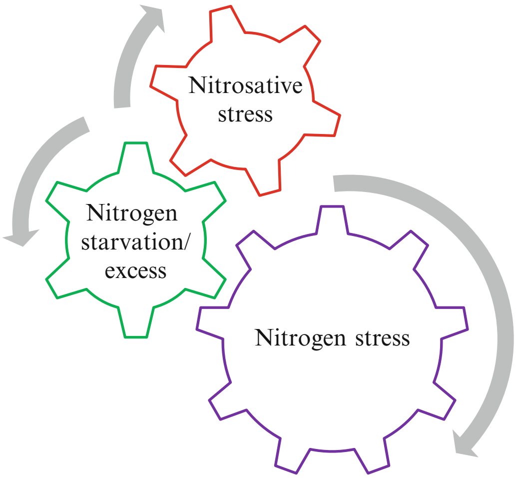


Fig. 12.3 Nitrogen stress caused by nitrogen starvation/ excess and nitrosative stress

12.2.2 Nitrogen Excess

In natural and in well-managed agricultural soils, excess N concentrations are rarely found for long, due to microbial conversions, surface runoff, volatilization, or leaching, apart from plant uptake. For example, although urea application in excess of 100 kg/ha is very common in intensively cultivated areas, its effective concentrations are often <70 mM in agricultural soils (Wang et al. 2008). Reaching far higher concentrations that contribute to nonspecific osmotic stress effects is only possible when other solutes are also high, such as in saline soils. This is also true for the conversion products of urea, viz., nitrate/nitrite/ammonium, whose ionic effects saturate in the millimolar range, and they rarely reach 100-fold levels needed to have any osmotic effect on their own. However, they can influence the pH of the soil temporarily, though the extent and duration of that influence on the soil as well as on the plant depend on the soil type/conditions and the plant itself. In any case, the ionic/ pH/osmotic effects are indirect and generic effects that are not specific to N and therefore cannot be strictly considered as N stress. Terms such as N “sufficiency” or “excess” or “high N” have to be understood in this context, as they are often used interconvertibly, mainly to contrast with N limitation/starvation (Fig. 12.3).

Genes from transporter families have shown altered gene expression during chronic N stress probably because plants need to adjust to the varying levels of N available to plants. In the high concentration range, the activity of low-affinity transport systems (LATS) from the large family of transporters for nitrate (NRT1) and peptides (PTR) plays a major role (Tsay et al. 2007). Unlike with the HATS, the LATS-mediated nitrate or ammonia uptake (or influx) does not saturate and shows a generally linear increase with increasing external concentration (Touraine and Class 1997; Nacry et al. 2013). But they could accumulate in plant cells, unless their assimilation can match the uptake. This necessitates a mechanism to regulate cytosolic nitrate concentration, which is provided by a nitrate-inducible efflux system that prevents excessive accumulation of nitrate in the cell (Miller et al. 2007). An efflux transporter, NAXT1, was recently identified belonging to the NRT1/PTR family of transporters (Segonzac et al. 2007; Chapman and Miller 2011).

Plants also have a capacity to store N in vacuoles as a way of balancing between uptake, assimilation, and translocation to other parts of the plant and minimize losses through efflux/volatilization. Some vegetables grown under excess N conditions have a tendency to accumulate N (Chen et al. 2004; Anjana et al. 2007) that enters our food chain. There is a growing attention toward the adverse health effects of excessive dietary exposure to nitrate and other forms of reactive N, including methemoglobinemia, gastric cancer, respiratory ailments, cardiac disease, etc. (Townsend et al. 2003; Anjana and Iqbal 2007).

12.2.3 Nitrosative Stress

Reactive nitrogen species (RNS) include NO and related molecules such as *S*-nitrosothiols (SNOs), *S*-nitrosoglutathione (GSNO), peroxynitrite ($ONOO^-$), dinitrogen trioxide (N_2O_3), and nitrogen dioxide (NO_2). During adverse environmental conditions, these molecules can cause stress to plants, which is designated as nitrosative stress (Fig. 12.3). This can combine with the stress caused by reactive oxygen species (ROS) to form nitro-oxidative stress in plants. Like ROS, the role of RNS signaling has been implicated in many abiotic stresses such as salinity, water stress, temperature stress, and UV radiation. Evidence for RNS signaling in certain abiotic stresses like salinity and heat stress was strictly species or treatment specific, and the literature is inconclusive, if not contradictory (Corpas et al. 2011). In other stresses such as UV-radiation and ozone stress, there is a marked increase in the activity of RNS species, which leads to cell death (Corpas et al. 2011). The role of RNS signaling needs to be studied further to understand the changes in plant physiology under stress.

