

Improving Barley (*Hordeum vulgare* L.) Yield through Deeper Root Architecture Impacted by Nitrogen Management in a Reengineered Kurosol

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Research Article

Keywords: Soil re-engineering, subsoil acidity, soil compaction, nutrient acquisition, root architecture

Posted Date: December 11th, 2025

DOI: <https://doi.org/10.21203/rs.3.rs-8326636/v1>

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Additional Declarations: The authors declare no competing interests.

Abstract

Subsoil acidity and compaction are major constraints to crop production on Kurosols, duplex soils, in Western Australia, limiting root proliferation, nutrient uptake, and grain yield. This study evaluated how soil re-engineering practices, particularly deep liming, interact with nitrogen (N) fertiliser application to influence barley (*Hordeum vulgare* L.) root system development, crop performance, and soil properties in an acidic Kurosol. A semi-controlled experiment was conducted using 80 cm reconstructed soil profiles using soils from a field site. Treatments included soil loosening with or without lime incorporation, combined with varying N rates in split applications. Results showed that lime incorporation significantly enhanced total root length, volumetric root length density, and root surface area, shifting root distribution deeper into the profile and reducing reliance on surface soils. Grain yield and biomass increased markedly under limed treatments, with responses to N remaining linear, whereas unlimed soils showed yield plateaus at higher N rates. Root surface area was the strongest predictor of yield and biomass ($R^2 > 0.8$), highlighting the importance of fine-root proliferation. Liming also increased subsoil pH, decreased extractable aluminium, and increased organic carbon. These findings show that effective management of acidic, compacted subsoils requires integrated amelioration and nutritional strategies, where liming enables sustained root development and efficient fertiliser use. While soil column studies provide mechanistic insights, validation under field conditions is needed to assess persistence, rainfall interactions, and the economic viability of deep liming combined with optimised N management for sustainable barley production in water-limited environments.

Introduction

A deeper, uniform and denser root system of annual crops can improve the capture of water (Materechera et al. 1993; Gregory 1997; Manschadi et al. 2006a) and nutrients (Wiesler and Horst 1994; Dunbabin et al. 2003; Azam et al. 2024b) and increase the productivity of field crops (Kell 2011). Water acquisition from deep subsoil horizons is particularly important in Australia's water-limited arid and semi-arid climates, where crops depend largely on seasonal rainfall (Kirkegaard et al. 2011). However, naturally occurring and human induced subsoil constraints such as compaction and acidity can restrict the development of deeper roots (Azam and Gazey 2021; Lynch 2022; Gregory and Eastham 1996).

Most of the highly weathered and extremely ancient Australian soils are characterised by acidic subsoils (Eldridge et al. 2018). Around 50% of the arable subsoils of south-western Western Australia (WA) are acidic, with a pH_{Ca} (measured in 0.01 M $CaCl_2$ extract; hereafter pH_{Ca}) below a minimum target pH_{Ca} of 4.8 (Gazey et al. 2019). Subsoil acidity occurs in most of the Arenosol soil order (with varying clay contents) and in the Kurosol soil order (which has a strong textural contrast between the A and B horizons - also referred to as duplex soil) (van Gool 2016; Isbell 2021). Acidic soils pose several challenges to root system growth such as toxicities of aluminium (Al) and manganese, and deficiencies of calcium and phosphorus (Lynch and Wojciechowski 2015). High soluble concentrations of Al (at soil pH_{Ca} values below pH_{Ca} 4.8) can damage root apices, root elongation (Delhaize and Ryan 1995) and reduce the roots' ability to forage for water and nutrients. Around 75% of arable lands in WA are also

affected or susceptible to soil compaction (Parker et al. 2021). Soil compaction or high mechanical impedance of soil can affect root elongation under both wet and dry conditions (Lynch and Wojciechowski 2015; Materechera et al. 1993). Soil acidity and compaction frequently co-occur (van Gool 2016; Gazey et al. 2019; Parker et al. 2021), compounding their adverse effects on root development and crop performance. Understanding the individual and combined impacts of these constraints on crop growth is essential for designing effective soil management strategies that enhance productivity and improve water-use efficiency.

Duplex soils can be defined as texture contrast soils where the B horizon is dominated by a texture class one and a half (or more) finer than the A horizon with a clear to sharp change between the two horizons that occur within 10 cm (Tennant et al. 1992). Half to two-thirds of Western Australian agricultural land is duplex and these soils in WA are often reported to have shallow crop roots (Siddique et al. 1990; Gregory et al. 1992; Gregory and Eastham 1996) with a small proportion of roots in the B horizon due to a combination of chemical and physical limitations, which may include sub-soil compaction, poor soil fertility and acidity (Tennant et al. 1992; Azam et al. 2024a). The soil penetration resistance of these soils often exceeds 2 MPa (Tennant et al. 1992). This limits plant access to subsoil water and nutrients. It has been reported that the grain yields of wheat and lupin crops on duplex soils are often below the potential yield (Gregory 1997).

Deep tillage and/or deep incorporation of soil amendments, such as lime, and inorganic or organic fertilisers (Doss et al., 1979; Fiskell and Calvert, 1975 and Azam and Gazey, 2022) have been used to ameliorate subsoil constraints facilitating the deepening of the root zone. However, most of these previous interventions were confined to a maximum depth of 40–50 cm and the improvement in soil properties and grain yield have never reached the optimum level or full potential. Therefore, a series of field experiments on “soil re-engineering” is underway in Western Australia, where an acidic duplex soil profile has been ameliorated by loosening the soil and incorporating different ameliorants to a depth of 80 cm to benchmark the theoretical water-limited yield potential on these soils. In the field experiments, deep incorporation of a high rate of inorganic fertiliser, lime, and clay resulted in a significant grain yield increase in barley, wheat and canola compared to the surface application of these ameliorants (Azam et al. 2024a). However, experiments conducted by Azam et al. (2024a) were unable to determine the optimum rate of nutrients, especially nitrogen (N), required for maximising productivity once a soil is reengineered.

To address these challenges, we conducted an experiment in a semi-controlled environment using an acidic Kurosol from Meenar, WA (31.64° S, 116.89° E) to evaluate the combined effects of soil re-engineering and N fertilisation on the root development and crop performance of barley. The experiment compared intact and reconstructed soil columns to assess the interactive effects of lime incorporation and various rates of surface-applied N. We hypothesised that: (i) once soil compaction is removed, deep incorporation of lime would alleviate subsoil acidity and aluminium toxicity, enabling deeper and more extensive root systems, (ii) the positive effects of N fertilisation on root growth and grain yield would be maximised only in ameliorated soils, and (iii) root architectural traits, particularly root surface area,

would serve as the strongest predictors of crop productivity in re-engineered duplex soils. The experiment was expected to provide mechanistic insights into how integrated chemical and nutritional management strategies could enhance barley performance on a constrained Kurosol.

Methods

This experiment was conducted during the winter growing seasons of 2023 in a screen/shade house research facility at Department of Primary Industries and Regional Development (DPIRD), Northam, Western Australia (31.65° S, 116.70° E). The soil used in the experiment was collected from a cultivated farmer's paddock near Meenar, Western Australia (31.64° S, 116.89° E). This soil was classified as an acidic Kurosol in the Australian Soil Classification (Isbell 2021). Three intact soil cores with a diameter of 4 cm were sampled to a depth of 80 cm and sectioned at 10 cm intervals. For pre-trial assessment, three samples from the corresponding depths of the three cores were combined and analysed for soil chemical and physical properties as shown in Table 1. The soil was characterised as consisting of a weakly water repellent sandy layer over clay-loam subsoil. Soil pH_{Ca}) ranged from 4.3 to 4.7, and soil organic carbon (SOC) content ranged from 0.23 to 1.29%. Inorganic N, P_{Colwell}, and K_{Colwell} were at optimal levels in the 0–10 cm soil layer but were very low or below the detection limit in the deeper subsoil. The soil had a very low cation exchange capacity (CEC) in the soil layers below 10 cm (0.52–1.16 cmol kg⁻¹).

Table 1
Soil properties in 0–80 cm depths at the soil excavation site at Meenar cultivated field.

Depth (cm)	Sand (%)	Silt (%)	Clay (%)	pH _{Ca}	SOC (%)	Inorganic N (mg kg ⁻¹)	P _{Colwell} (mg kg ⁻¹)	K _{Colwell} (mg kg ⁻¹)	CEC (cmol kg ⁻¹)
0–10	90.8 ± 0.4	2.3 ± 0.2	6.9 ± 0.3	4.7 ± 0.06	1.29 ± 0.17	17.0 ± 1.0	18.7 ± 1.4	25.3 ± 3.8	2.79 ± 0.09
10–20	87.7 ± 1.9	2.3 ± 0.3	10.0 ± 1.6	4.4 ± 0.1	0.66 ± 0.06	4.7 ± 1.5	16.7 ± 2.6	16.3 ± 1.3	1.16 ± 0.06
20–30	80.7 ± 2.8	2.8 ± 0.5	16.5 ± 2.3	4.4 ± 0.1	0.39 ± 0.02	4.3 ± 0.3	14.3 ± 2.8	17.3 ± 1.4	0.52 ± 0.08
30–40	71.1 ± 5.7	3.1 ± 0.4	25.8 ± 5.4	4.3 ± 0.1	0.29 ± 0.02	5.7 ± 0.3	14.3 ± 4.1	19.0 ± 0.6	0.53 ± 0.07
40–50	62.9 ± 6.3	3.4 ± 0.4	33.7 ± 6.0	4.4 ± 0.1	0.23 ± 0.01	6.0 ± 0.0	8.3 ± 1.6	na	0.85 ± 0.14
50–60	57.6 ± 1.1	3.9 ± 1.1	38.5 ± 1.8	4.3 ± 0.0	0.18 ± 0.05	6.0 ± 1.0	3.3 ± 0.6	na	1.09 ± 0.07
60–70	57.6 ± 1.10	3.4 ± 0.4	33.7 ± 6.0	4.3 ± 0.1	0.23 ± 0.01	6.0 ± 0.0	8.3 ± 1.6	na	1.09 ± 0.07
70–80	62.9 ± 6.3	3.5 ± 0.3	33.6 ± 5.8	4.4 ± 0.1	0.23 ± 0.01	6.0 ± 0.0	8.3 ± 1.7	na	0.85 ± 0.14

na; values below the method detection limit

Experimental designs

In this experiment, we used a set of reconstructed soil columns to evaluate the effects of deep soil loosening and the deep incorporation of lime. This experiment consisted of a factorial combination of two different re-engineering treatments and four N rates (all applied as surface applications) (Table 2).

