The Responses of Maize Roots to Nitrogen Supply

By

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Abstract

Substantial quantities of costly nitrogen (N) fertilisers are applied to cereal crops each year to maximise yields, but only approximately half of the N is captured by cereals, providing scope to increase root N uptake. However, our understanding of how the nitrate (NO_3^-) uptake system is regulated and how it could be improved is limited. Furthermore, the changes to root morphology in response to NO_3^- supply are not well understood, in this case due to the difficulties associated with phenotyping roots in soil.

To investigate how the NO_3^- uptake system is up-regulated, maize (*Zea mays* var. B73 and Mo17) was grown hydroponically with low or sufficient NO_3^- supply, and a range of physiological parameters associated with NO_3^- uptake were measured across the transition from seed N use, to external N capture. This transition provides an ideal system to clarify how the NO_3^- uptake system up-regulates as this is when plants first rely on increasing root N capture to meet demand. Across both lines and treatments, concentrations of shoot N and free amino acids in roots and shoots rapidly decrease as seed N reserves exhaust. Once free amino acid concentrations decrease to a critical level, root NO_3^- uptake capacity rapidly increased, corresponding with a rise in transcript levels of putative NO_3^- transporter genes *ZmNRT2.1* and *ZmNRT2*.2. As NO_3^- uptake capacity reached maximum levels, shoot N concentrations stabilised. Despite shoot N concentrations stabilising, B73 was unable to maintain net N uptake and shoot growth in low NO_3^-, relative to sufficient NO_3^-. Conversely, Mo17 maintained shoot growth and net N uptake, and increased root mass in low NO_3^- relative to sufficient NO_3^-. The effects of NO_3^- limitation on growth were reflected by an increased root:shoot, which emerged just prior to shoot N concentrations stabilising.

In order to understand how root morphology may reflect the NO_3^- treatments differences observed in growth and net N uptake, morphological root traits were quantified

across seedling development. Analysis showed that although B73 achieved greater absorption area per unit root mass than Mo17, its morphology was unresponsive to NO_3^- supply. Conversely, Mo17 responded to NO_3^- limitation by increasing lateral and axial root length before increasing root mass or volume. Subsequently, 11 putative quantitative trait loci (QTL) associated with morphological root traits corresponding with shoot growth or N uptake were detected across low and sufficient NO_3^-, with one major QTL for lateral root length and surface area being identified in low NO_3^- on chromosome 5.

These results provide insight into the processes involved in up-regulating root NO_3^- uptake capacity and how root morphology can adapt to NO_3^- supply. These findings identify potential control points in the regulation of NO_3^- uptake capacity and root morphology, which may be investigated further via global transcriptional analysis or fine-mapping of identified QTL respectively. Ultimately, this work may lead to identification of candidate regulatory genes that could be either manipulated to generate new lines with enhanced N uptake efficiencies, or allow the identification of germplasm with this trait.

List of Abbreviations

ANOVA	analysis of variance
AvgD	average root diameter
AvgLRL	average lateral root length
AvgLRSA	average lateral root surface area
AvgLRV	average lateral root volume
AvgSM	average seed mass
AxR	axial root
B	boron
BLUPs	best linear unbiased predictors
C	carbon
Ca	calcium
CLC	chloride channel
Cu	copper
d	day
DAI	days after imbibition
DW	dry-weight
EDDHA	ethylenediamine-N,N'-bis(2-hydroxyphenylacetic acid)
EDTA	Ethylenediaminetetraacetic acid
Fe	iron
Fig	figure
FW	fresh-weight
Gln	glutamine
Glu	glutamate
GOGAT	glutamate synthase
GS	glutamine synthetase
h	hour
ha	hectares
HATS	high-affinity transport system
IBM	intermated B73 x Mo17
IcM	IBM centimorgans
IRIL	intermated recombinant inbred line

K	potassium
kg	kilograms
kg	kilogram
LATS	low-affinity transport system
LOD	logarithm of odds
LR	lateral root
Mg	magnesium
min	minute
Mn	manganese
Mo	molybdenum
MQ	milli-Q
Mt	megatonne
N	nitrogen
NAOH	sodium hydroxide
NAR	nitrate assimilation related
NH_4^+	ammonium
NiR	nitrite reductase
NO_2^-	nitrite
NO_3^-	nitrate
NPF	nitrate transporter 1/peptide transporter family
NR	nitrate reductase
NRT	nitrate transporter
NRT	nitrate transporter
NUE	nitrogen use efficiency
NUpE	nitrogen uptake efficiency
NUtE	nitrogen utilisation efficiency
P	phosphorous
PTR	peptide transporter
Q-PCR	quantitative polymerase chain reaction