

1 **The effect of tillage depth and traffic management on soil properties and**  
2 **root development during two growth stages of winter wheat (*Triticum***  
3 ***aestivum L.*)**

4  
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9 **4.1 Abstract**

10 The management of agricultural soils during crop establishment can affect root development by changes to soil  
11 structure. This paper assesses the influence of tillage depth (250 mm, 100 mm & zero) and traffic management  
12 (conventional tyre pressure, low tyre pressure & no traffic) on wheat root system architecture during winter wheat  
13 (*Triticum aestivum L.*) tillering and flowering growth stages (GS) on a long-term tillage trial site. The study  
14 revealed that zero-tillage systems increased crop yield through significantly greater root biomass ( $P < 0.001$ ), root  
15 length density and deeper seminal rooting analysed using X-ray Computed Tomography (CT) ( $P < 0.001$ )  
16 compared to trafficked treatments. In general, conventional pressure trafficking had a significant negative  
17 influence on crop yield ( $P < 0.01$ ), root development ( $0.001$ ), bulk density ( $P < 0.05$ ) and total soil porosity ( $P <$   
18  $0.05$ ) of deep and shallow tillage conventional pressure systems compared no traffic zero and deep tillage systems.  
19 Visual improvements in soil structure under zero tillage may have improved crop rooting in zero tillage treatments  
20 through vertical pore fissures (biopores), enhancing water uptake during the crop flowering period. This study  
21 highlights the increasing implications of soil structural damage on root system architecture created by compaction  
22 machinery trafficking in crop production. Although tillage method was less important, theThe constricted root  
23 systems found inwere more pronounced in conventional pressure shallow tillage and zero and deep tillage  
24 systems. trafficked regimes-emphasizinges the importance of using controlled traffic farming methodstechnology  
25 to improve soil management and reduce the trafficked areas of agricultural fields.

26 **1.4.2 Introduction**

27 Soil resources are under significant pressure from anthropogenic activities especially conventional tillage. The  
28 resulting soil degradation has significant implications for food security globally (Lal, 2010). Changing weather  
29 patterns from prolonged rain to drought periods are being experienced on a global scale, substantiating the  
30 challenges faced by food producers. In 2018, worldwide wheat production fell by 34.5 million ton due to  
31 prolonged droughts across Europe, Australia, and Canada. Soil compaction from field traffic is a well-recognized  
32 problem in many parts of the world (Chan et al., 2006; Arvidsson and Keller, 2007; Naderi-Boldaji et al., 2018  
33 (Chan et al., 2006; Arvidsson and Keller, 2007; Naderi-Boldaji et al., 2018) affecting 33 million hectares in  
34 Europe alone (Akker and Canarache, 2001). Soil compaction is a form of physical degradation caused by short  
35 crop rotations and heavy farm machinery working on low organic matter soils in wet conditions resulting in the  
36 loss of pore space due to an externally applied load, forcing soil aggregates together (Defossez and Richard, 2002).

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37 The resulting anaerobic high density soils have significantly reduced capacity to store water and nutrients required  
38 by growing crops (Hamza and Anderson, 2005) and severely compacted soils prevent soil exploration from root  
39 growth (Tracy et al., 2012a).

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40 Soil compaction is due in part to the pressure to complete field operations such as harvesting or drilling  
41 often in short windows of good weather, which is exacerbated by the increasing use of larger machinery with  
42 increasing axle loads designed to improve operational efficiencies. Common agricultural operations are conducted  
43 using wheeled farm machinery which has tripled in weight and power since 1966 with wheel loads rising by a  
44 factor of six (Chamen, 2006). When soils are cultivated in moist or wet conditions, soils can not withstand the  
45 compressive forces applied post cultivation by heavy farm machinery traffic during operations such as seeding  
46 (Raper, 2005), resulting in soil degradation (Batey, 2009). When soil is wet, tyre stress can propagate a greater  
47 distance down through the soil profile. The depth and severity of soil stress is related to soil moisture, traction  
48 device applied (track or tyre), track size, tyre inflation pressure and wheel load (Naderi-Boldaji et al., 2018).

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49 Reforming the approach to soil management to mitigate challenges such as soil compaction and soil  
50 erosion offer significant financial and environmental benefits compared to conventional agriculture. Cultivation  
51 practice using minimal, or zero tillage techniques are widespread across many climatic conditions from semi-arid  
52 Canadian plains to the temperate climates of Western Europe. In conventional tillage, the soil is either inverted  
53 >200 mm using a mouldboard plough or deeply ripped using tines. The soil is then cultivated again to break down  
54 soil aggregates to a crumb structure or fine tilth that is suitable to plant seeds (Morris et al., 2010). Conservation  
55 tillage, also known as non-inversion tillage or reduced tillage, has been used for decades to improve soil structure  
56 and health (Skaalsveen, Ingram and Clarke, 2019). Under conservation tillage, soil is disturbed to a lesser extent  
57 (<100 mm using tines or discs) or not disturbed at all such as under zero tillage which involves the direct placement  
58 of seed into undisturbed crop residues (Soane et al., 2012).

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59 The successful adaption of reduced tillage systems is not universally guaranteed with factors such as soil  
60 texture and drainage, crop type and weather influencing successful implementation (Soane et al., 2012). In  
61 northern Europe, crop yields under reduced cultivation systems rarely exceed those achieved by ploughing  
62 (Arvidsson, 2010). The exception under drier arid climates such as Spain, no tillage improved crop yields by  
63 moisture retention in below average rainfall years (Muñoz-Romero et al., 2010). Higher bulk density and  
64 penetration resistance are typically found throughout the formerly tilled or "plough pan" layer in no tillage soils  
65 within the first two years of adoption, resulting in root mechanical impedance (V. Boguzas et al., 2006). Yet, over  
66 time, long term zero tillage has shown to attribute improvements in soil pore architecture and continuity  
67 throughout the soil profile by bioturbation, suggesting roots could penetrate to lower soil horizons (Cooper et al.,  
68 2021).

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69 To date, studies have focused on how tillage influences physical soil properties (bulk density, cone  
70 penetrometer, soil aeration) with root and crop yield responses (Whalley et al., 2008; Pires et al., 2017; Czyż,  
71 2004). Soil types and tillage systems have a considerable influence on the structural integrity of soil which controls  
72 rooting potential (Morris et al., 2017a). Studies have shown that low pressure tyres can reduce surface compaction  
73 compared to high tyre pressure (Soane et al., 1980; Boguzas and Hakansson, 2001). As trafficking increases soil  
74 strength and reduces a plant root's ability to penetrate soil layers, it is important to understand the relationship

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75 between tillage depth and root system architecture during the growing season in response to trafficking. A dearth  
76 of information exists on how tillage depth and tyre pressure affect rooting properties and crop yield on longer  
77 term field sites. Yield reduction by soil surface compaction can increase abiotic stress in plants in three ways. It  
78 reduces soil aeration, increases mechanical impedance of roots which in turn reduces root exploration of soil thus,  
79 mitigating the extraction of water and nutrients from the soil resource (Chamen, 2011).

80 Quantitative measurement of root system architecture in three dimensions (3D) has become tractable  
81 using X-ray CT in pot experiments (Mairhofer et al., 2017). Few examples of root studies using high resolution  
82 X-ray computed tomography have been successfully conducted in field trials using undisturbed soil cores. Many  
83 studies have focused on measuring soil structural properties such as porosity, soil pore size and distribution and  
84 the influence of tillage method and trafficking (Millington et al., 2017; Rab et al., 2014). However, studying root  
85 development and architecture in three-dimensional field structured soils remains challenging with X-ray CT due  
86 to a bottleneck of rapid and standardized root extraction methods available, insufficient resolution and inability to  
87 segment similarities in grey scale values between root and organic materials (Zhou et al., 2021; Mooney et al.,  
88 2012; Pfeifer et al., 2015).

