N uptake and distribution in crops: an agronomical and ecophysiological perspective

F. Gastal[1] and G. Lemaire

Unité d'Ecophysiologie des Plantes Fourragères, INRA-UEPF, 86600 Lusignan, France

Abstract

The rate of N uptake of crops is highly variable during crop development and between years and sites. However, under ample soil N availability, crop N accumulation is highly related to crop growth rate and to biomass accumulation. Critical N concentration has been defined as the minimum N concentration which allows maximum growth rate. Critical N concentration declines during crop growth. The relationship between critical N concentration and biomass accumulation over the growth period of a crop is broadly similar within major C_3 and C_4 cultivated species. Therefore, the critical N concentration concept is widely used in agronomy as the basis of the diagnosis of crop N status, and allows discrimination between situations of sub-optimal and supra-optimal N supply. The relationship between N and biomass accumulation in crops, relies on the interregulation of multiple crop physiological processes. Among these processes, N uptake, crop C assimilation and thus growth rate, and C and N allocation between organs and between plants, play a particular role. Under sub-optimal N supply, N uptake of the crop depends on soil mineral N availability and distribution, and on root distribution. Under ample N supply, N uptake largely depends on growth rate via internal plant regulation. Carbon assimilation of the crop is related to crop N through the distribution of N between mature leaves with consequences for leaf and canopy photosynthesis. However, although less commonly emphasized, carbon assimilation of the crop also depends on crop N through leaf area development. Therefore, crop growth rate fundamentally relies on the balance of N allocation between growing and mature leaves. Nitrogen uptake and distribution also depends on C allocation between organs and N composition of these organs. Within shoots, allocation of C to stems generally increases in relation to C allocation to the leaves over the crop growth period. Allocation of C and N between shoots and roots also changes to a large extent in relation to soil N and/or crop N. These alterations in C and N allocation between plant organs have implications, together with soil availability and carbon assimilation, on N uptake and distribution in crops. Therefore, N uptake and distribution in plants and crops involves many aspects of growth and development. Regulation of nitrogen assimilation needs to be considered in the context of these interregulatory processes.

Introduction

Agriculture, previously dominated by productivity, now has multiple objectives. The environmental impact of crops and cropping systems, the quality of crop products, the low cost of production and hence increased N use efficiency, are among these objectives.

Understanding the processes that govern N fluxes, particularly N uptake and distribution in crops, is of major importance with respect to both environmental concerns and the quality of crop products. Nitrogen uptake and accumulation in crops represent two major components of the N cycle in the agrosystem. Nitrate ions not taken up by a crop, may potentially be leached to underground water. Modelling N uptake together with soil water transfers is, therefore, key in quantifying and preventing nitrate leaching. Although outside the scope

---

[1] To whom correspondence should be addressed. Fax: +33 5 49 55 60 68. E-mail: gastal@lusignan.inra.fr

of this paper, understanding N uptake and assimilation is also necessary in any attempt to improve the N use efficiency of crops through the adaptation of N fertilization strategies. Nitrogen use efficiency of crops can also potentially be improved through manipulating the N use efficiency of the individual plants. A key question which then arises is which plant process should and could be improved. Given that individual plants within crops are exposed to complex environmental conditions, due to competition for light above ground and for nitrogen and water below ground, it is necessary to evaluate the consequences of manipulating a particular process at the plant level on the overall N use efficiency of the crop.

Past agronomical research into N uptake, and its relation to yield, has mostly been considered empirical. The evaluation of crop N requirements and the analysis of yield components in relation to N supply were mostly directed toward the general objective of satisfying crop N demand (Greenwood *et al.*, 1986). A more functional approach linking plant productivity with N supply was developed in plant ecology, however, this approach was mostly directed toward a search for adaptive strategies of species under contrasting natural environments (Lambers and Poorter, 1992). Research conducted in the last 15 years has allowed the development of a functional approach for crops based on the ecophysiology of N uptake, N distribution and growth (Lemaire, 2001). The first objective of this paper is to review and provide a framework for an agronomical and ecophysiological basis for such a functional approach. The second objective is to illustrate that the processes involved in crop N uptake and utilization are highly interrelated and can, therefore, only be fully understood through a dynamic and integrated approach.

An agronomical approach of crop growth: the concept of critical N concentration

Nitrogen uptake of field crops is highly variable within a single year, between years, between sites, and between crops, even when the N supplies from both the soil and additional fertilizer inputs are plentiful. In situations with a generous N supply, the variability in the dynamics of N accumulation in crops, under different climatic conditions and for different species, can be greatly reduced when crop N content (amount of crop N per unit ground area) is related to crop biomass rather than time (Greenwood *et al.*, 1986; Lemaire and Salette, 1984). This suggests that with an adequate soil N supply, crop N uptake is to a large extent determined by crop growth rate. However, the increase in crop N content with crop mass is not linear, additional N uptake per unit of additional biomass declines as a crop gets bigger. This nonlinear relationship between crop N content and crop biomass appears to be a general phenomenon for vegetative crops as it has been reported for many species (Greenwood and Barnes, 1978; Greenwood, 1982; Penning de Vries, 1982; Lemaire and Salette, 1984; Charles-Edwards *et al.*, 1986; Greenwood *et al.*, 1990; Angus and Moncur, 1985). In several instances, crop N content has been related to crop biomass through the allometric relationship $N_{content} = aW^b$, where W represents crop biomass per unit ground area (Lemaire and Salette, 1984; Greenwood *et al.*, 1990). It unequivocally follows that $N\% = a'W^{1-b}$, where N concentration ($N\%$) is the percentage of plant mass which is N, and where a and a' differ by a factor dependent on the units adopted for $N_{content}$ and for W. Critical N concentration ($N\%_{critical}$) is defined as the minimum crop N concentration required to achieve maximum crop growth rate (Greenwood *et al.*, 1986, 1991), and is analogous to the critical N content (minimum N content necessary to achieve maximum crop growth rate). The introduction of this concept has allowed rationalization of the analysis of field data. A statistical approach allowing the determination of critical N concentration of field-grown crops has been proposed (Justes *et al.*, 1994). Critical N concentration is not a goal in itself to follow during crop growth, but rather is a fundamental reference at any growth stage and in any environment, which allows the determination of whether crop N nutrition is supra-optimal (i.e. actual N content is in excess compared to the N content required for maximum growth rate), or sub-optimal with respect to crop growth rate (Fig. 1A). The discrepancy between actual $N\%$ and the corresponding critical $N\%$ at the same shoot biomass indicates the intensity of the N deficiency (or excess) experienced by a crop. Several authors have demonstrated that the reduction in crop growth rate is proportional to the ratio $N\%_{actual}/N\%_{critical}$ and have proposed the use of this ratio as a nitrogen nutrition index (Lemaire and Gastal, 1997). The usefulness of such an index is emphasized by the fact that the relationship between critical N concentration and crop mass does not vary substantially with major environmental factors, other than those affecting soil N supply (Lemaire and Salette, 1984; Lemaire and Denoix, 1987; Lemaire and Gastal, 1997).