12.3 Nitrogen in Abiotic Stresses

Nitrogen availability depends on plant–soil–microbe interactions, whereas nitrogen acquisition is basically driven by transpiration, which is in turn affected by temperature and levels of CO_2. Climate change models predict that elevated levels of green house gases (GHGs) like CO_2, CH_4, and N_2O cause increased variability in temperature, humidity, precipitation, wind velocity, and photosynthetically active radiation, all leading to major abiotic stresses such as drought, heat/cold stress, waterlogging, etc. (IPCC 2007; Bloom et al. 2010). All these stresses affect plant phenology and also alter nitrogen availability and its uptake/retention (Fig. 12.4). For example, Borner et al. (2008) observed that the snow depth in

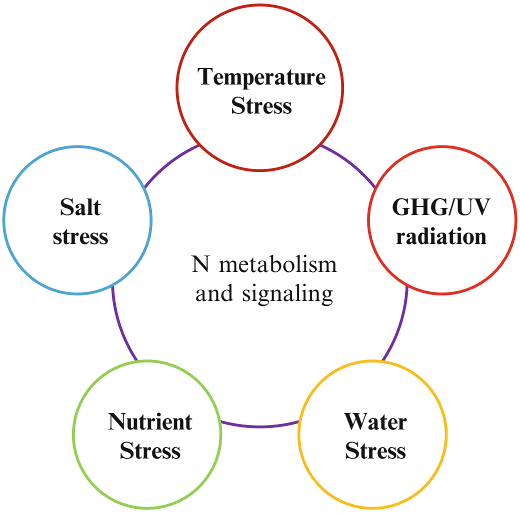


Fig. 12.4 N metabolism and signaling play an integral role in various abiotic stresses

tundra regions affect N mineralization directly. Volatilization of reactive N from the plant further contributes to GHG accumulation, completing the vicious cycle. The role of reactive N in plant stress can be understood in the context of specific stresses as given below or integrated into a network of multiple interacting stresses, as elaborated later in this chapter.

12.3.1 N in Elevated CO_2 and NO_2

Continuous exposure to elevated atmospheric CO_2 may result in stomatal closure, adversely affecting the rate of transpiration and therefore nutrient uptake of the plants, leading to nutrient N deficiency stress. Decrease in activities of assimilatory enzymes such as nitrate reductase (Ferrario-Méry et al. 1997) and RuBisCO (Bloom et al. 2010) was also observed. However, brief exposure to elevated CO_2 showed enhanced activities of NR and GSA in cucumber and sunflower leaves (Aguera et al. 2006). Inhibition of photorespiration-dependent nitrate assimilation (Rachmilevitch et al. 2004) is also observed at higher levels of CO_2. Elevated CO_2 is known to enhance photosynthesis in C3 plants and improve NUE (Shimono and Bunce 2009). The form of N used under various CO_2 concentrations affect the nutrients and their distribution in the plant (Natali et al. 2009). For example, wheat plants supplied with ammonium salts as a source of N were more N responsive under elevated CO_2 concentrations, in terms of nutrient accumulation, yield, and yield components, as compared to those supplied with nitrate (Carlisle et al. 2012 and references therein).

Nitrogen dioxide can be absorbed and utilized by the plants in small quantities for assimilation (Mokhele et al. 2012) and therefore elevated NO_2 causes some increase in intracellular nitrate concentration (Qiao and Murray 1998). On the other hand, reduction of ambient NO_2 level has no effect on the organic N content of the plants or on the amount or rate of N uptake in the plants.

12.3.2 N in Water and Salt Stress

Drought stress alone is projected to double in future, which will lead to loss of yield (IPCC 2007). While photosynthesis can be maintained under fluctuations of water supply (Lightfoot et al. 2007), water deficit can alter the C and N transformations by bringing about changes in the soil–microbe interactions such as reducing the activity of nitrifying bacteria (StClair and Lynch 2010 and references therein). This is evident from the inhibition of nitrogen fixation in legume crops during C and N fluxes under drought (Ladrera et al. 2007; Rogers et al. 2009).

The effect of drought on leaf N status remains uncertain, as it was reported to increase in *Malus domestica* (Jie et al. 2010), decrease in *Prunus persica* (Dichio et al. 2007), and be unaffected in *Quercus* (Li et al. 2013). However, a recent transcriptome study suggests the interactive effects on the genome-wide impact of drought and N limitation in maize (Humbert et al. 2013). It studied 30 conditions involving three major parameters such as organ (leaf, root, or stem), nitrogen supply (optimal or chronic limitation), and water supply (optimal supply, mild water stress by withdrawal for 3 days, severe water stress by withdrawal for 5 days, and recovery from severe stress by rewatering for 2 h or 5 h). The impact of severe stress was more extensive in root and stem than in leaf, in terms of the number of spots/genes affected. The pathways most affected were sucrose and starch metabolism, Calvin cycle, proline, and asparagine biosynthesis. Both photosynthetic assimilation and nitrate assimilation were shown to be downregulated. The effect of water withdrawal and nitrogen limitation on ammonium assimilation was tissue specific; the transcripts for glutamine and glutamate synthases were more in leaf as compared to stem and root. This study also shows that while nitrogen limitation has very little impact on the transcriptome on its own (0.2 % of the spots), even mild water stress makes the plant more vulnerable to N limitation, affecting the expression of a much larger number of genes (Humbert et al. 2013). These observations need to be validated in other plants before wider generalizations could be made. However, studying such interactive effects could be useful to optimize NUE along with water-use efficiency (WUE) (Di Paolo and Rinaldi 2008). On the other hand, heavy use of N fertilizers regardless of the water regime can be detrimental on grain filling and drought tolerance (Humbert et al. 2013 and references therein).