89 The purpose of this paper was to identify the in-situ relationship the effect and interaction of machinery  
90 traffic and between tillage depth and using commercial crop establishment methods. on R root architecture, soil  
91 physical structure and crop yield under different traffic methods were studied during two key growth stages of  
92 winter wheat. X-ray CT was deployed to show if root architecture behaviors could be captured in-situ to the soil  
93 structural environment created by the tillage method. Three cultivation practices and traffic management systems  
94 were studied: Deep tillage (250 mm), shallow tillage (100 mm) and zero tillage, under no traffic, low tyre pressure  
95 and conventional tyre pressure. The objectives of this study were to (i) assess the relationship between of traffic  
96 management and three tillage depths and its effects on root system architecture and soil physical properties (ii)  
97 Utilise 3D image analysis along with 2D destructive methods to verify rooting properties responsible for crop  
98 yield.

### 99 2.4.3 Materials and Methods

#### 100 2.4.3.1 Site and soils

101 The study took place during the 2018/19 growing season. The experimental site is 3.12 ha, located at Harper  
102 Adams University (HAU), Edgmond, Newport, England (52.779738 N, -2.426886 W). The HAU site is a loamy  
103 sand soil consisting of the Olerton and Salwick series soils (Eutric Endogleyic Arenosol and Chromic Endostagnic  
104 Luvisol respectively) (Millington et al., 2017). Further details of the soil properties are described in Table 1. To  
105 highlight if any site variability existed across the site, soil properties were examined for fertility (pH and nutrient  
106 levels), bulk density, soil strength and soil moisture. Particle size analysis (Gee and Or, 2002) was conducted to  
107 determine soil texture classifications. The trial site was established in 2011 for previous studies with plots and  
108 treatments carried out in the same location.

110 The experimental site was established in 2011 with each plot receiving the same tillage and traffic  
111 treatment as this study. In the year prior to this study, it was necessary to plant a break crop (2017/18) as part of  
112 a standard crop rotation to improve soil conditions and reduce diseases such as take all (*Gaeumannomyces*

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113 ~~graminis var. tritici). A field bean (*Vicia Fabia*) break crop was planted, and yields were assessed to ensure the~~  
 114 ~~trial site was uniform with no underlying issues. During the trial period, the site was treated with a standard~~ Since  
 115 ~~the trial site began, the~~ crop rotation ~~has been first~~with winter wheat (*Triticum aestivum L.*) harvested in 2012  
 116 followed winter wheat in 2013, winter barley (*Hordeum vulgare L.*) 2014, winter barley 2015, followed by a cover  
 117 crop “TerraLife-N-Fixx” (DSV United Kingdom Ltd, 2015); Spring oats 2016, spring wheat 2017 and winter  
 118 beans 2018. In the year prior to this study, it was necessary to plant a break crop (2017/18) as part of a standard  
 119 crop rotation to improve soil conditions and reduce diseases such as take all (*Gaeumannomyces graminis var.*  
 120 tritici). A field bean (*Vicia Fabia*) break crop was planted, and yields were assessed to ensure the trial site was  
 121 uniform with no underlying issues. For this trial, winter wheat (*Triticum aestivum L. cv. Graham*) followed the  
 122 bean crop and was drilled early October 2018 when the soil was dry, friable and soil temperatures >6 °C. The  
 123 seeding rate was 250 seeds per m<sup>2</sup> and drilling took place on the 5<sup>th</sup> of October. This is in line with local normal  
 124 farming practice.

126 **Table 4.1:** Description of the topsoil (0-300 mm) properties for Harper Adams University trial site, Shropshire,  
 127 UK.

Property	Units	
Location	Latitude	52.779738 N
	Longitude	-2.426886 W
Soil type	Landis group*	Argillic brown earths, brown sands
	Landis series*	Salwick, Ollerton
	FAO	Luvisol & Arenosol
Sand (2000-65µm)	g g <sup>-1</sup> dry soil	0.743
Silt (63-2µm)	g g <sup>-1</sup> dry soil	0.115
Clay (<2µm)	g g <sup>-1</sup> dry soil	0.143
Texture	SSEW class	Loamy sand
Organic matter (LOI)	g g <sup>-1</sup> dry soil	0.044

128 \*Landis Soil guide (University, 2021).

129 LOI, Loss of Ignition.

130

131 2.4.3.2 Experiment design

132 The experiment was a randomised 3 x 3 factorial arrangement of 9 treatments in four complete replicate blocks.  
 133 Each plot was 4 m wide x 84 m long with exception of block 4. Block 4 is 78.2 m long for operational reasons.  
 134 Tramlines were at a 90° angle to plots with 24 m spacing for fertilising and spraying operations throughout the  
 135 growing season. A split-plot design was used, half the plot (30 m) designated for sampling and the other half was  
 136 undisturbed for yield data collection. The half plot for sampling was sub-divided for the two sampling stages,  
 137 ensuring sampling did not occur near the same location as the previous sample. Cultivation for spring beans in  
 138 2017 was performed at three depths, 250 mm for deep tillage, 100 mm for shallow tillage and direct into stubble

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139 for zero tillage. In the winter wheat trial, soil cores were collected at tillering (Growth stage (GS) 25) and the  
140 flowering stage (GS 61-69) (Zadoks et al., 1974) in July 2019.

141 Three commercial crop establishment systems were used consisting of three different tillage depths. The  
142 following tillage treatments are denoted as: Treatment 1 = Deep tine cultivator at 250 mm (DT) for deep tillage  
143 similar to (Ren et al., 2019), treatment 2 = shallow disc cultivation at 100 mm (ST) and treatment 3 = zero tillage  
144 using a direct seed drill (ZT). In combination with the different tillage depths, three traffic regimes were used in  
145 this study no traffic (NT), conventional tyre pressure (CP) and low tyre pressure (LP). Tillage depths were  
146 combined with traffic management practices for the 9 treatments (DTNT, DTCP, DTLP, STNT, STCP, STLP,  
147 ZTNT, ZTCP & ZTLP). Using GPS guidance and markers, trafficked areas of each plot were marked out to ensure  
148 samples were taken from the correct location. A GPS (Trimble FMX display unit) was used to apply all tillage  
149 and drilling applications. All wheelings from cultivation and drilling occurred in the same traffic lanes for the  
150 duration of the trial. During drilling, the drill coulters directly behind the tractor wheeling were marked to aid  
151 identification of trafficked crop rows. During harvest, it was necessary to avoid driving on non-trafficked areas  
152 with the plot combine restricted the wheelway zones. This ensured a CTF system was replicated.

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#### 157 2.4.3.2.23 Tillage equipment and tyres

158 Primary cultivations in HAU involved a rigid tine and conical disc cultivator (Vaderstad Topdown) at 250 mm  
159 depth to cut surface residues, loosen, mix, and consolidate the seedbed. The same implement was used for shallow  
160 tillage treatments with tines adjusted upwards to reduce tillage depth (100 mm). A 290 hp Massey Ferguson 8480  
161 with a track width of 2.1 m was used. Increased flexion AxioBib tyres were fitted IF 650/85 R38 179D TL on the  
162 rear axle and (IF 600/70 R30 159D TL) at the front. A pneumatic disc seed drill (Vaderstad Spirit) was used to  
163 sow the crop with 167 mm row spacing. The same drill was used to sow the zero tillage plots with the tines and  
164 discs lifted to minimise disturbance (Kaczorowska-Dolowy et al., 2019a).