Table 2
Description of the variables used in the experiment.

Variables (Factors)	Treatment code	Treatments
Soil re-engineering treatment	L	Soil loosening – no lime
	LL	Soil loosening + lime (1 g of lime kg ⁻¹ of soil)
Nitrogen treatment	N1	0 kg ha ⁻¹
	N2	75 kg ha ⁻¹ (at seeding)
	N3	150 kg ha ⁻¹ (75 kg ha ⁻¹ at seeding and 4 weeks after seeding)
	N4	225 kg ha ⁻¹ (75 kg ha ⁻¹ at seeding, 4 and 8 weeks after seeding)
Intact core	IC	No soil treatments and no N, P and K supply
¹ For lime treatment, 1g of lime kg ⁻¹ of soil was added to the soil mixture (Azam and Gazey 2023)		

Based on the physico-chemical properties presented above, three bulk soil samples were excavated from topsoil (0–10 cm), midsoil (10–40 cm), and subsoil (40–80 cm) from the field. All soils were air-dried, sieved (< 4 mm), and homogenised separately prior to the reconstruction of soil columns to replicate reengineered soil profiles from a nearby field trial (Azam et al. 2024a).

Polyvinyl chloride (PVC) tubes (internal diameter 15.1 cm; height 85 cm) with perforated basal caps were used as column containers. A wick was inserted through the base to facilitate drainage (Zhu et al. 2003), and a lubricant (Kincrome NLGI2 Lithium Complex, Kincrome Australia Pty Ltd., Scoresby, Victoria) was applied to the inner walls of PVC tubes as a layer to minimise preferential root growth along the column edges. Soil was packed in three layers (0–10 cm, 10–40 cm and 40–80 cm). For the lime incorporation treatment, soils were thoroughly mixed, using a rotary cement mixture, with lime before column packing. Soils were packed into the PVC tubes to targeted bulk densities of 1.4 g cm⁻³ for the topsoil layer and 1.5 g cm⁻³ for the subsoil layers, with an approximate soil strength of ~ 1 MPa. The bulk densities used to repack the soil columns were similar to those achieved through soil loosening in the field re-engineering trial (Azam et al. 2024a). However, due to limitations of the dry filling technique used in column reconstruction, it was difficult to replicate the much higher bulk densities (~ 1.8 g/cm³) observed in the field control plots that had not undergone soil loosening. Figure 1 shows a reconstructed soil column removed from a PVC tube at the conclusion of the experiment, consisting of topsoil (0–10 cm), midsoil (10–40 cm), and subsoil (40–80 cm).

Extraction of intact soil columns

To preserve the natural soil structure and compaction of the soil profile, intact monolithic columns were hydraulically extracted directly from the field near the bulk soil excavation site in April 2023. Steel cylinders, matching the dimensions of the PVC tubes, were driven into pre-wetted soil (wetted 48h prior to soil extraction), and intact columns were carefully extracted with minimal disturbance or compaction. These intact cores, which more accurately represent field conditions (e.g., with natural macropores), were included as a baseline for comparative purposes, but they were not part of the main factorial design involving the nitrogen and soil amendment factors (Table 2).

Seeding and crop management

Soil columns were initially wetted to near saturation, and they were then allowed to drain to field capacity before seeding while preventing evaporation from topsoil surface using a thin plastic film. Following crop emergence, the columns were irrigated twice weekly using an overhead sprinkler system to maintain soil moisture of approximately 75% of field capacity.

A barley crop (variety Maximus) was grown (Fig. 2). Five seeds were sown manually at a depth of 3 cm on 1 June 2023. Two uniform plants per column (equivalent to ~ 112 plants m^{-2}) were retained after thinning at 13 days after seeding (DAS). Columns were randomised in a block design with four replicates and grown under a screenhouse. The crop was grown for 22 weeks (from 1 June to 3 November 2023). A basal fertiliser consisting of phosphorus (16 kg ha^{-1}) as single superphosphate ($16\% \text{ P}_2\text{O}_5$) and potassium (50 kg ha^{-1}) as muriate of potash ($60\% \text{ K}_2\text{O}$) were banded 5 cm below the surface before sowing. The N treatments (applied as urea) were as mentioned in Table 2.

Irrigation volumes were adjusted to 75% of field capacity based on pre-irrigation weighing of the columns. A total of 500 mm of irrigation was applied during crop growth. However, to simulate a terminal moisture stress during the grain filling period, irrigation was withheld from the pots during the final 31 days. The aim of applying terminal moisture stress during grain-filling was to examine how rooting depth, influenced by soil re-engineering interventions, affected the plant's ability to access subsoil water when topsoil moisture was depleted, and how this ultimately impacted grain production.

Measurements

Soil inorganic N (nitrate plus ammonium) was measured in 0–80 cm intact cores as well as mixed soil using Rayment and Lyons (2011) Method 7C2b. This gave an estimate of the baseline N level before the application of any fertiliser N.

At physiological maturity (156 DAS), shoot biomass was determined by harvesting shoots to the soil surface, oven-drying at 60°C to constant mass, and weighing. Grain heads were counted, threshed manually, and grain N concentration was determined using the Dumas combustion method described by Rayment and Lyons (2011) Method 7A5.

After harvest, PVC columns of three replicates per treatment were laid horizontally and the PVC walls were sliced longitudinally to expose the soil profile (Fig. 1). Soil was sectioned into discrete depth intervals (0–10, 10–20, 20–30, 30–40, 40–60, and 60–80 cm). Roots were extracted by wet sieving (0.7 mm mesh), washed carefully to avoid damage, and stored in water at 3–4°C until scanning. Root systems were scanned submerged in distilled water using a flatbed scanner (Epson Perfection V800; Epson, Japan) at 400 dpi. Image analyses were performed using WinRHIZO Pro 2007a (Regent Instruments, Québec, Canada), determining variables such as total root length, average diameter, and volume using the global threshold method. Volumetric root length density, a measure of the total length of roots per unit volume of soil ($vRLD$, $cm\ cm^{-3}$) was determined using the method of Wacker et al. (2024).

The remaining one replicate per treatment was used to visualise the entire root systems using a modified pinboard method after Kano-Nakata et al. (2011). Soil was gently washed away with a water jet while the whole soil column with the entire root system was pinned to a wire mesh, preserving the roots' spatial configuration for photographic documentation. Digital photographs of washed roots were taken against a black grid background under uniform indoor light conditions. Images were processed to quantify root colour using the CIE $L^*a^*b^*$ color space. Photographs were first converted from RGB to $L^*a^*b^*$ colour space using the Open CV library in Python (v 3.13). To isolate the root pixels from the background grid, images were converted into HSV colour space and a colour threshold was applied to capture the yellow–brown range characteristic of roots (H: 10–35°, S: 40–255, V: 40–255). The resulting binary mask was used to extract only root pixels. For each image, mean L^* , a^* , and b^* values were calculated by averaging across all masked pixels, providing an overall estimate of root lightness (L^*) and chromatic components (a^* , b^*). These relative Lab* values were used to compare root colours as some visual colour differences were observed between roots from treatments with and without lime incorporation.

A bulk soil sample (around 300 g) from each depth was collected during wet sieving the three replicates for root extraction. A portion of this sample was used to analyse moisture content immediately, i.e., residual moisture at the harvest, by drying at 105 °C. The remaining soil samples were dried at 40 °C and passed through a 2 mm sieve. Soil pH and aluminium were measured in a solution of 0.01M $CaCl_2$ (pH_{Ca} and Al_{Ca} respectively). The concentration of aluminium in the soil extract was determined using the method described by Bromfield (Bromfield 1987). Soil organic carbon was measured using Rayment and Lyons (2011) Method 6A1, while ammonium and nitrate N were measured using Rayment and Lyons (2011) Method 7C2b. Electrical conductivity ($EC_{1:5}$) was measured in a water extract at a 1:5 soil to solution ratio using Rayment and Lyons (2011) Method 4A1. Colwell extractable P ($P_{Colwell}$) and K ($K_{Colwell}$) was measured using a 0.5 M sodium bicarbonate extract as described by Rayment and Lyons (2011) using Method 9B2 and 18A1, respectively.

Statistical analyses

All statistical analyses were conducted using GenStat statistical software (Version 22, VSN International, Oxford, UK). A two-way analysis of variance (ANOVA) was performed to evaluate the effects of main variables (N and soil re-engineering) and their interactions on soil parameters, vRLD, and other plant growth parameters. Fisher's protected least significant difference (LSD) test was applied at a significance level of $P \leq 0.05$ to identify significant differences among treatments. Regression analyses were performed to assess the strength and direction of linear relationships between residual soil moisture content in the whole profile and grain yield, as well as total root length versus the grain yield of triticale. In each regression analysis, r - and r^2 -values were reported to evaluate the direction and strength of the relationships, while a probability (P) value was provided to indicate the significance of the relationship between the parameters. A relationship was considered significant at $P \leq 0.05$. Regression tree analyses were performed to assess the relative and interactive effect of N rate, soil pH_{Ca} , Al_{Ca} and clay content on total root length and grain yield.