Drought tolerance genes contribute to greater NUE because they improve biomass production over an extended range of soil moisture availability and weather conditions (Harrigan et al. 2009). The traits for drought stress include yield potential, WUE, harvest index (HI), improved transpiration efficiency, and deep root penetration (to access water and nutrients), all of which are relevant to NUE as well.

Flooding is another form of water stress that is expected to increase due to climate change. Waterlogging affects ~10 % of the global land area and an estimated 10 million hectares of land in developing countries. It can cause a wide variety of symptoms that can affect yield either directly or indirectly, through affecting leaf senescence, tiller number, and reduced plant height. While N availability could increase under situations in which floods bring silt and nutrients along, it could also decrease in situations where topsoil and fertilizer N are lost or diluted out. Other parameters such as temperature could also result in interactive effects.

Salinity is one of the major abiotic stresses that lower the yield and usually is accompanied by water stress. The impact of salt stress is dependent on the cultivar/organ/developmental stage and the degree of salt stress. Salinity is known to alter the activities of various enzymes from the N assimilatory pathway like nitrate reductase in leaves than in roots (Mokhele et al. 2012).

12.3.3 N in Heat and Cold Stress

Accumulation of greenhouse gases (carbon dioxide, methane, and nitrous oxide) in the earth's atmosphere is expected to warm up the earth's surface by 1.8–4 °C by the end of this century (IPCC 2007). Rising temperatures of both soil and air could alter the rate of water and nitrogen uptake due to their effect on rate of transpiration and soil moisture respectively (Dong et al. 2001). Elevated temperatures also alter N allocation, reduce foliar N concentration and carbohydrate content (Tjoelker et al. 1999), damage photosynthetic membranes and cause chlorophyll loss decreasing leaf photosynthetic rate, increase embryo abortion, lower grain number, and decrease grain-filling duration and rates resulting in lower grain yield.

Globally, leaf N concentrations have been found to vary along altitudinal and latitudinal temperature gradients across plant species or functional groups (Reich and Oleksyn 2004). Foliar N content increases from the tropics to the cooler and drier midlatitudes due to "temperature-related plant physiological stoichiometry and biogeographical gradients in soil substrate age, as well as cold temperature effects on biogeochemistry at high latitudes" (Reich and Oleksyn 2004).

Cold stress can produce undesirable responses to nitrogen fertilizers that are often applied in high concentrations to increase yield. For example, high nitrogen supply before/during pollen development aggravates the effect of pollen sterility in rice in extreme cold conditions (Gunawardena et al. 2003). Low temperatures are also known to inactivate RuBisCO carboxylase by *S*-nitrosylation (Corpas et al. 2011).

12.3.4 N in UV and Other Stresses

Enhanced exposure to UV is one of the consequences of climate change. Plant N uptake and assimilation are significantly inhibited at high levels of UV-B radiation. Excessive UV light along with nutrient deficiencies can lead to photo-oxidative stress, which is worsened further in other environmental stresses such as metal toxicity (Lynch and StClair 2004 and the references therein). There is a significant increase in the activity of RNS species under UV-radiation stress as well as under ozone stress, which could lead to cell death (Corpas et al. 2011).

12.3.5 N in Multiple Interacting Stresses

Most of the plant stress studies were done by changing or observing a single variable factor, i.e., changing CO_2, temperature, or water concentrations, but very few studies considered the combinatorial effect of these stresses. Studies show increased concentrations of CO_2 can increase the demand for nutrient, but increase in temperatures can influence the length of growing season and in turn reduce the demand for nutrients (Nord and Lynch 2009; Mittler and Blumwald 2010). Rice plants exposed to increases levels of CO_2 have shown increased sensitivity to cold stress (Shimono and Bunce 2009). Both drought and heat stresses together affect N availability storage and remobilization in pine trees (Rennenberg et al. 2009; Huang et al. 2012). The activity of many nitrate-regulated genes is hypothesized to be regulated by light, and evidence shows that both NRT2 and NR activity is dependent on light as well as nitrogen (Lillo 2004, 2008; Chapman and Miller 2011). Thus, mounting evidence suggests that the effect of various abiotic stresses can lead to unanticipated changes in plant growth and development.