165 For the tyre pressure treatment, the conventional tyre treatments were inflated to 1 bar for front and rear  
166 tyres during cultivations. Low tyre pressure treatments and controlled traffic farming (CTF) plots operated on 0.7  
167 bar front and 0.8 bar on the rear axle. A front weight block of 540 kg was applied to the tractor for tillage primary  
168 cultivation. All operations were performed under the same wheel-ways to keep traffic free zones for CTF plots.  
169 During harvest, a Claas Dominator combine operated on a 4-m header, matching plot sizes (Smith, 2016). Crop  
170 husbandry was carried out in accordance to the AHDB guidelines and soil fertility test analysis (AHDB, 2018).

#### 171 2.4.3.3.14 Soil physical properties

172 Soil bulk density samples were also collected within the trafficked and non-trafficked area of the plot, to represent  
173 the bulk density of the tillage treatments. Samples were replicated three times. Each core sample was 50 mm in

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174 width and 300mm in length. An Eijkelkamp® soil corer was used to take bulk densities samples. Each bulk density  
175 sample was taken within 0.5 m of the location of the soil cores taken for X-ray CT. The objective was to represent  
176 the physical constraints (or lack of) for root growth in each plot examined. The method used in this study involved  
177 splitting the bulk density sample into three 100 mm sections (0-100 mm, 100-200 mm and 200 – 300 mm) similar  
178 to (Smith, 2016). The corer was opened in the field and split using a knife and ruler.

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179 The core sections were stored in resealable bags and labelled before transporting to the laboratory for  
180 analysis. Intact fresh soil cores were weighed prior to drying to record sample fresh weights. Samples were placed  
181 into an oven at 105°C for 24 h and reweighed to determine moisture % as per equation 1 and dry bulk density as  
182 per equation 2 (Campbell and Henshall, 2000).

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183 Moisture % = fresh weight(g) – dry weight (g) / dry weight(g) \*100 Equation 1

184 Dry bulk density (Mg m<sup>-3</sup>) = dry soil weight (Mg)/ soil volume (m<sup>-3</sup>)

185 Equation 2.

#### 186 24.3.3.2.5 Penetration resistance (PR)

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187 Soil penetration resistance data were collected on each plot (in the wheel-ways and in the centre of the plot) down  
188 to 450 mm with a depth increment of 25 mm between each recorded penetrometer reading. A cone penetrometer  
189 (Data Field, Ukraine) was used, recording soil strength in kPa, the location and depth via built-in GPS device.  
190 Only the PR samples were recorded at 450 mm to complete a reading on the data logger. It is also widely known  
191 that roots penetrate past “tillage pans” (Bengough et al., 2011) (Bengough et al., 2011) . Five penetrations were  
192 made both under and between the wheel ways on each plot at GS 25 sampling to represent each treatment. PR  
193 was measured when soil conditions were at field capacity to ensure accuracy of each reading.

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#### 195 24.3.3.3.6 Soil porosity analysis

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196 Before soil porosity analysis on ImageJ software (version 1.52) (Schneider et al., 2012) could commence, an  
197 image stack was created in VG Studio Max® for each scan. The contrast was adjusted to improve the uniformity  
198 and visibility of the soil pores. The register object tool corrected scan discrepancies for soil core angle.  
199 Straightening the scan allowed a cylindrical shape to be cropped and the tube edges and air space outside of the  
200 soil core removed. This enabled soil data to be captured throughout the soil core. A new volume was selected and  
201 extracted from the original. This created a separate cropped image volume to work from. The surface  
202 determination tool in VG Studio Max® was used to threshold pore spaces within the solid matrix. The tool defines  
203 the contour of objects, separating 3D data into regions, providing meaningful soil data (Borges de Oliveira et al.,  
204 2016). The image was then inverted to remove the extracted variables from the image and highlighting the pore  
205 spaces in the soil core. The processed image was exported as an \*.TIFF image stack for further analysis using  
206 ImageJ software.

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207 Soil pore characteristics were measured using X-ray CT to establish information about the 3D soil  
208 environment for root growth without disrupting the structural integrity of the soil core. The original grey-scale X-  
209 ray CT images were analysed using ImageJ software. The scale was set for each dataset to define to spatial scale

210 of the active image. The unit of length was set in millimeters and the known distance was 0.045mm (45µm). Each  
211 scanned core was cropped to remove the area outside of the soil column. The action of soil coring during sampling  
212 had the effect of loosening the bottom 20 mm of the core, therefore 415 slices at the bottom of each scan were  
213 discarded to remove the loosening effect due to the sampling process. The downward movement of the PVC pipe  
214 also caused a smearing effect on the soil at the outside edge of the core and this area was also removed by cropping.

215 The processed image was 1220 x 1220 pixels in size. Applying the contrast enhancement filter helped  
216 normalize all slices. The filter reduces the differences in pixel grey-level between slices known as beam hardening  
217 (Wildenschild et al., 2002). The ImageJ Huang automatic threshold algorithms were used for each scan to create  
218 binarized images and separate the air-filled pores from the background region. The binarized scans were de-  
219 speckled twice to remove unwanted noise within each scanned image, improving analysis and accuracy of the  
220 investigated pores. The Look Up Table (LUT) was inverted to change the white pores to black, ensuring analysis  
221 calculated the air-filled pores and not the soil matrix. The resulting binary images were analysed using the Analyze  
222 Particles tool which provided information for average pore size, total area and percentage porosity for each  
223 individual image.

#### 224 2.4.14.3.6 Soil core sampling

225 Field soil core size was chosen to capture as much root material growing in the field as possible while minimizing  
226 the trade-off that exists with the X-ray CT technology between image resolution and core size (Mooney et al.,  
227 2012; Zhou et al., 2021). The core dimensions were consistently 70 x 300 mm (diameter x depth) for each sample.  
228 Soil cores were extracted from the field sites at GS 25 in February and again at GS 61 in June. Sampling was  
229 carried out at GS 61 during wheat anthesis, when root growth is at its peak (Gregory et al., 1978). Due to high  
230 moisture deficits in HAU (43 mm) during sampling at GS 61 in early July, the soil sample area was wetted with  
231 2.5 L of water and allowed to infiltrate. This lubricated the soil, reduced soil fracturing, and allowed tube insertion  
232 and soil core extraction to take place as smoothly as possible. Polyvinyl chloride (PVC) drainage pipes were cut  
233 to size (70 x 300 mm) and these tubes were used to collect soil cores (as per Millington et al, 2017).

234 A single wheat plant sample was located at random in each plot. The selected plant was cut at the base  
235 of the stem with a scissors and the above ground biomass discarded The PVC tube was placed (plant centred)  
236 directly over the remaining plant stubble to maximise root system capture. Tubes were inserted into the soil using  
237 a mallet in the crop rows in the centre of the plots between the wheel tracks (not trafficked by wheel) for  
238 untrafficked samples for no traffic samples. A second core was taken in the wheel way for the tyre pressure  
239 treatments. A small block of timber was used when hammering in the tube to protect tubes and soil cores from  
240 damage. A total of 72 samples were extracted on each sampling occasion and examined in this study. The PVC  
241 tubes were inserted into the soil to a depth of 300 mm. The soil core was extracted carefully using a spade and the  
242 sample locations were backfilled with soil. Following sampling, cores were sealed (top and bottom) using tape,  
243 labelled, and carefully placed into boxes protected with bubble wrap. Cores were tightly packed and insulated to  
244 minimise movement and drying of samples during transit to the laboratory for analysis. Samples were transferred  
245 to refrigerated storage (<4°C) to prevent and reduce compositional changes to the soil through biological  
246 degradation.

