



Figure S1: <2 mm pine biochar (left) and >4 mm pine biochar (right).

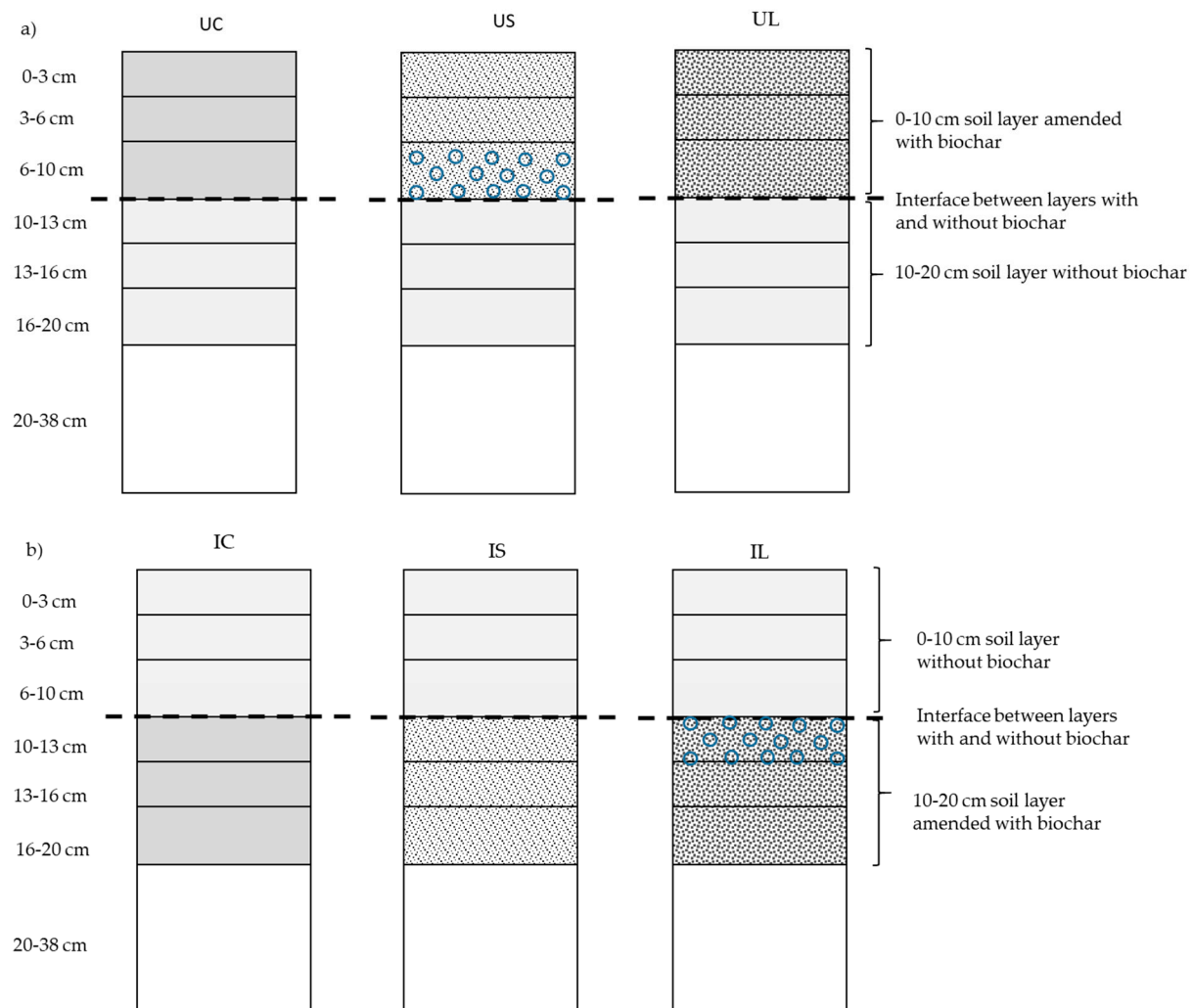


Figure S2: The soil in cores was packed as follows: a) Un-inverted soil; UC-control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – Un-inverted 0-10 cm with >4 mm biochar; b) Inverted soil; IC- Control inverted; IS- Inverted 0-10 cm with <2 mm biochar; IL - Inverted 0-10 cm with >4 mm biochar.

