Response of AVP1-OX Romaine Lettuce to Phosphorus

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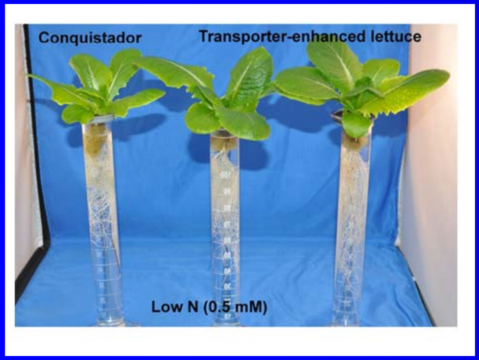


Concerns

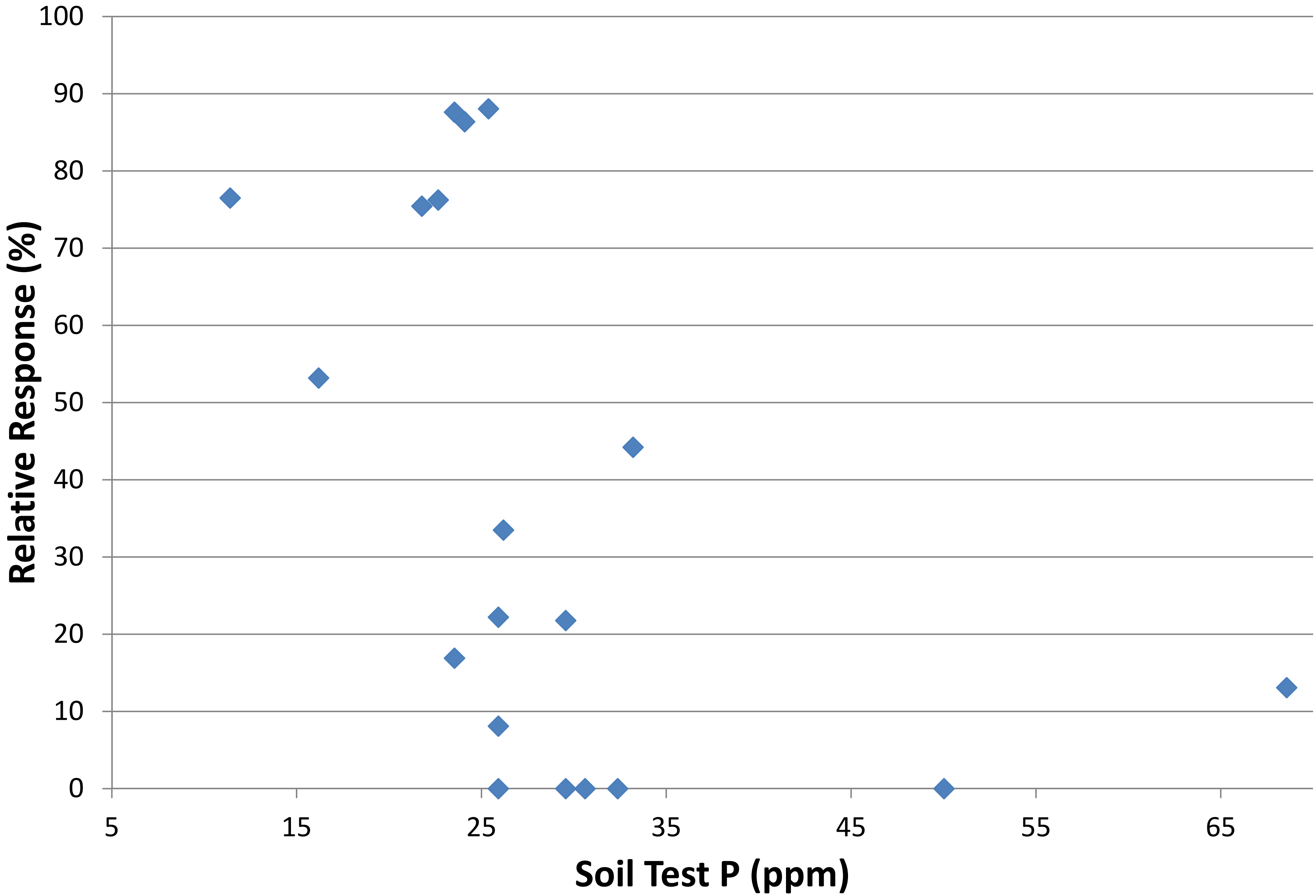
- P in runoff and drainage often has adverse ecological impacts on surface waters.
- World P reserves are rapidly declining and there is concern that a shortage of P fertilizers will ultimately compromise world food production (Vaccari, 2009).
- P fertilizer reserves are concentrated in a few nations.

P reactions in calcareous soils

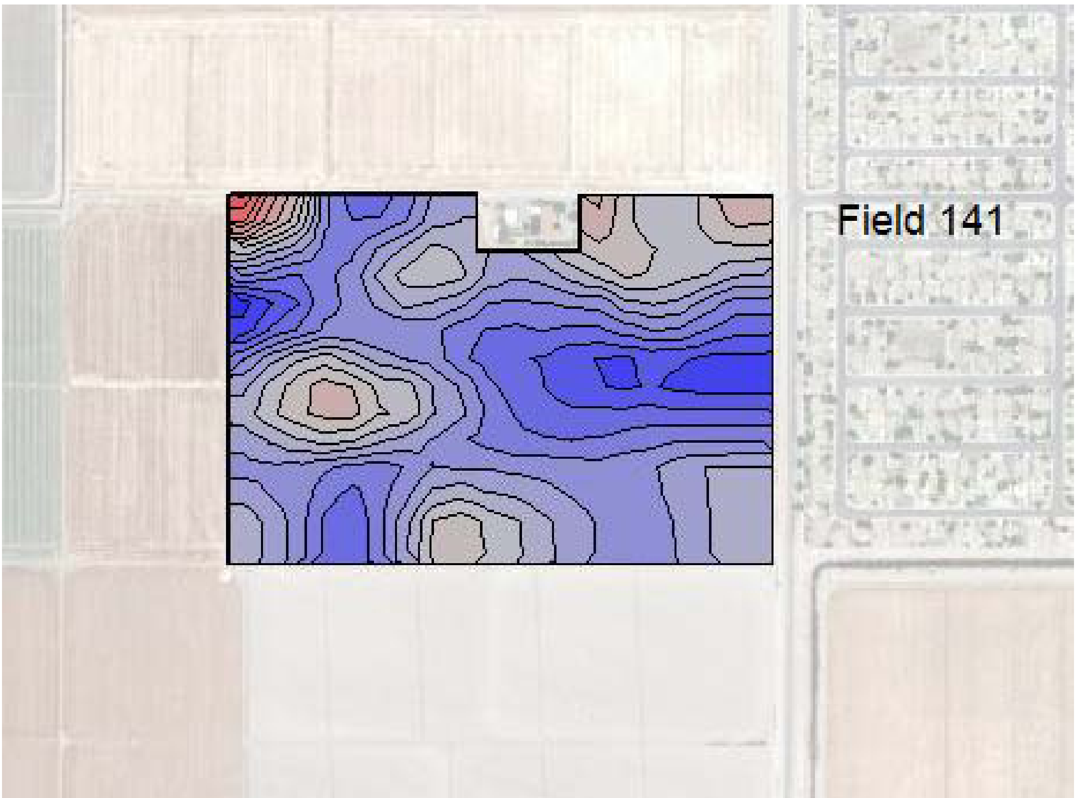
- **The reaction of P with $CaCO_3$ consist of initial sorption reactions followed by precipitation with increasing concentrations of P (Cole, 1953; Griffin and Jurinak, 1973; Holford and Mattingly, 1975).**
- **Most added P would precipitate initially as dicalcium phosphate dihydrate (DCPD) and dicalcium phosphate (DCP) (Lindsay, 1979).**
- **These products undergo a slow conversion to such compounds as octacalcium phosphate (OCP), tricalcium phosphate, (TCP) or one of the apatites (Lindsay and Moreno, 1960).**

Strategies for Improving P Efficiency

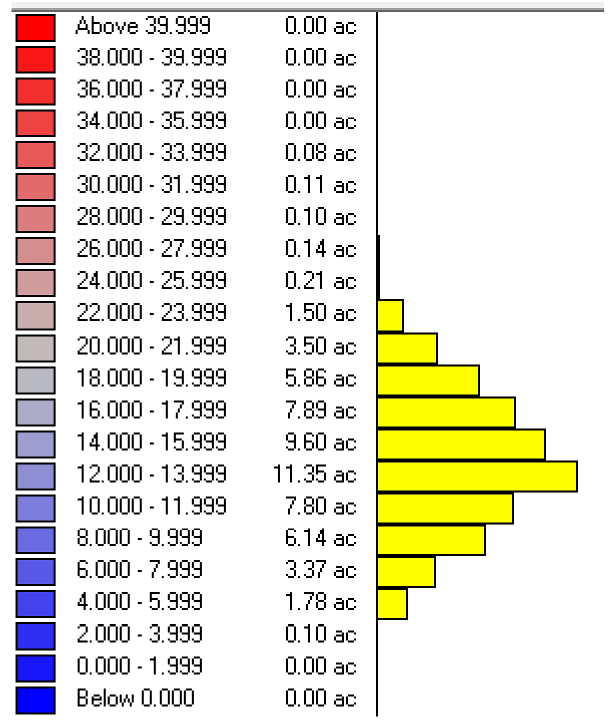
- Soil Testing and Plant Analysis
- P Placement
- Fertilizer technology
- Genetic modification



