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MODELLING NITROGEN UPTAKE IN WINTER OILSEED RAPE BY USING INFLUX KINETICS OF NITRATE TRANSPORT SYSTEMS

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Abstract

A mechanistic model was proposed in order to predict nitrogen uptake by a culture of oilseed rape (*Brassica napus* L.), using independently measured characteristics of plants growing in hydroponic or under field conditions.Uptake kinetics of the different components (Constitutive and Inducible) of the Low and High Affinity Transport Systems of nitrate (CLATS, ILATS, CHATS and IHATS, respectively) were determined by $^{15}NO_3^-$ labelling in controlled conditions. The use of kinetic equations of transport systems and the experimental field data from the INRA-Châlons rape databank allowed to model NO_3^- uptake during the plant growth cycle. The study of different factors such as root temperature, day/night cycle and ontogenetic stages on NO_3^- uptake rate has been undertaken in order to improve the model prediction.Model outputs show that the high affinity transport system (HATS) accounted for about 90 % of total NO_3^- uptake (20 and 70 % for CHATS and IHATS without fertilization, respectively). The low affinity transport system (LATS) accounted for a minor proportion of total N uptake, and its activity was restricted to the early phase of the growth cycle. However, N autumnal fertilization increased the duration of its contribution (from 67 to 100 days) to total N uptake.

Keywords: *Brassica napus* L., High and low Affinity Transport Systems, modelling, nitrate uptake

Introduction

It is now well established that at least two nitrate uptake systems are involved in root nitrate uptake, their activity being affected by substrate concentrations : a low (LATS) and a high affinity transport system (HATS) which have two components one constitutive and the other inducible. Few models exist which establish relationships between root uptake kinetics of these different transport systems and final nitrate taken up by a plant culture under field conditions (Càrdenas et al., 1999).

Consequently, a mechanistic model for nitrate uptake by rape accounting for the four putative nitrate transporters and controlled by N supply was developed. Rape was chosen for modelling because of the availability of the experimental field data obtained from the INRA Châlons Rape database. Data used from this internet site (http://www-bioclim.grignon.inra.fr) were i) nitrate concentrations at different soil depths, ii) root biomass, iii) temperature and iv) total plant N content; all of them having been quantified at weekly intervals during the growth cycle and for different N fertilization. Integration of spatio-temporal scales from short times studies (uptake per se, transport systems activities) to long time period (plant cycle) was obtained by following nitrate uptake variations during the nychthemeral cycle and ontogenetic stages. Regulations of transport processes by exogenous and/or endogenous factors were formalised in the model and allow to simulate a regulated nitrate uptake capacity by the plants.

Material and Methods

Physiological experiments

Rape seeds (*Brassica napus* L.) were germinated and grown hydroponically in a greenhouse on a complete nutrient solution (Lainé et al., 1993) containing or not 1 mM KNO_3.

Uptake kinetics of nitrate transporters by 16-day-old plants grown previously with or without NO_3^- have been determined by measurements of $^{15}NO_3^-$ influx for a range of concentrations from 0 to 10 mM KNO_3. Effects of different factors such as day/night cycle (16h/8h) or ontogenetic stages, application of low temperatures (from 4 to 24 °C) to the roots have been tested on the activities of HATS and LATS (by measuring influx at 100 µM and 5 mM respectively). $^{15}NO_3^-$ influx was assessed by transferring plants on a solution containing $K^{15}NO_3$ (^{15}N excess of 99,8 %) during five minutes as described in Faure et al. (2000). Equations relating uptake rates of the four NO_3^- transporters and temperatures, time in the day/night cycle, or developmental stages have been ajusted to experimental data by using sigma plot software. Forms for the equations were chosen for their ability to describe realistically individual physiological processes and were of Michaelis Menten type for CHATS and IHATS and linear for CLATS and ILATS when activities were plotted against substrate concentration (Faure et al., 2000).

Model description

The model simulates the total nitrogen taken up by rape culture from the root transport processes formalised by kinetic equations of the four different nitrate transport systems.

The three input variables: nitrate concentrations in different depths of the soil, soil temperatures and root biomass needed to run the model were obtained from Grignon-Châlons rape databank. Lateral root biomass was estimated from taproot weight given in the databank and from lateral root/taproot ratio, quantified with plant grown in controlled conditions. Three auxiliary variables were introduced to integrate environmental factor regulations on N uptake

and their effects were deduced from the above described experiments : temperature, nychthemeral cycle and ontogenetic stages. This model was developed with Model Maker software (Cherwell Scientific).