Critical $N\%$–biomass curves have been evaluated for several major crops. Comparing a range of cultivated species, Greenwood *et al.* concluded that the major factor which discriminates critical $N\%$ curves between species was their metabolic type, C_3 or C_4 (Greenwood *et al.*, 1990). The C_4 species were found to have a lower critical $N\%$–biomass curve than C_3 species (Fig. 1B), presumably related to a lower content of photosynthetic proteins. Within the C_3 species, it was observed that legumes have critical $N\%$–biomass curves almost identical to the critical curves of other C_3 species (lucerne in Lemaire *et al.*, 1985; french beans in Greenwood *et al.*, 1990; pea in
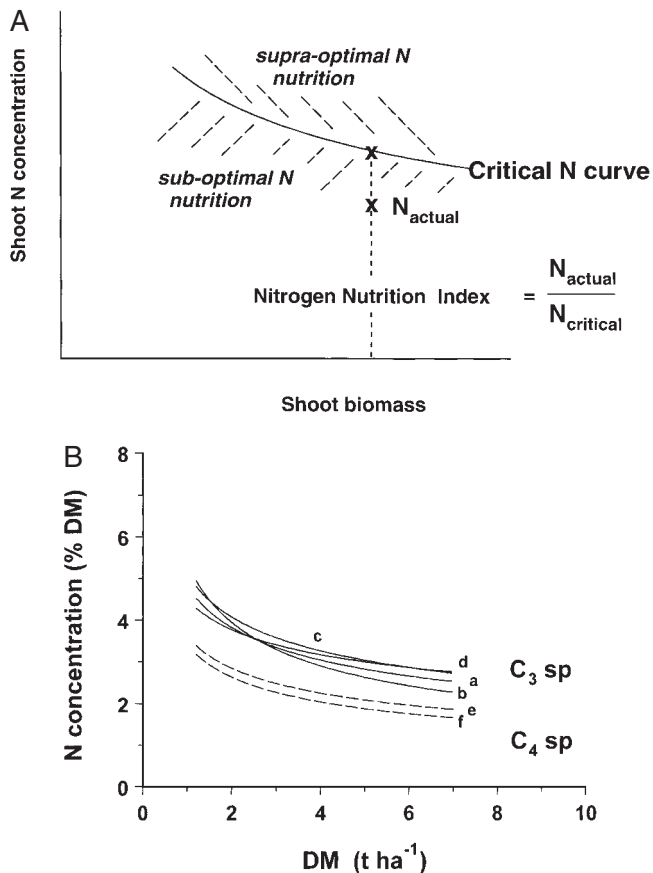
Fig. 1. (A) Critical N concentration curves: principles. (B) Relations available in the literature for different crops: a, perenial grasses ($N\%_c = 4.8W^{-0.33}$; Lemaire and Salette, 1984); b, wheat ($N\%_c = 5.35W^{-0.442}$; Justes *et al.*, 1994); c, pea ($N\%_c = 5.08W^{-0.32}$; Ney *et al.*, 1997); d, winter rape ($N\%_c = 4.48W^{-0.25}$; Colnenne *et al.*, 1998); e, maize, sorghum ($N\%_c = 4.1W^{-0.5}$; Greenwood *et al.*, 1990); f, maize ($N\%_c = 3.4W^{-0.37}$; Plénet and Lemaire, 1999).

Ney *et al.*, 1997). Since the early work (Greenwood *et al.*, 1990), critical N concentration has been evaluated in many cultivated species. The equations of the critical $N\%$–biomass curves now currently available in the literature (Fig. 1B), confirm the earlier observation (Greenwood *et al.*, 1990) that the differences in curve parameters between various C_3 species are small in comparison to the difference between C_3 and C_4 species. Indeed as parameter determination relies on a statistical approach, it is still debatable whether the value of the parameters reported for the C_3 species differ significantly or not.

What can be learnt from this critical N concentration approach? Firstly, it suggests that the N uptake rate of field-grown crops is regulated not only by soil availability but also by the crop growth rate. This is an important point, because crop N uptake has often been considered in relation either to soil availability (N supply approach), or to crop growth (N demand approach), rarely to both simultaneously. Secondly, as N uptake per unit biomass

decreases as crop mass increases, it suggests that the dependence between N uptake and growth is probably complex. This will be examined using the basis of fundamental physiological and ecophysiological processes in the following paragraphs.

Crop N uptake in relation to soil N

Nitrogen acquisition of crop plants is usually dominantly by the uptake of NO_3^- and NH_4^+, although soil organic nitrogen can be taken up by plants and may represent a significant proportion of total N absorption under particular ecological situations (acidic soils, low temperature environments). The mineral nitrogen content is generally greater in upper compared with lower soil layers, probably due to more favourable conditions for N mineralization in the upper part of the soil (higher content in organic matter; higher O_2 diffusion). This appears contrary to a dependence of crop N uptake on rooting depth. However, rooting depth determines the ability of a crop to intercept nitrate during periods of leaching and hence may be important from an environmental perspective. In this respect, not only the rooting depth of mature crops but also the rate at which roots of seedlings develop at depth will be important, particularly for crops which have an early phase of development during winter, the period where water drainage occurs most frequently. Rooting depth varies greatly between species and therefore each species requires individual evaluation. Several studies have indicated that soil N availability, although strongly altering shoot growth, does not significantly affect the dynamics of root growth at depth (Gabrielle *et al.*, 1998). The relatively small effect of N supply observed on rooting depth, in comparison to the large effect on shoot growth, probably relies on the decrease in root : shoot ratio observed with increasing N supply (Brouwer, 1962). Both root density and architecture also vary to a large extent between species (Fitter, 1991). In several species it has been observed that local NO_3^- application induces root proliferation due to an increased growth of laterals (Drew and Saker, 1975; Granato and Raper, 1989; Lambers *et al.*, 1982; Samuelson *et al.*, 1992; Robinson, 1994; Zhang and Forde, 2000). However, root growth responses to a localized N supply differ between species. In addition, a large range of root morphological plasticity in response to non-uniform distribution in soil N exists (Grime *et al.*, 1991; Robinson, 1994). As recently suggested, the impact of root proliferation on N uptake may be limited and more critical for plant-to-plant competition in N uptake, than for N uptake of a whole plant population such as a crop (Hodge *et al.*, 1999).

It has been reported in hydroponic studies that uptake of NO_3^- (or NH_4^+) depends on the NO_3^- (or NH_4^+)

concentration in the nutrient solution in a hyperbolic relationship often with multiphasic kinetics implying a complex regulation of uptake (Rao and Rains, 1976; Macduff *et al.*, 1989; Tischner, 2000; Glass *et al.*, 2002). Recently this has also been shown to apply to plants in the field throughout crop development (Devienne-Barret *et al.*, 2000). Interestingly, the latter study also showed that soil NO_3^- concentration regulates crop N uptake, not only under situations of low but also under situations of high soil NO_3^- concentration, when crop N is above its critical N concentration and where excess N accumulation in plants occurs. The regulation of whole plant and crop N uptake in heterogeneous soil remains poorly understood.

The amount of N taken up by the crop has a major impact on overall crop growth rate. The dependence of crop growth on crop N relies on several processes which will now be examined: leaf photosynthesis–N relationships, the distribution of N between leaves, leaf expansion and positioning and subsequent impacts on light interception (Novoa and Loomis, 1981; Sinclair and Shiraiwa, 1993).