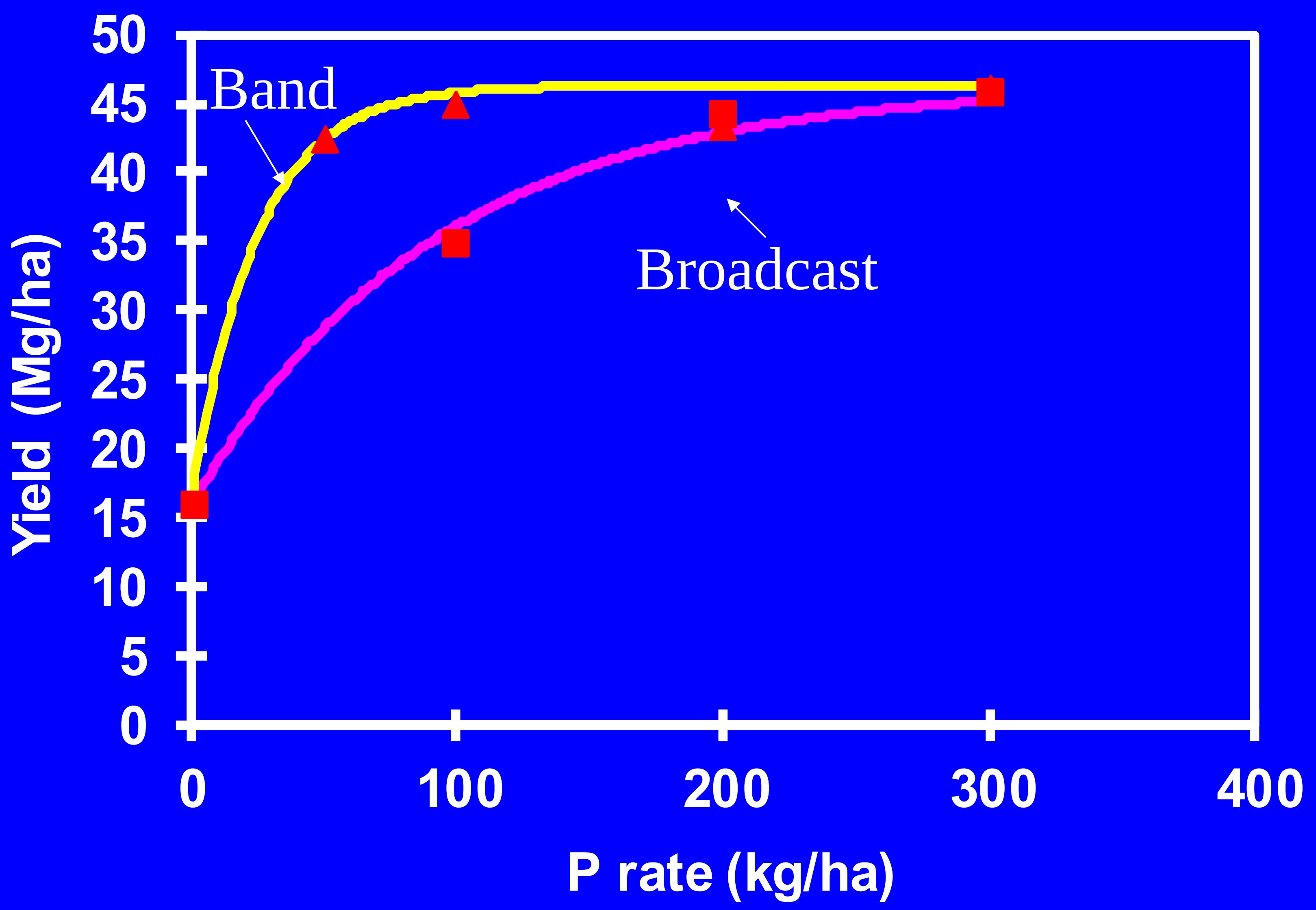
Relative Response (%)
100
90
80
70
60
50
40
30
20
10
0
5
15
25
35
45
55
65
Soil Test P (ppm)



