

Soil inversion, good for yield but what happens to soil microbes?

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Key messages

- Soil inversion decreased soil pH in the surface 0-10cm and increased plant yield.
- Soil inversion initially altered the distribution of microbial populations in the soil profile but by harvest the effects of a one-off cultivation were negligible.
- Soil inversion has implications for soil nitrogen cycling by moving ammonia oxidizing bacteria that are responsible for nitrification deeper into the soil profile and moving ammonia oxidizing archaea upwards into the 0-10cm soil layer.

Aims

- To determine if a one-off soil inversion would increase plant yield.
- To quantify the distribution of bacteria and archaea in the soil profile under wheat cropping.
- To determine the effect of a single soil inversion on soil microorganisms, specifically bacteria, archaea, ammonia oxidising bacteria and ammonia oxidising archaea.

Methods

Field Trial

A field trial was established in 2013 at Dandaragan using a strip-plot design with randomisation and three replicate plots per treatment. Previous research (Scanlon et al. 2014) at this site has indicated that the yield benefit from a one-off soil inversion was enough to off-set the cost. The main treatments tested here were: 1. Control with no soil inversion (not spaded treatment); 2. Soil inverted using a rotary spader (spaded treatment).

An Imants 37SX rotary spader was used for soil inversion which achieved a cultivation depth of 35 cm. The soil is classified as a deep yellow sand (Orthic-Tenosol, Australian Soil Classification) with moderate soil fertility. Soil was collected at pre-seeding (May 2013) and harvest (November 2013) with three soil cores being collected from each replicate plot ($n = 3$) of each treatment. Soil from the three soil cores were combined to produce one sample per field plot at each of the following depth intervals (in cm): 0–5, 5–10, 10–20, 20–30, 30–40 and 40–60 and stored in a cooled container prior to transport back to the laboratory. Sub-samples for DNA extraction were frozen immediately upon collection in a portable freezer and transferred to $-20\text{ }^{\circ}\text{C}$ within 1 h.

pH

Soil pH was measured by adding 20 mL of 0.01 M CaCl_2 to 4 g of sieved ($< 2\text{ mm}$) air-dried soil and shaking for 1 h. The extractant was allowed to settle for 30 min before measuring pH with an electronic probe (Rayment and Higginson, 1992).

DNA extraction and microbial abundance

Bacteria and archaea are two groups of soil organisms that play an important role in soil ecosystem functioning (especially nutrient cycling) with the abundance of these soil microbial populations known to be strongly dependent on depth. Nitrification is performed by microorganisms that convert ammonia to nitrate. It is the major pathway by which nitrogen is lost from soil. Ammonia oxidising bacteria (AOB) and ammonia oxidising archaea (AOA) produce the key enzyme responsible for nitrification – ammonia monooxygenase (*amoA*). This enzyme is the rate-limiting enzyme involved in nitrification. In soil, the availability of inorganic nitrogen (ammonium and nitrate) is important for plant nutrition. Nitrogen loss by nitrification decreases the efficiency of nitrogen fertiliser use and can result in large economic losses. Nitrogen can be lost by nitrate leaching into waterways and contribution to groundwater pollution and through the conversion of nitrate to nitrous oxide which contributes to soil greenhouse gas emissions.

DNA was extracted from duplicate 800 mg sub-samples using PowerSoil™ DNA Extraction Kit (MoBio Laboratories Inc., Carlsbad, CA, USA). The abundance of total bacteria and archaea as well as organisms responsible for nitrification in soil, specifically ammonia oxidising bacteria (AOB) and ammonia oxidising archaea (AOA), was quantified using a Vii7 qPCR machine (Thermo-Fisher). To determine the abundance of bacteria,

the bacterial *16S rRNA* gene primers Eub338 and Eub518 were used. To determine the abundance of archaea, the archaeal *16S rRNA* gene primers Parch519F and Arch915R were used. To determine the abundance of ammonia oxidising bacteria, the bacterial *amoA* gene primers amoA-1F and amoA-2R were used. To determine the abundance of ammonia oxidising archaea, the archaeal *amoA* gene primers Arch-amoAF and Arch-amoAR were used. Standard curves were generated for each reaction with r^2 values greater than 0.99. Efficiencies for all quantification reactions were 80–100 %.