Leaf N and leaf photosynthesis

The response of leaf photosynthesis to irradiance is largely dependent on the leaf N content. Photosynthetic proteins, including large amounts of Rubisco and, to a lesser extent, light harvesting complex proteins, represent a large proportion of total leaf N (Evans, 1983, 1989*a*; Field and Mooney, 1986). The physiological basis of the photosynthesis–leaf N relationship are reviewed in detail elsewhere (Lawlor, 2002). Numerous studies have reported that leaf photosynthesis at saturating light intensity (A_{max}) increases either linearly, or more generally asymptotically, with leaf N content (Evans, 1983, 1989*a*; Field and Mooney, 1986; see Grindlay, 1997, for a more comprehensive review). The A_{max}–leaf N relationship holds true whether leaf N content and A_{max} are expressed per unit leaf area or per unit leaf mass. The A_{max}–leaf N relationship shows a positive intercept on the N content axis, indicating that when leaf photosynthesis is theoretically zero leaves would still contain significant amounts of N, probably due to non-photosynthetic leaf N (Field and Mooney, 1986; Anten, 1995). The A_{max}–N relationship varies substantially in a wide range of plant species and plant types (Evans, 1989*a*). The variation is still significant within herbaceous species (Poorter and Evans, 1998), and at an intra-specific level (Nelson, 1988), but seems more limited within cultivated C_3 species (Evans, 1989*a*; van Keulen *et al.*, 1989), possibly as a result of selection pressure for yield.

By contrast to the large number of experimental reports on the A_{max}–N relationship, data concerning the impact of leaf N content on light use efficiency are scarce. In part this may be due to no significant effect of leaf N content being observed on many occasions (Anten, 1995). However, a significant effect of leaf N content on light use efficiency has been reported in several studies (Hirose and Werger, 1987; Pons *et al.*, 1989; Muchow and Sinclair, 1994; Drouet, 1998; Gastal and Lemaire, 1997). The influence of leaf N on leaf light use efficiency has a very limited impact on instantaneous canopy photosynthesis especially under a high incident light intensity and when leaf area is low. However, its impact on canopy photosynthesis when integrated over a day or longer growth periods becomes more important, due to the increased duration of periods of low light intensity, and also under a crop of high leaf area index when a greater proportion of the leaves are shaded. The magnitude of the effect of leaf N content on light use efficiency of the leaf is likely to be species-dependent, possibly in relation to observed species differences in the partitioning of leaf N between Rubisco and light harvesting complex proteins (Evans, 1989*a*). The effect of leaf N content on light use efficiency may be more important in field studies than in studies under artificial lamps, as has been observed for tall fescue (F Gastal, unpublished data). This may also partly explain the limited attention devoted to the effect of N on light use efficiency, as many leaf photosynthesis studies have been conducted on plants grown under artificial light.

Nitrogen distribution between leaves of the canopy

A large body of data indicates that the nitrogen distribution between the leaves of a canopy is not uniform (Grindlay, 1997). Individual leaves in a canopy experience different light environments due to shading by upper leaves; they also differ in age. In addition, different leaves in the canopy may develop under different conditions of N supply because of fluctuations in soil N supply during crop growth whilst leaf production remains continuous. All these aspects will potentially lead to the observed non-uniform N distribution.

Leaves exhibit a structural and functional acclimation of the photosynthetic apparatus to the light intensity experienced during their growth (Reyss and Prioul, 1975; Prioul *et al.*, 1980*a*; Bjorkman, 1981). This occurs even when light intensity is modulated on only part of the shoot (Evans, 1989*b*). The light acclimation is largely reversible within a few days (Prioul *et al.*, 1980*b*).

As a result of this light acclimation process, when light intensity varies with leaf position or is manipulated artificially through stand density, the leaves of a canopy show an increase in N content per area basis (N_L) and

an increase in A_{max} with increased light intensity received (on herbaceous species: Hirose and Werger, 1987; Pons *et al.*, 1993; Shiraiwa and Sinclair, 1993; Schieving *et al.*, 1992; Evans 1993*a*, *b*; Lemaire *et al.*, 1991; Anten *et al.*, 1995*a*, *b*; see Grindlay, 1997, for a thorough review and analysis). The effect of leaf age on N_L and A_{max} appears to be more limited than the light acclimation effect, though it is recognized that both effects occur concurrently (Hikosaka *et al.*, 1994; Schieving *et al.*, 1992).

Species with relatively horizontal leaves show a steeper gradient in N_L, as a function of leaf area index, than species with more erect leaves (Anten *et al.*, 1995*a*). Several studies have reported that the gradient in N_L with leaf area index is exponential, in parallel with the light gradient, (Hirose and Werger, 1987; Pons *et al.*, 1989; Anten *et al.*, 1995*a*), however, other reports found a more linear relationship (Lemaire *et al.*, 1991; Evans, 1993*a*, *b*; Shiraiwa and Sinclair, 1993; Grindlay *et al.*, 1995; Dreccer *et al.*, 2000). This variation in the leaf area index–N_L relationship suggests that, despite the general and broad N_L response to light, species may differ in their acclimation responses. Other aspects such as leaf age or N remobilization capacity of the leaf may interfere with the light acclimation resulting in variations in the N distribution profile. A further complication arises with monocot species during vegetative development; they have large standing leaves across the canopy without any specific stem tissue. Leaves thus experience large changes in radiation from their base to their tip. With *Carex acutiformis* a gradient in N_L was observed along the lamina in relation to received light intensity, but this was less steep than on a dicot grown in the same experiment (Schieving *et al.*, 1992). This may result from a larger proportion of N invested in non-photosynthetic tissues of lamina bases to physically support upper lamina portions (Grindlay, 1997).

It has been reported in lucerne (Lemaire *et al.*, 1991, 1992) and in sunflower (Sadras *et al.*, 1993) that lamina N concentration decreases to a smaller extent than N_L, both during crop development and from the upper to the lower leaves in a canopy. The difference between leaf N_L and leaf $N\%$ profiles is related to variations in specific leaf area, which increases with canopy development and within a canopy from the upper to the lower laminas. In grasses, specific leaf area appeared to increase to a lesser extent from top to bottom of the canopy than in dicots (on sorghum: Charles-Edwards *et al.*, 1986; in *Lolium perenne* and *Festuca arundinaceae*: F Gastal, unpublished data), or even was found to decrease in *Carex acutiformis* (Pons *et al.*, 1993). As mentioned previously, grasses have a relatively large mass of structural tissue (midribs, sclerenchymae) in lower portions of laminae. This explains the limited increase or even the decrease in specific leaf area from top to bottom of the

canopy, and is also related to the more limited differences in N_L than $N\%$ profiles.

In general, N supply significantly alters the leaf area index–N_L profile in canopies (Fig. 2). However, several studies show that the effect of N supply on N_L at the top of the canopy is somewhat limited (Shiraiwa and Sinclair, 1993; Grindlay *et al.*, 1995; Dreccer *et al.*, 2000). The decline in N_L with increasing cumulative leaf area index from canopy top or in relation to transmitted light within the canopy, tends to be steeper under low N supply, though differences between slopes of the regressions are not always significant (Grindlay *et al.*, 1995; Anten *et al.*, 1995*b*; Dreccer *et al.*, 2000; Fig. 2). The leaf N content (N_L) at the bottom of the canopy is often as low under ample N supply as under limited supply (Fig. 2), because of the substantially larger leaf area index achieved in well fertilized canopies. Thus, overall, under increasing crop N supply, the amplitude of variation in N_L from top to bottom of the canopy is larger than under a low N supply. The limited number of field studies where the distribution of leaf N has been examined in relation to N supply, contrasts with the large number of studies on leaf allocation in relation to light and photosynthesis. There are indications that N supply may affect leaf area index–N_L profiles differently according to the developmental stage of the crops (Dreccer *et al.*, 2000); particularly during grain filling (Sadras *et al.*, 1993). Further work is needed to understand how N supply
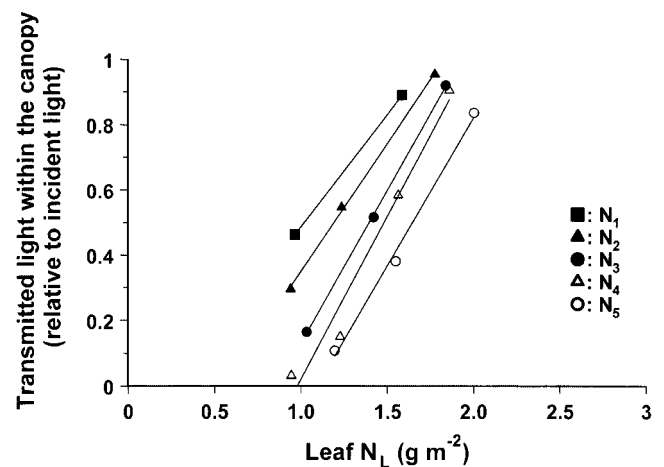