Field 141

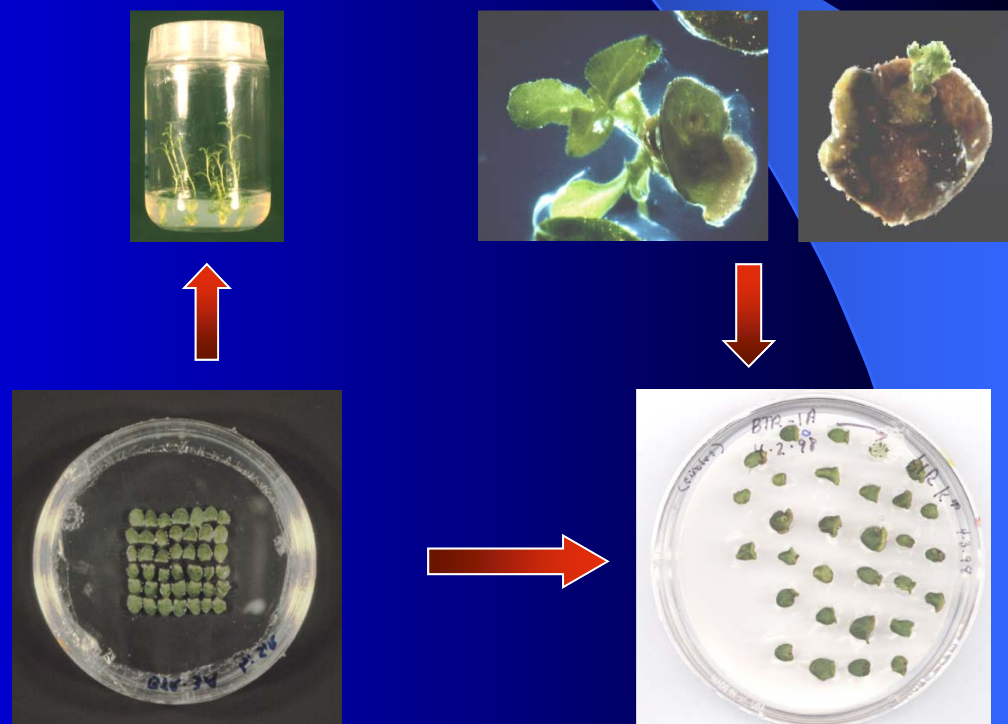


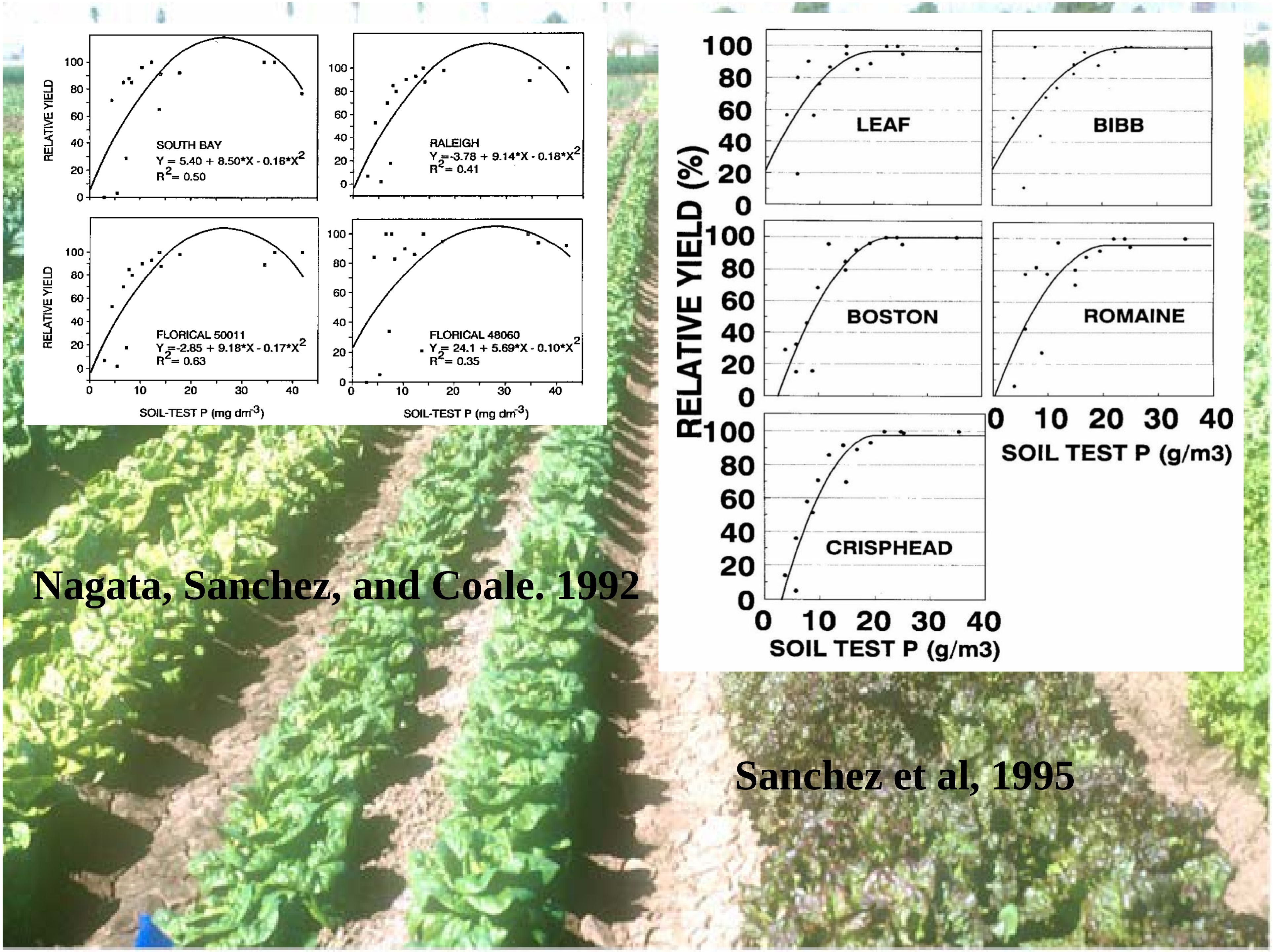
Above 39.999 0.00 ac
38.000 - 39.999 0.00 ac
36.000 - 37.999 0.00 ac
34.000 - 35.999 0.00 ac
32.000 - 33.999 0.08 ac
30.000 - 31.999 0.11 ac
28.000 - 29.999 0.10 ac
26.000 - 27.999 0.14 ac
24.000 - 25.999 0.21 ac
22.000 - 23.999 1.50 ac
20.000 - 21.999 3.50 ac
18.000 - 19.999 5.86 ac
16.000 - 17.999 7.89 ac
14.000 - 15.999 9.60 ac
12.000 - 13.999 11.35 ac
10.000 - 11.999 7.80 ac
8.000 - 9.999 6.14 ac
6.000 - 7.999 3.37 ac
4.000 - 5.999 1.78 ac
2.000 - 3.999 0.10 ac
0.000 - 1.999 0.00 ac
Below 0.000 0.00 ac



Sanchez et al. 1990

For over a century we have been changing the soil for the plant. Why not change the plant for the soil?



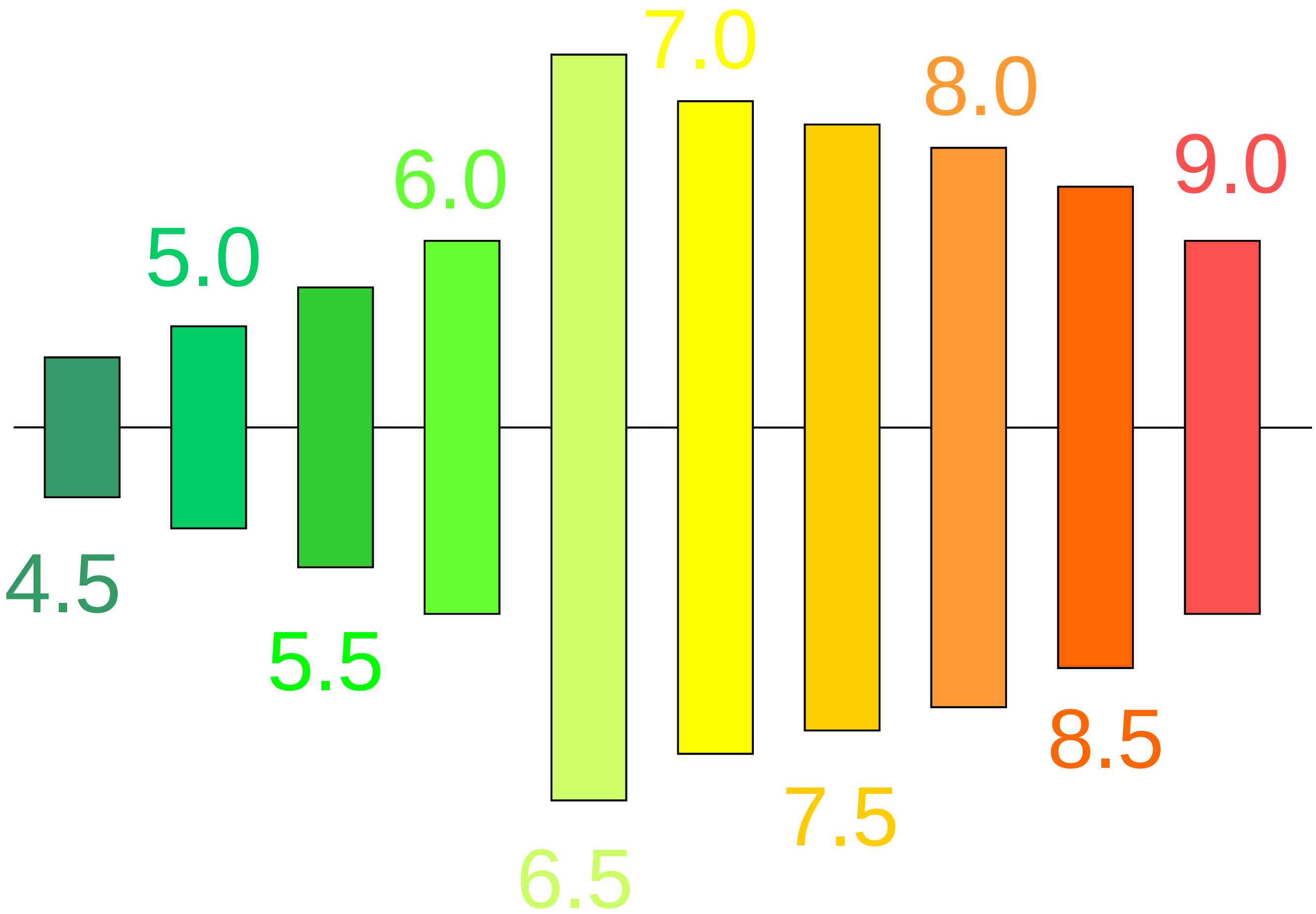


RELATIVE YIELD
SOUTH BAY
Y = 5.40 + 8.50*X - 0.16*X2
R2= 0.50
RALEIGH
Y = -3.78 + 9.14*X - 0.18*X2
R2= 0.41
FLORICAL 50011
Y = -2.85 + 9.18*X - 0.17*X2
R2= 0.63
FLORICAL 48060
Y = 24.1 + 5.69*X - 0.10*X2
R2= 0.35
SOIL-TEST P (mg dm-3)
Nagata, Sanchez, and Coale. 1992
RELATIVE YIELD (%)
LEAF
BIBB
BOSTON
ROMAINE
CRISPHEAD
SOIL TEST P (g/m3)
Sanchez et al, 1995

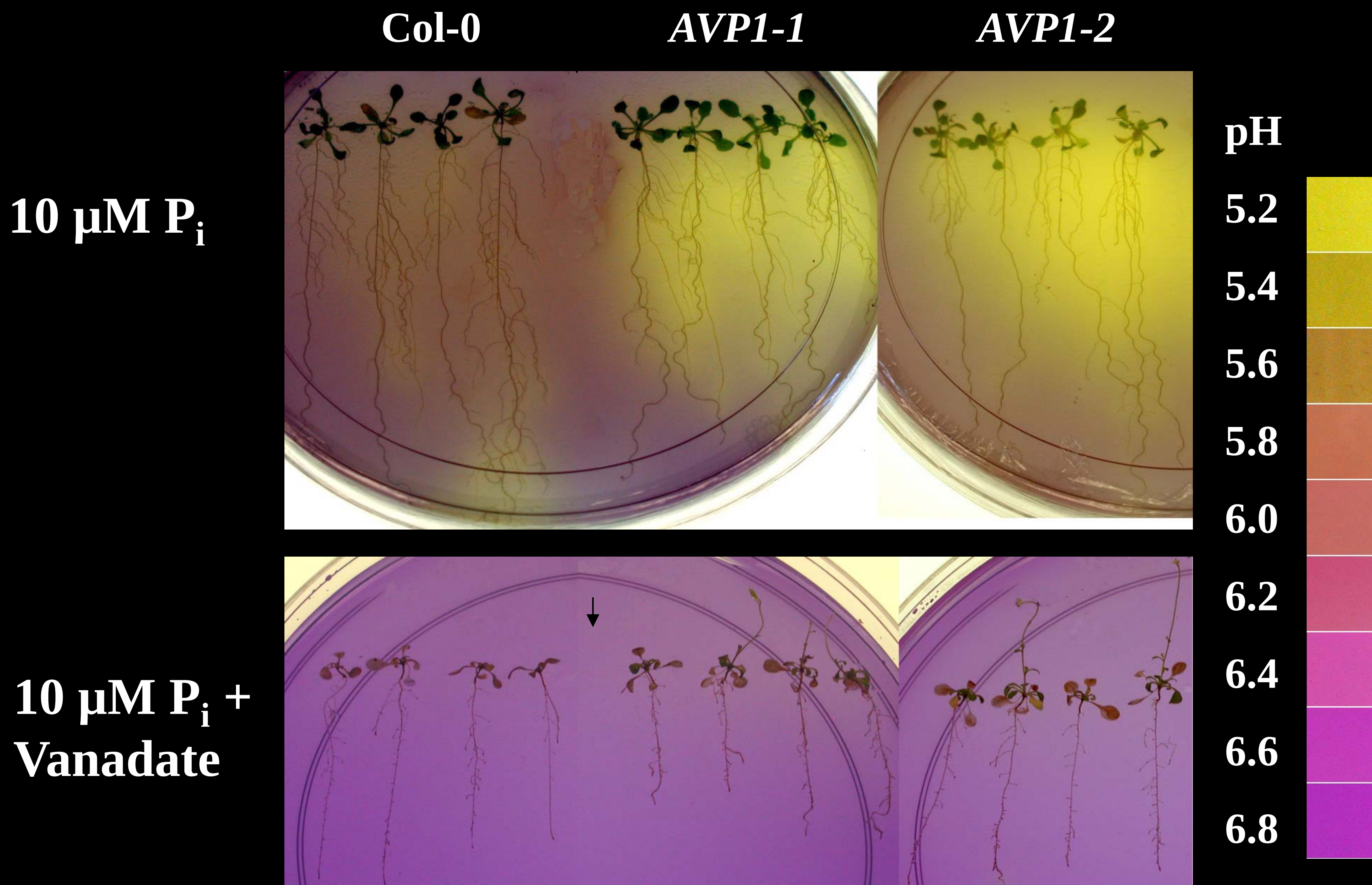
Type I H+ PPase Arabidopsis vacuolar pyrophosphatase (AVP1-OX; cDNA of At1g15690 from the Caulifower Mosaic Virus 35S Promoter)