Results

Root development and growth

The interaction between soil re-engineering treatments and N rates was visually apparent in whole root systems extracted from nine soil profiles, as shown in Fig. 3. Root colour differed between the treatments with deep lime incorporation (LL) and all the treatments without no lime incorporation (L treatments and IC) at crop maturity. Barley roots in the LL treatment (four panels on the right) were lighter and more yellowish in colour than those in the L and IC treatments (five panels on the left) (Fig. 3). As the colour information on root systems, two variables in the CIELAB colour space: b^* , and L^* were measured as described in the methodology. The average L^* value (lightness, ranging from 0 to 100) of root systems without lime incorporation was 47.3, while the average b^* value (blue to yellow scale) was 2.42. In contrast, the LL root systems showed higher values, with an average L^* of 59.2 and a b^* value of 9.7.

The main effect of soil re-engineering treatments and nitrogen (N) rates and their interactions (Table 3) significantly enhanced root surface area (Fig. 4a) and volumetric root length density (vRLD) (Fig. 4b). Soil columns with deep incorporated lime and applied nitrogen at 75, 150, or 225 kg N ha⁻¹ (LL-N2, LL-N3, and LL-N4) showed greater root surface area and volumetric root length density (vRLD) than all combinations of soil loosening alone (L) with varying N rates, as well as the limed treatment without N (LL-N1) (Fig. 4a and 4b). The application of lime (the LL treatment) increased root surface area by 1.5-fold and vRLD by 1.9-fold compared to the L treatment. The root surface area and vRLD were also increased by rates of N application: N3 and N4 had the highest root surface area (Fig. 4a) and N4 had the highest vRLD (Fig. 4b). The N4 rate increased vRLD by 1.8-fold compared to the nil N (N1) (Fig. 4b). As the nitrogen (N) rate increased, both vRLD and root surface area also increased. The highest root surface area was observed at N3 and N4 rates (Fig. 4a), whereas the highest vRLD was observed at N4 (Fig. 4b).

Table 3

Summary of the two-way analyses of variance (ANOVA) for soil re-engineering (RE) treatments and nitrogen (N) rates as fixed factors on below- and aboveground barley growth parameters. Fisher's protected least significant difference (LSD) test ($P \leq 0.05$) was used to separate means. Significance levels: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$; NS = not significant.

Variates	Factors	P-value	Level of significance	LSD
Volumetric root length density (cm cm^{-3})	RE	< 0.001	***	0.17
	N rate	< 0.001	***	0.23
	RE x N rate	0.018	*	0.34
Root surface area ($\text{m}^2 \text{pot}^{-1}$)	RE	< 0.001	***	0.009
	N rate	< 0.001	***	0.013
	RE x N rate	0.004	**	0.018
Grain yield (g pot^{-1})	RE	< 0.001	***	1.03
	N rate	< 0.001	***	1.45
	RE x N rate	0.002	**	2.05
Shoot biomass (g pot^{-1})	RE	< 0.001	***	2.14
	N rate	< 0.001	***	3.02
	RE x N rate	0.013	*	4.28
1000-grain weight (g)	RE	0.259	NS	3.74
	N rate	0.686	NS	5.29
	RE x N rate	0.953	NS	7.47
Protein yield (g pot^{-1})	RE	0.155	NS	0.16
	N rate	< 0.001	***	0.22
	RE x N rate	0.234	NS	0.31

The proportional distribution of the vRLD along the depths (root profile distribution) were affected by the main effect of soil re-engineering and N rate treatments (Fig. 4c and 4d), but not by their interactions. The L treatment had a greater proportion of vRLD in the topsoil (0–10 cm) compared to the LL treatments, whereas the LL treatments showed higher vRLD at 10–40 cm depth than the L treatment (Fig. 4c). There was no difference between L and LL treatments in proportional vRLD at 40–80 cm depth. Application of any N (N2, N3 and N4), significantly increased proportional vRLD compared to the nil N

(N1), but the difference was only observed at 0–10 cm depth (Fig. 4d). The distribution of vRLD in the L and LL treatments contrasted with the intact soil column, where 93% of the roots were concentrated in the upper 10 cm of soil and 99% root occurred in the top 20 cm of the soil (Fig. 4c and 4d).

Shoot biomass, grain yield and yield parameters

The soil re-engineering treatments and nitrogen (N) rates and their interactions were significant on grain yield and shoot biomass (Table 3; Fig. 5a and 5b). The main effect of soil re-engineering was not significant on seed size and protein yield (Table 3; Fig. 5c and 5d). The main effect of N rates was not significant on seed size (Fig. 5c), but it was significant on protein yield (Table 3; Fig. 5d). The interaction of soil re-engineering treatments and nitrogen (N) rates were not significant on seed size (Table 3; Fig. 5c) and protein yield (Table 3; Fig. 5d).

For improving yield and shoot biomass, the application of N2 with lime (LL-N2) had the same influence as that of the two higher rates of N without lime (L-N3 and L-N4) (Fig. 5a and 5b). Similarly, nil N with lime (LL-N1) had the same effect as that of N2 without lime (L-N2). With lime, the application of the two higher rates of N significantly increased grain yield and shoot biomass compared to any other treatment combinations, where LL-N4 was superior to LL-N3. Protein yield increased with every increment in N rates (Fig. 5d). Increase in grain yield was not related to the seed size (Fig. 5e). Increase in grain yield was strongly related to the increase in protein yield (Fig. 5f).

Relationships between root traits and crop performance

The vRLD was strongly and positively related to barley grain yield ($R^2 = 0.74$, $P < 0.001$; Fig. 6a) and shoot biomass ($R^2 = 0.82$, $P < 0.001$; Fig. 6b). However, barley grain yield ($R^2 = 0.84$, $P < 0.001$) and shoot biomass ($R^2 = 0.89$, $P < 0.001$) were more strongly and positively related to the root surface area (Fig. 6c and 6d) than the vRLD (Fig. 6a and 6b). In the LL treatment, the increasing rate of N improved both below-ground (e.g. vRLD and surface area) and above-ground (shoot biomass and grain yield) growth parameters linearly. By comparison, in the L treatment, the effect of N rate on above mentioned plant growth parameters were plateaued at N3 (150 kg N ha⁻¹).

Soil moisture and other properties at harvest

The interaction between re-engineering treatments and nitrogen (N) rates significantly affected the residual soil water content at barley harvest, 31 days after irrigation ceased (Fig. 7). Lime incorporation (LL) resulted in lower residual soil water levels compared with the loosened (L) treatment. Nitrogen rate effects were evident only in the absence of lime incorporation, where the N3 and N4 treatments had significantly lower soil water contents in the deeper layers compared with N0 and N1 (Fig. 7a). In contrast, N rate had no significant effect on residual soil water where lime was incorporated (Fig. 7b).

The interactions of the soil amelioration and N rates were not significant to influence soil pH_{Ca}, Al_{Ca}, EC, OC and N, P and K levels (Fig. 7). The applied N also did not affect any of the above-mentioned soil

properties. Only soil amelioration treatments affect all the properties, and the effect varies at with depth for a particular soil property.

Unsurprisingly, soil pH_{Ca} increased due to lime incorporation (Fig. 8a) and decreased Al_{Ca} (Fig. 8b) throughout the depth. Surprisingly, the increase in soil $\text{pH}_{\text{CaCl}_2}$ was smaller in the sandy layers (0–40 cm) compared to the sandy clay layer (40–80 cm). Soil columns with plants had less ammonium N in the topsoil (0–10 cm) than the ICs without plants (Fig. 8c). Conversely, soil columns with plants had more ammonium N in the subsoil than in the ICs without plants. The ICs without plant had higher nitrate N throughout the depths compared to all other treatments (Fig. 8d). The LL treatments had significantly less nitrate N at deepest depth than any other treatments.

The LL treatment had higher EC than the L treatment at all depths, however, LL treatment was not different from ICs with and without plants (Fig. 8e). Surprisingly, both L and LL treatments had higher OC in the deeper depths compared to the ICs with and without plants (Fig. 8f). Intact cores without plants had higher $\text{P}_{\text{Colwell}}$ than the ICs with plant, L and LL treatments at 0–40 cm depths (Fig. 8g). At 0–30 cm depth, ICs with plants also had higher $\text{P}_{\text{Colwell}}$ than the L and LL treatments. The treatment effects were more distinguished for the $\text{K}_{\text{Colwell}}$ in the order of $\text{LL} > \text{L} > \text{IC with plants} > \text{IC without plants}$ (Fig. 8h).

Discussion

The present study examined how soil re-engineering practices (to a depth of 80 cm), particularly liming interact with N fertiliser application to influence barley root system architecture, crop performance, and soil properties in acidic and compacted soils. By integrating detailed root measurements along with barley yield, water use and soil properties data, our findings provide new insights into the mechanistic pathways through which amelioration improves subsoil exploration, nutrient-use efficiency, and long-term soil health. The following discussion interprets these outcomes in the context of both the Australian and international literature, with emphasis on the roles of soil acidity, aluminium toxicity, soil compaction, nitrogen dynamics, and root architectural traits in shaping up barley yield on duplex soil with multiple soil constraints throughout the profile.

Liming alleviates acidity constraints and unlocks deeper rooting in barley

Lime incorporation substantially improved barley root development, enhancing total root length (TRL), volumetric root length density (vRLD), and root surface area while shifting roots from a shallow, topsoil-concentrated profile toward deeper soil horizons. In the unlimed treatments, more than 50% of the roots were confined to the 0–10 cm layer, reflecting typical responses of barley to acidic soils (Tang et al. 2013). In contrast, limed profiles showed up to a 15-fold increase in root length in the 40–80 cm layer. These results are consistent with Australian field trials which demonstrated that lime enhances root exploration and water capture in sandy and duplex soils (Li et al. 2019; Azam et al. 2024b; Enesi et al. 2023).

The mechanistic basis lies in reduced aluminium (Al) toxicity, as indicated by strong negative relationships between TRL and Al_{Ca} . International studies have established that Al rapidly inhibits root elongation by binding to cell walls and disrupting cell division and expansion (Kochian et al. 2015). Barley is particularly sensitive to Al, yet tolerance can be conferred by the HvAACT1 gene, which encodes a MATE transporter that effluxes citrate to chelate toxic Al^{3+} (Furukawa et al. 2007; Fujii et al. 2012). In our study, liming alleviated Al toxicity and allowed sustained root elongation, explaining why TRL increased more strongly with declining Al_{Ca} than with rising pH_{Ca} alone. Similarly, Al_{Ca} is a better predictor of wheat grain yield response to lime application than pH_{Ca} (Anderson and Bell 2019). This reinforces the importance of chemical amelioration in enabling barley roots to penetrate hostile acidic subsoils.