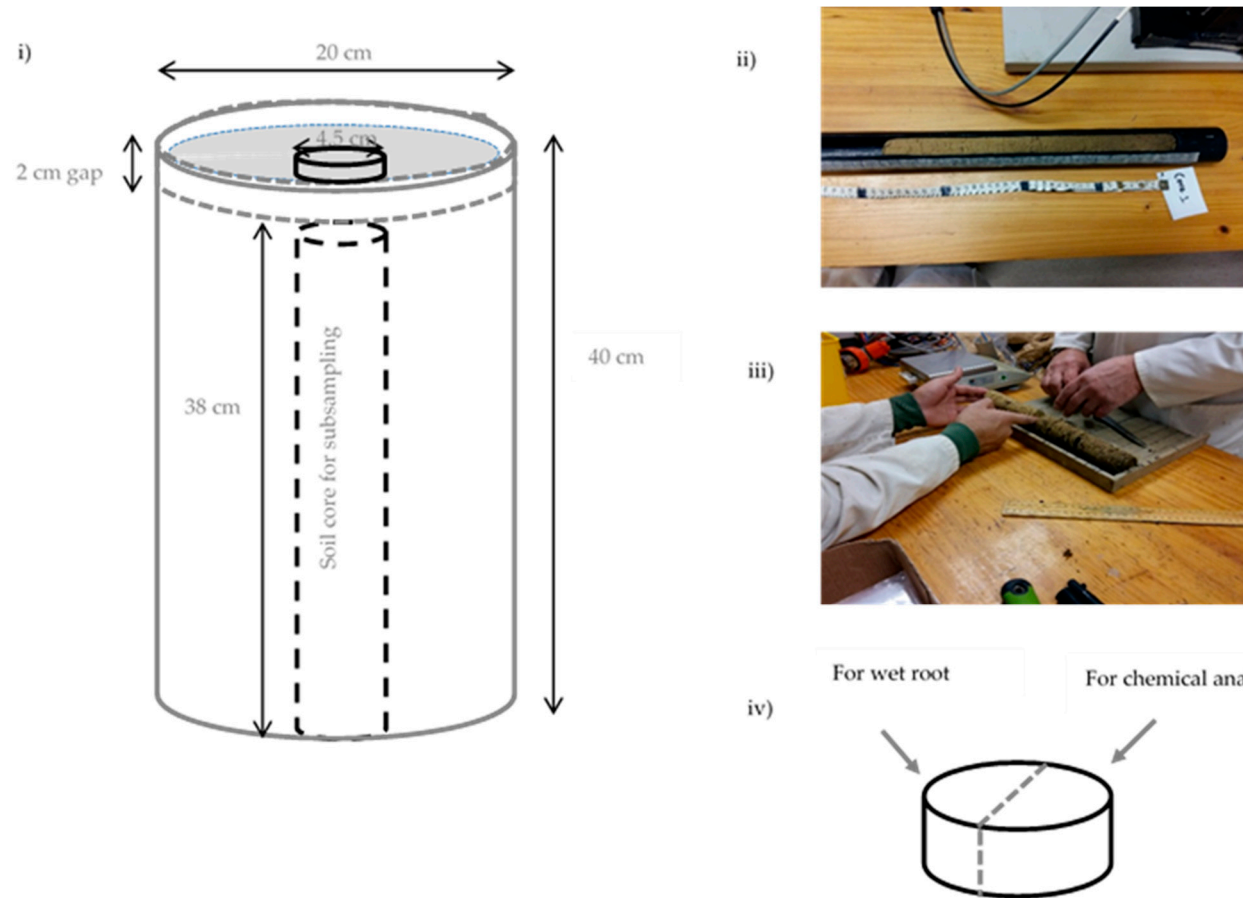


Figure S3: End-of-trial sampling; i) A soil core (subsample) was taken out from each core using a corer (diameter of 45 mm) from 0-38 cm depth; ii) Each core was scanned with NIRS; iii) soil was sliced according to designated depth; iv) each sliced soil was cut into half and separated for analysis.