Fig. 2. Incidence of N supply on leaf N_L profile (N content per leaf area basis) from top to bottom of the canopy, plotted in relation to transmitted light within the canopy (F Gastal, unpublished data). Data were obtained on canopies of *Lolium perenne* in the field at the end of a spring growth period. The canopies received five N fertilizations rates at the beginning of the spring (50, 75, 100, 125, and 150 kg N ha^{-1} referred to as N_1, N_2, N_3, N_4, and N_5, respectively). The canopies were sampled by 10 cm deep layers from top to bottom of the canopy. Nitrogen content and leaf area were determined on leaves by canopy layer. (Transmitted PAR at depth z was calculated as $I_z/I_0 = \exp(-kLAI)$, where I_0 refers to incident PAR above the canopy, *LAI* refers to leaf area index and where k was taken from Bélanger *et al.*, 1992.)

affects leaf N distribution at different stages of crop growth and development.

Impact of leaf N distribution and photosynthesis–N relations on crop growth

From the top to the bottom of a canopy, individual leaves require progressively less N to maximize their carbon assimilation, due to light attenuation within the canopy and due to the lower amount of N needed to maximize leaf photosynthesis under lower light intensities. Hence it has been suggested that leaf N gradients observed in canopies represent a way to maximize carbon assimilation by the canopy (Mooney and Gulmon, 1979; Field, 1983; Hirose and Werger, 1987). In fact, it has been shown that the actual non-uniform N distribution between leaves in a canopy increases the daily canopy carbon assimilation by 20–40% (depending on species, daily radiation and model assumptions) compared with a theoretical uniform N distribution (Hirose and Werger, 1987; Sinclair and Shiraiwa, 1993; Anten, 1995; Chen *et al.*, 1993; Schieving *et al.*, 1992; Drouet, 1998; Warren and Adams, 2001). However, several studies conducted on various herbaceous species concluded that leaf N gradients experimentally observed in canopies are less steep than gradients that would maximize canopy photosynthesis (Hirose and Werger, 1987; Pons *et al.*, 1989; Evans 1993*b*; Anten *et al.*, 1995; Drouet, 1998). The adaptation of leaf N content to light intensity during leaf growth, and subsequent N mobilization related to changes in light environment of the leaf during canopy development, may apparently interfere with other physiological aspects or constraints (leaf age; N mobilization capacity of mature or senescing leaves).

Leaf N distribution within the canopy with its consequences for leaf and canopy photosynthesis, are major determinants of the radiation use efficiency (RUE) of a crop (Sinclair and Shiraiwa, 1993). Thus the relationships between N supply, leaf N distribution, and leaf and canopy photosynthesis discussed above, provide a fundamental basis to explain the almost systematic effect of crop N supply on RUE (Muchow and Davis, 1988; Sinclair and Horie, 1989; Bélanger *et al.*, 1992; Muchow and Sinclair, 1994; Bélanger and Richards, 1997). Most of these studies allow the relative effect of N on RUE and on light interception to be evaluated. The effect of N on light interception of the canopy is often substantial and additional to its effect on RUE (Muchow and Davies, 1988; Bélanger *et al.*, 1992; Muchow and Sinclair, 1994). Thus the effect of N on crop growth through its effect on leaf growth is generally as important as its effect on photosynthesis. However, the situation can arise where significant crop N shortage only occurs after canopy closure and thus after maximum light interception is achieved. In these situations, as illustrated by a study with maize where favourable soil N mineralization and rapid leaf area expansion occurred early in crop development despite a limited N fertilization rate (Plénet, 1995), the effect of N on crop growth is predominantly through its effect on RUE.

Since the effect of N on crop growth is generally due to both an effect of N on leaf photosynthesis and on light interception via leaf growth, it follows that there is a trade-off between allocation of N to maintain N_L and thus photosynthesis of existing leaves, and allocation of N to develop additional leaf area, as pointed out previously (Sinclair and Horie, 1989). Although the first aspect has been covered extensively, the balance between allocation of N to maintain leaf photosynthesis and allocation of N to production of new leaves is poorly investigated and, as a consequence, species strategies are almost not documented.

Crop N and leaf growth

Whatever the relative impact of N on crop growth via leaf area and leaf and canopy photosynthesis, N supply does have a systematic and large effect on leaf growth. As illustrated in Fig. 3, it has been shown in several instances, that N supply increases the leaf area of plants and canopies to a greater extent than leaf and canopy photosynthesis (Robson and Deacon, 1978; Gastal and Saugier, 1986; MacDonald *et al.*, 1986). The increase in leaf area of plants and canopies is brought about by a large effect of N supply on the expansion of individual leaves and on branching, or tillering in grasses (Wilman and Pearse, 1984; Gastal and Lemaire, 1988; Trapani and Hall, 1996; Taylor *et al.*, 1993; Vos and Biemond, 1992; Vos *et al.*, 1996). The rate of leaf appearance and
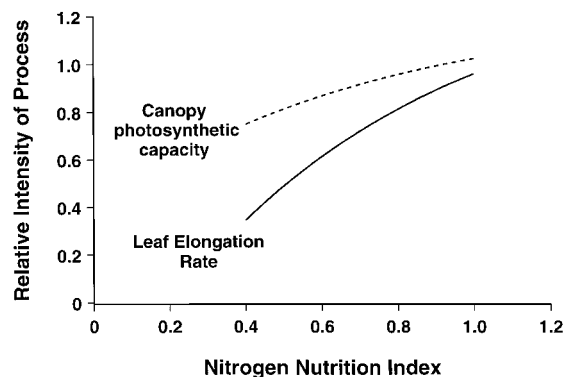
Fig. 3. Comparison of the sensitivity to nitrogen nutrition of leaf elongation and canopy photosynthetic capacity (canopy photosynthesis at a similar leaf area index for various N supply rates). (Redrawn from Gastal *et al.*, 1992, and Gastal and Bélanger, 1993.)

the duration of leaf expansion was little affected by N in many species (Gastal and Lemaire, 1988; Trapani and Hall, 1996; Taylor *et al.*, 1993; Vos and Biemond, 1992; Vos *et al.*, 1996). However, a number of other species do show a significant effect of N on the rate of leaf appearance (Cruz and Boval, 2000). Thus, the marked effect of N supply on leaf expansion and on branching appears a general response, whereas the effect on leaf appearance and leaf expansion duration is more species-specific.