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247 2.5.14.3.7 X-ray computed tomography (CT) – Root analysis

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248 Soil cores were transferred to the University College Dublin (UCD) X-ray CT facility at the Rosemount  
249 Experimental Research Station at Belfield Campus, UCD, Ireland. The soil cores were scanned using a Phoenix®  
250 Vertex M 240 kV scanner (GE Measurement and Control solution, Wunstorf, Germany). The Vertex M  
251 was set at a voltage of 90 kV and current of 400 µA to optimize contrast between background soil and root material.  
252 A voxel resolution of 45 µm was achieved by using the ‘Multi Scan option’ to scan in 4 segments. A total of 1800  
253 projection images per section were taken at 200 m/s per image using the ‘Fast Scan option’, which has the default  
254 values of an image averaging of 1 and 0 skip. No filters were used during scanning. The total scan time per core  
255 was 24 minutes or 6 minutes per section. Once scanning was complete, the images were reconstructed using  
256 Phoenix datosx2 rec reconstruction software, the four scans were assembled into one 3D volume for the whole  
257 core. Core samples were scanned within a week of the sampling date, the scanned core was 300 mm in length and  
258 70 mm diameter. The software corrected movements during the scanning process and removed noise from scanned  
259 images.

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260 2.5.2 X-ray CT root segmentation

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261 Image analysis for X-ray CT images was performed using the software VGStudioMax®, version 3.2 (Volume  
262 Graphics GmbH, Heidelberg, Germany) to segment roots and soil porosity. Roots were segmented by setting seed  
263 points and using selected threshold values in the “Region grower” that enabled fast and accurate selection of grey-  
264 scale voxels (3D pixels) pertaining to root materials. The root system was extracted from the greyscale CT image  
265 of soil using the VGStudioMax® semi-automated local adaptive thresholding “Region Growing” selection tool,  
266 similar to (Tracy et al., 2013). Root volumes were calculated by segmenting the root region of interest (ROI).  
267 Once the roots were segmented from the image, erosion and dilation tool was selected at 1 pixel using the Region  
268 Growing tool. Root system architecture parameters such as root vertical depth, root volume and root surface area  
269 were measured from the segmented root systems. Root vertical depth was calculated on the Z axis in  
270 VGStudioMax® from the length of a complete root from the base seed point.

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272 2.5.34.3.9 Destructive 2D root analysis

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273 After the soil cores were scanned, the soil and root material were separated by root washing gently with a water  
274 jet hose. Two sets of sieves with a mesh size of 2 mm and 1 mm collected root material. Roots were washed and  
275 soil material removed before the roots were placed into a sealed and labelled bag filled with water. The washed  
276 root samples were placed into a freezer until scanning and analysis with WinRHIZO™ scanning and software  
277 (version 2016a Regent Instruments, Canada) commenced. The root samples were thawed before scanning with  
278 the WinRHIZO™ software. Large root stumps were removed from the sample prior to placing it inside the tray to  
279 reduce root misrepresentation (Wang and Zhang, 2009). Roots were placed onto a clear transparent tray (30 cm x  
280 20 cm) with water. A pair of plastic forceps were used to spread out root seminal and lateral roots. Images were  
281 scanned at a resolution of 600 dpi (42 µm pixel size) with an Epson Perfection V800 scanning system. Root  
282 images were measured for root length, root surface area, average root diameter and root volume for the total soil  
283 core. This output was used to verify the 3D root outputs from VGStudioMax® (Flavel et al., 2017; Tracy et al.,

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284 2012a). The WinRHIZO™ software enabled rapid assessment of root parameters. It calculated the root volume by  
285 determining the average root diameter and root length by pixel counting the 2D root image and then assuming the  
286 root shape was cylindrical. The WinRHIZO™ used a skeletonization method for characterizing root systems  
287 (Himmelbauer, Loiskandl and Kastanek, 2004). The software uses greyscale values in \*.TIFF file format. The  
288 output of the images was distinguished by global thresholding analyses for root diameter while root length was  
289 validated by skeleton images. After WinRHIZO™ scanning, the roots were removed from the scanning tray using  
290 forceps. The root samples were dried at 70°C for 24 hours and the root biomass samples were weighed.

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291 2.6.4.3.10 Soil Moisture Deficit Model

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292 Soil Moisture Deficit (SMD) was calculated based on the SMD hybrid model for Irish grassland (Schulte et al.,  
293 2005). The model is based on weather parameters and soil drainage classes. The inputs of the model include data  
294 on maximum and minimum temperatures, rainfall (mm), wind speed (m/s), sunshine hours, ~~maximum and~~  
295 ~~minimum temperature which~~ data were taken from the nearest weather station located in Newport, Shropshire 6km  
296 from the site (Met office, 2019).

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298 4.3.1.2.7 Statistics

299 Data from the scanned (destructive and non-destructive) images and root biomass were not normally distributed.  
300 Non-normal data do not meet the assumptions underpinning ANOVA (Analysis of Variance); therefore, all data  
301 underwent log transformation (in Microsoft Excel) before being exported to Minitab 18® where analysis of  
302 variance (ANOVA) was performed to homogenize the variances of the compared means (Poorter and Garnier,  
303 1996). A two-way ANOVA was performed using the general linear model using the minitab software package.  
304 All means were analysed for normality before the test was run. When significant effects of rooting were detected,  
305 regression analysis was utilised to observe the relationship between the variables. For linear regression analysis,  
306 residuals of data were made to ensure that the assumptions of the analysis were met (normal distribution, constant  
307 variance, etc). Normality was tested using the Anderson-Darling test in Minitab 18®.

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309 3.4.4 Results

310 4.4.1.3.1 Growing conditions during crop season

311 In 2018, crops were established at low soil moisture levels, which may have reduced soil compaction caused by  
312 tillage operations across all site locations. From January to August (2019), 418.6 mm of rainfall was recorded at  
313 HAU, 68 mm in total for January and February. Soil moisture deficits reached 66.2 mm in HAU (Supplementary  
314 Figure 4.10) by early June 2019. High soil moisture deficits were recorded from early April to June, causing  
315 drought stress during rapid growth periods (Met office, 2019).

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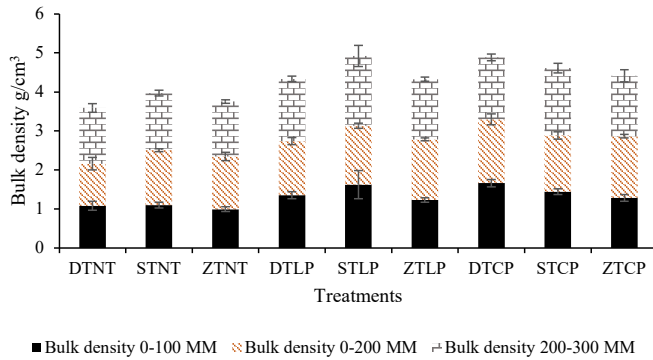
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316 [4.4.3.2.12](#) Soil properties – Bulk density & Penetrometer resistance  
 317 The calculated probability (*P*-value) and standard error of the mean (SEM) from one-way ANOVA analysis is  
 318 given in Figure 4.1, while using two way linear model analysis (ANOVA) results are shown in Table 24.4 in the  
 319 supplementary section of this paper. Bulk density presented for 0-100 mm, 100-200 mm, and 200-300 mm  
 320 measurements in Figure 4.1. In the top 0-100 mm, bulk density was significantly higher in DTCP (1.66 Mg m<sup>-3</sup>)  
 321 and STCP (1.44 Mg m<sup>-3</sup>) treatments compared to ZTNT (0.994 Mg m<sup>-3</sup>) and DTNT (0.97 Mg m<sup>-3</sup>) (*P*<0.01). STNT  
 322 (1.09 Mg m<sup>-3</sup>) was significantly higher than ZTNT and DTNT and only significantly lower than DTCP. In the  
 323 middle horizon (100-200 mm), a significant interaction between trafficking treatment was found. Bulk density  
 324 was significantly lower in DTNT (1.07 Mg m<sup>-3</sup>) compared to DTCP (1.63 Mg m<sup>-3</sup>) and ZTCP (1.58 Mg m<sup>-3</sup>)  
 325 treatments (*P*<0.05). In the bottom 200-300 mm layer measured, no significant tillage x traffic interaction was  
 326 found (*P*>0.05). Table 24.4 shows a significant traffic effect on soil bulk density in the 0-100mm layer and 100-  
 327 200mm layer (*P* < 0.01). No traffic in both layers revealed a lower bulk density compared to trafficked treatments.