Table S1: Time scale of the experiment.

| Date | Week | Event |
|---------------------------|------------------|--|
| 26/9/2014 | 1 | Finish setting up all cores and first irrigation to reach 50% WFPS- topsoil (0-10 cm) has been mixed with basic fertilizer to stimulate seedlings (NPK; 60 kg N ha ⁻¹ ; 50 kg K ha ⁻¹ and 60 kg P ha ⁻¹) N from urea, K from KCl and P from SSP. |
| 10/10/2014 | 3 | Sowing ryegrass seeds |
| 4/11/2014 | 7 | Thinning to 20 seedlings |
| 13/11/2014 | 8 | First harvest, cut to 5 cm length |
| 24/11/2014 | Week 10 & onward | Subsequent harvests |
| 15/12/2014 | 13 | Gas measurement to measure N ₂ O emission prior to urine application: sampling (day 0)- summer trial |
| 16/12/2014 | 13 | First application of cow urine equal to 354 kg N ha ⁻¹ , 3.5 mm of urine applied using syringe onto 1/3 of total area of soil core. |
| 17/12/2014 until 3/2/2015 | Weeks 13-20 | Gas measurement day 1 to day 49 |
| | Weeks 21-28 | Gap |
| 9 and 10/4/2015 | 29 | Leaching before 2 nd cow urine application |
| 27/4/2015 | 32 | Gas measurement to measure N ₂ O emission prior to urine application: sampling (day 0)-autumn trial |
| 28/4/2015 | 32 | Second application of cow urine equal to 563 kg N ha ⁻¹ |
| 29-4-2015 until 16-6-2015 | Weeks 32-39 | Gas measurement day 1 to day 49 |
| 16-9-2015 | 52 | Experiment setup dismantled. Soil cores were kept in the chiller room for further analysis. |

Table S2. Soil pH of each layer measured at the end of the experiment.

| Soil depth (cm) | Treatments | | | | | | | |
|--------------------|------------------|-------|-------|-----------------|---------------|-------|-------|-----------------|
| | Un-inverted soil | | | | Inverted soil | | | |
| | UC | US | UL | <i>p</i> -value | IC | IS | IL | <i>p</i> -value |
| Layer A (0-3 cm) | 5.74a | 5.72a | 5.68a | 0.906 | 5.68a | 5.67a | 5.62a | 0.411 |
| Layer B (3-6 cm) | 5.65a | 5.34a | 5.43a | 0.068 | 5.43a | 5.39a | 5.38a | 0.875 |
| Layer C (6-10 cm) | 5.52a | 5.43a | 5.34a | 0.481 | 5.29a | 5.38a | 5.37a | 0.728 |
| Layer D (10-13 cm) | 5.59a | 5.30a | 5.34a | 0.105 | 5.30a | 5.42a | 5.38a | 0.745 |
| Layer E (13-16 cm) | 5.54a | 5.25b | 5.34b | 0.034 | 5.29a | 5.42a | 5.37a | 0.387 |
| Layer F (16-20 cm) | 5.53a | 5.25a | 5.38a | 0.203 | 5.40a | 5.43a | 5.36a | 0.647 |

UC-control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – Un-inverted 0-10 cm with >4 mm biochar; IC- Control inverted; IS- Inverted 0-10 cm with <2 mm biochar; IL - Inverted 0-10 cm with >4 mm biochar. Different letters denote significant differences at $p < 0.10$ between treatments ($n = 4$) for each layer. In the un-inverted soil biochar was at 0-10 cm depth; in the inverted soil biochar was at 10-20 cm depth.

Table S3. N₂O emission fluxes measured during summer and autumn trial (data showed for selected days only).

| Summer Trial | | | | |
|--|----------------|------------|-----------------------------|-----------------------------|
| Day | Soil Inversion | Treatments | | |
| | | Control | Small-particle size biochar | Large-particle size biochar |
| Day prior to urine application | Un-inverted | 0.12bB | 1.51aA | 0.50bA |
| | Inverted | 2.08aA | 1.63aA | 0.19aA |
| Day after urine application with maximum N ₂ O peak | Un-inverted | 30.5aA | 60.7aA | 54.2aA |
| | Inverted | 13.9aA | 25.4aA | 29.1aA |
| Day 10 after urine application | Un-inverted | 3.11aA | 3.18aA | 4.41aA |
| | Inverted | 2.70aA | 15.9aA | 7.90aA |
| Autumn Trial | | | | |
| Day | Soil Inversion | Treatments | | |
| | | Control | Small-particle size biochar | Large-particle size biochar |
| Day prior to urine application | Un-inverted | 0.76bB | 4.61aA | 3.24aB |
| | Inverted | 21.8aA | 4.36bA | 24.8aA |
| Day after urine application with maximum N ₂ O peak | Un-inverted | 40.9aA | 85.7aA | 69.7aA |
| | Inverted | 66.9aA | 59.7aA | 93.8aA |
| Day 10 after urine application | Un-inverted | 14.6aA | 28.6aA | 14.1aA |
| | Inverted | 9.43aA | 23.4aA | 22.7aA |

Within a specific season, (i) different small letters within a row indicate differences between biochar treatments ($n = 4$) and $p < 0.10$), and (ii) different uppercase letters within a column indicate differences between un-inverted and inverted treatments ($n = 4$ and $p < 0.10$).