12.4 Conclusions and Prospects

There is a growing demand for developing crops with resilience to climate change, abiotic stress, and N-use efficiency for global food security and environmental sustainability. While studies on individual stresses and the signaling mechanisms involved in the plant's response to them have made impressive progress, integrative studies are needed that can model the complex interactions between various abiotic stresses and the signaling and/or regulatory events involved in them. Similarly, in the area of N-response and N-use efficiency, integration of crop genetics and functional genomics approaches have begun to make rapid strides, but the links between nitrogen and stress remain peripheral, especially at the level of signaling and regulatory interface of stress and N, except in the area of nitrosative stress. This calls for new synergies to be forged between research on abiotic stress and resource-use efficiency in general and N-use efficiency in particular, to identify some common signaling aspects or regulatory targets for developing not only stress-resistant and climate-resilient crops but also N-use-efficient or resource-use-efficient crops.

Crop improvement through QTL mapping and marker-assisted selection/breeding seems to be a promising route in this regard. Extensive studies in various plants have lead to the mapping of many agronomic traits such NUE, yield, biomass, N uptake, and remobilization (Habash et al. 2007; Fontaine et al. 2009). The possibility of co-localization of multiple agronomically important QTLs is an increasingly attractive avenue to explore in this regard. For example, in tropical maize, QTLs for grain yield and secondary traits were identified under varying N and water supply, some of which were found to be co-localized (Ribaut et al. 2007). More efforts in this direction can be enabled by suitable national policies and intergovernmental cooperation for germplasm exchange and collaboration. The role of public sector may prove to be at least as crucial as that of the private sector in facilitating affordable access to such technologies for the farmers, breeders, and consumers alike.

References

Aguera E, Ruano D, Cabello Haba P (2006) Impact of atmospheric CO_2 on growth, photosynthesis and nitrogen metabolism in cucumber (Cucumis sativus L) plants. J Plant Physiol 163:809–817

Alvarez JM, Vidal EA, Gutiérrez RA (2012) Integration of local and systemic signaling pathways for plant N responses. Curr Opin Plant Biol 15(2):185–191. doi:10.1016/j.pbi.2012.03.009

Amiour N, Imbaud S, Clément G, Agier N, Zivy M, Valot B, Balliau T, Armengaud P, Quilleré I, Cañas R (2012) The use of metabolomics integrated with transcriptomic and proteomic studies for identifying key steps involved in the control of nitrogen metabolism in crops such as maize. J Exp Bot 63:5017–5033

Anjana SU, Iqbal M (2007) Nitrate accumulation in plants, factors affecting the process and human health implications—a review. Agron Sustain Dev 27:45–57

Anjana SU, Iqbal M, Abrol YP (2007) Are nitrate concentrations in leafy vegetables within safe limits? Curr Sci 92:355–360

Bi YM, Wang RL, Zhu T, Rothstein SJ (2007) Global transcription profiling reveals differential responses to chronic nitrogen stress and putative nitrogen regulatory components in Arabidopsis. BMC Genomics 8:281. doi:10.1186/1471-2164-8-281

Bloom AJ, Burger M, Rubio Asensio JS, Cousins AB (2010) Carbon dioxide enrichment inhibits nitrate assimilation in wheat and Arabidopsis. Science 328(5980):899–903. doi:10.1126/science.1186440

Borner AP, Kielland K, Walker MD (2008) Effects of simulated climate change on plant phenology and nitrogen mineralization in Alaskan arctic Tundra. Arct Antarct Alp Res 40:27–38

Bouguyon E, Gojon A, Nacry P (2012) Nitrate sensing and signaling in plants. Semin Cell Dev Biol 23(6):648–654. doi:10.1016/jsemcdb.2012.01.004

Cai H, Lu Y, Xie W, Zhu T, Lian X (2012) Transcriptome response to nitrogen starvation in rice. J Biosci 37(4):731–747. doi:10.1007/s12038-012-9242-2

Carlisle E, Myers S, Raboy V, Bloom A (2012) The effects of inorganic nitrogen form and CO_2 concentration on wheat yield and nutrient accumulation and distribution. Front Plant Sci 3(195):1–13. doi:10.3389/fpls.2012.00195

Castaings L, Camargo A, Pocholle D, Gaudon V, Texier Y, Boutet-Mercey S, Krapp A (2009) The nodule inception-like protein 7 modulates nitrate sensing and metabolism in Arabidopsis. Plant J 57(3):426–435. doi:10.1111/j.1365-313X.2008.03695.x