328 **Table 24.4:** Variance of analysis for bulk density x traffic, tillage and tillage x traffic.

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HAU	Deep	Shallow	Zero	Mean
<b>Traffic/ Tillage 0 – 100 mm</b>				
No traffic	0.971	1.099	0.994	1.058 <sub>b</sub>
LP	1.351	1.625	1.230	1.401 <sub>a</sub>
CP	1.661	1.444	1.282	1.462 <sub>a</sub>
P < 0.01				
<b>Traffic/Tillage 100 – 200 mm</b>				
No traffic	1.079	1.406	1.353	1.279 <sub>b</sub>
LP	1.389	1.509	1.552	1.483 <sub>a</sub>
CP	1.637	1.437	1.583	1.553 <sub>a</sub>
P < 0.01				
<b>Traffic/Tillage 200 – 300 mm</b>				
No traffic	1.429	1.466	1.404	1.433
LP	1.593	1.787	1.619	1.666
CP	1.537	1.548	1.548	1.544

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335 Penetration resistance (PR) was recorded in February 2019 when the soil was at field capacity.  
336 Measurements were grouped into three groups, 0-150 mm, 150-300 mm, and 300-450 mm depth layers [and a](#)  
337 [linear variance of analysis between tillage, traffic and tillage x traffic interaction conducted and shown in Table](#)  
338 [3](#). [Figure 4.2 depicts the combined three layers grouped into one 0-450 mm graph. The one-way ANOVA](#)  
339 [analysis revealed highly significant differences for each layer. In the 0-150 mm layer, DTNT recorded the lowest](#)  
340 [kPa \(kilopascals\) readings and was significantly lower than ZTCP, STCP, STLP, ZTLP and ZTNT \(P< 0.000\).](#)  
341 [DTCP and DTLP were significantly lower kPa than ZTLP, STLP, STCP and ZTCP. ZTCP recorded the highest](#)  
342 [kPa reading and was significantly higher than ZTLP, ZTNT, STNT, DTLP, DTCP and DTNT. In the second layer](#)  
343 [\(150-300 mm\), similar trends were found and highly significant \(P<0.000\). STCP showed the highest kPa \(3193.5](#)  
344 [kPa\) and was significantly higher than STNT, ZTNT, DTNT, DTLP and DTCP. In contrast, DTNT recorded the](#)  
345 [lowest reading \(1268.4 kPa\) and was significantly lower than ZTNT, STNT, ZTLP, ZTCP, STCP and STLP.](#)  
346 [STNT revealed significantly lower kPa than STLP, ZTCP and STCP. ZTNT penetrometer readings were](#)  
347 [significantly lower than all trafficked ZT and ST treatments. In the lower depth \(300-450 mm\), DTNT was](#)  
348 [significantly lower than STLP, STCP, ZTCP, ZTLP and STNT \(P<0.000\). ~~An analysis of variance table is~~](#)  
349 [presented in section 4.7 of this thesis for penetrometer resistance \(Table 4.5\).The results revealed a significant](#)  
350 [traffic interaction for 0 – 150mm \(P < 0.001\), 150 – 300mm \(P < 0.000\) and 300 – 450 mm \(P < 0.000\). Again,](#)  
351 [no traffic PR was significantly lower than trafficked treatments. When tillage was measured, a significant effect](#)  
352 [was observed for each layer studied \(0 – 150mm = P < 0.000, 150 – 300mm = P < 0.000, 300 – 450mm = P <](#)  
353 [0.000\). In the top layer, deep tillage was significantly lower than shallow and zero. Further, shallow tillage was](#)  
354 [significantly lower than zero tillage PR. In the second and third layer, deep tillage was lower than shallow and](#)  
355 [zero. A tillage v traffic interaction was observed in the first layer \(0 – 150mm\) \(P < 0.001\) but the second and](#)  
356 [third layer were not significant \(P < 0.067, P < 0.313 respectively\).](#)

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**Table 3:** Analysis of variance table for penetration resistance x tillage, traffic and tillage x traffic interactions between 0 – 150 mm, 150 – 300 mm and 300 – 450 mm

	0 - 150 mm 2019	150 - 300 mm 2019	300 - 450 mm 2019
<b>Tillage</b>			
No traffic	432.3 <sub>a</sub>	164.5 <sub>a</sub>	3729.5 <sub>a</sub>
CT	519.8 <sub>a</sub>	261.4 <sub>a</sub>	3754.8 <sub>a</sub>
LP	616.7 <sub>a</sub>	212.2 <sub>a</sub>	3655.7 <sub>a</sub>
P value	<0.001	<0.001	<0.001
<b>Traffic</b>			
None	298 <sub>a</sub>	1676.2 <sub>b</sub>	2416.7 <sub>b</sub>
Light	322.4 <sub>a</sub>	2547.6 <sub>b</sub>	3296 <sub>b</sub>
Heavy	418.8 <sub>a</sub>	2774.7 <sub>b</sub>	3674.2 <sub>b</sub>
P value	<0.001	<0.001	<0.001
<b>Tillage x traffic</b>			
None	349 <sub>a</sub>	2467	352

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\*Significant difference between means is represented by different letters

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**Commented [DH2]:** Results of the Penetrometer resistance provided in table format with clearer description of the results.

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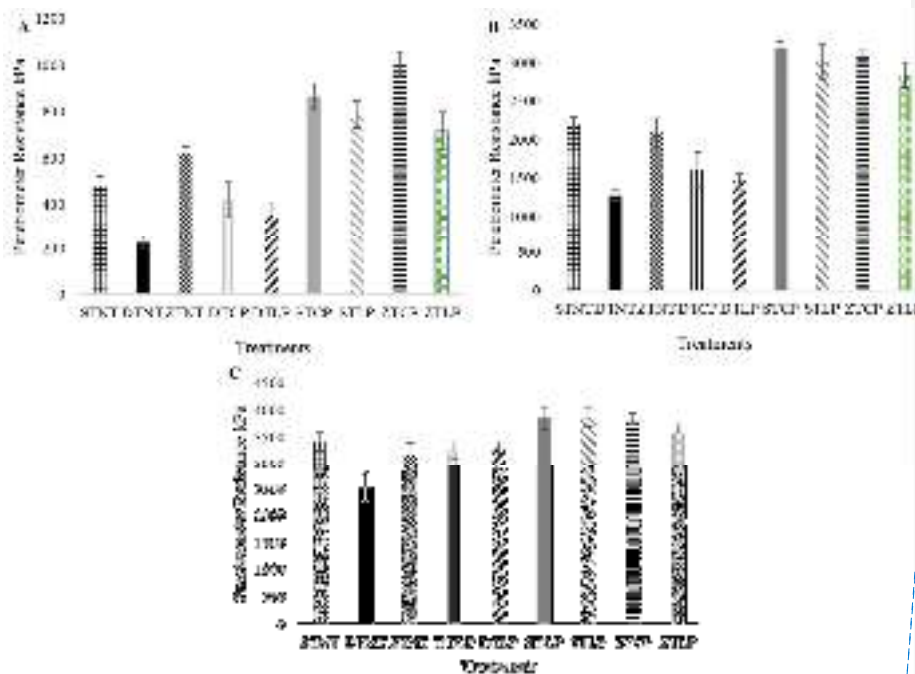
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368 **Figure 4.12:** Penetration resistance for three layers (a) 0-150 mm ( $P < 0.000$ ), (b) 150-300 mm ( $P < 0.000$ ) and (c)  
 369 300-450 mm ( $P < 0.000$ ) during wheat tillering (GS25). Soil moisture conditions were at field capacity during  
 370 sampling. Bars represent the standard error of the mean.