Table S2: Treatment effects on the amount of NO₃-N measured for every leaching event.

| Leaching event | NO ₃ -N leachate (NO ₃ -N g m ⁻² / % of added N leached as NO ₃ -N) | | | | | | | |
|-----------------|---|-------------|-------------|-----------------|-------------|-------------|-------------|-----------------|
| | UC | US | UL | <i>p</i> -value | IC | IS | IL | <i>p</i> -value |
| 1 st | 0.073/0.21a | 0.036/0.10a | 0.066/0.19a | 0.680 | 0.030/0.08a | 0.035/0.10a | 0.036/0.10a | 0.933 |
| 2 nd | 0.015/0.04a | 0.026/0.07a | 0.018/0.05a | 0.683 | 0.013/0.04a | 0.060/0.17a | 0.021/0.06a | 0.132 |
| 3 rd | 0.015/0.03a | 0.022/0.04a | 0.015/0.03a | 0.634 | 0.020/0.03a | 0.025/0.04a | 0.036/0.06a | 0.426 |

UC - control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – un-inverted 0-10 cm with >4 mm biochar; IC - control inverted; IS - inverted 0-10 cm with <2 mm biochar; IL - inverted 0-10 cm with >4 mm biochar. Different letters denote significant differences at $p < 0.10$ between treatments at each leaching event ($n = 4$).

Table S3: Soil bulk density (g/cm³) at each depth measured at the end of the experiment.

| Soil depth (cm) | Treatments | | | | | | | |
|--------------------|------------------|-------|-------|-----------------|---------------|-------|-------|-----------------|
| | Un-inverted soil | | | | Inverted soil | | | |
| | UC | US | UL | <i>p</i> -value | IC | IS | IL | <i>p</i> -value |
| Layer A (0-3 cm) | 0.92a | 0.93a | 0.84a | 0.115 | 1.16a | 1.06a | 1.12a | 0.271 |
| Layer B (3-6 cm) | 1.04a | 0.99a | 0.94a | 0.106 | 1.14a | 1.19a | 1.18a | 0.792 |
| Layer C (6-10 cm) | 0.97a | 1.06a | 0.99a | 0.101 | 1.05a | 1.17a | 1.08a | 0.186 |
| Layer D (10-13 cm) | 1.18a | 1.18a | 1.12a | 0.741 | 0.92a | 0.95a | 1.00a | 0.327 |
| Layer E (13-16 cm) | 1.17a | 1.10a | 1.11a | 0.599 | 1.00a | 0.94a | 0.92a | 0.417 |
| Layer F (16-20 cm) | 1.18a | 1.10a | 1.08a | 0.183 | 1.00a | 0.91a | 0.93a | 0.267 |

UC - control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – un-inverted 0-10 cm with >4 mm biochar; IC - control inverted; IS - inverted 0-10 cm with <2 mm biochar; IL - inverted 0-10 cm with >4 mm biochar. Different letters denote significant differences at $p < 0.10$ between treatments ($n = 4$) for each layer. In the un-inverted soil biochar was at 0-10 cm depth; in the inverted soil biochar was at 10-20 cm depth.