- Increased root proliferation.
- Rhizophere acidification.
- Transport processes

Soil pH and Phosphorus Availability



AVP1-Enhanced Root Architecture Results in Higher Rhizosphere Acidification



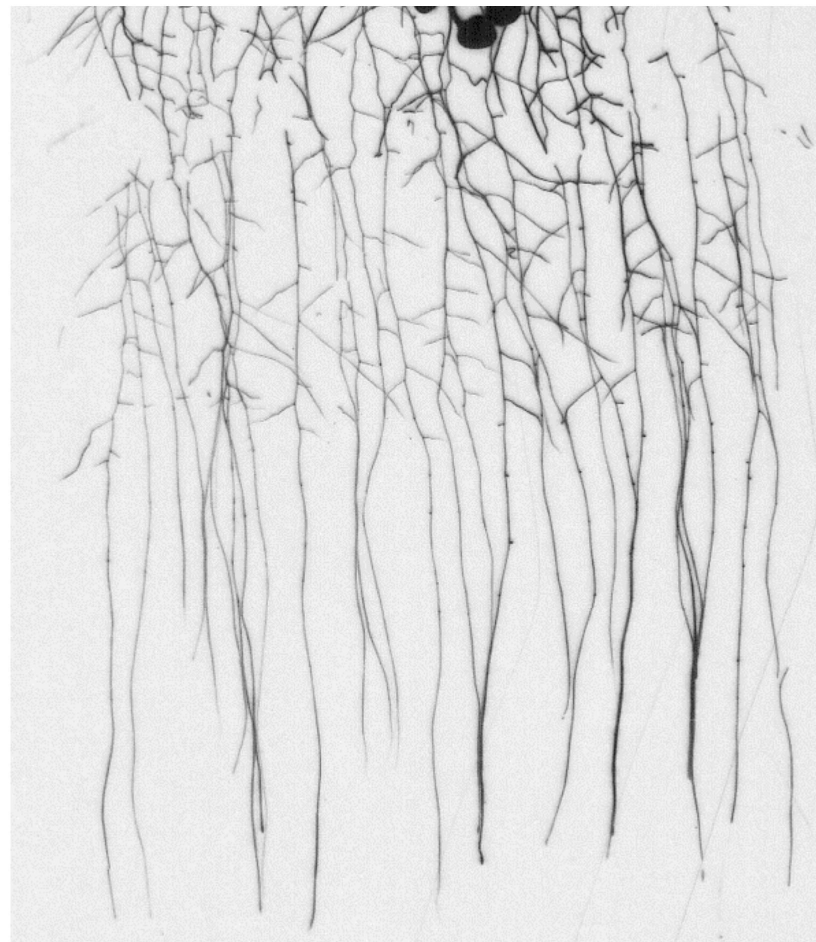
Bromocresol Purple

*AVP1*OX Root Systems Respond More Vigorously than Controls to Limiting P_i Conditions

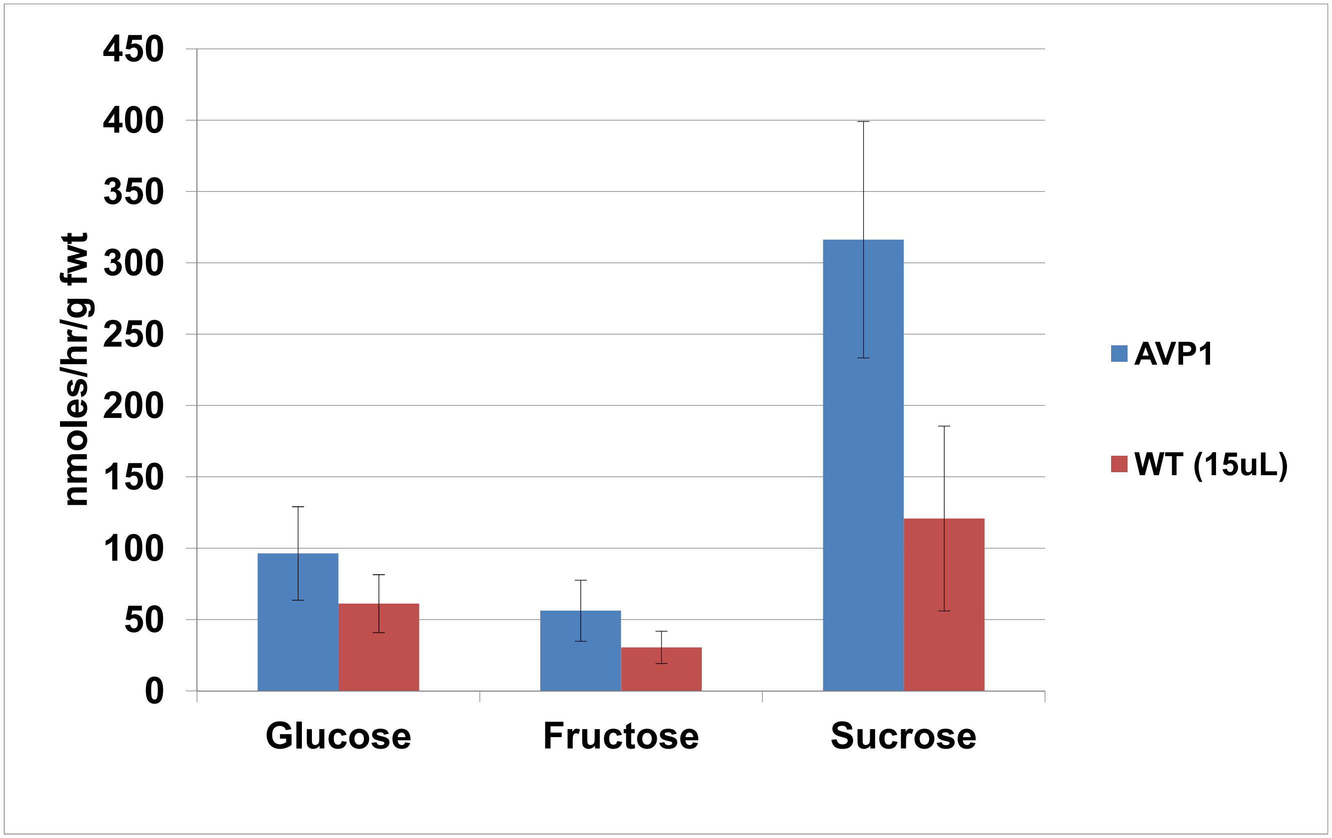
Col-0 **_AVP1-1_** **_AVP1-2_**

These results suggest that up-regulation of *AVP1* enhances the response capacity of the plants to limiting P_i.