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372 3.2.24.4.3 Soil porosity

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373 The results of the ANOVA analysis of the CT-measured porosity (0-220 mm) are presented in Table 42. Soil  
 374 porosity results were split into two soil layers of 0-100 mm and 100-200 mm respectively. In the top 0-100 mm  
 375 layer, DTNT showed significantly higher total pore space ( $P < 0.01$ ) compared to all other treatments except ZTNT.  
 376 Tillage had a significant effect on soil porosity in the no traffic samples in the 0-100 mm layer ( $P < 0.05$ ). Deep  
 377 tillage with no traffic had higher soil porosity (22.72%) than in shallow tillage (no traffic) (10.58%). There was  
 378 no significant difference between soil porosity under zero tillage and shallow tillage in the no traffic samples.  
 379 Trafficking had a significant effect on overall porosity. In deep tillage treatments, overall porosity 22.72% (no  
 380 traffic) was reduced to 8.08% (under low tyre pressure) and 6.50% under conventional tyre pressure. Traffic had  
 381 little effect on shallow and zero tillage porosity in the top 0-100 mm when compared to the no traffic samples  
 382 with small reductions in porosity. In the second examined layer, 100-200 mm zone, tillage and traffic were not  
 383 significantly different ( $P < 0.487$ ). The percentage porosity shown in Table 42, indicate a sharp decline in the lower  
 384 depth with only 9.02% in DTNT. DTCP treatments recorded the lowest porosity (3.96%).

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388 **Table 4.2:** Soil porosity for tillage x traffic for two soil layers.

ImageJ soil porosity % 0-100mm	n	Conventional tyre pressure		
		No traffic	low tyre pressure	pressure
Deep	4	22.72 <b>a</b>	8.08 <b>b</b>	6.50 <b>b</b>
Shallow	4	10.58 <b>b</b>	8.64 <b>b</b>	7.23 <b>b</b>
Zero	4	10.77 <b>ab</b>	8.41 <b>b</b>	8.49 <b>b</b>
P<0.01				
ImageJ Soil porosity % 100-200mm	n			
Deep	4	9.02	6.16	3.96
Shallow	4	4.06	6.44	5.32
Zero	4	2.895	6.44	5.32
P<0.487				

389 \*Significant differences between means are represented by different letters.

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391 3.3.14.4.4 Destructive 2D root analysis

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392 The interaction between tillage system and trafficking protocols using destructive root measuring methods (WinRHIZO™) are shown in Figure S24.3 for GS 25 and Figure S3.4.4 for GS 61 in the supplementary section.

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393 The variance of analysis results are for WinRHIZO™ are presented shown in Table 4.6.5 of the supplementary section. At GS25, no significant differences were found between tillage, traffic, and traffic x tillage

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394 interactions. However, the WinRHIZO™ analysis revealed a tendency towards increased root growth in no traffic treatments. At the later growth stage (GS61), Figure 4.4 Table 5 depicts the results showing highly significant

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395 interactions between trafficking and tillage systems on root length density (RLD) (P<0.001) and root length (P<0.001), root surface area (P<0.002) and a traffic effect on root volume (P<0.05). Variance of analysis results

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396 showed a significant effect of tillage system (P < 0.01) for deep tillage compared to shallow and zero tillage. When trafficking was considered, an even greater significance under was recorded (P < 0.000) for root length

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397 with no traffic significantly greater than conventional (CP) and low pressure (LP) treatments. For root surface area (mm<sup>2</sup>), a significant tillage (P < 0.05) and traffic (P < 0.000) was found. Deep tillage showed significance

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398 over shallow tillage but not zero. Root surface area was significantly lower in CP than no traffic but not LP. Root volume (mm<sup>3</sup>) showed a significant traffic effect, but tillage was not significant. Indeed, no traffic was

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399 significantly greater than CP but not LP. For RLD mm<sup>3</sup>, a significant tillage (P < 0.01) and trafficking effect (P < 0.000) was found. Deep tillage established greater RLD compared to zero and shallow tillage while no traffic

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400 was significantly greater than CP and LP.

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401 When traffic x tillage interactions were compared, no significant difference was found (Table 5.4.6). However, individually, treatments were significantly greater than others. For example, DTNT showed significantly higher

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402 RLD, root surface area and root length compared to ZTCP, STCP and STLP. Root volume was significantly higher

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412 in DTNT over ZTCP and STCP. DTNT produced nearly double the root length compared to ZRCP. In contrast to  
 413 DTCP, root surface area reduced by 36% compared to untrafficked areas (no traffic samples). In shallow and zero  
 414 tillage, root surface area was reduced by 32% and 63.6% respectively in conventional pressure samples compared  
 415 to untrafficked samples. There was no significant difference for root diameter and between all tillage and  
 416 trafficking regimes. The results demonstrate that there was no significant difference in RLD at the tillering stage,  
 417 nor could trends be found as roots were undeveloped. However, at anthesis, the RLD was significantly higher  
 418 under non-trafficked tillage treatments when compared to DTCP, STCP and ZTCP (Figure S4-3b).

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421 **Table 5: WinRhizo results of Tillage, traffic and Tillage x Traffic interactions with root traits at HAU during**  
 422 **tillering (GS25) and anthesis (GS61). P values represent level of significance and 'NS' indicates non-significance.**  
 423 **d.f. = degrees of freedom]**

Root trait	Term	d.f.	GS25	GS61
Root length mm	Tillage	2	NS	< 0.01
	Traffic	2	NS	< 0.000
	Tillage x Traffic	35	NS	NS
Surface area mm <sup>2</sup>	Tillage	2	NS	< 0.05
	Traffic	2	NS	< 0.000
	Tillage x Traffic	35	NS	NS
Root diameter mm	Tillage	2	NS	NS
	Traffic	2	NS	NS
	Tillage x Traffic	35	NS	NS
Root volume mm <sup>3</sup>	Tillage	2	NS	NS
	Traffic	2	NS	< 0.05
	Tillage x Traffic	35	NS	NS
RLD mm <sup>3</sup>	Tillage	2	NS	< 0.01
	Traffic	2	NS	< 0.000
	Tillage x Traffic	35	NS	NS

