Biological amelioration of subsoil acidity via excess anion uptake by wheat

Zhe (Han) Weng[1], Peter Sale[1], Guangdi Li[2], Caixian Tang[1]

[1]Department of Animal, Plant & Soil Sciences, Centre for AgriBioscience, La Trobe University, Melbourne, Vic 3086, Australia
[2]NSW Department of Primary Industries, Wagga Wagga Agricultural Institute, PMB, Pine Gully Road, Wagga Wagga, NSW 2650, Australia
Corresponding authors: h.weng@latrobe.edu.au; c.tang@latrobe.edu.au

Abstract
Excess anion uptake in the form of nitrate has been shown to reduce soil acidification at depth. This root-induced alkalization of rhizosphere soil can be extended to bulk soil. This study investigated ways to maximize the alkalinizing effect of calcium nitrate in reducing subsoil acidity in wheat (ET8) and canola (44Y90). A controlled environment experiment was carried out over 35 days. A Chromosol with topsoil (0-8 cm, pH_{CaCl2} 5.4 and 1.5 mg Al kg^{-1}) and subsurface soil (8-15 cm, pH_{CaCl2} 4.8 and 2.9 mg Al kg^{-1}) layers was used. The soil was reconstructed as 0-10 cm topsoil and 10-50 cm subsurface soil in columns (15 cm in diameter, 60 cm in height). Air-dried soils (<2 mm) were treated with three N fertilizers: 1) control, 2) urea and 3) $Ca(NO_3)_2$ with and without phosphorus fertilizer at three depths: 1) 0-10 cm, 2) 10-20 cm and 3) 20-30 cm. All N and P fertilizers were applied at 134 kg N ha^{-1} and 56 kg P ha^{-1} at sowing. Uptake of $Ca(NO_3)_2$ increased pH up to 0.2 units of bulk soil in the 0-10 cm layer compared with the urea application regardless of the placement of the treatments. Rhizosphere alkalization was greater at the depth where nitrate and P were combined compared with those with the urea treatment. We highlighted the importance of balancing nutrient supply at depth in encouraging anion uptake by plants, which enhances rhizosphere alkalization in acid subsoil.

Keywords
Root-soil interactions, form of N fertilizers, subsoil fertilization, acidity management

Introduction
Application of nitrogen (N) fertilizer in the form of nitrate (NO_3^-) can enhance anion uptake by plants which increases the pH of the rhizosphere soil via the release of OH^- ions (Tang et al., 2013). The alkalization through the uptake of NO_3^- was reported to be 0.5-0.8 units in rhizosphere soil in laboratory (Weligama et al., 2010) and 0.3 pH units in bulk soil and field conditions (Conyers et al., 2011; Tang et al., 2011). However, there is no information on how to maximize the alkalinizing effects of NO_3^--based fertilizer in crops grown in acid subsoil. In this study, we aim to quantify and compare the effectiveness of calcium nitrate alone and in combination with P compared with urea in ameliorating subsoil acidity by crops with fertilizers placed at various depths. Here we highlight the results of wheat.

Methods
Column experiment

A controlled environment experiment was established on 14 November 2018. The factorial experiment consisted of 1 crop (wheat) × 3 placements (0-10, 10-20, and 20-30 cm) × 2 N fertilisers × 2 P fertilisers (+/-P) × 3 replicates = 36 columns.

Topsoil (0-8 cm) and subsurface soil (8-15 cm) were collected from a Chromosol at Rutherglen, Victoria. Soil pH_{CaCl_2} was 5.4 with 1.5 mg Al kg^{-1} in the topsoil and pH_{CaCl_2} was 4.8 with 2.9 mg Al kg^{-1} in the subsurface soil. The soil was re-packed as 0-10 cm with topsoil and 10-50 cm with subsurface soil in columns (15 cm in diameter and 60 cm in height). Air-dried soils (sieved <2 mm) were treated with two N types of fertilizer (urea and $Ca(NO_3)_2$ at 237 mg N $column^{-1}$, equivalent to 134 kg ha^{-1}), two rates of P fertilisers (0 and 99 mg P $column^{-1}$, equivalent to 56 kg ha^{-1}) with nil N control and non-planted N control. All N and P fertilisers were applied at sowing. For the N+P treatment, P was banded with N fertiliser and no P applied at the remaining of the column. For the N-P treatment, a basal P (45 mg NaH_2PO_4 kg^{-1}) was added to every 10-cm layer except for N-treated layer. The total basal P per column was equivalent to the P applied in the N+P treatment. Additional 136 mg Ca kg^{-1} was added to the urea-treated layer to match the Ca loading as in the $Ca(NO_3)_2$-treated layer. The following types and amounts (mg kg^{-1}) of basal nutrients were added in solution and mixed thoroughly in the soils: $CaCl_2.2H_2O$, 180; K_2SO_4, 120; $MgSO_4.7H_2O$, 50; $ZnSO_4.7H_2O$, 9; $CuSO_4.5H_2O$, 6; $Na_2MoO_4.2H_2O$, 0.4.