Results and Discussion

Soil pH

At pre-seeding and harvest soil pH was significantly ($p < 0.05$) higher in the surface 0-10cm compared to the sub-surface (10-60cm). At pre-seeding spading altered the soil pH profile and decreased pH in the surface from 6.50 for not-spaded plots to 5.25 in spaded plots. At harvest soil pH was still significantly ($p < 0.05$) lower in spaded plots when compared to not-spaded plots although the difference was not as great as at pre-seeding.

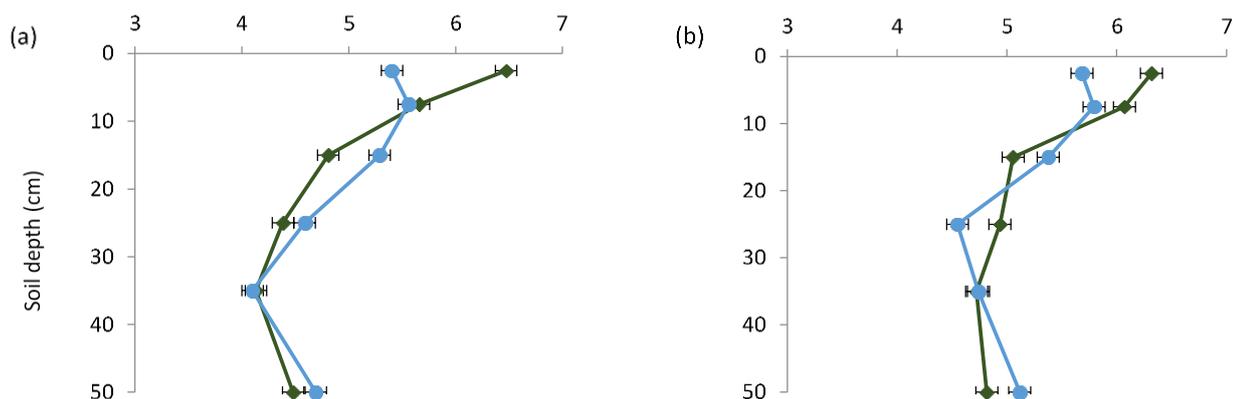


Figure 1 Mean pH at (a) pre-seeding and (b) harvest for spaded and not-spaded treatments. Green lines represent plots that were not spaded and blue lines represent spaded plots. Standard errors of the means ($n=3$) are shown.

Microbial populations

At this field site, bacteria were generally more abundant than archaea, and AOB were generally more abundant than AOA (Fig 2 and Fig 3) which is consistent with previous reports for WA soils (Banning et al. 2015). The field site exhibited distinct depth profiles for each of the soil microbial populations assessed as has been demonstrated in previous studies (Banning et al. 2015).

At pre-seeding, where no spading was applied, bacteria and archaea were both significantly more abundant ($p < 0.05$) in the top 0-10cm compared to the sub-soil (10-60cm). This contrasts with the spaded plots where bacteria and archaea were significantly ($p < 0.05$) less abundant in the top 0-10cm compared to the sub-soil (Fig 2a and Fig 3a). This indicates that spading moved both bacteria and archaea from the top 0-10cm down deeper in the soil profile. At harvest, the abundance of bacteria and archaea in spaded and not-spaded plots was not significantly different ($p > 0.05$) in the top 0-10cm. The spaded plots exhibited a similar depth profile of bacteria and archaea to the not-spaded plots indicating that the bacteria and archaea had re-established in the top 0-10cm over the course of the season (Fig 2c and Fig 3c).