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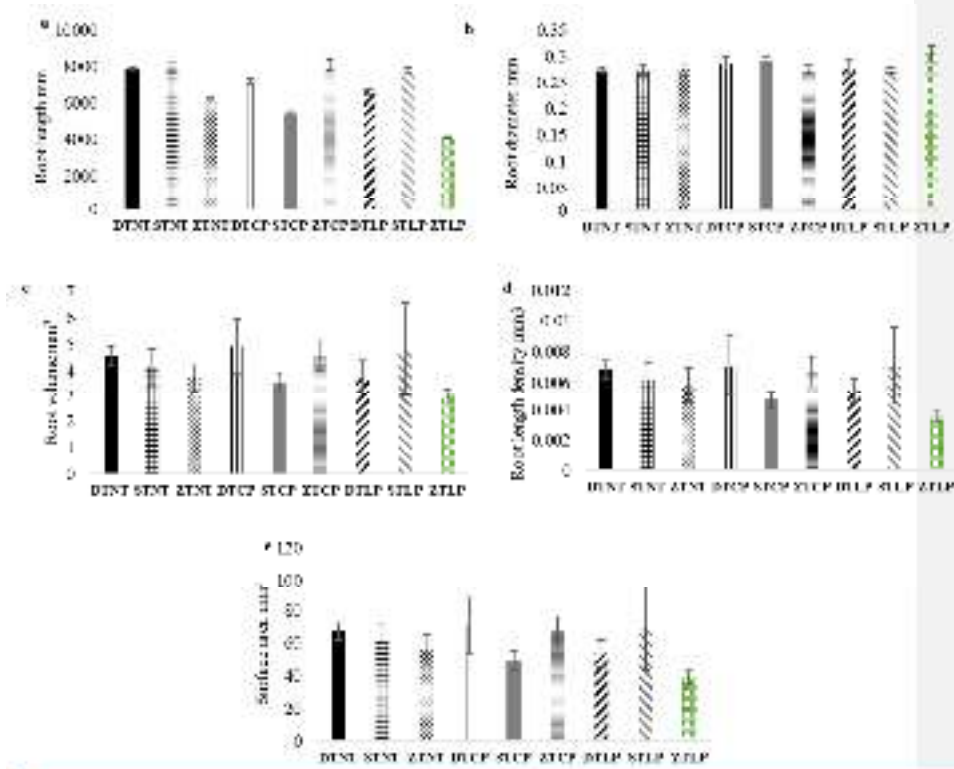
**Commented [DH3]:** Variance of analysis added for WinRHIZO results – replacing figure 3 & 4. Figure 3 & 4 added to the supplementary data

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428 **Figure 4.3:** Tillering (GS25) root system architecture using destructive root method. (a) Root length (mm), (b) ←  
 429 Root diameter (mm) (c) Root volume (mm<sup>3</sup>), (d) Root length density (mm<sup>3</sup>), (e) Root surface area (mm<sup>2</sup>).

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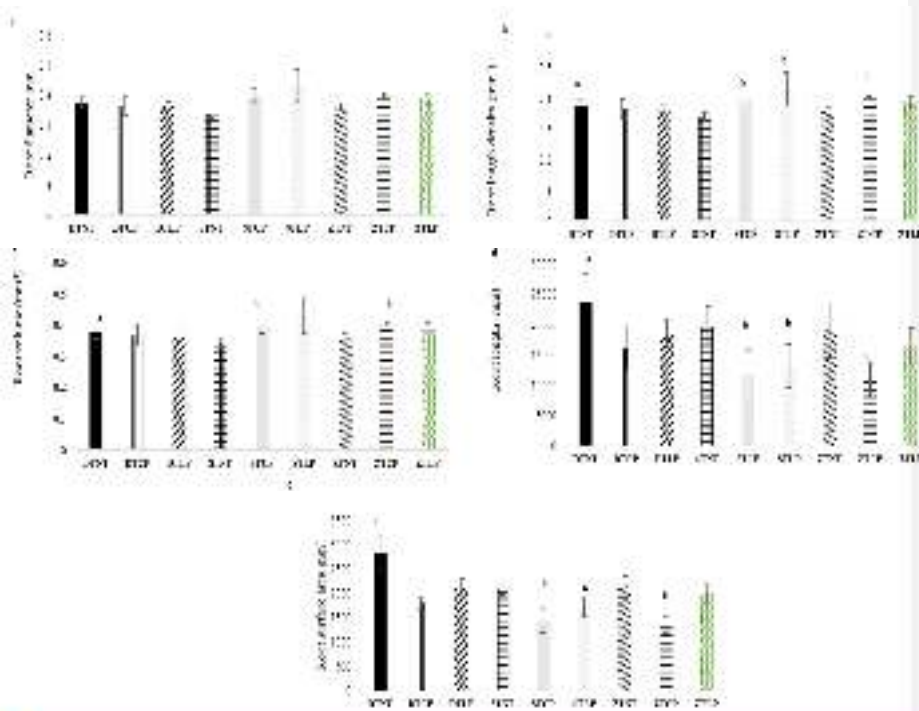
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439 **Figure 4.4:** Flowering growth stage 61 root system architecture using destructive root method. (a) Root diameter,  
 440 (b) Root length density (mm<sup>3</sup>), (c) Root volume (mm<sup>3</sup>), (d) root length (mm), (e) Root surface area (mm<sup>2</sup>). **Table**  
 441 **4.6:** WinRhizo results of Tillage, traffic and Tillage x Traffic interactions with root traits at HAU during tillering  
 442 (GS25) and anthesis (GS61). P values represent level of significance and 'NS' indicates non-significance. d.f. =  
 443 degrees of freedom.

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446 **3.3.24.4.5** X-ray CT root analysis results **Formatted:** Font: Not Italic

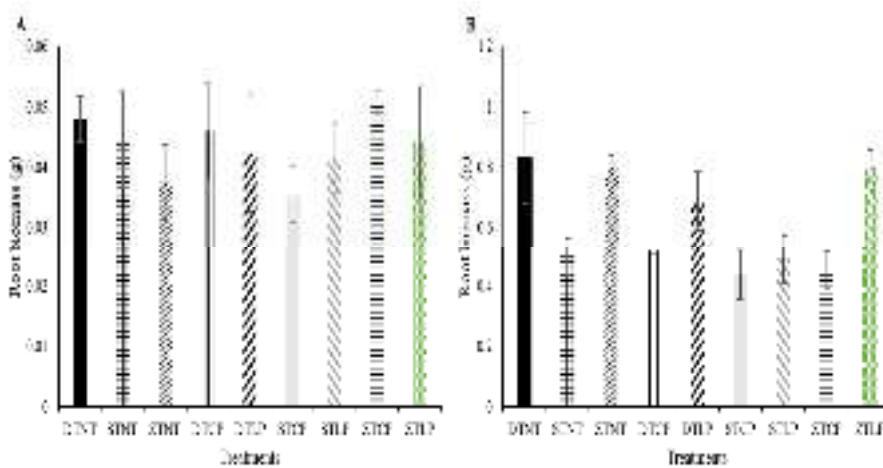
447 Significant differences were found between trafficking treatments at GS61 for RLD and vertical root depth using **Formatted:** Font: 10 pt

448 non-destructive VGStudioMax 3.2 (Table 4.3). The X-ray CT scans revealed significantly longer vertical rooting  
 449 (measured via the Z axis in VGStudioMax®) in ZTNT (112.7 mm) compared to DTCP (60.44 mm), DTLP (66.96  
 450 mm), STLP (65.39 mm) treatments (P<0.001). ZTNT showed significantly greater RLD (0.000098 mm/m<sup>3</sup>) over  
 451 DTCP (0.000052 mm/m<sup>3</sup>), DTLP (0.000058 mm/m<sup>3</sup>), STLP (0.000058 mm/m<sup>3</sup>) and ZTCP (0.000060 mm/m<sup>3</sup>)  
 452 treatments (P<0.001). Root volume and surface area showed no significant difference using X-ray CT. However,  
 453 similar trends were found to the conventional WinRHIZO™ method. Trafficking had more of an influence on  
 454 rooting than tillage method which did not have any significant effect on root parameters. As RLD is an important  
 455 root trait commonly measured to estimate water uptake (White et al., 2015), linear regression was used to verify  
 456 the relationship between root depth and RLD. A significant relationship (P < 0.001) was found with a coefficient  
 457 of determination R<sup>2</sup> = 0.54 (Supplementary Fig. S42).

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472 **Figure 34.5:** Root biomass at tillering (GS25) and flowering (GS61) for traffic and tillage treatments. Treatments  
 473 represented by initials (Tillage: D = Deep, S = Shallow, Z = Zero), (Traffic: NT = No traffic, LP = Low pressure  
 474 tyre, CP = Conventional pressure tyre).