Chapman N, Miller T (2011) Nitrate transporters and root architecture. In: Geisler M, Venema K (eds) Transporters and pumps in plant signaling. Signaling and communication in plants, vol 7. Springer, Berlin, p 388. doi:10.1007/978-3-642-14369-4

Chardon F, Barthélémy J, Daniel-Vedele F, Masclaux-Daubresse C (2010) Natural variation of nitrate uptake and nitrogen use efficiency in Arabidopsis thaliana cultivated with limiting and ample nitrogen supply. J Exp Bot 61:2293–2302

Chardon F, Noël V, Masclaux-Daubresse C (2012) Exploring NUE in crops and in Arabidopsis ideotypes to improve yield and seed quality. J Exp Bot 63:3321–3323. doi:10.1093/jxb/err353

Chen BM, Wang ZH, Li SX, Wang GX, Song HX, Wang XN (2004) Effects of nitrate supply on plant growth, nitrate accumulation, metabolic nitrate concentration and nitrate reductase activity in three leafy vegetables. Plant Sci 167(3):635–643. doi:10.1016/j.plantsci.2004.05.015

Coque M, Gallais A (2006) Genomic regions involved in response to grain yield selection at high and low nitrogen fertilization in maize. Theor Appl Genet 112:1205–2017

Corpas FJ, Leterrier M, Valderrama R, Airaki M, Chaki M, Palma JM, Barroso JB (2011) Nitric oxide imbalance provokes a nitrosative response in plants under abiotic stress. Plant Sci 181(5):604–611. doi:10.1016/j.plantsci.2011.04.005

Daniel-Vedele F, Krapp A, Kaiser WM (2010) Cellular biology of nitrogen metabolism and signaling. In: Hell R, Mendel R-R (eds) Cell biology of metals and nutrients. Springer Berlin, Heidelberg, pp 145–172

Das SK, Pathak RR, Choudhury D, Raghuram N (2007) Genomewide computational analysis of nitrate response elements in rice and Arabidopsis. Mol Genet Genomics 278(5):519–525. doi:10.1007/s00438-007-0268-3

Di Paolo E, Rinaldi M (2008) Yield response of corn to irrigation and nitrogen fertilization in a Mediterranean environment. Field Crop Res 105(3):202–210

Dichio B, Xiloyannis C, Sofo A, Montanaro G (2007) Effects of post-harvest regulated deficit irrigation on carbohydrate and nitrogen partitioning, yield quality and vegetative growth of peach trees. Plant Soil 290:127–137

Dong S, Scagel CF, Cheng L, Fuchigami LH, Rygiewicz PT (2001) Soil temperature and plant growth stage influence nitrogen uptake and amino acid concentration of apple during early spring growth. Tree Physiol 21(8):541–547

Ferrario-Méry S, Thibaud MC, Betsche T, Valadier MH, Foyer CH (1997) Modulation of carbon and nitrogen metabolism, and of nitrate reductase, in untransformed and transformed Nicotiana plumbaginifolia during CO_2 enrichment of plants grown in pots and in hydroponic culture. Planta 202:510–521

Fontaine JX, Ravel C, Pageau K, Heumez E, Dubois F, Hirel B, Le Gouis J (2009) A quantitative genetic study for elucidating the contribution of glutamine synthetase, glutamate dehydrogenase and other nitrogen-related physiological traits to the agronomic performance of common wheat. Theor Appl Genet 119(4):645–662

Gallais A, Hirel B (2004) An approach to the genetics of nitrogen use efficiency in maize. J Exp Bot 55(396):295–306. doi:10.1093/jxb/erh006

Galloway J, Raghuram N, Abrol YP (2008) A perspective on reactive nitrogen in a global, Asian and Indian context. Curr Sci 94(11):1375–1381

Gojon A, Krouk G, Perrine-Walker F, Laugier E (2011) Nitrate transceptor (s) in plants. J Exp Bot 62(7):2299–2308

Good AG, Shrawat AK, Muench DG (2004) Can less yield more? Is reducing nutrient input into the environment compatible with maintaining crop production? Trends Plant Sci 9(12):597–605. doi:10.1016/j.tplants.2004.10.008

Gunawardena TA, Fukai S, Blamey FPC (2003) Low temperature induced spikelet sterility in rice. I. Nitrogen fertilisation and sensitive reproductive period. Aust J Agr Res 54:937–946

Habash DZ, Bernard S, Schondelmaier J, Weyen J, Quarrie SA (2007) The genetics of nitrogen use in hexaploid wheat: N utilisation, development and yield. Theor Appl Genet 114(3):403–419

Harrigan GG, Ridley WP, Miller KD, Sorbet R, Riordan SG, Nemeth MA, Reeves W, Pester TA (2009) The forage and grain of MON 87460, a drought-tolerant corn hybrid, are compositionally equivalent to that of conventional corn. J Agric Food Chem 57:9754–9763