The soil columns were watered to 90% field capacity and allowed to stand in a controlled environment room (CER) at 25˚C. Twenty pre-germinated seeds of wheat (Al-tolerant ET8) or canola (44Y90) were sown evenly in each column. The seedlings were thinned to 15 plants at 7 days after sowing (DAS) and 10 at 15 DAS. Columns were weighed every three days and sufficient amount of water was added to maintain 90% of field capacity. The plant shoots were harvested on 35 DAS. The shoots were washed once in 0.1 M HCl and rinsed three times in deionised water, dried at 70˚C for two days and weighed.

Soil sampling and analysis

The soil was sampled by slicing the soil column into 0-10, 10-20, 20-30, 30-40 and 40-50 cm soil layers. The roots were carefully separated from the soil. The rhizosphere soil was collected by shaking soil adhered to roots into a bag. The remaining soil was considered as the bulk soil. Roots were cleaned with distilled water and scanned for root length and diameter using WinRHIZO (Regent Instruments, Canada, 2017). Roots were then oven-dried at 70°C for two days for dry matter determination. The pH of bulk and rhizosphere soil was measured using a Thermo Orion 720 pH meter after extraction in 0.01 M $CaCl_2$ solution (1:5 w/v soil: solution ratio) using an end-over-end shaker for 1 h.

For each depth, a three-way Analysis of Variance (ANOVA) was performed in a completely randomised design to evaluate the effects of N form, P addition, placement depth and their interactions on soil pH using the statistical language environment R 3.2.3. A least significant difference (LSD) at P= 0.05 was calculated to test differences between means.

Results

Localized alkalization at the depth

There was up to 0.2 unit increase in bulk soil pH in the $Ca(NO_3)_2$-treated soils compared with those treated with urea (P< 0.05, Table 1). The P application did not affect soil pH in the nitrate-treated soil. Soil pH was increased at the depth where the nitrate was placed compared with the urea treatment. Bulk soil pH did not change in the non-planted nitrate/urea-treated subsurface soil (non-planted N control, 4.82 pH units) compared with the non-planted non-amended control (nil N control, 4.85 pH units). Calcium nitrate increased soil pH up to 0.3 units in 0-10 cm (where 50-70% of root were distributed) *c.f.* the nil N control regardless of the placement depth. Rhizosphere pH was further increased by the uptake of $Ca(NO_3)_2$ at the treated depths compared with the urea treatments (data not shown). Uptake of calcium nitrate was effective in ameliorating subsurface soil acidity through rhizosphere alkalization compared with urea.

Combined nitrate and phosphate application increased shoot and root biomass

For biomass, N+P treatments had greater shoot and root biomass compared with N-P treatments regardless of depths of treatment (Figure 1). Nitrate fertiliser treatments increased plant shoot and root biomass by up to 8% and 20%, respectively, compared with urea fertiliser treatments. The trend can be summarized as greater shoot and root when fertilizer applied in 0-10 cm > 10-20 cm > 20-30 cm and NO_3+P> Urea+P > NO_3-P > Urea-P (Figure 1). Changes in root distribution were found at all treated depths, *i.e.* more root biomass at the treated layers (P< 0.05, data not shown). The most notable increase in root biomass was $NO_3 \pm$P treatments at 10-20 cm.

Table 1. Bulk soil pH at four depths (0-10, 10-20, 20-30 and 30-40 cm) in Chromosol columns amended with Urea-P, Nitrate-P, Urea+P and Nitrate+P. Results of soil grown with wheat were presented. Standard errors (SE) of the mean were tabulated (n=3).