Table S4: Volumetric soil moisture contents (%) at different matric potentials including the available water content (AWC) measured after the end of the experiment.

| | | Treatments | | | | | | | |
|------------------|-----------------|------------------|-------------|-------------|-----------------|---------------|-------------|-------------|-----------------|
| Matric potential | Soil depth (cm) | Un-inverted soil | | | | Inverted soil | | | |
| | | UC | US | UL | <i>p</i> -value | IC | IS | IL | <i>p</i> -value |
| -15bar | 0-3 | 14.79±0.60 | 14.35±0.94 | 14.12±0.91 | ns | 14.79±0.85 | 14.02±0.12 | 14.61±0.86 | ns |
| | 3-6 | 19.78±0.67 | 19.36±0.78 | 18.58±0.77 | ns | 17.25±0.15 | 20.34±2.16 | 19.27±2.61 | ns |
| | 6-10 | 17.45b±0.01 | 19.37a±0.62 | 19.11a±0.03 | 0.058 | 18.51±6.70 | 18.25±0.60 | 17.25±2.09 | ns |
| | 10-13 | 18.85±1.06 | 19.98±0.10 | 16.46±1.91 | ns | 14.73b±0.90 | 16.22b±0.50 | 19.68a±0.35 | 0.024 |
| | 13-16 | 20.45±0.45 | 21.38±0.15 | 19.32±3.11 | ns | 20.28±0.21 | 16.21±0.82 | 18.52±1.08 | ns |
| | 16-20 | 20.79±0.62 | 20.81±0.12 | 19.49±1.75 | ns | 20.73±0.75 | 17.32±0.91 | 18.64±1.94 | ns |
| -1bar | 0-3 | 21.62±0.85 | 21.40±1.82 | 19.82±1.28 | ns | 19.98±1.34 | 19.22±0.17 | 19.96±0.98 | ns |
| | 3-6 | 26.17±0.27 | 24.89±2.21 | 23.45±1.02 | ns | 22.72±2.07 | 24.67±2.50 | 23.15±2.81 | ns |
| | 6-10 | 22.81b±0.14 | 27.81a±0.88 | 24.01b±0.37 | 0.016 | 15.43±1.02 | 22.15±1.01 | 20.74±2.52 | ns |
| | 10-13 | 26.16±1.93 | 26.96±0.88 | 22.37±2.60 | ns | 20.43b±0.95 | 21.66b±0.81 | 24.77a±0.28 | 0.053 |
| | 13-16 | 24.20±0.22 | 25.72±0.12 | 22.79±3.63 | ns | 24.00±1.04 | 20.99±1.53 | 22.86±1.18 | ns |
| | 16-20 | 24.05±1.46 | 21.13±2.53 | 21.92±2.31 | ns | 24.71±0.61 | 20.76±0.92 | 23.13±1.70 | ns |
| -0.3 bar | 0-3 | 27.31±1.08 | 26.99±2.18 | 24.58±1.67 | ns | 26.36±1.99 | 24.92±0.01 | 26.15±1.01 | ns |
| | 3-6 | 34.22±0.49 | 32.06±1.72 | 30.68±0.01 | ns | 27.66±0.25 | 32.72±2.87 | 30.71±3.15 | ns |
| | 6-10 | 28.76b±0.41 | 34.04a±1.24 | 30.06b±0.02 | 0.032 | 19.57±1.43 | 28.07±1.01 | 26.49±3.00 | ns |
| | 10-13 | 32.16±5.09 | 29.68±1.03 | 25.44±3.24 | ns | 23.52b±1.25 | 25.24b±0.72 | 28.99a±1.13 | 0.074 |
| | 13-16 | 29.07±0.27 | 29.23±0.08 | 26.85±4.42 | ns | 29.45±0.40 | 25.15±1.99 | 26.74±1.42 | ns |
| | 16-20 | 30.51±0.98 | 28.32±0.51 | 27.51±2.83 | ns | 28.89±0.50 | 26.61±1.01 | 27.57±2.37 | ns |
| AWC | 0-3 | 12.52±0.48 | 12.64±1.24 | 10.46±0.76 | ns | 11.58±1.14 | 10.90±0.12 | 11.55±0.16 | ns |
| | 3-6 | 14.45±0.19 | 12.69±0.94 | 12.10±0.76 | ns | 10.42±0.10 | 12.39±0.72 | 11.44±0.54 | ns |
| | 6-10 | 11.31±0.40 | 14.68±1.86 | 10.94±0.05 | ns | 4.38±1.94 | 9.82±0.41 | 9.24±0.91 | ns |
| | 10-13 | 13.31±4.04 | 9.70±0.93 | 8.98±1.32 | ns | 8.79±0.35 | 9.02±0.22 | 9.31±0.78 | ns |
| | 13-16 | 8.63±0.19 | 7.85±0.23 | 7.53±1.32 | ns | 9.17±0.20 | 8.94±1.17 | 8.21±0.35 | ns |
| | 16-20 | 9.73±0.36 | 7.51±0.39 | 8.02±1.08 | ns | 8.17±0.25 | 9.30±0.10 | 8.93±0.44 | ns |