Nitrogen application modifies root traits but interacts strongly with soil amelioration

Nitrogen (N) addition altered root morphology, decreasing diameter and reducing TRL at higher rates in unlimed soils, likely due to rhizosphere acidification from ammonium transformation (Guo et al. 2010). However, in limed soils, N enhanced vRLD and root surface area, showing that chemical amelioration is a prerequisite for efficient fertiliser use. Similar responses have been reported in Australian soils, where lime mitigated Al toxicity and improved nutrient use efficiency (Kirkby et al. 2014).

Globally, barley often shows shallow root proliferation around fertiliser bands in sandy and duplex soils (Gregory and Eastham 1996; McDonald et al. 2018), limiting its ability to exploit deep soil resources. In our study, liming facilitated deeper root proliferation, enabling the effective utilisation of N available throughout the 0–80 cm profile. The contrasting plateau in biomass and yield responses under loosened-only soils compared with the linear responses under limed soils shows that without amelioration, higher N rates cannot compensate for subsoil acidity constraints. Thus, integrating liming with appropriate nutrient strategies is critical for sustainable intensification of cropping system.

Soil compaction reshapes barley root system architecture, but chemical and nutritional interventions mitigate impacts

Soil compaction and high bulk density restrict root elongation, reduce fine root proliferation, and force lateral rather than vertical growth. International studies show that barley root length can be reduced by more than 50% under compaction, primarily through the loss of fine roots, which are critical for nutrient uptake (Kristoffersen and Riley 2005; Mulholland et al. 1996). Compaction also decreases stomatal conductance and shoot growth, even in the absence of visible drought stress (Pfeifer et al. 2014). Our results on a texture-contrast Kurosol mirrored these findings, with intact cores showing that > 90% of root systems were confined to the top 10 cm. However, lime incorporation enabled deeper rooting into the 10–40 cm layer, partly overcoming physical impedance and alleviating chemical stress. Nitrogen further promoted vRLD, though its effects on depth distribution were modest compared to lime. This suggests that while compaction exerts strong physical constraints, improving the chemical environment

of the subsoil is equally important for sustaining root penetration and compensatory growth responses (Azam et al. 2024b). Such interactions between mechanical and chemical constraints have also been observed in Australian duplex soils, where deep ripping combined with lime or gypsum enhanced root depth and yield (Davies et al. 2020; Azam et al. 2023). Similarly, the soil profile's RLD is influenced by the soil environment, growing seasons and climatic conditions (Zuo et al. 2006). For example, wheat's RLD were observed to range from the lowest RLD in Zou et al. (2006) data based on growing on a compacted deep sand, which was N-deficient due to highly leaching rainfall compared to highest RLD following deep ripping and adequate N nutrition due to dry seasonal conditions (Anderson et al. 1997).

Reengineered soils with improved root system architecture and better nitrogen management led to more than fivefold increase in barley yield

In our study, the combined removal of subsoil acidity and compaction along with optimised N application and substantial improvement in root architecture had improved barley yield by more than fivefold, which is far greater than what is achieved through standard soil amelioration across the world (Schneider et al. 2017; Davies et al. 2024). The standard soil amelioration might not provide full water-limited yield potential due to their limited efficiency in complete removal of subsoil constraints (Isbister et al. 2024; Scanlan and Davies 2019). Treatment responses in our study reinforce that once subsoil acidity and compaction are reengineered, progressive N increments increased both below- and above-ground traits linearly, while in unlimed soils, responses plateaued at moderate N rate. Similar findings from China and Brazil indicate that crop responses to fertilisation are contingent upon liming to sustain fine-root growth and surface area (Guo et al. 2010; Rao et al. 2016). Thus, liming is a prerequisite for fertiliser efficiency in acidic, compact soils.

Our findings indicate that root surface area, rather than vRLD alone, was the strongest predictor of shoot biomass and grain yield (r^2 up to 0.89). This is consistent with international evidence that surface area, reflecting fine-root integrity and rhizosphere contact, is the key trait underpinning nutrient and water acquisition (Lynch 2019). Studies on wheat and barley have shown that increased fine-root surface area improves water extraction in drying soils and enhances yield stability under drought (Wasson et al. 2012; Manschadi et al. 2006b). In compacted soils, however, root length increases are often superficial and functionally ineffective unless fine-root expansion accompanies them (Kristoffersen and Riley 2005; Pfeifer et al. 2014).

Beyond yield, liming raised the soil pH_{Ca} , reduced Al_{Ca} , and enhanced organic carbon in subsoil layers. Comparable improvements in soil chemistry and nutrient cycling have been reported in long-term Australian experiments (Scott et al. 2000; Li et al. 2019). Overall, these results emphasise that soil re-engineering practices such as deep liming not only enhance crop productivity but also underpin long-term soil health and sustainability.

Conclusions

This study has shown that the amelioration of a Kurosol subsoil with lime substantially enhanced barley root development, enabling deeper and more evenly distributed root systems that alleviate the limitations imposed by acidity and compaction. Liming increased total root length, volumetric root length density, and root surface area, with the latter emerging as the strongest predictor of grain yield and biomass. Nitrogen fertilisation further enhanced root and shoot traits, but only where acidity and compaction had been ameliorated, emphasising the need for integrated nutrient and soil management.

The results also suggest that root surface area, rather than length alone, is the critical trait governing nutrient capture and yield resilience in constrained soils. International evidence supports this, showing that fine-root proliferation and rhizosphere contact underpin efficient acquisition of nitrogen, phosphorus, and water under stress conditions. Beyond crop productivity, liming improved soil pH_{Ca} , reduced Al_{Ca} , and increased organic carbon and potassium availability in subsoil layers, pointing to broader soil health benefits.

As this was a semi-controlled column study, the magnitude of responses observed here must be validated under field conditions, where spatial soil heterogeneity, seasonal rainfall, and machinery-based amelioration influence outcomes. On-farm research is required to: (i) confirm the yield responses, (ii) quantify the persistence of deep-rooting responses over multiple seasons, (iii) evaluate interactions with variable rainfall and fertiliser placement, (iv) assess economic trade-offs of deep liming and fertiliser strategies, and (v) develop machinery approaches that can achieve uniform lime incorporation at depth. Further genetic studies are also warranted to identify and deploy barley cultivars with robust fine-root traits that complement soil amelioration. Such efforts will ensure that the benefits demonstrated in this study can be translated into scalable, farmer-ready practices that deliver both productivity gains and long-term soil health improvements.

Declarations

Acknowledgements

The authors would like to thank the Department of Primary Industries and Regional Development (DPIRD), as well as the Grains Research and Development Corporation (GRDC), for supporting this research under projects DAW1902_003RTX and DAW2407-001SPX. We sincerely appreciate the contribution of Ty Fulwood, who allowed us to collect soil samples from their properties and provided seeds for growing plants. We would also like to thank the technical staff (particularly Shahinur Rahman, Hasinur Rahman, Trey Beeson, Mousumi Banik, Pete Grey and Steve Rossi) at DPIRD, WA. The authors thank Dr Andrew van Burgel for his advice on trial design. We also thank Dr Edward G. Barrett-Lennard, Dr Geoff Anderson and Wayne Parker for the internal review of the manuscript before submission.

Conflict of Interest

The authors declare no conflicts of interest.

Declaration of Funding

This research was funded by Grains Research and Development Corporation (GRDC), under project DAW1902_003RTX.

Data Availability

The datasets used and/or analysed during the current study are available from the corresponding author upon reasonable request.

Author's contribution

Kanchana Wickramarachchi: Trial design, execution, root image collection and analysis, data analysis and manuscript preparation.

Gaus Azam: Fund sourcing, trial design, data analysis and manuscript preparation.

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Figures

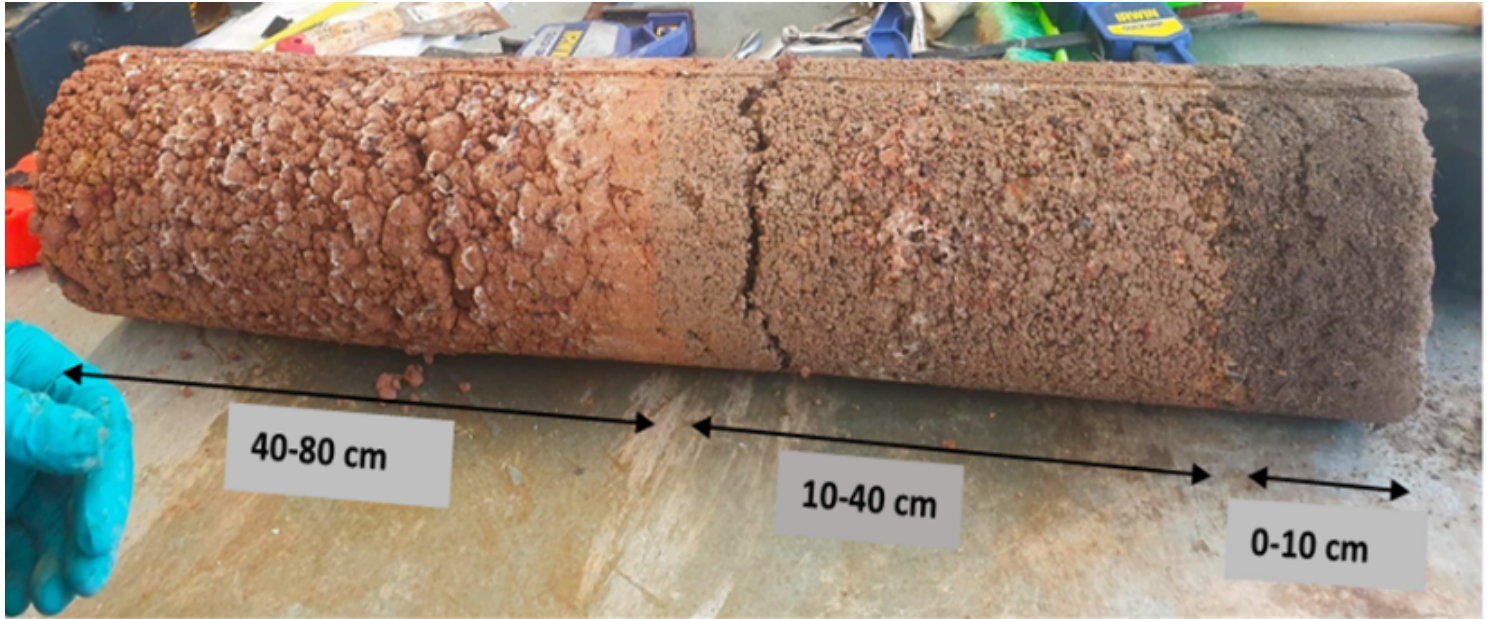


Figure 1

Reconstructed soil column with topsoil (0–10 cm), midsoil (10–40 cm) and subsoil (40–80 cm) layers at the end of the semi-controlled environment experiment.