Placement depth	N fertilizer	P	pH (1:5 $CaCl_2$)				
			0-10 cm	10-20 cm	20-30 cm	30-40 cm	40-50 cm
0-10 cm	Urea	No P	5.46	4.92	4.85	4.84	4.85
		P	5.50	4.90	4.88	4.86	4.78
	Calcium nitrate	No P	5.63	4.89	4.87	4.85	4.77
		P	5.66	4.89	4.88	4.87	4.82
10-20 cm	Urea	No P	5.40	4.91	4.86	4.85	4.68
		P	5.41	4.93	4.87	4.85	4.75
	Calcium nitrate	No P	5.67	5.02	4.85	4.86	4.70
		P	5.65	5.05	4.88	4.84	4.83
20-30 cm	Urea	No P	5.49	4.87	4.88	4.84	4.79
		P	5.50	4.90	4.94	4.86	4.77
	Calcium nitrate	No P	5.72	4.90	4.96	4.86	4.77
		P	5.70	4.88	4.99	4.86	4.79
	Nil	No P	5.45	4.85	4.85	4.78	4.75
		P	5.41	4.82	4.88	4.78	4.77
		LSD (*P*=0.05)	0.08	0.07	0.03	0.02	0.04
	Significance level						
		N	***	ns	**	*	ns
		P	ns	ns	ns	ns	*
		Depth	**	***	***	ns	ns
		N × P	ns	ns	ns	ns	*
		N × P × Depth	ns	ns	***	**	ns

Not significant (ns), *, ** and *** indicate, $P>0.05$, $P<0.05$, $P<0.01$ and $P<0.001$, respectively

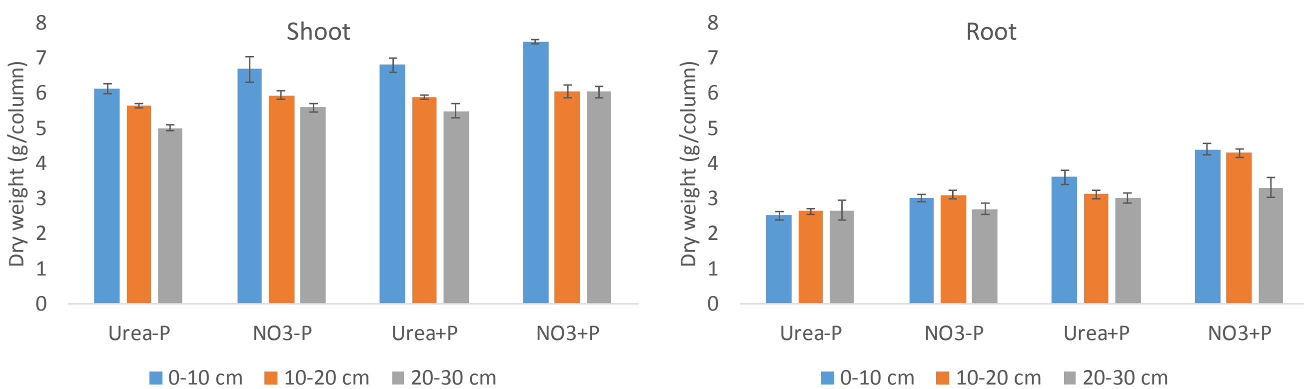

Figure 1. Dry weight for wheat shoots (left) and roots (right). N fertilizers alone and in combination with P and other nutrients were applied at 0-10 cm (blue), 10-20 cm (orange) and 20-30 cm (grey). Bars indicate standard errors of the mean (n=3).

Conclusion

We highlighted the importance of the N form in ameliorating subsoil acidification. Localized alkalisation of the soil by nitrate treatment was evident at all treated layers regardless of P application compared with the urea treatment. Combined nitrate and phosphate application increased shoot biomass and root proliferation of wheat at all treated depths compared with the nitrate-alone application. We showed that calcium nitrate increased alkalisation of the soil in the top soil (0-10 cm) regardless of the treatment depths compared with the urea treatment. It is important to balance nutrient supply at depth to improve excess anion uptake by plants, which in turn can extend rhizosphere alkalization to bulk soil in acid subsoil.

Reference

Conyers MK, Tang C, Poile GJ, Liu D, Chen DL, Nuruzzaman Z (2011) A combination of biological activity and the nitrate form of nitrogen can be used to ameliorate subsurface soil acidity under dryland wheat farming. Plant Soil 348 (1-2):155-166. doi:10.1007/s11104-011-0827-6

Tang C, Conyers MK, Nuruzzaman M, Poile GJ, Liu DL (2011) Biological amelioration of subsoil acidity through managing nitrate uptake by wheat crops. Plant Soil 338 (1-2):383-397. doi:10.1007/s11104-010-0552-6

Tang C, Weligama C, Sale P (2013) Subsurface soil acidification in farming systems: Its possible causes and management options. Molecular Environmental Soil Science. doi:10.1007/978-94-007-4177-5_13

Weligama C, Sale PWG, Conyers MK, Liu DL, Tang C (2010) Nitrate leaching stimulates subsurface root growth of wheat and increases rhizosphere alkalisation in a highly acidic soil. Plant Soil 328 (1-2):119-132. doi:10.1007/s11104-009-0087-x