Roots	au / mm
WT	731 ±222
AVP1-OX	1374 ±360

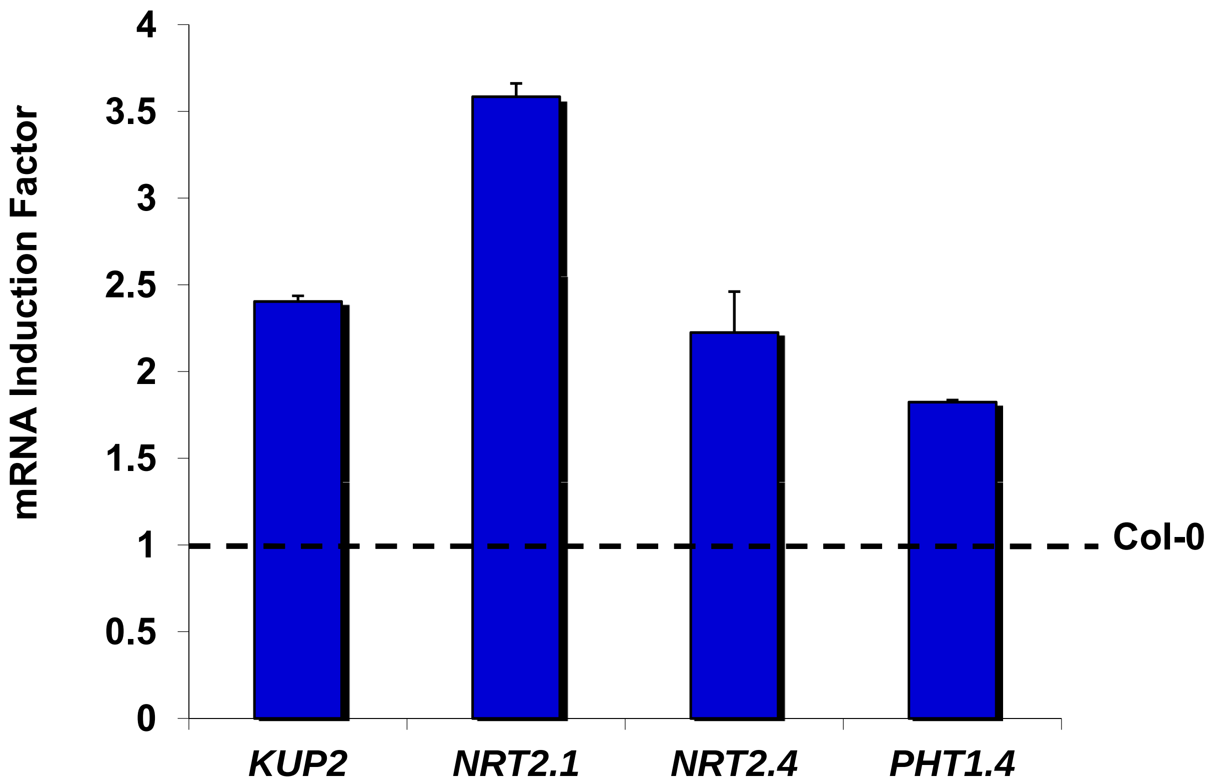


AthAVP1-OX plants have higher phloem sucrose content

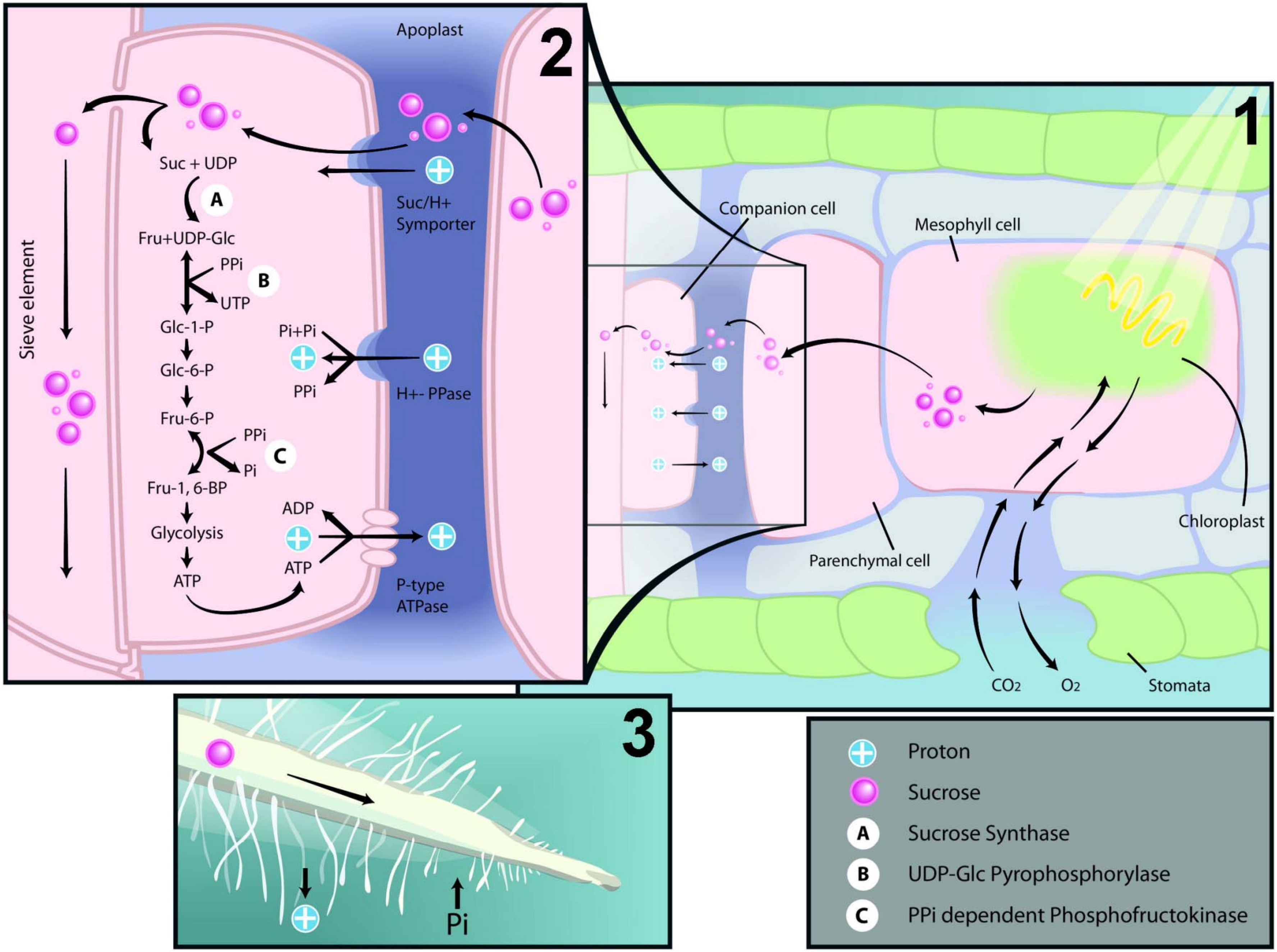


Paez-Valencia, J. *et al*, in preparation

The expression of sugar-induced ion transporters is up-regulated in roots of *AthAVP1*-OX plants



Paez-Valencia, J. *et al*, in preparation


2
Apoplast
Suc + UDP
A
Fru+UDP-Glc
PPi
B
UTP
Glc-1-P
Glc-6-P
Fru-6-P
PPi
C
Pi
Fru-1, 6-BP
Glycolysis
ATP
Sieve element
Suc/H+
Symporter
Pi+Pi
PPi
H+- PPase
ADP
ATP
P-type
ATPase
1
Companion cell
Mesophyll cell
Parenchymal cell
Chloroplast
CO2
O2
Stomata
3
Pi
Proton
Sucrose
A Sucrose Synthase
B UDP-Glc Pyrophosphorylase
C PPi dependent Phosphofructokinase