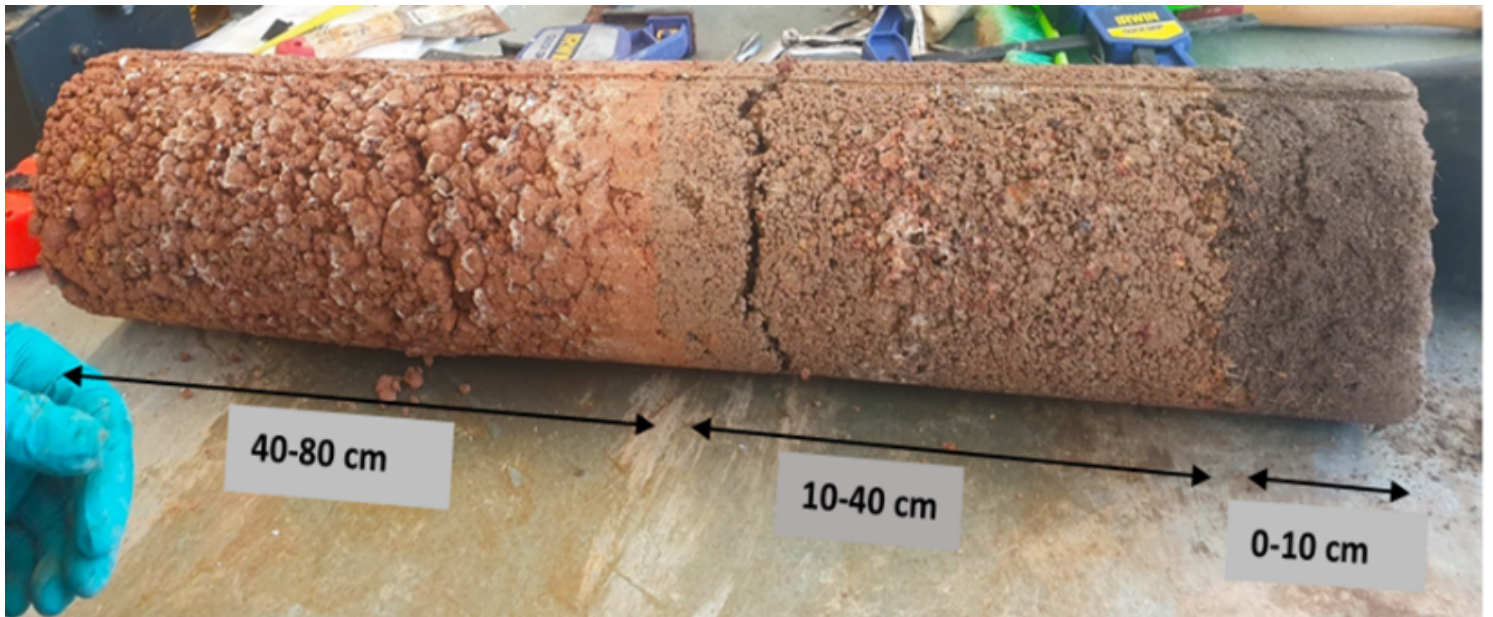


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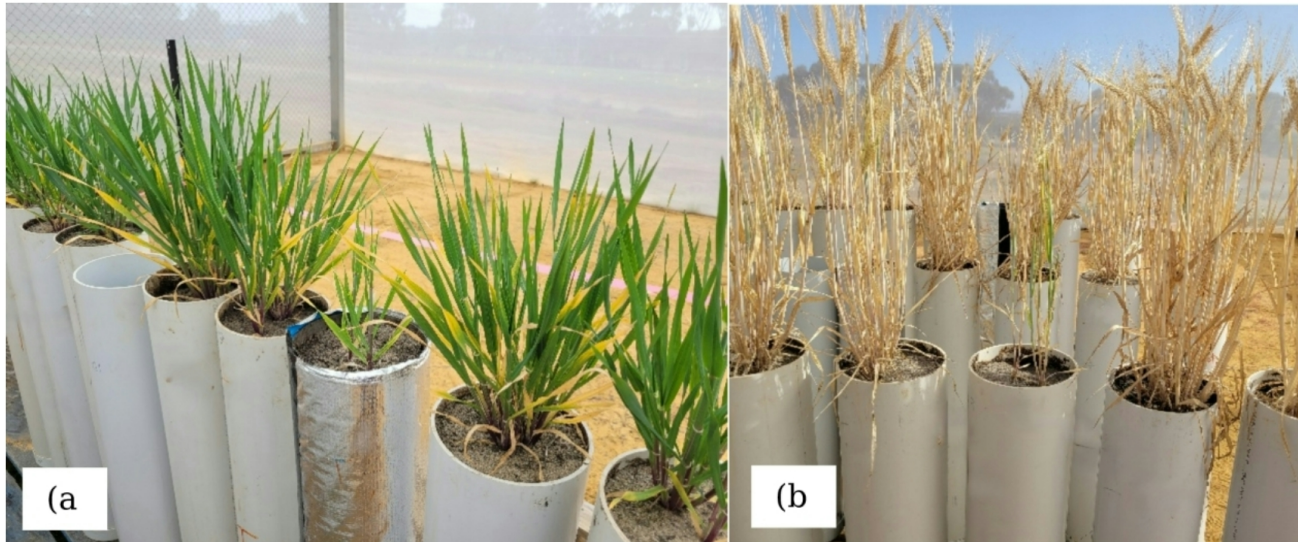


Figure 2

Barley (var. Maximus) growing in 80 cm-deep soil columns: (a) 82 days and (b) 141 days after seeding, in both reconstructed (PVC tubes) and intact columns (in metallic tubes wrapped with insulation foil).

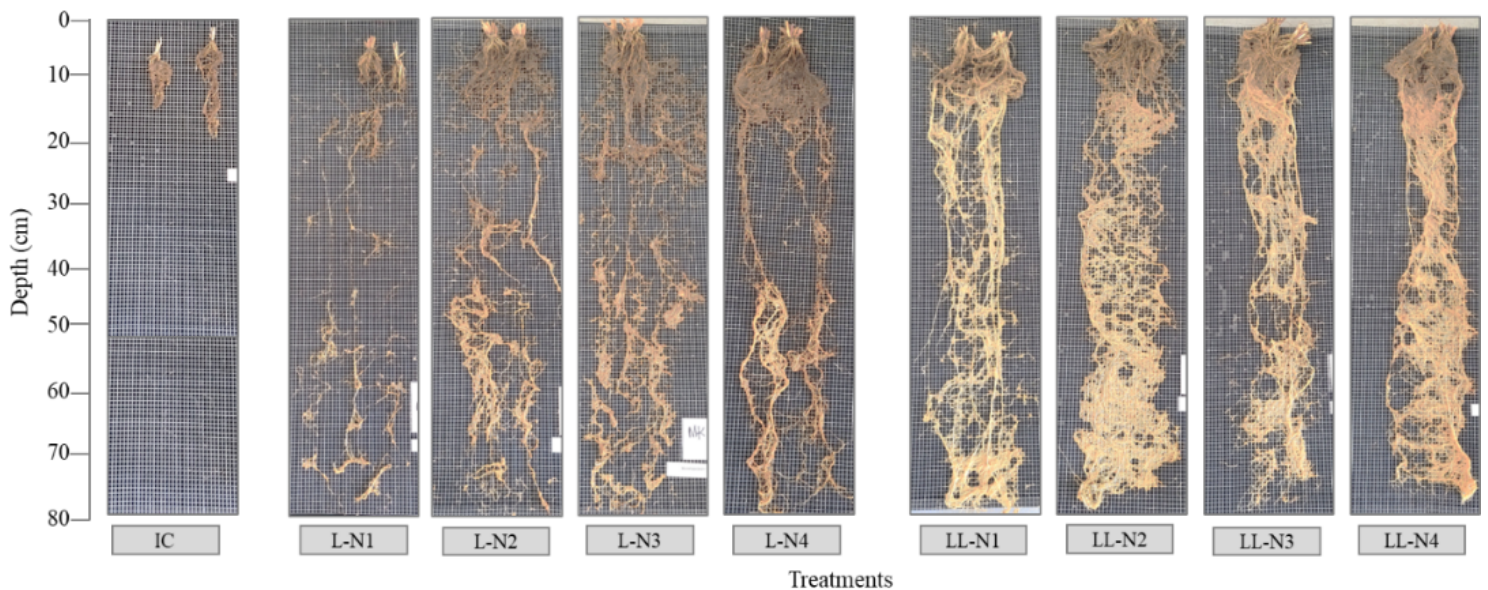


Figure 3

Images of barley (*Hordeum vulgare* L. var. Maximus) roots from various soil re-engineering treatments and nitrogen (N) rates in a Kurosol from Meenar, Western Australia. Soil RE treatments were IC = intact core, L = loosen and LL = loosen + lime. N rates were: N1 = 0, N2 = 75, N3 = 150 and N4 = 225 kg N ha⁻¹.

