



Genotypic variation and associated loci for lateral root density and length in rice (*Oryza sativa* L.)

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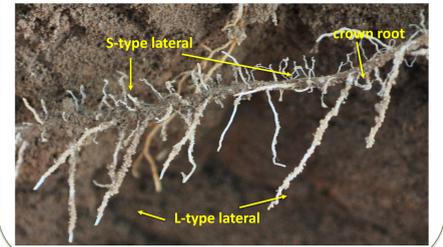
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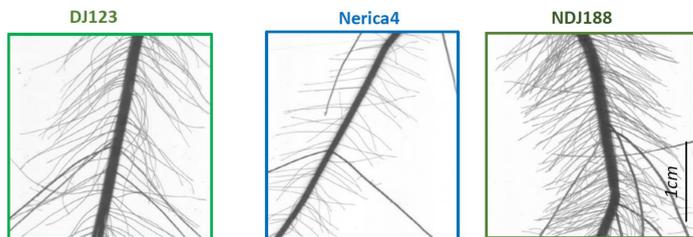
Introduction

The rice root system consists of two lateral root types, indeterminate larger **L-types** capable of further branching, and determinate, short, unbranched **S-types**. **L-types** correspond to the typical lateral roots of cereals whereas **S-types** are unique to rice. Both types contribute to nutrient and water uptake and our objectives were to assess whether genotypic variation for density and length of these laterals could be exploited in rice improvement to enhance adaptations to nutrient and water-limited environments.

2 types of lateral roots



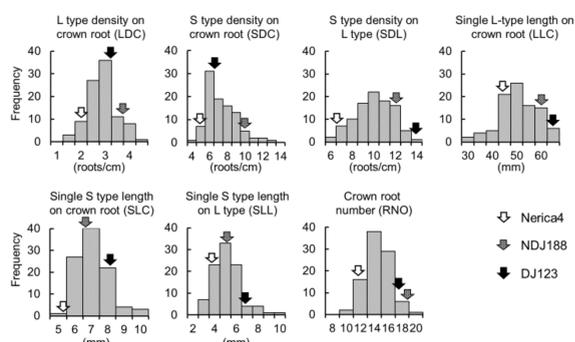
Genotypic variation for lateral density and length



Variation exists for density and length with parent NDJ 188 being superior to Nerica 4

QTL mapping population

Nerica4 x DJ123
 NDJ-188 x Nerica 4
 BC1F-5 (n=135)
 135 BC₁F₅ lines were phenotyped in the field on a P-deficient soil

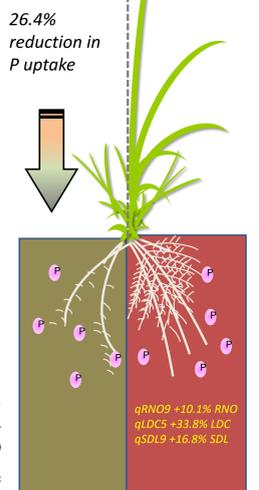
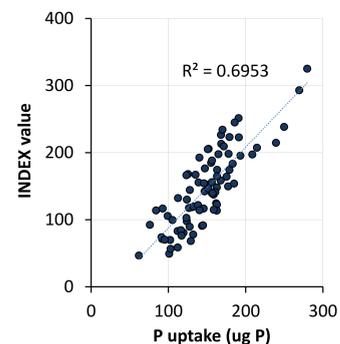


Lines of the QTL mapping population show ample variation, which is largest for SLL and LDC, but smaller for LLC and SLC

QTL identification & effects on P uptake

| Trait | QTL | Chromo some | Position (Mb) | LOD | R ² | Effect (%) |
|--------------------------------------|--------------|-------------|---------------|------|----------------|------------|
| L-type density on crown root (LDC) | <i>qLDC5</i> | 5 | 2.62 | 11.4 | 0.46 | 33.8 |
| S-type density on crown root (SDC) | <i>qSDC1</i> | 1 | 29.26 | 6.3 | 0.17 | 22.9 |
| S-type density on L-type (SDL) | <i>qSDL9</i> | 9 | 8.12 | 4.0 | 0.17 | 16.8 |
| Single S-type length on L-type (SLL) | <i>qSLL1</i> | 1 | 40.87 | 5.5 | 0.24 | 4.2 |
| Root Number (RNO) | <i>qRNO9</i> | 9 | 12.92 | 3.6 | 0.17 | 10.1 |

Stepwise linear regression analysis indicated **LDC** and **RNO** were most influential traits, followed by **SDL**. We derived an **INDEX** for each line of the mapping population based on its variation for these 3 traits. This index explained 70% of the variation for P uptake within the mapping population. Selecting for the 3 associated QTL therefore is promising to improve P uptake.