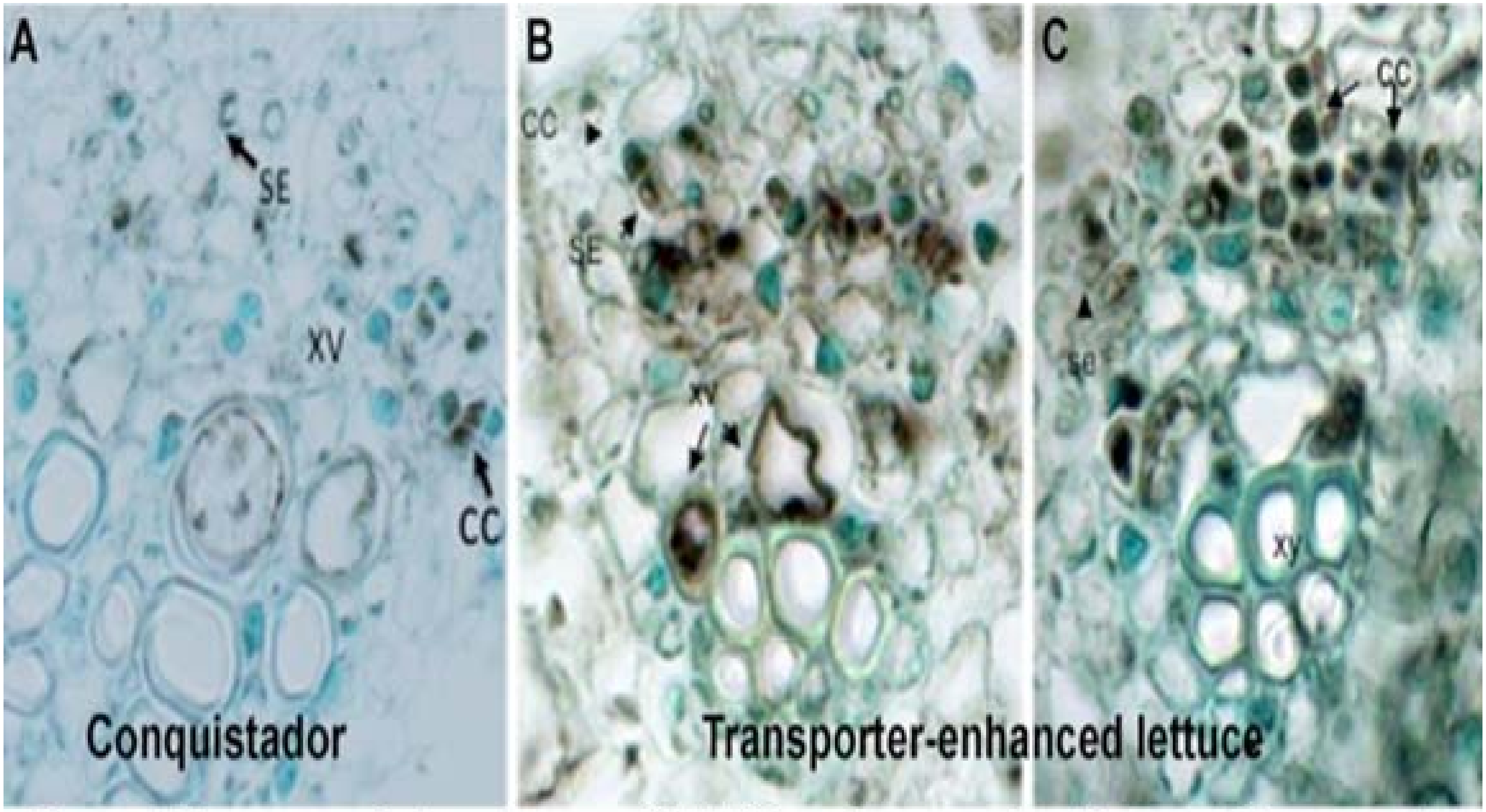
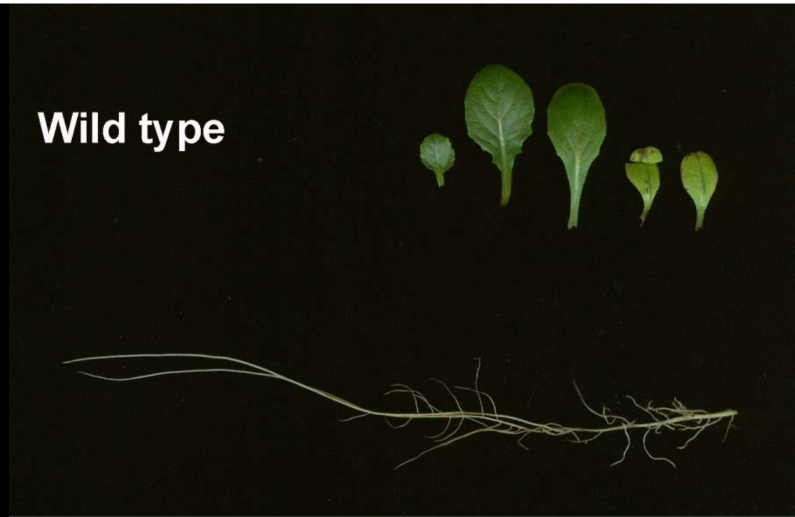
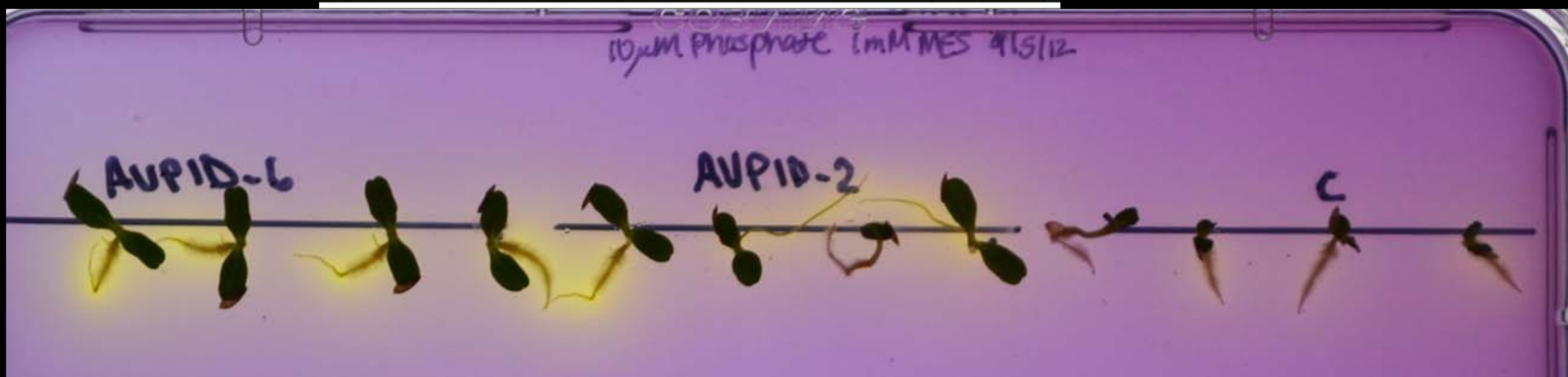
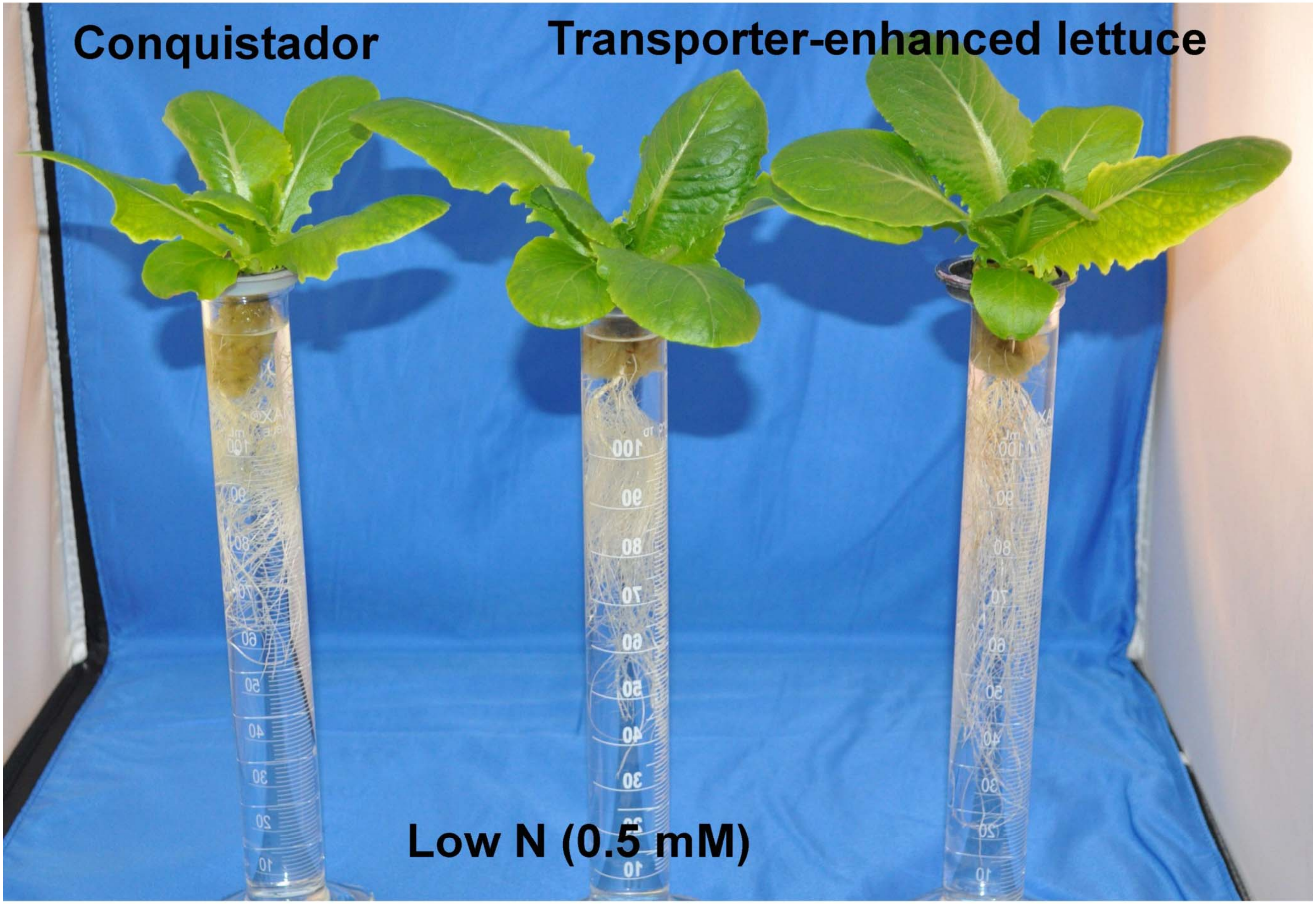


Figure 4. Increased abundance of H^+-PPase in vascular tissue of transporter-enhanced lettuce leaves. Immunohistochemical localization of H^+-PPase in leaf cross sections of conventional Conquistador (A) and transporter-enhanced Conquistador (B and C) incubated with antisera raised against H^+-PPase.

