

# PHENOTYPIC VARIATION, MAPPING AND QTL ANALYSIS OF ADVENTITIOUS ROOTING UNDER LOW PHOSPHORUS STRESS IN COMMON BEAN (*Phaseolus vulgaris* L.)

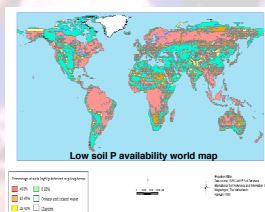
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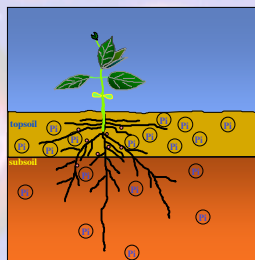
## Problem

- Over half of global bean production occurs on extremely P-deficient soils.
- Phosphorus distribution is highly heterogeneous in most soils, generally being greatest in surface horizons and decreasing with depth.
- P efficiency is a typical quantitative trait that is subject to great environmental variability.
- Evaluations of root traits associated to mechanisms of tolerance are laborious and time consuming.



## Approach

- Adventitious roots are the shallowest portion of bean root system and might explore topsoil horizons more efficiently than other root types (Lynch and Brown 2001).
- Genotypic variation was observed for both inherent ability to produce adventitious roots as well as the plasticity of adventitious rooting in response to phosphorus stress (Miller, Ochoa et al. 2003).
- Recombinant Inbred Lines (RILs) from contrasting parents for adventitious rooting will permit the genetic mapping of this trait, which will be important in understanding its genetic basis, and will enable the generation of DNA markers to assist breeding programs.



## Objectives

- To characterize the phenotypic variation and genetic regulation of adventitious rooting in phosphorus limiting environments
- To develop a genetic linkage map and identify putative QTLs for adventitious root traits under high and low P conditions

## Materials and Methods

- Locations:
  - Field (F): Darien, Colombia (6-week-old plants, 3 reps)
  - Greenhouse (GH): PSU, PA, USA (2-week-old plants, 5 reps)
- Phosphorus treatments:
  - High P: 45 kg P<sub>2</sub>O<sub>5</sub> (F) or 100 µM buffered Alumina-P (GH).
  - Low P: 7.5 kg P<sub>2</sub>O<sub>5</sub> (F) or 1.8 µM buffered Alumina-P (GH).
- Plant Material
  - Parents
    - G2333: small seeded, race M from Mesoamerican gene pool.
    - G19839: large seeded, race N from Andean gene pool.
  - Mapping population: 84 F<sub>8</sub> Recombinant Inbred Lines (RILs).
- Root image analyses: Adventitious roots for the whole RIL population were counted, scanned with WinRhizo program for getting root length, and oven-dried for biomass determination.
- Map construction: A genetic linkage map based on 149 genetic markers including 76 SSR, 60 RAPD, 8 SCAR, 3 STS, 1 morphological (flower color), and 1 biochemical marker (phaseolin) was constructed using MAPMAKER/EXP 3.0 (Lander et al., 1987) and MAPDISTO (Lorieux, 2000) computer software programs to assign markers order.
- QTL identification: Putative QTLs for the traits were identified by employing interval mapping analysis and QTL Cartographer software (Basten et al., 1999).

## References

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## Results and Discussion

Table 1. Mean values, probabilities between parents and among RILs, and heritability values (h<sup>2</sup>).

| Adventitious root traits                  | Parents                           | Mean         | Range        | P <sub>int</sub> | h <sup>2</sup> |
|---|-----------------------------------|--------------|--------------|------------------|----------------|
| Field                                     |                                   |              |              |                  |                |
| Number                                    | HP 34.6 ± 2.6<br>LP 33.0 ± 2.6    | 31.8 ± 0.7   | 12.0 - 52.0  | ***              | 0.67           |
| Biomass (mg)                              | HP 7.0 ± 2.0<br>LP 7.8 ± 2.7      | 11.0 ± 0.4   | 3.4 - 19.4   | ***              | 0.49           |
| Length (m)                                | HP 153.1 ± 17.4<br>LP 81.1 ± 14.2 | 117.5 ± 22.3 | 22.3 - 306.9 | ns               | 0.19           |
| Specific Root Length (m g <sup>-1</sup> ) | HP 2.1 ± 0.8<br>LP 1.7 ± 0.6      | 3.9 ± 0.2    | 0.4 - 10.9   | ***              | 0.64           |
| Greenhouse                                |                                   |              |              |                  |                |
| Number                                    | HP 34.6 ± 2.6<br>LP 33.0 ± 2.6    | 31.8 ± 0.7   | 12.0 - 52.0  | ***              | 0.70           |
| Biomass (mg)                              | HP 7.0 ± 2.0<br>LP 7.8 ± 2.7      | 11.0 ± 0.4   | 3.4 - 19.4   | ***              | 0.58           |
| Length (m)                                | HP 153.1 ± 17.4<br>LP 81.1 ± 14.2 | 117.5 ± 22.3 | 22.3 - 306.9 | ns               | 0.19           |
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\*, \*\*, \*\*\* Significant differences between the two parents (P<sub>0</sub>) or among RILs (P<sub>int</sub>) at P=0.10, 0.05, 0.01, 0.001, respectively. ns Not significant at P=0.10.