In grasses, the reduction in leaf expansion due to a decreased N supply is often accompanied by an increase in fructans, both at the level of the whole plant and within leaf meristems (Volenec and Nelson, 1984), whilst in dicots starch often accumulates (MacDonald *et al.*, 1986; Paul and Stitt, 1993). The accumulation of non-structural carbohydrates in N-deficient plants suggests that carbohydrate supply is not the cause of the leaf growth reduction under low N supply, though this remains to be demonstrated definitively. Nitrogen supply alters the rates of cell division and cell expansion in growing leaves. In grasses, N supply greatly stimulates cell production rate, and the final cell length is little affected (MacAdam *et al.*, 1989; Gastal and Nelson, 1994; Fricke *et al.*, 1997). Whilst some studies have reported an increased cell elongation rate and a decreased duration of cell elongation in response to N supply (Gastal and Nelson, 1994; Fricke *et al.*, 1997), both processes appeared unaffected in other studies (MacAdam *et al.*, 1989; Gastal and Nelson, 1994). In all instances the impact of N on leaf expansion rate of grasses was related more to the effect of N on cell production than cell elongation rate. In dicots, early studies concluded that the impact of N supply on leaf growth was mostly due to an increased cell growth rate, because a larger final cell size was observed and also because N supply seemed to increase leaf water potential (Radin and Parker, 1979; Radin and Ackerson, 1981; Radin *et al.*, 1982). However, it was shown more recently that N supply does not significantly increase the turgor pressure of growing leaf tissue in grasses (Durand *et al.*, 1994; Fricke *et al.*, 1997), suggesting more an effect of N on wall expansion properties. Whether the response of leaf growth to N differs between grasses and dicots, as suggested earlier (Radin, 1983) remains an open question. It was recently shown in *Ricinus communis* that the relative impact of N on cell division and cell expansion depends on the developmental stage of the leaf (Roggatz *et al.*, 1999). A large effect of N on cell division was observed on this dicot species, similar to the observations on grasses, provided that N starvation was initiated early in leaf development, in the period of intense cell division (Roggatz *et al.*, 1999).

All the above elements underline that current understanding of the regulation of leaf growth in relation to N supply is extremely limited.

Nitrogen allocation to non-photosynthetic vegetative organs

As discussed above, much attention has been paid to the allocation of N to leaves, and even more specifically to lamina, within canopies. However, allocation of N to other vegetative tissues is also quantitatively important. Green leaf N content represented only 53% of shoot N in a lucerne crop (Lemaire *et al.*, 1992), and only 30% of shoot N in a wheat crop at the beginning of the grain-filling period (Grindlay *et al.*, 1995).

The leaf:stem ratio generally decreases as crop biomass increases (Lemaire *et al.*, 1992; Bélanger and McQueen, 1999; Bélanger and Richards, 2000). Therefore, a progressively greater proportion of C and N is allocated to the stem over the crop developmental period. In lucerne (Lemaire *et al.*, 1992) and reproductive ryegrass (F Gastal, unpublished data), the decrease in leaf:stem ratio over the growth period was accompanied by a much larger decrease in N concentration of stems than the limited decrease in N concentration of laminas reported earlier. As discussed earlier, in vegetative grasses where the stem is not developed, lamina bases contain a higher proportion of structural tissue and thus structural mass of lower N concentration, compared with lamina tips. Hence it is a common feature of both dicots and grasses, that an increasing proportion of shoot C and N is allocated to non-photosynthetic tissues during the growth of the crop, irrespective of whether this is achieved through alteration in the leaf:stem ratio or through changes in the proportion of photosynthetic and structural tissues within laminas.

The relative increase in the proportion of structural tissues is greater when plants are growing in dense canopies (Lemaire and Chartier, 1992; Lemaire and Gastal, 1997). Stem growth is enhanced under low blue light in dicots. In grasses, either a low level of light (Davies *et al.*, 1983; Wilson and Laidlaw, 1985; Casey *et al.*, 1999) or a reduction in the red/far-red ratio (Skinner and Simmons, 1993) at the leaf base, such as occurs during light competition, induces an increase in leaf growth and results in longer leaves with a higher cell wall content than shorter leaves (Duru *et al.*, 1999). There is growing evidence that increasing plant density leads to plant photomorphological reactions which determine to a large extent the observed changes in the allocation of C and N between photosynthetic and non-photosynthetic tissues. These considerations bring experimental support to the theory originally developed by Caloin and Yu (Caloin and Yu, 1984) and later by Charles-Edwards *et al.* (Charles-Edwards *et al.*, 1987), that the decline in shoot N concentration over the plant growth period, as observed in the relationship between critical N concentration and crop biomass, is determined by a decrease in the proportion of shoot material directly associated

with growth. Thus, the increase in leaf area during crop growth, achieved by building and positioning new leaves in the light, necessitates proportionally more structural tissues of low N content. The already mentioned trade-off between allocation of N to maintain photosynthetic activity of existing leaves, and allocation of N to build new leaf tissue in order to increase leaf area, is complicated by the additional cost in structural C and N.

Nitrogen supply also induces large changes in C and N allocation between roots and shoots. As discussed previously (Brouwer, 1962), increased N supply decreases the root:shoot ratio. Since this has been repeatedly verified in many studies, it will not be discussed further here, other than to mention that substantial variation between species in the intensity of the root:shoot response to nitrogen supply exists (Robinson, 1994). Root growth in response to N supply is also discussed in an earlier section of this paper.

Conclusion

Since soil N availability, N uptake and distribution, and finally crop growth are permanently interrelated during crop development and growth, the traditional view that crop N uptake and crop growth are either regulated by soil N supply or, alternatively, are regulated by crop demand, needs to be reconsidered and replaced by a more dynamic approach. The concept of critical N concentration has been developed extensively in the last decade. It allows the N status of crops to be precisely and dynamically quantified over their development, and thus represents a unique tool for the study of the various agronomical and ecophysiological aspects underlying the N–growth relationship. Critical N curves are now available on a number of major crops. On an ecophysiological point of view, researches conducted in the last decade have substantially improved current knowledge in several areas: response of root growth to local N supply; N partitioning between leaves of the canopy in relation to light environment and the consequences for plant and canopy photosynthesis; the allocation of N to non-photosynthetic organs, and particularly within shoots. Reviewing these recent advances in the context of a dynamic and integrated view of N uptake and distribution in crops also leads to the identification of a number of areas where current knowledge is particularly limited. The regulation of N uptake under heterogeneous soil N conditions, the regulation of leaf growth in relation to N supply and N uptake, and the regulation of N allocation between mature and growing leaves in relation to N supply, are among these areas where future research would improve an overall understanding of N uptake and distribution in plants and crops.

References

Angus JF, Moncur MW. 1985. Models of growth and development of wheat in relation to plant nitrogen. *Australian Journal of Agricultural Research* **36,** 537–544.

Anten NPR. 1995. Canopy structure and patterns of leaf nitrogen distribution in relation to carbon gain. Utrecht: Universiteit Utrecht.

Anten NPR, Schieving F, Werger MJA. 1995*a*. Patterns of light and nitrogen distribution in relation to whole canopy carbon gain in C_3 and C_4 mono- and dicotyledonous species. *Oecologia* **101,** 504–513.

Anten NPR, Schieving F, Medina E, Werger MJA, Schuffelen P. 1995*b*. Optimal leaf area indices in C_3 and C_4 mono- and dicotyledonous species at low and high nitrogen availability. *Physiologica Plantarum* **95,** 541–550.

Bélanger G, McQueen RE. 1999. Leaf and stem nutritive value of timothy grown with varying N nutrition in spring and summer. *Canadian Journal of Plant Sciences* **79,** 223–229.