Hirose T (2011) Nitrogen use efficiency revisited. Oecologia 166(4):863–867. doi:10.1007/s00442-011-1942

Ho CH, Tsay YF (2010) Nitrate, ammonium, and potassium sensing and signaling. Curr Opin Plant Biol 13(5):604–610. doi:10.1016/j.pbi.2010.08.005

Ho CH, Lin SH, Hu HC, Tsay YF (2009) CHL1 functions as a nitrate sensor in plants. Cell 138(6):1184–1194. doi:10.1016/j.cell.2009.07.004

Huang Y, Drengstig T, Ruoff P (2012) Integrating fluctuating nitrate uptake and assimilation to robust homeostasis. Plant Cell Environ 35(5):917–928. doi:10.1111/j.1365-3040.2011.02462.x

Humbert S, Subedi S, Cohn J, Zeng B, Bi Y, Chen X, Rothstein SJ (2013) Genome-wide expression profiling of maize in response to individual and combined water and nitrogen stresses. BMC Genomics 14(1):1. doi:10.1186/1471-2164-14-3

IPCC (Intergovernmental Panel on Climate Change) (2007) Climate change: changes in atmospheric constituents and in radiative forcing. In: Forster P, Ramaswamy V, Artaxo P, Berntsen T, Betts R, Fahey DW, Haywood J, Lean J, Lowe DC, Myhre G, Nganga J, Prinn R, Raga G, Schulz M, Dorland RV, Solomon S, Qin D, Manning M, Chen Z, Marquis M, Averyt KB, Tignor M, Miller HL (eds) The physical science basis. Cambridge University Press, Cambridge

Jie Z, Yuncong Y, Streeter JG, Ferree DC (2010) Influence of soil drought stress on photosynthesis, carbohydrates and the nitrogen and phosphorus absorbed in different section of leaves and stems of Fugi/M9EML, a young apple seedling. Afr J Biotechnol 9:5320–5325

Kant S, Bi YM, Rothstein SJ (2011) Understanding plant response to nitrogen limitation for the improvement of crop nitrogen use efficiency. J Exp Bot 62(4):1499–1509. doi:10.1093/jxb/erq297

Kichey T, Hirel B, Heumez E, Dubois F, Le Gouis J (2007) In winter wheat (Triticum aestivum L), post-anthesis nitrogen uptake and remobilisation to the grain correlates with agronomic traits and nitrogen physiological markers. Field Crop Res 102:22–32

Kojima S, Bohner A, Gassert B, Yuan L, von Wiren N (2007) AtDUR3 represents the major transporter for high-affinity urea transport across the plasma membrane of nitrogen-deficient Arabidopsis roots. Plant J 52:30–40

Konishi M, Yanagisawa S (2010) Identification of a nitrate-responsive cis-element in the Arabidopsis NIR1 promoter defines the presence of multiple cis-regulatory elements for nitrogen response. Plant J 63(2):269–282. doi:10.1111/j.1365-313X.2010.04239.x

Konishi M, Yanagisawa S (2011) The regulatory region controlling the nitrate-responsive expression of a nitrate reductase gene, NIA1, in Arabidopsis. Plant Cell Physiol 52(5):824–836. doi:10.1093/pcp/pcr033

Kraiser T, Gras DE, Gutiérrez AG, González B, Gutiérrez RA (2011) A holistic view of nitrogen acquisition in plants. J Exp Bot 62(4):1455–1466. doi:10.1093/jxb/erq425

Krapp A, David LC, Chardin C, Girin T, Marmagne A, Leprince A, Meyer C (2014) Nitrate transport and signalling in Arabidopsis. J Exp Bot 65(3):789–798. doi:10.1093/jxb/eru001

Krouk G, Lacombe B, Bielach A, Perrine-Walker F, Malinska K, Mounier E, Gojon A (2010) Nitrate-regulated auxin transport by NRT11 defines a mechanism for nutrient sensing in plants. Dev Cell 18(6):927–937. doi:10.1016/j.devcel.2010.05.008

Krouk G, Ruffel S, Gutiérrez RA, Gojon A, Crawford NM, Coruzzi GM, Lacombe B (2011) A framework integrating plant growth with hormones and nutrients. Trends Plant Sci 16(4): 178–182. doi:10.1016/j.tplants.2011.02.004

Ladrera R, Marino D, Larrainzar E, Gonzalez EM, Arrese-Igor C (2007) Reduced carbon availability to bacteroides and elevated ureides in nodules, but not in shoots, are involved in the nitrogen fixation response to early drought in soybean. Plant Physiol 145:539–546

Li MH, Cherubini P, Dobbertin M, Arend M, Xiao WF, Rigling A (2013) Responses of leaf nitrogen and mobile carbohydrates in different Quercus species/provenances to moderate climate changes. Plant Biol 15(s1):177–184