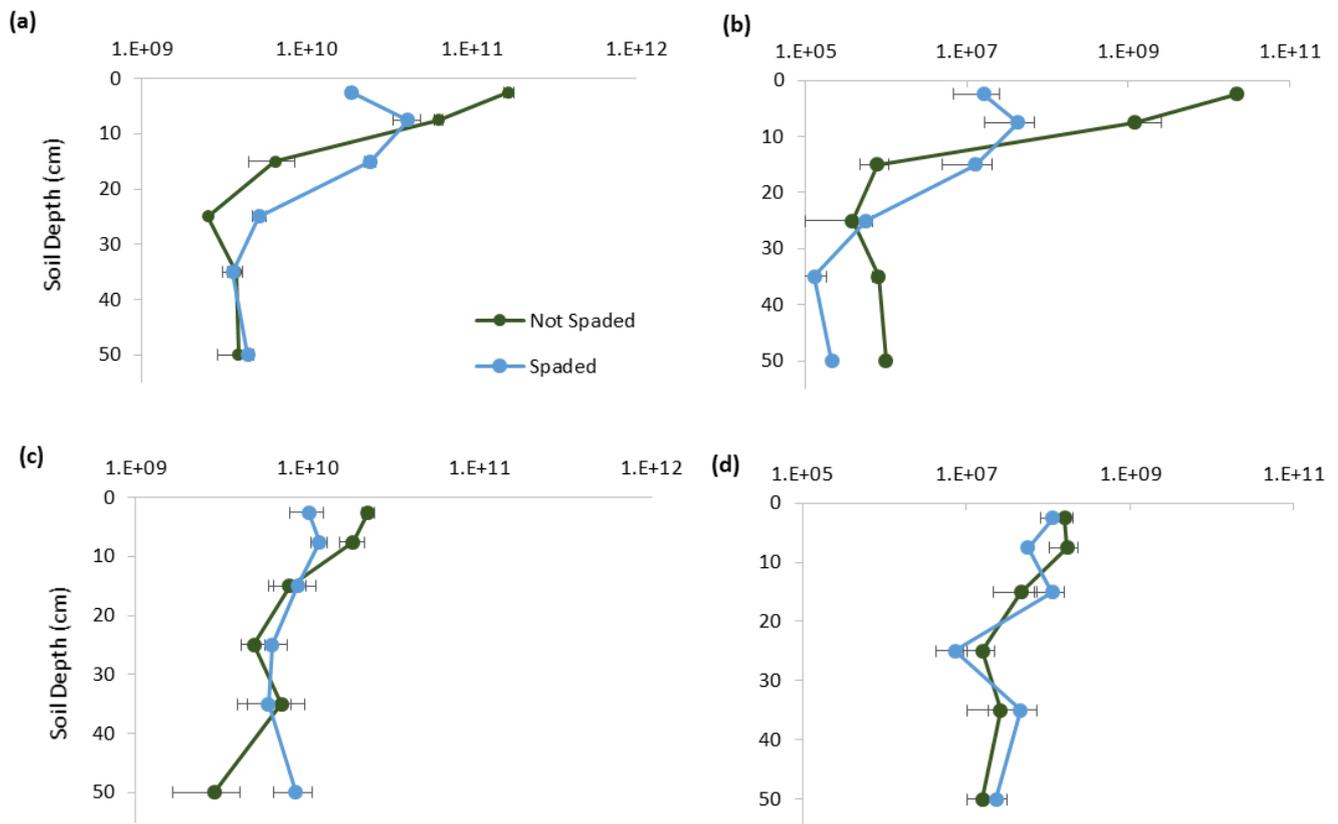


Figure 2 Abundance of (a) total bacteria (b) ammonia oxidising bacteria at pre-seeding (c) total bacteria (d) ammonia oxidising bacteria at harvest. Standard errors of the means (n=3) are shown.

At pre-seeding, where no spading was carried out, AOB were significantly more abundant ($p < 0.05$) than AOA in the top 0-10cm (Fig 2b and Fig 3b). This is consistent with other reports for WA soils where AOB dominate over AOA (Gleeson et al. 2010; Barton et al. 2013; Banning et al. 2015). Where spading was applied, the AOB were significantly less abundant ($p < 0.05$) in the top 0-10cm compared to the sub-soil (Fig 2b). At harvest, the abundance of AOB in spaded and not-spaded plots was not significantly different ($p > 0.05$) in the top 0-10cm (Fig 2d). The spaded plots exhibited a similar depth profile to the not-spaded plots indicating that the AOB had re-established in the top 0-10cm over the course of the season (Fig 2d).

Where spading was applied, the AOA were significantly more abundant ($p < 0.05$) in the top 0-10cm compared to the sub-soil (Fig 3b) indicating that spading had moved AOA from deeper in the soil profile into the top 0-10cm. At harvest the abundance of AOA in the top 0-10cm of spaded plots was still significantly greater ($p < 0.05$) than the not-spaded plots (Fig 3d) although the difference was not as great as at pre-seeding.

Previous research has shown that in WA soils AOB rather than AOA are responsible for nitrification (Barton et al. 2013; Banning et al. 2015). Moving AOB from the surface to deeper in the soil profile has the potential to increase nitrification in the sub-surface and thus this will need consideration in nitrogen fertiliser management, particularly at seeding when the effect of spading is most pronounced.