Bélanger G, Richards JE. 1997. Growth analysis of timothy grown with varying N nutrition. *Canadian Journal of Plant Sciences* **77,** 373–380.

Bélanger G, Richards JE. 2000. Dynamics of biomass and N accumulation of alfalfa under three N fertilization rates. *Plant and Soil* **219,** 177–185.

Bélanger G, Gastal F, Lemaire G. 1992. Growth analysis of a tall fescue sward fertilized with different rates of nitrogen. *Crop Science* **32,** 1371–1376.

Bjorkman O. 1981. Responses to different quantum flux densities. In: Lange OL, Nobel PS, Osmond CB, Ziegler P, eds. *Encyclopedia of plant physiology*, New series, Vol. 12A. *Physiological plant ecology*. I. *Responses to the physical environment*. Berlin: Springer Verlag, 57–107.

Brouwer R. 1962. Distribution of dry matter in the plant. *Netherlands Journal Agricultural Science* **10,** 361–376.

Caloin M, Yu O. 1984. Analysis of the time-course of change in nitrogen content in *Dactylis glomerata* L. using a model of plant growth. *Annals of Botany* **54,** 69–76.

Casey IA, Brereton AJ, Laidlaw AS, McGilloway DA. 1999. Effects of sheath tube length on leaf development in perennial ryegrass (*Lolium perenne* L.). *Annals of Applied Biology* **134,** 251–257.

Charles-Edwards DA, Doley D, Rimmington GM. 1986. *Modelling plant growth and development*. Sydney: Academic Press.

Charles-Edwards DA, Stutzel H, Ferraris R, Beech DF. 1987. An analysis of spatial variation in the nitrogen content of leaves from different horizons within a canopy. *Annals of Botany* **60,** 421–426.

Chen JL, Reynolds JF, Harley PC, Tenhunen JD. 1993. Coordination theory of leaf nitrogen distribution in a canopy. *Oecologia* **93,** 63–69.

Colnenne C, Meynard JM, Reau R, Justes E, Merrien A. 1998. Determination of a critical nitrogen dilution curve for winter oilseed rape. *Annals of Botany* **81,** 311–317.

Cruz P, Boval M. 2000. Effect of nitrogen on some morphogenetic traits of temperate and tropical perennial forage grasses. In: Lemaire G, Hodgson J, de Morales A de F, Carvalho PC, Nabinger C, eds. *Grassland ecophysiology and grazing ecology*. Cambridge: University Press, 151–168.

Davies A, Evans ME, Exley JK. 1983. Regrowth of perennial ryegrass as affected by simulated leaf sheaths. *Journal of Agricultural Science* **101,** 131–137.

Devienne-Barret F, Justes E, Machet JM, Mary B. 2000. Integrated control of nitrate uptake by crop growth rate

and soil nitrate availability under field conditions. *Annals of Botany* **86**, 995–1005.

Dreccer MF, Van Oijen M, Schapendonk HCM, Pot CS, Rabbinge R. 2000. Dynamics of vertical leaf nitrogen distribution in a vegetative wheat canopy. Impact on canopy photosynthesis. *Annals of Botany* **86**, 821–831.

Drew MC, Saker LR. 1975. Nutrient supply and the growth of the seminal root system in barley. II. Localized compensatory increases in lateral root growth and rates of nitrate uptake when nitrate supply is restricted to only part of the root system. *Journal of Experimental Botany* **26**, 79–90.

Drouet JL. 1998. Variations de la disposition spatiale et de la teneur en azote des feuilles d'un peuplement de maïs en phase végétative. Etude par simulation de l'influence de ces variations sur la photosynthèse potentielle du couvert. Thèse INA-PG.

Durand JL, Onillon B, Gastal F. 1994. Turgor in the growth zone of tall fescue leaves at low water and nitrogen availibility: driving or driven force? Colloque inter disciplinaires du CNRS. *Biomécanique des Végétaux, France* **94**, 57–58.

Duru M, Feuillerac E, Ducrocq H. 1999. *In vitro* digestibility response of cocksfoot (*Dactylis glomerata* L.) to growth and defoliation: a simple model. *Journal of Agricultural Science* **133**, 379–388.

Evans JR. 1983. Nitrogen and photosynthesis in the flag leaf of wheat (*Triticum aestivum* L.). *Plant Physiology* **72**, 297–302.

Evans JR. 1989*a*. Photosynthesis and nitrogen relationships in leaves of C₃ plants. *Oecologia* **78**, 9–19.

Evans JR. 1989*b*. Partitioning of nitrogen between and within leaves grown under different irradiances. *Australian Journal of Plant Physiology* **16**, 533–548.

Evans JR. 1993*a*. Photosynthetic acclimation and nitrogen partitioning within a lucerne canopy. I. Canopy characteristics. *Australian Journal of Plant Physiology* **20**, 55–67.

Evans JR. 1993*b*. Photosynthetic acclimation and nitrogen partitioning within a lucerne canopy. II. Stability through time and comparison with a theoretical optimum. *Australian Journal of Plant Physiology* **20**, 69–82.

Field C. 1983. Allocating leaf nitrogen for the maximization of carbon gain: leaf age as a control on the allocation program. *Oecologia* **56**, 341–347.

Field C, Mooney HA. 1986. The photosynthesis–nitrogen relationship in wild plants. In: Givnish TJ, ed. *On the economy of plant form.* Cambridge: University Press, 25–53.

Fitter AH. 1991. The ecological significance of root system architecture: an economic approach. In: Atkinson D, ed. *Plant root growth. An ecological perspective.* Oxford: Blackwell Scientific Publications, 229–242.

Fricke W, McDonald AJS, Matson-Djos L. 1997. Why do leaves and leaf cells of N-limited barley elongate at reduced rates? *Planta* **202**, 522–530.

Gabrielle B, Justes E, Denoroy P. 1998. Modelling of temperature and nitrogen effects on the rooting dynamics of winter oilseed rape. *Actes du 16ème Congrès Mondial de Science du Sol, France* **98**, 7p.

Gastal F, Bélanger G. 1993. The effects of nitrogen fertilization and the growing season on photosynthesis of field-grown tall fescue (*Festuca arundinacea* Schreb.) canopies. *Annals of Botany* **72**, 401–408.

Gastal F, Bélanger G, Lemaire G. 1992. A model of the leaf extension rate of tall fescue in response to nitrogen and temperature. *Annals of Botany* **70**, 437–442.

Gastal F, Lemaire G. 1988. Study of a tall fescue sward grown under nitrogen deficiency conditions. *Proceedings of the XIIth General Meeting of European Grassland Federation, Ireland* **88**, 323–327.

Gastal F, Lemaire G. 1997. Nutrition azotée et croissance des peuplements végétaux cultivés. In: Morot-Gaudry JF, ed. *Assimilation de l'azote chez les plantes: aspects physiologiques, biochimiques et moléculaires.* INRA-Editions, Collections 'Mieux Comprendre', 355–367.

Gastal F, Nelson CJ. 1994. Nitrogen use within the growing leaf blade of tall fescue. *Plant Physiology* **105**, 191–197.

Gastal F, Saugier B. 1986. Alimentation azotée et croissance de la fétuque élevée. I. Assimilation du carbone et répartition entre organes. *Agronomie* **6**, 157–166.