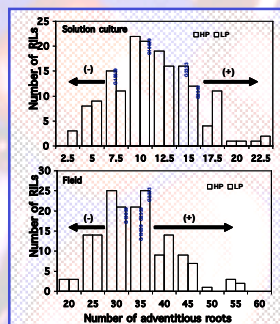


Figure 1. Frequency distribution of number of adventitious roots

Table 2. Genomic location, percentage of phenotypic variation, and genetic effect of QTLs for adventitious root traits grown under high and low P conditions in the field (F) and greenhouse (G).

| Adventitious Traits | Linkage group | Closest QTL Marker | Position (cM) | LOD  | R <sup>2</sup> | Additivity |
|---------------------|---------------|--------------------|---------------|------|----------------|------------|
| High P              |               |                    |               |      |                |            |
| Number (F)          | 7             | V100.29            | 102.8         | 3.68 | 0.14           | -3.53      |
|                     | 9             | P070.45            | 60.9          | 4.21 | 0.20           | 3.67       |
| Biomass (F)         | 2             | P101.6             | 52.1          | 3.95 | 0.18           | -2.66      |
|                     | 6             | P010.6             | 37.4          | 3.74 | 0.24           | -2.99      |
|                     | 9             | M130.34            | 57.9          | 4.47 | 0.23           | 3.14       |
| Biomass (G)         | 8             | W090.36            | 121.8         | 4.02 | 0.18           | 0.93       |
| Length (F)          | 2             | P101.6             | 49.1          | 3.66 | 0.18           | -2.88      |
|                     | 9             | M130.34            | 57.9          | 5.55 | 0.25           | 3.63       |
|                     | 11            | Bmy002             | 127.9         | 3.74 | 0.15           | 2.96       |
| Length (G)          | 8             | W090.36            | 121.8         | 4.38 | 0.18           | 1.20       |
| SRL (F)             | 2             | Q171.7             | 104.4         | 3.59 | 0.15           | 9.45       |
|                     | 7             | Phe                | 16.2          | 3.30 | 0.18           | 9.44       |
| SRL (G)             | 6             | U190.35            | 75.9          | 3.24 | 0.11           | 16.44      |
|                     | 7             | Bm185              | 4.7           | 3.20 | 0.11           | 15.35      |
|                     | 10            | M090.18            | 90.5          | 3.32 | 0.11           | 16.67      |
| Low P               |               |                    |               |      |                |            |
| Number (F)          | 2             | P101.6             | 57.3          | 3.37 | 0.13           | -2.86      |
|                     | 9             | P070.45            | 60.9          | 6.63 | 0.36           | 4.84       |
| SRL (F)             | 2             | Bm142              | 66.4          | 5.14 | 0.25           | -8.48      |
|                     | 4             | Bm140              | 50.2          | 4.36 | 0.21           | 13.41      |
| SRL (G)             | 4             | Bm027              | 56.9          | 5.65 | 0.23           | 14.25      |
|                     | 8             | Bm153              | 93.4          | 4.09 | 0.18           | 13.35      |

## Mapping and QTL analysis

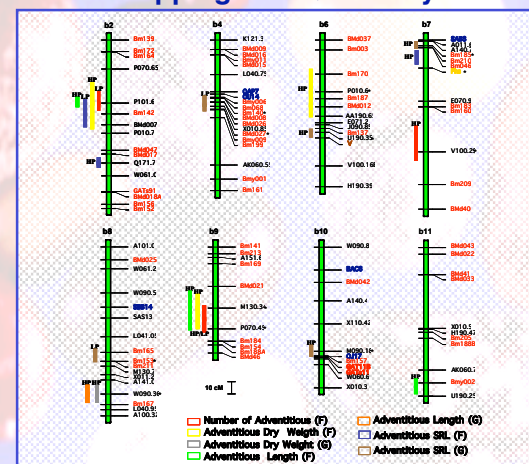


Figure 2. Molecular Markers and map location of putative QTLs for several adventitious root traits in G2333/G19839 RIL population under high and low P conditions in the field and greenhouse.

- One hundred forty nine markers were assigned into 11 linkage groups with a total cumulative map length of 1175 cM. These markers included 76 SSR, 60 RAPD, 8 SCAR, 3 STS, 1 morphological (flower color), and 1 biochemical marker (phaseolin).
- A total of 21 QTL were detected for number, biomass, length and specific root length in 8 out of 11 linkage groups mapped (Fig 2).
- These genomic regions accounted for 13 to 65% of the total phenotypic variation for those adventitious traits in the field and 18% to 34% under greenhouse conditions (Table 2).
- Under field conditions two major QTLs in linkage groups b7 and b9 in high P and two additional QTLs in linkage groups b2 and b9 in low P accounted for 34% and 49% of the total phenotypic variation observed for this trait respectively (Table 2).

## Conclusions

- There is genetic variation and transgressive segregation for adventitious rooting that could be useful for dissecting its physiological function under low phosphorus conditions
- A molecular linkage map has been developed, which allowed for the identification of QTLs for adventitious root traits specifically related to low phosphorus adaptation.
- Some QTLs for adventitious rooting differed depending on P treatment (high vs. low) and growing conditions (solution culture vs. field).
- Adventitious rooting under low P is a feasible target for bean breeding.
- Our map will allow the identification of some other root traits involved in low phosphorus adaptation mechanisms or other characteristics by which these two parents are contrasting (Checa et al., 2004; Blair et al., 2004).
- Simultaneous improvement of adventitious root parameters may be possible using marker assisted selection.

## Acknowledgments

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