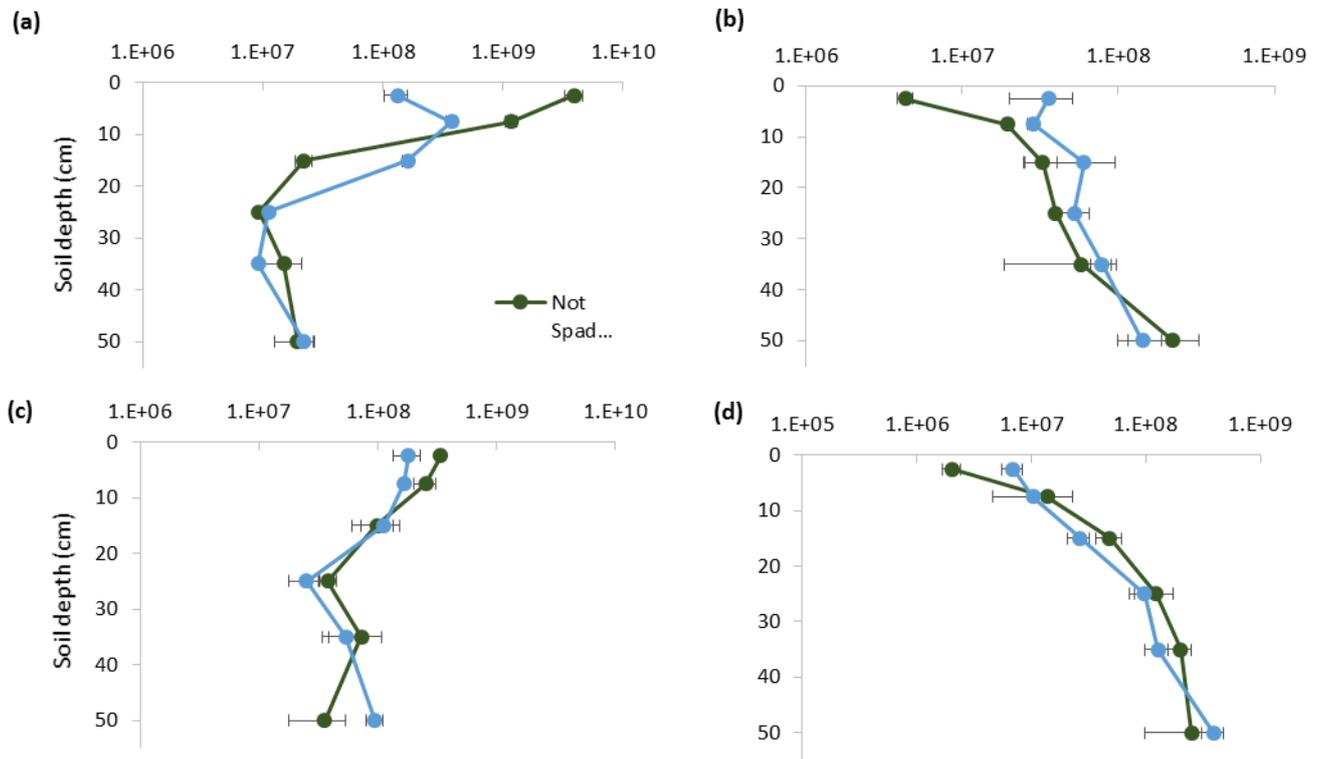


Figure 3 Abundance of (a) total archaea (b) ammonia oxidising archaea at pre-seeding (c) total archaea (d) ammonia oxidising archaea at harvest. Standard errors of the means (n=3) are shown.

Conclusions

- Spading decreased pH in the top 0-10cm as lower pH soil from the sub-surface was moved into the surface 0-10cm.
- AOA were more abundant and AOB less abundant at lower pH. This is consistent with reports of AOB being more pH sensitive than AOA.
- In the spaded plots only at pre-seeding AOB were less abundant while AOA were more abundant in the surface 0-10cm compared to the not-spaded plots where AOB outnumbered AOA which is consistent with other findings in WA.
- Spading has implications for soil nitrogen cycling as ammonia oxidising bacteria that are responsible for nitrification are moved deeper into the soil profile.
- At harvest the effect of on spading on soil microbes was negligible.

Key words

Soil inversion; bacteria; archaea; ammonia oxidising bacteria; ammonia oxidising archaea

Acknowledgments

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