Glass ADM, Britto DT, Kaiser BN, Kronzucker HJ, Kumar A, Okamoto M, Rawat SR, Siddiqi MY, Silim SM, Vidmar JJ, Zhuo D. 2001. Nitrogen transport in plants, with emphasis on the regulation of fluxes to match plant demand. *Zeitschrift für Pflanzenernährung Bodenkunde* **164**, 199–207.

Granato TC, Raper Jr CD. 1989. Proliferation of maize (*Zea mays* L.) roots in response to localized supply of nitrate. *Journal of Experimental Botany* **40**, 263–275.

Greenwood DJ. 1982. Modelling of crop response to nitrogen fertilizer. *Philosophical Transactions of the Royal Society, London*, Series B **296**, 351–362.

Greenwood DJ, Barnes A. 1978. A theoretical model for the decline in the protein content in plants during growth. *Journal of Agricultural Science, Cambridge* **91**, 461–466.

Greenwood DJ, Neeteson JJ, Draycott A. 1986. Quantitative relationships for the dependence of growth rate of arable crops on their nitrogen content, dry weight and aerial environment. *Plant and Soil* **91**, 281–301.

Greenwood DJ, Lemaire G, Gosse G, Cruz P, Draycott A, Neeteson JJ. 1990. Decline in percentage N of C₃ and C₄ crops with increasing plant mass. *Annals of Botany* **66**, 425–436.

Greenwood DJ, Gastal F, Lemaire G, Draycott A, Millard P, Neeteson JJ. 1991. Growth rate and % N of field grown crops: theory and experiments. *Annals of Botany* **67**, 181–190.

Grime JP, Campbell BD, Mackey JML, Crick JC. 1991. Root plasticity, nitrogen capture and competitive ability. In: Atkinson D, ed. *Plant root growth. An ecological perspective.* Oxford: Blackwell Scientific Publications, 381–397.

Grindlay DJC. 1997. Towards an explanation of crop nitrogen demand based on the optimization of leaf nitrogen per unit leaf area. *Journal of Agricultural Science, Cambridge* **128**, 377–396.

Grindlay DJC, Sylvester-Bradley R, Scott RK. 1995. The relationship between canopy green area and nitrogen in the shoot. In: Lemaire G, Burns IG, eds. *Diagnostic procedures for crop N management.* INRA-Editions, Collections 'Les Colloques', 53–60.

Hikosaka K, Terashima I, Katoh S. 1994. Effects of leaf age, nitrogen nutrition and photon flux density on the distribution of nitrogen among leaves of a vine (*Ipomea tricolor* Cav.) grown horizontally to avoid mutual shading of leaves. *Oecologia* **97**, 451–457.

Hirose T, Werger MJA. 1987. Maximising daily canopy photosynthesis with respect to the leaf nitrogen allocation pattern in a canopy. *Oecologia* **72**, 520–526.

Hodge A, Robinson D, Griffiths BS, Fitter AH. 1999. Why plants bother: root proliferation results in increased nitrogen capture from an organic patch when two grasses compete. *Plant, Cell and Environment* **22**, 811–820.

Justes E, Mary B, Meynard JM, Machet JM, Thellier-Huché L. 1994. Determination of a critical nitrogen dilution curve for winter wheat crops. *Annals of Botany* **74**, 397–407.

Lambers H, Poorter H. 1992. Inherent variation in growth rate between higher plants: a search for physiological causes and ecological consequences. *Advances in Ecological Research* **23,** 187–261.

Lambers H, Simpson RJ, Beilharz VC, Dalling MJ. 1982. Growth and translocation of C and N in wheat (*Triticum aestivum*) grown with a split root system. *Physiologia Plantarum* **56,** 421–429.

Lawlor DW. 2002. Carbon and nitrogen assimilation in relation to yield: mechanisms are the key to understanding production systems. *Journal of Experimental Botany* **53,** 773–787.

Lemaire G. 2001. Ecophysiology of grassland: dynamic aspects of forage plant populations in grazed swards. *Proceedings of the XIX International Grassland Congress, Brazil* **01,** 29–37.

Lemaire G, Chartier M. 1992. Relationships between growth dynamics and nitrogen uptake for individual sorghum plants growing at different densities. *Proceeding of the 2nd ESA Congress, UK* **92,** 98–99.

Lemaire G, Denoix A. 1987. Croissance estivale en matière sèche de peuplements de fétuque élevée (*Festuca arundinacea* Schreb.) et de dactyle (*Dactylis glomerata* L.) dans l'Ouest de la France. I. Etude en conditions de nutrition azotée et d'alimentation hydrique non limitantes. *Agronomie* **7,** 373–380.

Lemaire G, Gastal F. 1997. N uptake and distribution in plant canopies. In: Lemaire G, ed. *Diagnosis on the nitrogen status in crops.* Heidelberg: Springer-Verlag, 3–43.

Lemaire G, Salette J. 1984. Relation entre dynamique de croissance et dynamique de prélèvement d'azote pour un peuplement de graminées fourragères. I. Etude de l'effet du milieu. *Agronomie* **4,** 423–430.

Lemaire G, Cruz P, Gosse G, Chartier M. 1985. Etude des relations entre la dynamique de prélèvement d'azote et la dynamique de croissance en matière sèche d'un peuplement de luzerne (*Medicago sativa* L.). *Agronomie* **5,** 685–692.

Lemaire G, Onillon B, Gosse G, Chartier M, Allirand JM. 1991. Nitrogen distribution within a lucerne canopy during regrowth: relation with light distribution. *Annals of Botany* **68,** 483–488.

Lemaire G, Khaity M, Onillon B, Allirand JM, Chartier M, Gosse G. 1992. Dynamics of accumulation and partitioning of N leaves, stems and roots of lucerne in a dense canopy. *Annals of Botany* **70,** 429–435.

MacAdam JW, Volenec JJ, Nelson CJ. 1989. Effects of nitrogen on mesophyll cell division and epidermal cell elongation in tall fescue leaf blades. *Plant Physiology* **89,** 549–556.

MacDonald AJS, Lohammar T, Ericsson A. 1986. Growth response to step-decrease in nutrient availability in small birch (*Betula pendula* Roth). *Plant, Cell and Environment* **9,** 427–432.

Macduff JH, Jarvis SC, Mosquera A. 1989. Nitrate nutrition of grasses from steady-state supplies in flowing solution cultures following nitrate deprivation and/or defoliation. II. Assimilation of NO_3^- and short-term effect on NO_3^- uptake. *Journal of Experimental Botany* **40,** 977–984.

Mooney HA, Gulmon SL. 1979. Environmental and evolutionary constraints on the photosynthetic characteristics of higher plants. In: Solbrig OT, Jain S, Johnson GB, Raven Ph, eds. *Topics in plant population biology.* New York: Columbia University Press, 316–337.

Muchow RC, Davis R. 1988. Effect of nitrogen supply on the comparative productivity of maize and sorghum in a semi-arid tropical environment. II. Radiation interception and biomass accumulation. *Field Crop Research* **18,** 17–30.

Muchow RC, Sinclair TR. 1994. Nitrogen response of leaf photosynthesis and canopy radiation use efficiency in field grown maize and sorghum. *Crop Science* **34,** 721–727.

Nelson CJ. 1988. Genetic associations between photosynthetic characteristics and yield: review of the evidence. *Plant Physiology and Biochemistry* **26,** 543–554.

Ney B, Doré T, Sagan M. 1997. Grain legumes. In: Lemaire G, ed. *Diagnosis on the nitrogen status in crops.* Heidelberg: Springer-Verlag, 107–117.