Lian X, Wang S, Zhang J, Feng Q, Zhang L, Fan D, Li X, Yuan D, Han B, Zhang Q (2006) Expression profiles of 10,422 genes at early stage of low nitrogen stress in rice assayed using a cDNA microarray. Plant Mol Biol 60(5):617–631

Lightfoot DA, Mungur R, Ameziane R, Nolte S, Long L, Bernhard K, Colter A, Jones K, Iqbal M, Varsa E, Young B (2007) Improved drought tolerance of transgenic Zea mays plants that express the glutamate dehydrogenase gene (gdhA)of E coli. Euphytica 156:103–116

Lillo C (2004) Light regulation of nitrate uptake, assimilation and metabolism. Plant Ecophysiol 3:149–184

Lillo C (2008) Signalling cascades integrating light-enhanced nitrate metabolism. Biochem J 415(1):11–19. doi:10.1042/BJ20081115

Lynch JP, StClair SB (2004) Mineral stress: the missing link in understanding how global climate change will affect plants in real world soils. Field Crop Res 90(1):101–115. doi:10.1016/j.fcr.2004.07.008

Marchive C, Roudier F, Castaings L, Bréhaut V, Blondet E, Colot V, Meyer C, Krapp A (2013) Nuclear retention of the transcription factor NLP7 orchestrates the early response to nitrate in plants. Nat Commun 4:1713

Marschner H (1995) Mineral nutrition of higher plants, 2nd edn. Academic, New York

Masclaux-Daubresse C, Carrayol E, Valadier MH (2005) The two nitrogen mobilization and senescence-associated GS1 and GDH genes are controlled by C and N metabolites. Planta 221:580–588

Miller AJ, Fan X, Orsel M, Smith SJ, Wells DM (2007) Nitrate transport and signaling. J Exp Bot 58(9):2297–2306. doi:10.1093/jxb/erm066

Mittler R, Blumwald E (2010) Genetic engineering for modern agriculture: challenges and perspectives. Annu Rev Plant Biol 61:443–462. doi:10.1146/annurev-arplant-042809-112116

Mokhele B, Zhan X, Yang G, Zhang X (2012) Review: nitrogen assimilation in crop plants and its affecting factors. Can J Plant Sci 92:399–405. doi:10.4141/cjps2011-135

Nacry P, Bouguyon E, Gojon A (2013) Nitrogen acquisition by roots: physiological and developmental mechanisms ensuring plant adaptation to a fluctuating resource. Plant Soil 370:1–29. doi:10.1007/s11104-013-1645-9

Näsholm T, Kielland K, Ganeteg U (2009) Uptake of organic nitrogen by plants. New Phytol 182(1):31–48. doi:10.1111/j.1469-8137.2008.02751.x

Natali SM, Sanudo-Wilhelmy SA, Lerdau MT (2009) Plant and soil mediation of elevated CO_2 impacts on trace metals. Ecosystems 12:715–727. doi:10.1007/s10021-009-9251-7

Nord EA, Lynch JP (2009) Plant phenology: a critical controller of soil resource acquisition. J Exp Bot 60(7):1927–1937. doi:10.1093/jxb/erp018

Pathak RR, Ahmad A, Lochab S, Raghuram N (2008) Molecular physiology of plant nitrogen use efficiency and biotechnological options for its enhancement. Curr Sci 94(11):1394–1403

Pathak RR, Lochab S, Raghuram N (2011) Comprehensive biotechnology, 2nd edn, vol 1. Elsevier, Oxford, pp 209–218. doi:10.1016/B978-0-08-088504-900472-4

Qiao Z, Murray F (1998) The effects of NO_2 on the uptake and assimilation of nitrate by soybean plants. Environ Exp Bot 10:33–40

Rachmilevitch S, Cousins AB, Bloom AJ (2004) Nitrate assimilation in plant shoots depends on photorespiration. Proc Natl Acad Sci U S A 101:11506–11510

Reich PB, Oleksyn J (2004) Global patterns of plant leaf N and P in relation to temperature and latitude. Proc Natl Acad Sci U S A 101(30):11001–11006. doi:10.1073/pnas.0403588101

Rennenberg H, Dannenmann M, Gessler A, Kreuzwieser J, Simon J, Papen H (2009) Nitrogen balance in forest soils: nutritional limitation of plants under climate change stresses. Plant Biol 11(s1):4–23

Ribaut JM, Fracheboud Y, Monneveux P, Banziger M, Vargas M, Jiang C (2007) Quantitative trait loci for yield and correlated traits under high and low soil nitrogen conditions in tropical maize. Mol Breed 20(1):15–29. doi:10.1007/s11032-006-9041-2