_AVP1_OX lettuce develop larger shoots and roots than controls when grown under limiting P_i (10 µM) conditions.







Conquistador
Transporter-enhanced lettuce
Low N (0.5 mM)

Main effect dry matter means in greenhouse P experiments to P rate and cultivar.

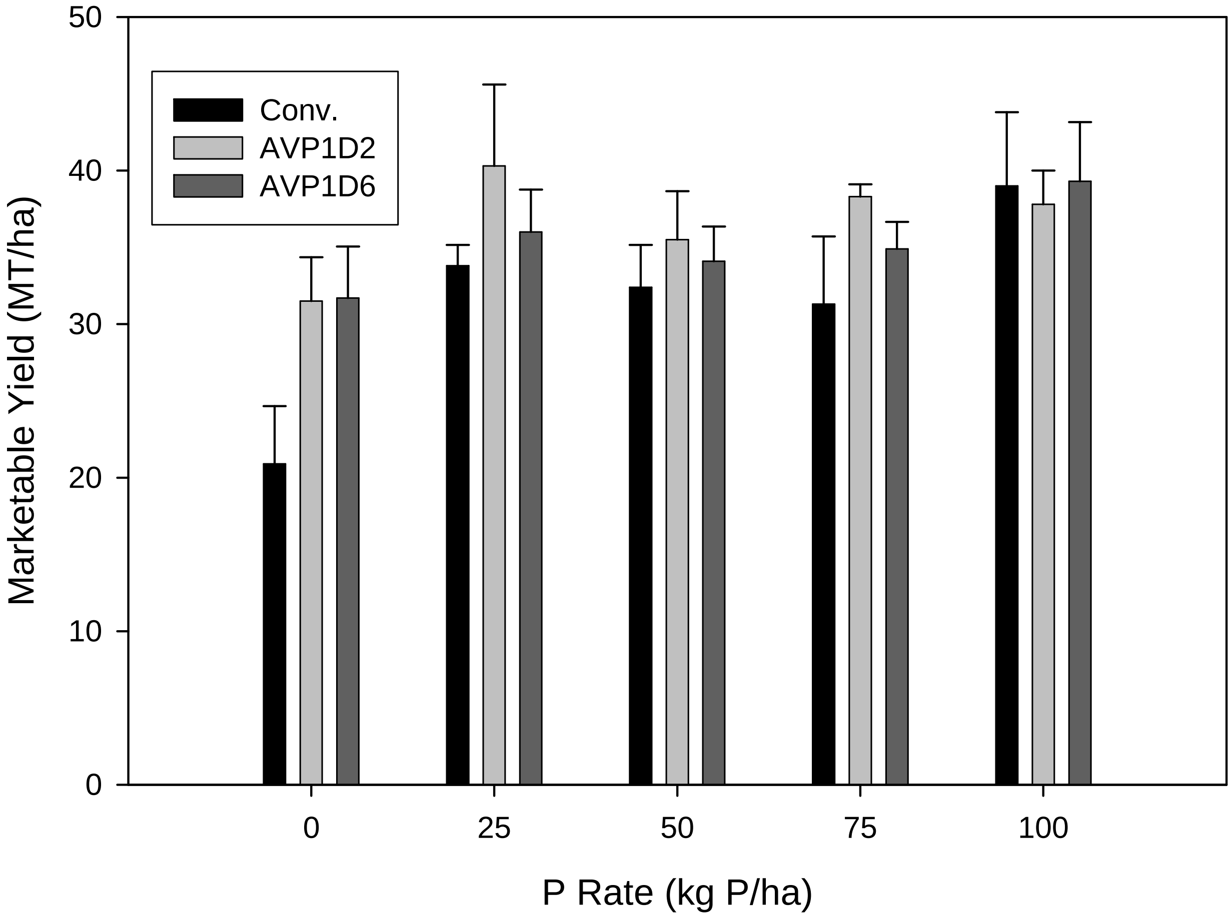
Treatments	Experiment			
P rate (g/pot)	1	2	3	4
Above-ground dry matter (g/pot)				
0	1.33	0.65	0.15	0.17
0.04	2.88	1.10	0.52	1.42
0.08	2.91	1.60	0.80	1.95
0.17	3.25	1.65	0.87	2.86
0.34	3.24	2.17	0.94	3.71
	L*Q**	L**	L**Q**	L**Q*
Cultivar				
Conventional	1.74a	1.06a	0.56a	1.52a
AVP1D2	3.19b	1.77b	0.71b	2.48b
AVP1D6	3.23b	1.47ab	0.69ab	2.06ab

Significant linear (L) and quadratic (Q) responses to N rate at P<0.01. Cultivar effect followed by same letter were not significant at P=0.05.

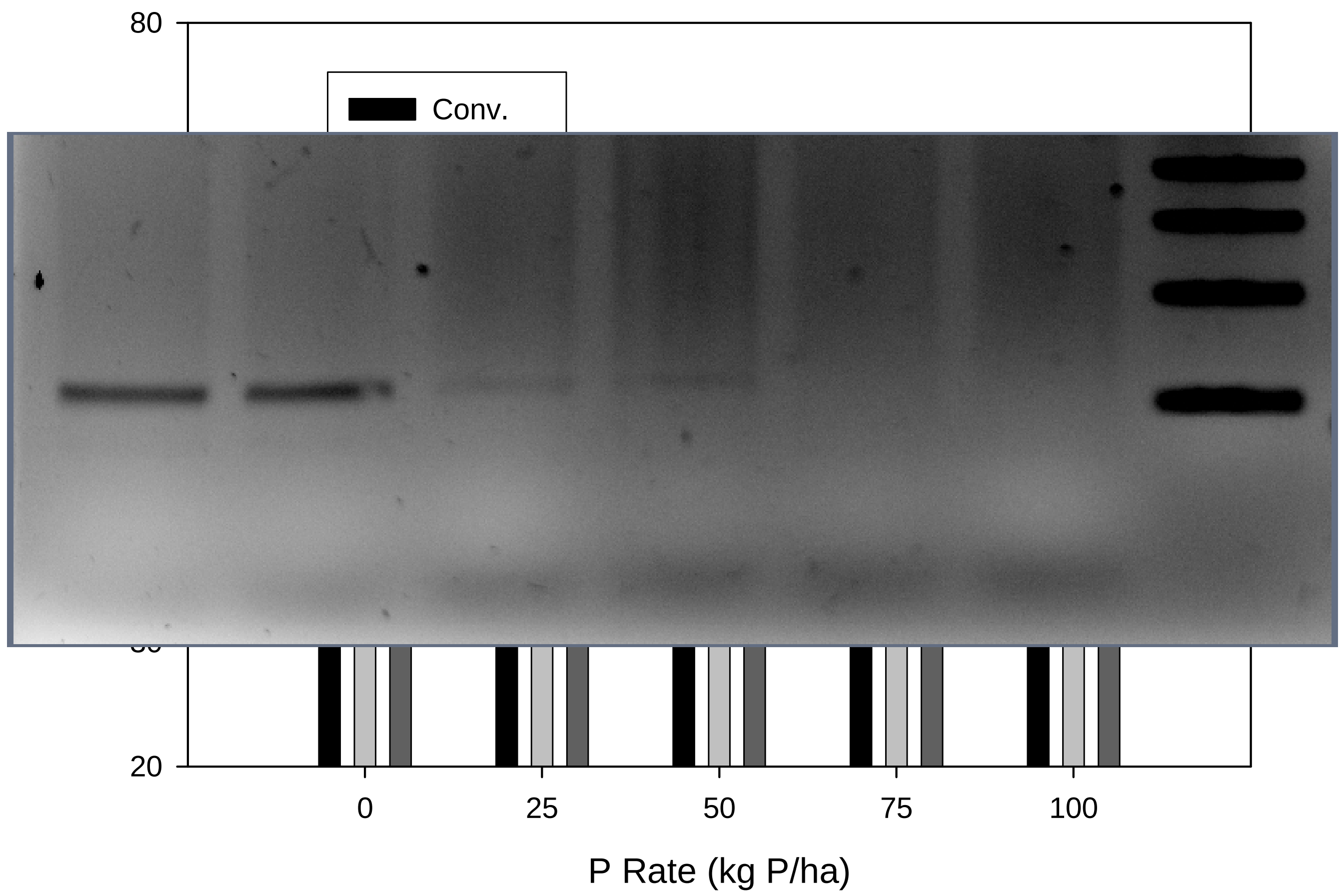
Main effect marketable yield means in field P experiments to P rate and cultivar.