Novoa R, Loomis RS. 1981. Nitrogen and plant production. *Plant and Soil* **58,** 177–204.

Paul MJ, Stitt M. 1993. Effects of nitrogen and phosphorus deficiencies on levels of carbohydrates, respiratory enzymes and metabolites in seedlings of tobacco and their response to exogenous sucrose. *Plant, Cell and Environment* **16,** 1047–1057.

Penning de Vries FWT. 1982. Crop production in relation to availability of nitrogen. In: Penning de Vries FWT, van Laar HH, eds. *Simulation of plant growth and crop production.* Wageningen: PUDOC, 213–221.

Plénet D. 1995. Fonctionnement des cultures de maïs sous contrainte azotée. Détermination et application d'un indice de nutrition. Thèse INPL-Nancy.

Plénet D, Lemaire G. 1999. Relationships between dynamics of nitrogen uptake and dry matter accumulation in maize crops. Determination of critical N concentration. *Plant and Soil* **216,** 65–82.

Pons TL, Schieving F, Hirose T, Werger MJA. 1989. Optimization of leaf nitrogen allocation for canopy photosynthesis in *Lysimachia vulgaris* L. In: Lambers H, Cambridge ML, Konings H, Pons TL, eds. *Causes and consequencces of variation in growth rate and productivity of higher plants.* The Hague: SPB Academic Press, 175–186.

Pons TL, van Rijnberk H, Scheurwater I, van der Werf A. 1993. Importance of the gradient in photosynthetically active radiation in a vegetation stand for leaf nitrogen allocation in two monocotyledons. *Oecologia* **95,** 416–424.

Poorter H, Evans JR. 1998. Photosynthetic nitrogen-use efficiency of species that differ inherently in specific leaf area. *Oecologia* **116,** 26–37.

Prioul JL, Brangeon J, Reyss A. 1980*a*. Interaction between external and internal conditions in the development of photosynthetic features in a grass leaf. I. Regional responses along a leaf during and after low-light or high-light. *Plant Physiology* **66,** 762–769.

Prioul JL, Brangeon J, Reyss A. 1980*b*. Interaction between external and internal conditions in the development of photosynthetic features in a grass leaf. II. Reversibility of light-induced responses as a function of developmental. *Plant Physiology* **66,** 770–774.

Radin JW. 1983. Control of plant growth by nitrogen: differences between cereals and broadleaf species. *Plant, Cell and Environment* **6,** 65–68.

Radin JW, Ackerson RC. 1981. Water relations of cotton plants under nitrogen deficiency. III. Stomatal conductance, photosynthesis and abscisic acid accumulation. *Plant Physiology* **67,** 115–119.

Radin JW, Parker LL. 1979. Water relations of cotton plant under nitrogen deficiency. I. Dependence upon leaf structure. *Plant Physiology* **64,** 495–498.

Radin JW, Parker LL, Guinn G. 1982. Water relations of cotton plants under nitrogen deficiency. V. Environmental control of abscisic acid accumulation and stomatal sensitivity to abscisic acid. *Plant Physiology* **70,** 1066–1070.

Rao KP, Rains DW. 1976. Nitrate absorption by barley. I. Kinetics and energetics. *Plant Physiology* **57,** 55–58.

Reyss A, Prioul JL. 1975. Carbonic anhydrase and carboxylase activities from plants (*Lolium multiflorum*) adapted to different light regimes. *Plant Science Letters* **5,** 189–195.

Robinson D. 1994. The responses of plants to non-uniform supplies of nutrients. *New Phytologist* **127,** 635–674.

Robson MJ, Deacon MJ. 1978. Nitrogen deficiency in small closed communities of S24 ryegrass. II. Changes in the weight and chemical composition of single leaves during their growth and death. *Annals of Botany* **42,** 1199–1213.

Roggatz U, McDonald AJS, Stadenberg I, Schurr U. 1999. Effects of nitrogen deprivation on cell division and expansion in leaves of *Ricinus communis* L. *Plant, Cell and Environment* **22,** 81–89.

Sadras VO, Hall AJ, Connor DJ. 1993. Light-associated nitrogen distribution profile in flowering canopies of sunflower (*Helianthus annuus* L.) altered during grain growth. *Oecologia* **95,** 488–494.

Samuelson ME, Eliasson L, Larsson C-M. 1992. Nitrate-regulated growth and cytokinin responses in seminal roots of barley. *Plant Physiology* **98,** 309–315.

Schieving F, Pons TL, Werger MJA, Hirose T. 1992. The vertical distribution of nitrogen and photosynthetic activity at different plant densities in *Carex acutiformis*. *Plant and Soil* **14,** 9–17.

Shiraiwa T, Sinclair TR. 1993. Distribution of nitrogen among leaves in soybean canopies. *Crop Science* **33,** 804–808.

Sinclair TR, Horie H. 1989. Leaf nitrogen, photosynthesis, and crop radiation use efficiency: a review. *Crop Science* **29,** 90–98.

Sinclair TR, Shiraiwa T. 1993. Soybean radiation-use efficiency as influenced by non-uniform specific leaf nitrogen distribution and diffuse radiation. *Crop Science* **33,** 808–812.

Skinner RH, Simmons SR. 1993. Modulation of leaf elongation, tiller appearance and tiller senescence in spring barley by far-red light. *Plant, Cell and Environment* **16,** 555–562.

Taylor G, McDonald AJS, Stadenberg I, Freer-Smith PH. 1993. Nitrate supply and the biophysics of leaf growth in *Salix viminalis*. *Journal of Experimental Botany* **44,** 155–164.

Tischner R. 2000. Nitrate uptake and reduction in higher and lower plants. *Plant, Cell and Environment* **23,** 1005–1024.

Trapani N, Hall AJ. 1996. Effects of leaf position and nitrogen supply on the expansion of leaves of field-grown sunflower (*Helianthus annuus* L.). *Plant and Soil* **184,** 331–340.

van Keulen H, Goudriaan J, Seligman NG. 1989. Modelling the effects of nitrogen on canopy development and crop growth. In: Russell G, Marshall LB, Jarvis PG, eds. *Plant canopies: their growth, form and function*. Cambridge University Press, 83–104.

Volenec JJ, Nelson CJ. 1984. Carbohydrate metabolism in leaf meristems of tall fescue. *Plant Physiology* **74,** 595–600.

Vos J, Biemond H. 1992. Effects of nitrogen on the development and growth of the potato plant. I. Leaf appearance, expansion growth, life span of leaves and stem branching. *Annals of Botany* **70,** 27–35.

Vos J, Biemond H, Struik PC. 1996. Dynamics of change of leaf attributes of brussels sprouts in response to switches between high and low supply of nitrogen. *Netherlands Journal of Agricultural Science* **44,** 31–42.

Warren CR, Adams MA. 2001. Distribution of N, Rubisco and photosynthesis in *Pinus pinaster* and acclimation to light. *Plant, Cell and Environment* **24,** 597–609.

Wilman D, Pearse PJ. 1984. Effect of applied nitrogen on grass yield, nitrogen content, tillers and leaves in field swards. *Journal of Agricultural Science, Cambridge* **103,** 201–211.

Wilson RE, Laidlaw AS. 1985. The role of the sheath tube in the development of expanding leaves in perennial ryegrass. *Association of Applied Biologists* **106,** 385–391.

Zhang H, Forde BG. 2000. Regulation of *Arabidopsis* root development by nitrate availability. *Journal of Experimental Botany* **51,** 51–59.