Rogers A, Ainsworth EA, Leakey ADB (2009) Will elevated carbon dioxide concentration amplify the benefits of nitrogen fixation in legumes? Plant Physiol 151(3):1009–1016. doi:10.1104/pp.109.144113

Scheible WR, Morcuende R, Czechowski T, Fritz C, Osuna D, Palacios- Rojas N, Schindelasch D, Thimm O, Udvardi MK, Stitt M (2004) Genome-wide reprogramming of primary and secondary metabolism, protein synthesis, cellular growth processes, and the regulatory infrastructure of Arabidopsis in response to nitrogen. Plant Physiol 136:2483–2499

Segonzac C, Boyer JC, Ipotesi E, Szponarski W, Tillard P, Touraine B, Sommerer N, Rossignol M, Gibrat R (2007) Nitrate efflux at the root plasma membrane: identification of an Arabidopsis excretion transporter. Plant Cell 19:3760–3777

Shimono H, Bunce JA (2009) Acclimation of nitrogen uptake capacity of rice to elevated atmospheric CO_2 concentration. Ann Bot 103:87–94

Song C, Zeng F, Feibo W, Ma W, Zhang G (2010) Proteomic analysis of nitrogen stress-responsive proteins in two rice cultivars differing in N utilization efficiency. J Integr Omics 1(1):78–87

StClair SB, Lynch JP (2010) The opening of Pandora's Box: climate change impacts on soil fertility and crop nutrition in developing countries. Plant Soil 335(1–2):101–115. doi:10.1007/s11104-010-0328-z

Stitt M (1999) Nitrate regulation of metabolism and growth. Curr Opin Plant Biol 2:178–186

Sutton MA, Bleeker A (2013) Environmental science: the shape of nitrogen to come. Nature 494(7438):435–437. doi:10.1038/nature11954

Tjoelker MG, Reich PB, Oleksyn J (1999) Changes in leaf nitrogen and carbohydrates underlie temperature and CO_2 acclimation of dark respiration in five boreal tree species. Plant Cell Environ 22:767–778

Touraine B, Class ADM (1997) NO_3^- and ClO_3^- fluxes in the chll-5 mutant of Arabidopsis thaliana. Plant Physiol 114:137–144

Townsend AR, Robert WH, Fakhri AB, Mary SB, Cory CC, Sharon KC, Andrew PD, Paul RE, Elisabeth AH, Dennis RK, Michael AM, Christine AR, Peter W, Amir HW (2003) Human health effects of a changing global nitrogen cycle. Front Ecol Environ 1(5):240–246

Trevisan S, Manoli A, Begheldo M, Nonis A, Enna M, Vaccaro S, Quaggiotti S (2011) Transcriptome analysis reveals coordinated spatiotemporal regulation of hemoglobin and nitrate reductase in response to nitrate in maize roots. New Phytol 192(2):338–352. doi:10.1111/j.1469-8137.2011.03822.x

Tsay YF, Chiu CC, Tsai CB, Ho CH, Hsu PK (2007) Nitrate transporters and peptide transporters. FEBS Lett 581(12):2290–2300. doi:10.1016/j.febslet.2007.04.047

Vert G, Chory J (2009) A toggle switch in plant nitrate uptake. Cell 138(6):1064–1066. doi:10.1016/j.cell.2009.09.005

Wang R, Guegler K, LaBrie ST, Crawford NM (2000) Genomic analysis of a nutrient response in Arabidopsis reveals diverse expression patterns and novel metabolic and potential regulatory genes induced by nitrate. Plant Cell 12(8):1491–1509

Wang Y, Garvin DF, Kochian LV (2001) Nitrate-induced genes in tomato roots. Array analysis reveals novel genes that may play a role in nitrogen nutrition. Plant Physiol 127:345–359

Wang R, Okamoto M, Xing X, Crawford NM (2003) Microarray analysis of the nitrate response in Arabidopsis roots and shoots reveals over 1,000 rapidly responding genes and new linkages to glucose, trehalose-6-phosphate, iron, and sulfate metabolism. Plant Physiol 132(2):556–567

Wang WH, Köhler B, Cao FQ, Liu LH (2008) Molecular and physiological aspects of urea transport in higher plants. Plant Sci 175(4):467–477. doi:10.1016/j.plantsci.2008.05.018

Wang YY, Hsu PK, Tsay YF (2012) Uptake, allocation and signaling of nitrate. Trends Plant Sci 17(8):458–467. doi:10.1016/j.tplants.2012.04.006

Wu X, Liu Y, Tian M, Chen R, Zheng Z, He C (2011) Genomics analysis of genes expressed reveals differential responses to low chronic nitrogen stress in maize. Afr J Biotechnol 10(6):939–949

Xu G, Fan X, Miller AJ (2012) Plant nitrogen assimilation and use efficiency. Annu Rev Plant Biol 63:153–182. doi:10.1146/annurev-arplant-042811-105532