The whole root system was extracted using a modified pinboard method, as described by Kano-Nakata et al. (2011).

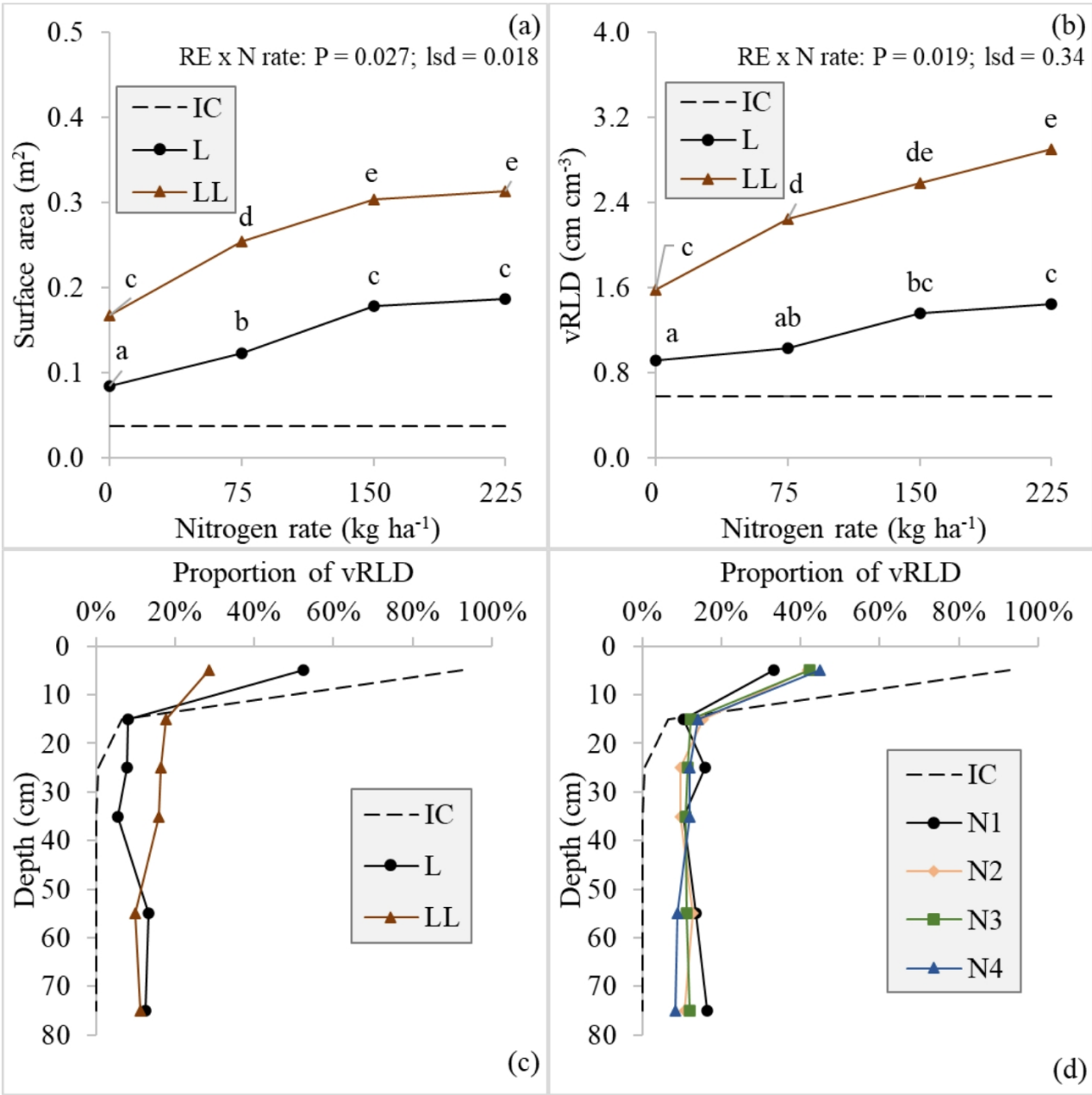


Figure 4

Effects of soil re-engineering (RE) treatments and nitrogen (N) rates on (a) root surface area, (b) volumetric root length density (vRLD) per column, (c-d) proportion of vRLD at each sampling depth of barley (*Hordeum vulgare* L. var. Maximus) crop in a Kurosol from Meenar, Western Australia. Soil RE treatments were IC = intact core, L = loosen and LL = loosen + lime. N rates were: N1 = 0, N2 = 75, N3 =

150 and N4 = 225 kg N ha⁻¹. Probability (P) and least significant difference (lsd) values are presented in each panel based on Fisher's protected lsd test.