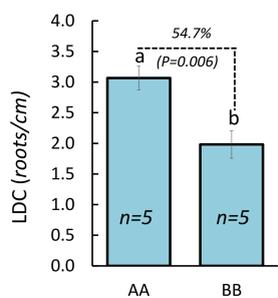


Estimation of QTL effects for P uptake

The OpenSimRoot rice model is built on DJ123 (Gonzalez et al. 2021) and effects of replacing DJ123 alleles with negative Nerica4 alleles were simulated. Estimated allelic effects at *qLDC5*, *qRNO9* and *qSDL9*, with resulting changes in root size and plant P uptake are shown for each QTL and the combination of all 3, in which case P uptake was affected by 26.4%

| Trait | Base | RNO (-10.1%)* | | LDC (-33.8%)* | | SDL (-16.8%)* | | RNO+LDC +SDL | |
|-------------------------------|--------|---------------|------|---------------|------|---------------|------|--------------|------|
| | | Value | % | Value | % | Value | % | Value | % |
| Total root surface area (RSA) | 257.5 | 230.1 | 89.3 | 222.0 | 86.2 | 251.4 | 97.6 | 206.6 | 80.2 |
| Total root length (RL) | 2888.8 | 2537.8 | 87.9 | 2079.2 | 72.0 | 2717.5 | 94.1 | 1916.3 | 66.3 |
| RL crown roots | 648.3 | 585.5 | 90.3 | 638.5 | 98.5 | 638.0 | 98.4 | 590.2 | 91.0 |
| RL L-type lateral roots | 1012.4 | 887.1 | 87.6 | 608.2 | 60.1 | 987.4 | 97.5 | 582.7 | 57.6 |
| RL S-type lateral roots | 1228.1 | 1065.2 | 86.7 | 832.6 | 67.8 | 1092.1 | 88.9 | 743.4 | 60.5 |
| P uptake day 30 (µg P) | 31.2 | 27.5 | 88.2 | 24.8 | 79.6 | 30.0 | 96.1 | 23.0 | 73.6 |

QTL confirmation



Having positive alleles at *qLDC9* increases LDC from 2 to 3 L-type roots per cm crown root.

Dry shoot weight (DSW) of contrasting groups of lines homozygous for the DJ123 (AA) or Nerica4 (BB) allele at QTL *qLDC5*, *qSDL9*, and *qRNO9*.

| QTL name | Allele | DSW (mg) | SD | n | |
|--|--------|----------------------|--------------------|------|----|
| <i>qLDC5</i> | AA | 390.8 ^{ab} | 79.2 | 10 | |
| | BB | 337.7 ^{bc} | 62 | 18 | |
| | % | 15.8 | | | |
| <i>qRNO9</i> | AA | 370.2 ^{ab} | 20.4 | 19 | |
| | BB | 315.84 ^{bc} | 31.6 | 10 | |
| | % | 17.3 | | | |
| <i>qLDC5</i> + <i>qRNO9</i> | AA | 433.4 ^a | 42.2 | 5 | |
| | BB | 307.5 ^{bc} | 17 | 6 | |
| | % | 41.1 | | | |
| <i>qLDC5</i> + <i>qSDL9</i> + <i>qRNO9</i> | AA | 433.4 ^a | 42.2 | 5 | |
| | BB | 307.5 ^{bc} | 17 | 6 | |
| | % | 41.1 | | | |
| Nerica4 | BB | 287.9 ^c | 51.1 | 10 | |
| | DJ123 | AA | 414.3 ^a | 42.4 | 19 |
| | | % | 43.9 | | |

Conclusions and outlook

- We identified a major QTL for L-type density on crown roots (*qLDC5*) that may increase P uptake by about 20%.
- Pyramiding *qLDC5* with *qRNO9* and *qSDL9* is possible given that trade-offs between traits were not detected. Phenotypic selection for RNO during variety development would be feasible. However, the costs of doing so reliably for lateral root density traits is prohibitive and markers identified here therefore provide the first opportunity to incorporate such traits into a breeding program.
- Breeding lines combining above QTL are currently being tested without supplementary irrigation in farmers' fields in Madagascar. One line has been accepted as a new variety in Madagascar (see right)
- The Aux/IAA family gene *IAA19* is located in proximity to *qLDC5*. Other auxin-related genes (*IAA5* and *IAA6*) are located near *qSDC1*. The potential role of these IAA homologs should be investigated further.



New upland rice variety to be released in Madagascar. It is derived from the Nerica4 x DJ123 mapping population and has superior root development on low-P soil. Picture © N Ranaivo

Check it out <https://sites.google.com/site/wissuwaresearchgroup/home?pli=1>

• Dinh LT, Ueda Y, Gonzalez D, ... Wissuwa M (2023) Novel QTL for lateral root density and length improve phosphorus uptake in rice (*Oryza sativa* L.). Rice 16:37 doi.org/10.1186/s12284-023-00654-z
 • Ranaivo NH, Dinh TL, Ueda Y, ... Wissuwa M (2022) QTL mapping for early root and shoot vigor of upland rice (*Oryza sativa* L.) under P deficient field conditions in Japan and Madagascar. Front Plant Sci: 101741919
 • Gonzalez D, Postma J, Wissuwa M (2021) Cost-benefit analysis of the upland-rice root architecture in relation to phosphate: 3D Simulations highlight the importance of S-type lateral roots for reducing the pay-off time. Frontiers in Plant Nutrition 12: doi.org/10.3389/fpls.2021.641835.