UC-control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – Un-inverted 0-10 cm with >4 mm biochar; IC- Control inverted; IS- Inverted 0-10 cm with <2mm biochar; IL - Inverted 0-10 cm with >4 mm biochar. SEM ($n = 4$) and significant differences were detected at $p < 0.10$. Different letters denote significant differences between treatments for each layer. In the un-inverted soil biochar was at 0-10 cm depth; in the inverted soil biochar was at 10-20 cm depth.

Table S5: Soil total C (%) of each layer measured at the end of the experiment.

| Soil depth (cm) | Treatments | | | | | | | |
|--------------------|------------------|--------|--------|-----------------|---------------|--------|--------|-----------------|
| | Un-inverted soil | | | | Inverted soil | | | |
| | UC | US | UL | <i>p</i> -value | IC | IS | IL | <i>p</i> -value |
| Layer A (0-3 cm) | 3.85 b | 5.64 a | 5.23 a | 0.000 | 2.49a | 2.47a | 2.33a | 0.187 |
| Layer B (3-6 cm) | 3.76 b | 5.18 a | 4.91 a | 0.000 | 2.27a | 2.34a | 2.35a | 0.176 |
| Layer C (6-10 cm) | 3.23 b | 4.93 a | 4.77 a | 0.001 | 2.32a | 2.30a | 2.32a | 0.900 |
| Layer D (10-13 cm) | 2.30a | 2.30a | 2.31a | 0.971 | 3.42 b | 5.04 a | 4.79 a | 0.003 |
| Layer E (13-16 cm) | 2.24a | 2.29a | 2.29a | 0.299 | 3.61 c | 5.20 a | 4.58 b | 0.000 |
| Layer F (16-20 cm) | 1.96a | 2.17a | 2.30a | 0.100 | 3.24 c | 5.22 a | 4.09 b | 0.000 |

UC - control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – un-inverted 0-10 cm with >4 mm biochar; IC - control inverted; IS - inverted 0-10 cm with <2 mm biochar; IL - inverted 0-10 cm with >4 mm biochar. Different letters denote significant differences at $p < 0.10$ between treatments ($n = 4$) for each layer. In the un-inverted soil, biochar was at 0-10 cm depth; in the inverted soil biochar was at 10-20 cm depth.

Table S6: Dry root (mg cm⁻³) in each soil layer measured at the end of the experiment.

| Soil depth (cm) | Treatments | | | | | | | |
|--------------------|------------------|--------|-------|-----------------|---------------|--------|--------|-----------------|
| | Un-inverted soil | | | | Inverted soil | | | |
| | UC | US | UL | <i>p</i> -value | IC | IS | IL | <i>p</i> -value |
| Layer A (0-3 cm) | 5.61a | 10.99a | 5.61a | 0.522 | 5.83a | 9.87a | 7.63a | 0.415 |
| Layer B (3-6 cm) | 4.04a | 1.35a | 3.59a | 0.318 | 1.57a | 0.90a | 1.79a | 0.539 |
| Layer C (6-10 cm) | 0.68a | 0.85a | 0.85a | 0.908 | 0.34 a | 1.88 b | 1.54 b | 0.078 |
| Layer D (10-13 cm) | 0.45a | 0.67a | 0.90a | 0.192 | 2.69 a | 1.12 b | 0.67 b | 0.009 |
| Layer E (13-16 cm) | 0.45a | 0.43a | 0.44a | 1.000 | 0.67a | 0.90a | 0.89a | 0.898 |
| Layer F (16-20 cm) | 0.34a | 0.33a | 0.51a | 0.829 | 0.34a | 0.51a | 0.85a | 0.563 |

UC - control un-inverted; US – Un-inverted 0-10 cm with <2 mm biochar; UL – un-inverted 0-10 cm with >4 mm biochar; IC - control inverted; IS - inverted 0-10 cm with <2 mm biochar; IL - inverted 0-10 cm with >4 mm biochar. Different letters denote significant differences at $p < 0.10$ between treatments ($n = 4$) for each layer. In the un-inverted soil, biochar was at 0-10 cm depth; in the inverted soil biochar was at 10-20 cm depth.