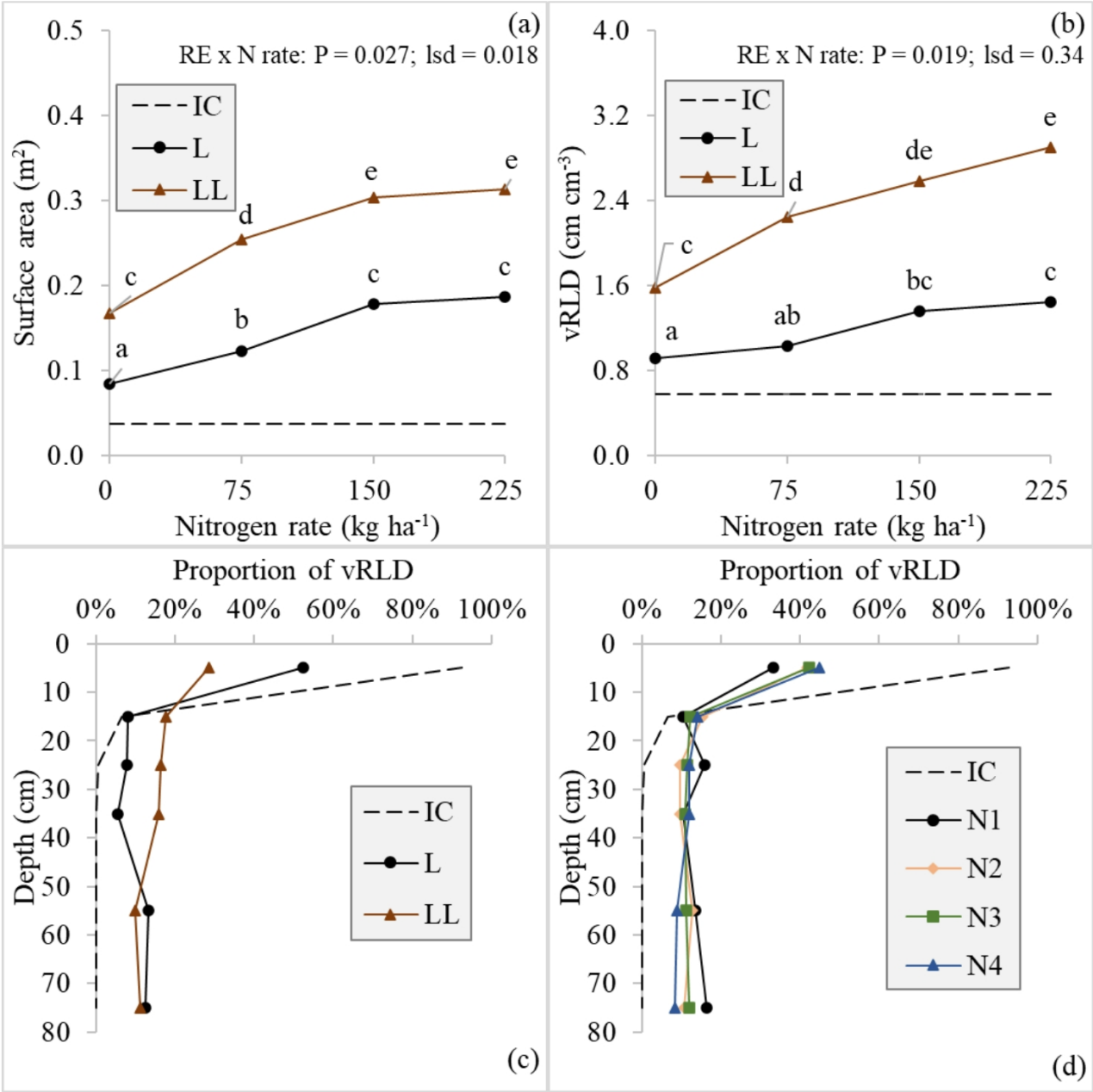


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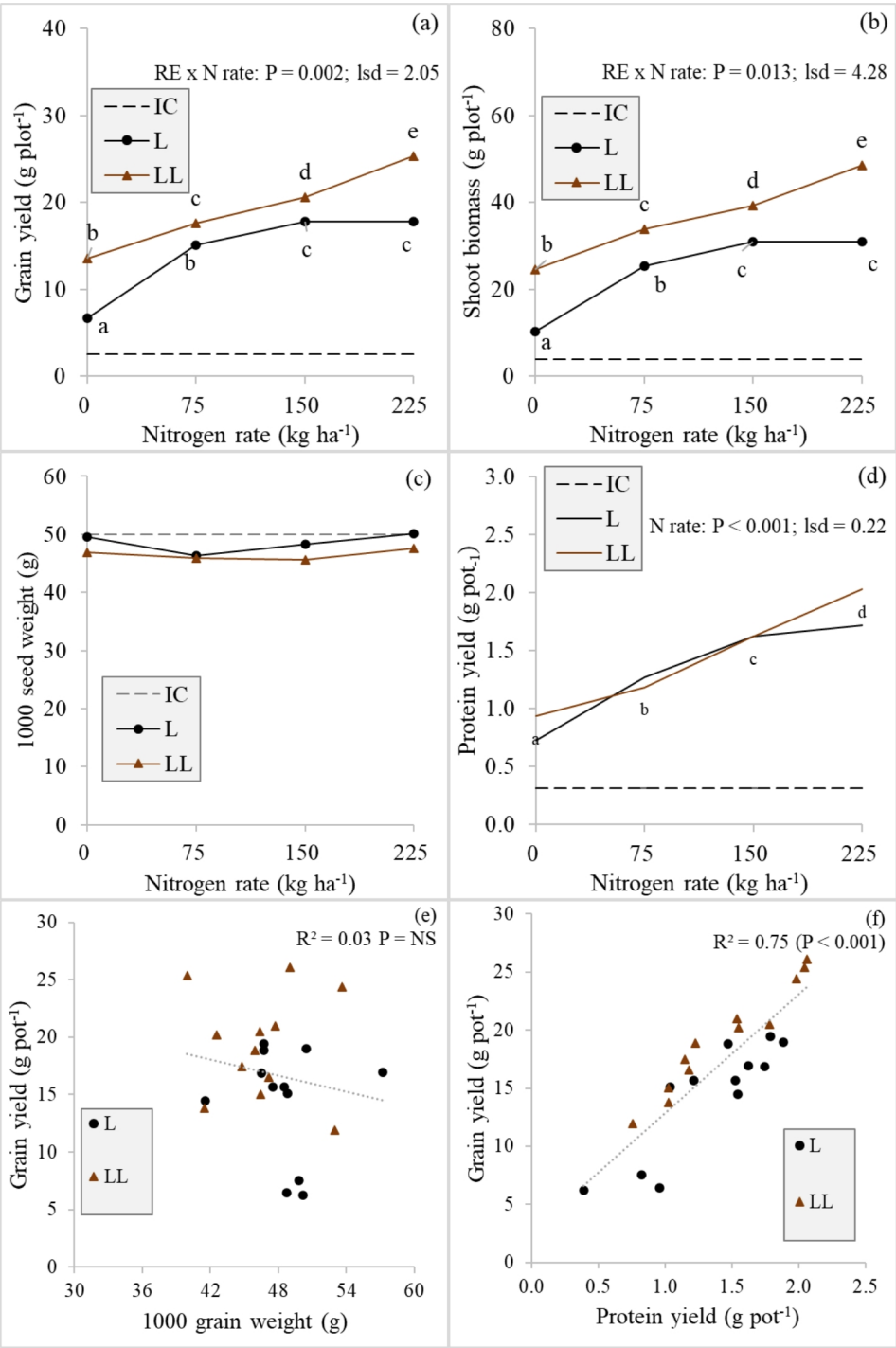


Figure 5

Effects of soil re-engineering (RE) treatments and nitrogen (N) rates on (a) grain yield, (b) shoot biomass, (c) harvest index, (d) 1000 grain weight, (e) relationship between grain size and grain yield, and (f) relationship between grain protein yield and grain yield of barley (*Hordeum vulgare* L. var. Maximus) crop

in a deep coarse sand from Meenar, Western Australia. Soil RE treatments were IC = intact core, L = loosen and LL = loosen + lime. Probability (P) and least significant difference (lsd) values are presented in each panel based on Fisher’s protected lsd test. R2 values are followed by the P values taken from a regression analysis where a significant relationship was determined at $P \leq 0.05$.

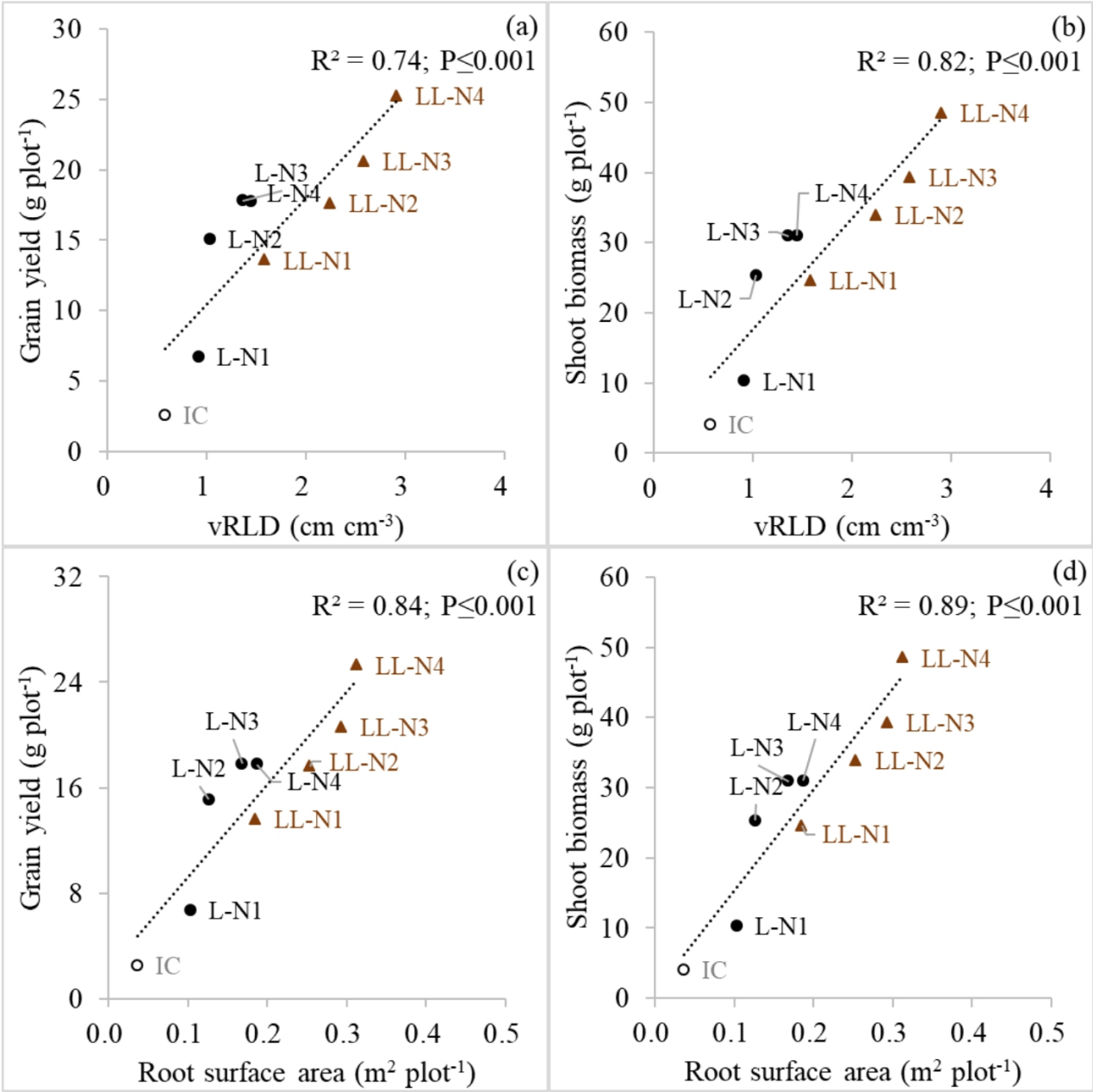


Figure 6

Effects of soil re-engineering (RE) treatments and nitrogen (N) rates on (a) relationship between vRLD and yield, (b) relationship between vRLD and shoot biomass, (c) relationship between root surface area and yield, and (d) relation between root surface area and shoot biomass of barley (*Hordeum vulgare* L.

var. Maximus) crop in a deep coarse sand from Meenar, Western Australia. Soil RE treatments were IC = intact core, L = loosen and LL = loosen + lime. N rates were: N1 = 0, N2 = 75, N3 = 150 and N4 = 225 kg N ha⁻¹. Probability (P) and least significant difference (lsd) values are presented in each panel based on Fisher's protected lsd test. R² values are followed by the P values taken from a regression analysis where a significant relationship was determined at $P \leq 0.05$.