Treatments	Experiment			
P rate (kg/ha)	1	2	3	4
Marketable yield MT/ha				
0	28.0	47.3	33.0	37.0
25	36.7	49.9	38.5	55.6
50	34.3	52.0	41.9	63.6
75	34.8	59.2	39.8	75.2
100	38.7	57.4	42.2	72.6
	L**	L**	L*	L**Q*
Cultivar				
Conventional	31.5a	47.5a	35.5a	51.9
AVP1D2	36.7b	52.2a	40.3ab	63.8
AVP1D6	35.2b	59.2b	41.4b	66.7

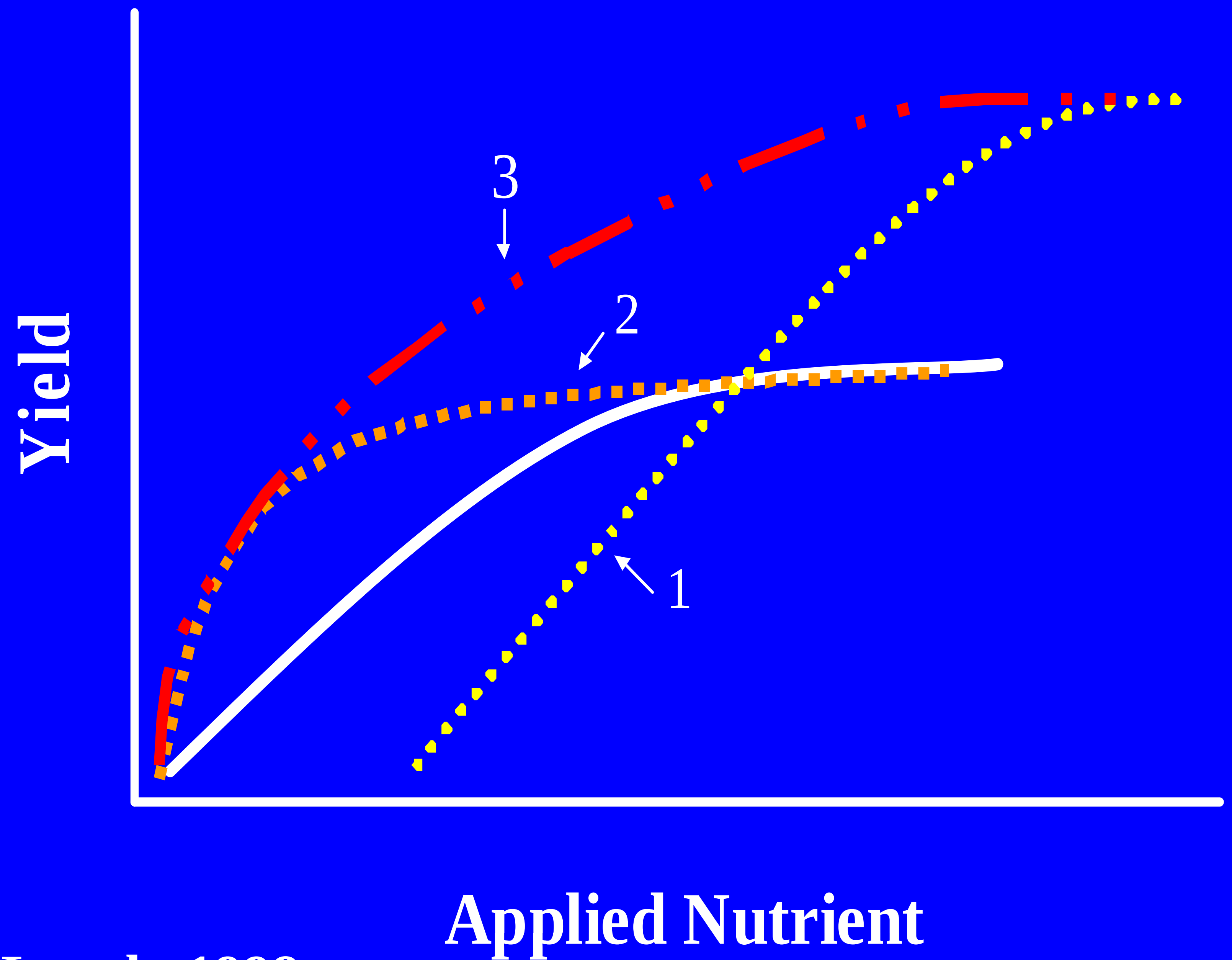
Significant linear (L) and quadratic (Q) responses to N rate at P<0.01. Cultivar effect followed by same letter were no significant at P=0.05.



Marketable Yield (MT/ha)
50
40
30
20
10
0
Conv.
AVP1D2
AVP1D6
0
25
50
75
100
P Rate (kg P/ha)



80
20
Conv.
0
25
50
75
100
P Rate (kg P/ha)



Lynch, 1998

How about roots vs. shoots.

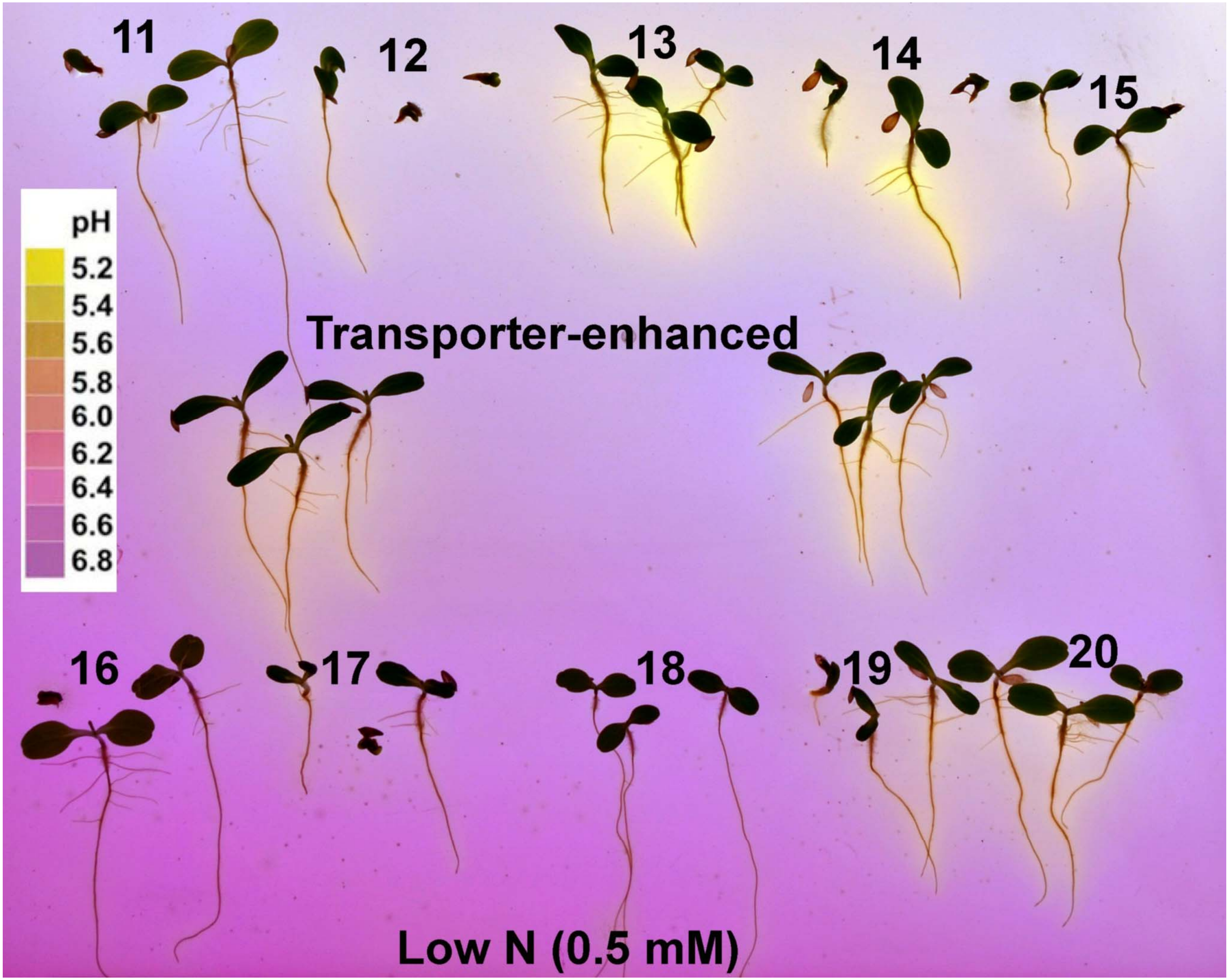
- There is concern that increased root growth is often at the expense of shoot growth (Lynch, 1995).
- Overall, enhanced shoot and fruit yields suggest that, under the conditions tested, the physiological costs incurred by the development of larger root systems did not jeopardize the 35Sp:*AVP1D* plants capacity to allocate sufficient photosynthates for shoot and fruit development.

Are there tradeoffs?

- Natural selection over millennia is unlikely to have missed simple, trade-off free improvements (Denison 2003).

Important Considerations

- Will the public accept GMO modified food?
- If not, is there another rout toward a similar goal.


11
12
13
14
15
pH
5.2
5.4
5.6
5.8
6.0
6.2
6.4
6.6
6.8
Transporter-enhanced
16
17
18
19
20
Low N (0.5 mM)

Acknowledgement

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