Results and Discussion

The simulated N exported of a rape culture (no fertilizer applied) without any endogenous and/or exogenous regulations on the different transport systems is presented in figure 1A. When total N uptake was only controlled by soil NO_3^- availability, the inducible and constitutive components of the high affinity transport systems are predominantly involved in N uptake. The IHATS and the CHATS accounted for 75 and 17% of total simulated NO_3^- uptake, respectively. LATS component was involved only during the first 67 days of the culture . Thereafter, soil NO_3^- concentrations were not high enough to allow a significant contribution of the low affinity systems.

The introduction in the model of the temperature effect decreased by about two fold the amount of simulated total N uptake, and the decrease was higher in winter than autumn or spring seasons (Fig. 1B). Low temperatures affected more the HATS (active transport system) than the LATS (putative channel system) which is consistent with the enzymatic nature of transporters. The insert in the figure 1 C shows that the predicted N uptake in the model when high fertilization was applied was overestimated in winter compared to the real N taken up by the culture. It may be hypothesized that low temperature affect directly N transporters activities (taken into account) but also indirectly through a reduction of shoot growth.

Integration of day/night variations of nitrate influx and the subsequent diurnal regulations of nitrate transport processes by assimilates shows that nitrate uptake by the four transport systems was reduced by about 2 fold during the plant growth cycle when this later regulation was taken into account (Fig. 1B). This reduction was of the same order of magnitude for the HATS and the LATS (data not shown).

The ontogenetic effects corresponding to the regulation of nitrate influx in plants at different developmental stages were only observed on N exported at the end of the plant growth cycle (Fig. 1B). This factor is related to the root age, state of development and nutrient status of the plant. In fact, influx decreased dramatically during the flowering period. Integration of these three factors in the model reduced the estimation of total N taken up by the culture by about 6,5 folds (Fig. 1C). The comparison between the real N and the simulated N uptake by the plants differed only by a factor of 1,7 at the end of the growth cycle.

The model was tested experimentally to compare observed and predicted N uptake by rape culture after two N fertilization treatments (N0 : 0 N $kg.ha^{-1}$; N2 : 270 kg N. ha^{-1}). From these results, it clearly appears that the model matched better to experimental data when N fertilizer is added (data not shown). In the fertilized treatment, N uptake increased slightly compared to the control treatment (N0). High fertilization rate induced an increase of nitrate uptake by all the components of the transport systems except for the CHATS. This result was a consequence of higher activity (about 1,5 fold) and duration of functioning (extended from 67 to 100 days) of both LATS components after autumnal fertilization (Table 1).

Our modelling approach in comparison to other models considers that the balance between N uptake and N demand depends only on exogenous (temperature) and endogenous factors which regulate the functioning of the HATS and LATS components during the plant growth cycle in short (day/night cycle) and long time scale (ontogenetic development). Amongst the different integrated factors, the temperature was the most limitant and its effect was probably underestimated on shoot growth for each level of tested fertilization treatments. The satisfactory predicted values for high levels of fertilization in the model indicates that this mechanistic approach is powerful and could be easily improved.

References

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Table 1 - Simulation of total N taken up during the growth cycle of rape by each individual four transporters (CHATS, IHATS, CLATS, ILATS) and their duration of functioning during a rape culture not fertilized (N0) or receiving 270 kg $N.ha^{-1}$ (N2). Respective contribution of each transporter to total N uptake is given between brackets.

	N exported ($kg.ha^{-1}$)		Duration (days)	
	N0	N2	N0	N2
CHATS	64 (20)	60 (15.8)	280	280
IHATS	225.9 (70)	261.1 (68.6)	280	280
CLATS	6.9 (2.1)	11.9 (3.1)	67	100
ILATS	27.4 (8.5)	47.8 (12.5)	67	100
TOTAL	324.2	380.8	-	-

Figure 1 - Observed (■) and simulated (—) nitrogen exported (kg.ha^{-1}) by a rape culture (*Brassica napus* L.) after N fertilization treatment (N2: 270 kg.ha^{-1}). A, Respective contribution of the HATS and LATS components in the simulated N taken up without regulations. B, Independent effects of each auxiliary variable on simulated N uptake. C, Cumulative effects of endogenous and exogenous factors on the N uptake. Insert represents experimental and simulated results for N uptake.