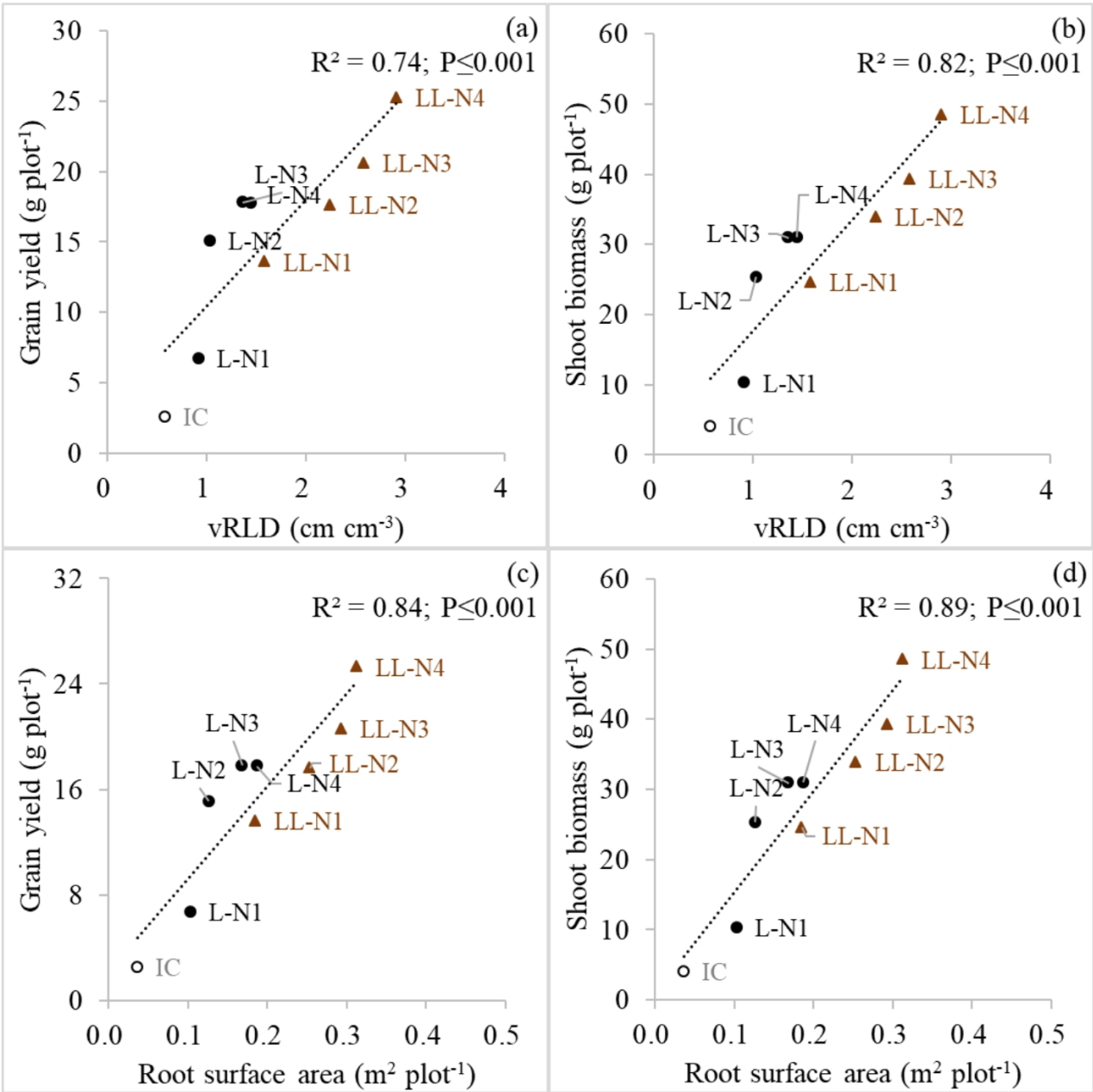


Figure 6

Effects of soil re-engineering (RE) treatments and nitrogen (N) rates on (a) relationship between vRLD and yield, (b) relationship between vRLD and shoot biomass, (c) relationship between root surface area

and yield, and (d) relation between root surface area and shoot biomass of barley (*Hordeum vulgare* L. var. Maximus) crop in a deep coarse sand from Meenar, Western Australia. Soil RE treatments were IC = intact core, L = loosen and LL = loosen + lime. N rates were: N1 = 0, N2 = 75, N3 = 150 and N4 = 225 kg N ha⁻¹. Probability (P) and least significant difference (Isd) values are presented in each panel based on Fisher's protected Isd test. R² values are followed by the P values taken from a regression analysis where a significant relationship was determined at $P \leq 0.05$.

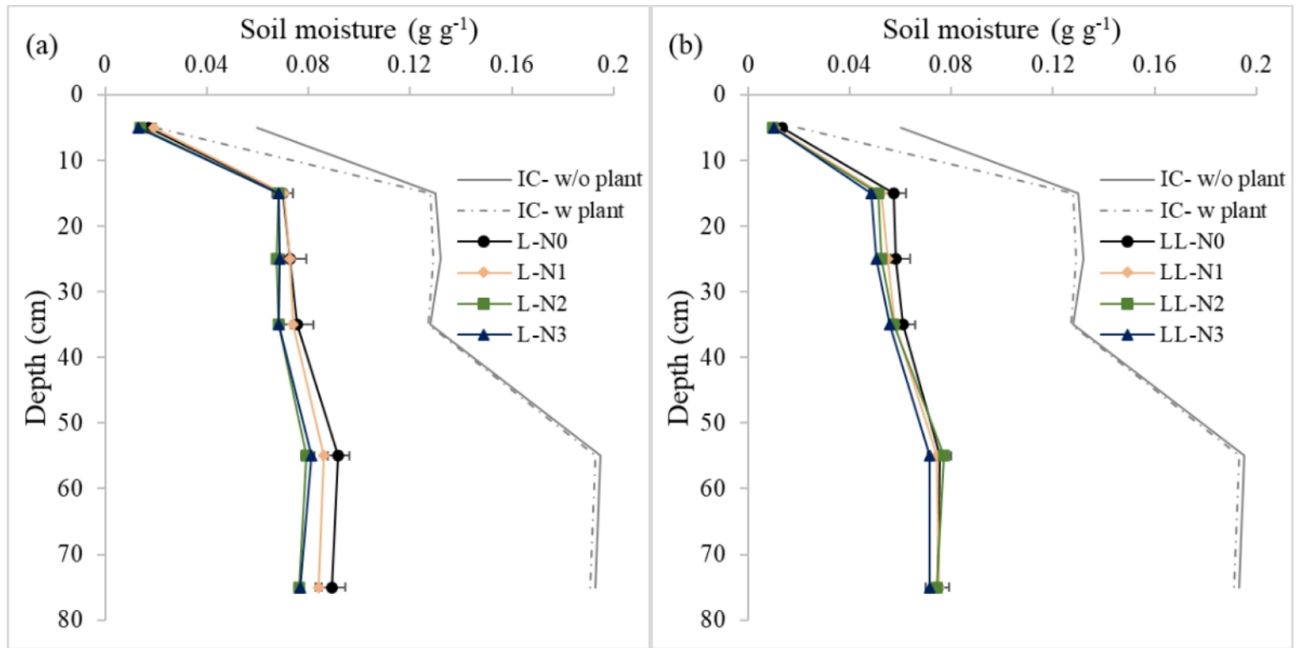


Figure 7

Effects of soil re-engineering (RE) treatments: (a) loosened, and (b) loosened + lime; and nitrogen (N) on soil water status in 80 cm soil profiles at the time of barley crop harvest. Soil RE treatments were IC = intact core, L = loosen, LL = loosen + lime and LLC = loosen + lime + clay. N rates were: N0 = 0, N1 = 75, N2 = 150 and N3 = 225 kg N ha⁻¹.

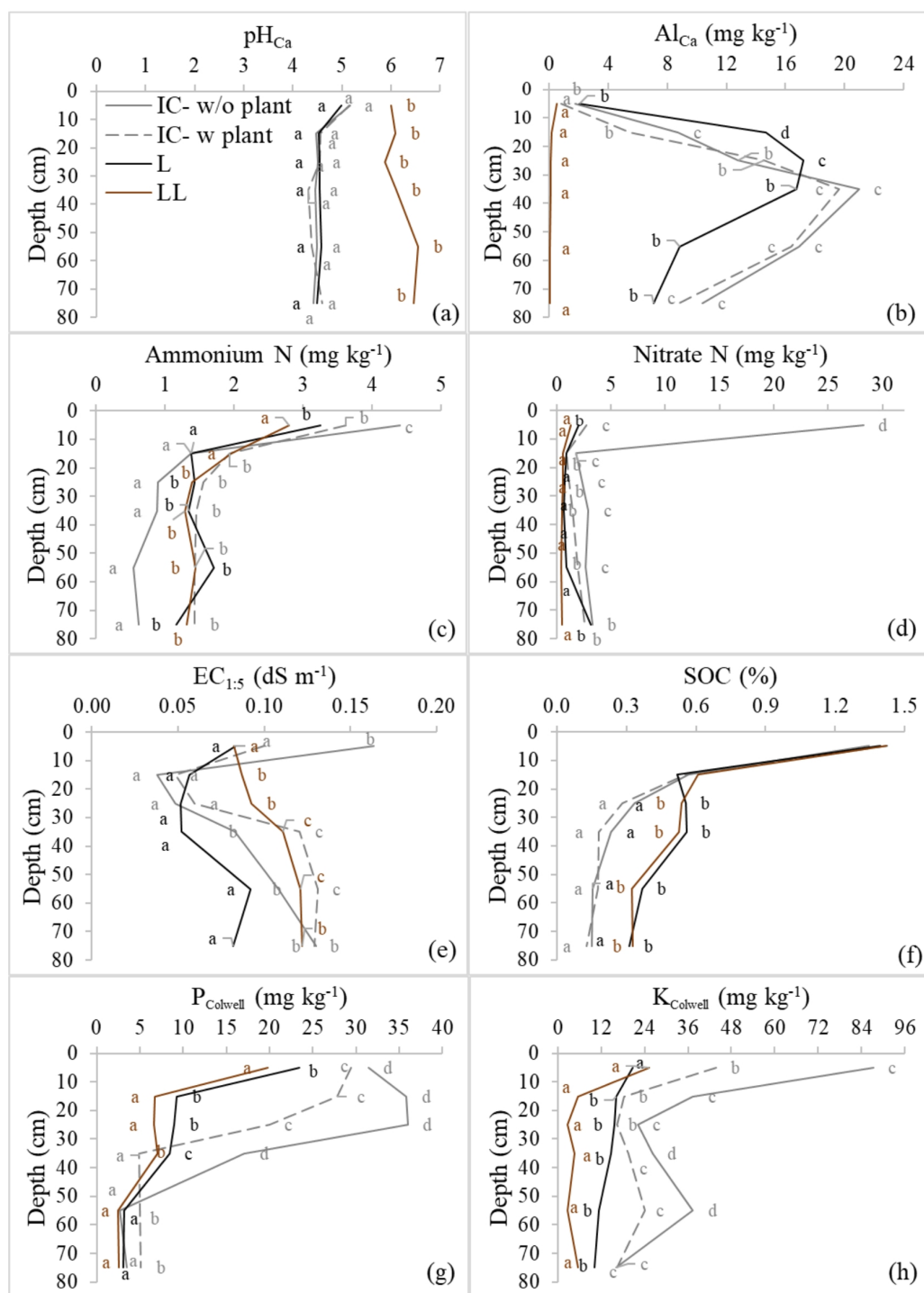


Figure 8

Effects of soil re-engineering (RE) treatments on soil (a) pH_{Ca} (b) Al_{Ca} , (c) ammonium nitrogen (N), (d) nitrate N, (e) $\text{EC}_{1:5}$, (f) soil organic carbon (SOC), (g) $\text{P}_{\text{Colwell}}$, and (h) $\text{K}_{\text{Colwell}}$. Soil samples were collected at the time of harvest. Soil RE treatments were IC = intact core (with and without plant), L = loosen and LL = loosen + lime. Different letters at the same sampling depth are statistically different at $P \leq 0.05$ based on Fisher's protected LSD test in the ANOVA procedure.