475

#### 476 3.4.4.4.6 Crop yield

477 Crop yield was highly significant between trafficking treatments and tillage ( $P < 0.01$ ) shown in Figure. 46. ZTLP  
 478 had the highest yield (11,385 kg ha<sup>-1</sup>) and was significantly greater than DTLP (10,757 kg ha<sup>-1</sup>), STCP (10,700  
 479 kg ha<sup>-1</sup>), STNT (10,678 kg ha<sup>-1</sup>), STLP (10,638 kg ha<sup>-1</sup>) and DTCP (10,613 kg ha<sup>-1</sup>). All three zero tillage  
 480 treatments trended higher than deep tillage and shallow tillage treatments. ZTLP showed a 500 kg ha<sup>-1</sup> yield  
 481 advantage over DTNT (NS) and between 628 - 772 kg ha<sup>-1</sup> over trafficked treatments and STNT with high  
 482 significance. In general, this study did not show a trend in yield between conventional and low tyre pressure  
 483 treatments. For deep tillage, conventional tyre pressure reduced crop yield compared to low tyre pressure by 144  
 484 kg ha<sup>-1</sup>) (1.34%). When compared to the no traffic sample, conventional tyre pressure consistently reduced yield  
 485 by 272 kg ha<sup>-1</sup>) (2.5%) in deep tillage. Although not significant, trafficking trended towards improving yield by  
 486 30 kg ha<sup>-1</sup>) (0.03%) using conventional tyre pressure and 340 kg ha<sup>-1</sup>) (3.07%) using low tyre pressure. No trends  
 487 were found in shallow tillage treatments. Linear regression of root depth using X-ray CT showed a significant  
 488 relationship to crop yield ( $P < 0.001$ ) and positive correlation ( $r = 0.54$ ). However, the coefficient of determination  
 489 was low  $R^2 = 0.3094$  (Figure S54+12). Moreover, regression analysis also showed a significant relationship  
 490 between root biomass and crop yield ( $P < 0.01$ ). However, the correlation between the two variables was weaker  
 491 ( $r = 0.43$ ) (coefficient of variance  $R^2 = 0.1859$ . This indicates that root depth is a stronger predictor of crop yield.

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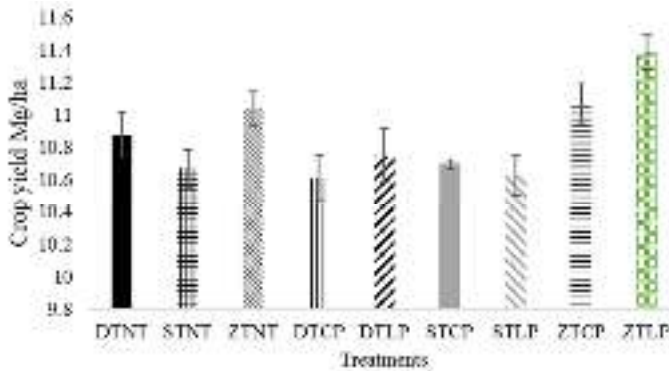


Figure 4.6: Crop yield in Mg/ha for traffic x tillage treatments.

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#### 4.5. Discussion

##### 4.5.1 Soil physical responses to tillage & trafficking

In line with this paper's hypothesis, trafficking effects were more influential on crop and root performance than tillage system. The presence of wheeled areas in both zero and deep cultivation treatments increased soil bulk density significantly in deep tillage treatments (Figure 4.1). Previous studies have shown that zero tillage systems increase in bulk density, penetration resistance and reduce in porosity in the early years of adoption from conventional tillage systems (Christian and Ball, 1994; Six et al., 2004; Mangalassery et al., 2014b; Smith, 2016). Vogeler et al., (2009) showed that bulk density is higher under conservation tillage methods in the top 100 mm layer during the first five years of adoption from conventional systems. Indeed, Soane et al., (2012) reported that significant regeneration of soil structure requires a three-year period from tillage depending on previous historic land management practice. Moreover, values decrease in the long term with multiple benefits including improved saturated conductivity, soil organic matter and air permeability in lower soil horizons. Arvidsson, 1998 showed that soils with <math>30 \text{ g kg}^{-1}</math> of organic matter were likely to suffer 11% higher crop yield loss due to compaction using uniaxial compression tests. It is plausible that the actions of soil fauna such as earthworms and old root channels could have reduced bulk density over time (Figure 54.7) as identified by (Angers and Caron, 1998). Roots promote soil structural formation through increasing soil aggregation. Root mucilage production, root hair formation, and localised wetting and drying cycles encourage a reduction in soil bulk density (Bengough, 2012).

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##### 4.1.2 Soil porosity in response to tillage

Sandy soils due to their adhesive and coarse grain nature, have reduced porosity, including lower levels of micropores compared to loamy soils (Arvidsson, 1998). The aggregation potential in this sandy loam soil is low. In the presence of plants, porosity and pore connectivity as shown to reduce further compared to clay cohesive

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519 soils which tend to increase in porosity through flocculation and aggregation (Bacq-Labreuil et al., 2018). Here,  
520 we found soil porosity to be low in general across all treatments. When comparing cultivation systems, we found  
521 that shallow tillage in the 0-100 mm layer had significantly lower porosity (10.58%) compared to deep tillage  
522 (22.72%). Although zero tillage recorded low porosity values also (10.72%), it was not significantly different to  
523 the other two systems.

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524 A key characteristic of zero tilled soils is a change in soil pore architecture with vertically orientated fissures  
525 connected down through the soil profile created by biopores (Figure 54-7). Similar findings have resulted in  
526 reduced CO<sub>2</sub> fluxes and increased saturated hydraulic conductivity by surface-connected porosity (Cooper et al.,  
527 2021). The same study found similar soil porosity levels between conventional and zero tillage with zero tillage  
528 total porosity ranging from <5%, 10% and 12% on average over 1-5, 6-10 and 11-15 years respectively. The  
529 significant increase in deep tillage soil porosity substantially increases soil respiration, resulting in up to 13.8  
530 times higher CO<sub>2</sub> emissions through increased oxidation and carbon breakdown (Reicosky et al., 1999). The lower  
531 porosities in zero and shallow tilled soils reduces space for gas exchange, reducing soil respiration and supporting  
532 carbon sequestration, thus increasing recalcitrant levels of carbon in soil. Mangalassery et al., (2014) found similar  
533 porosity results using X-ray CT methods to measure the effect of tillage method on greenhouse gas emissions,  
534 finding significantly higher porosity in tilled soil (13.6%) compared to zero tilled soil (9.6%) in the top 0-100 mm  
535 layer. However, in deeper soil horizons, no difference could be found between tillage system. The findings in this  
536 experiment agree with that study, showing both tillage methods did not differ significantly in the 100-200 mm  
537 layer with lower soil porosities recorded.

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#### 538 4.1.3 Root system architecture responses to tillage

539 The 'hidden half' (i.e. roots) of plants are difficult to interpret in field studies (Lynch and Brown, 2001). A large  
540 root system is characterized by large biomass, root length and root length density (Ehdaie et al., 2010; Hamblin  
541 and Tennant, 1987). Root biomass was an important indicator of root size, showing treatment effect at anthesis  
542 compared to the tillering stage. In general, root biomass had a positive relationship with grain yield. Zero tillage  
543 treatments both untrafficked and trafficked at low pressure had greater root biomass over all shallow tillage  
544 treatments and deep till trafficked at conventional pressure. Although deep tillage treatments had the highest root  
545 biomass by GS61, it did not achieve the highest yield. No significant difference in root biomass was found between  
546 tillage treatments, confirming that roots are more sensitive to trafficking than tillage method. RLD in shallow  
547 tillage treatments and zero tilled treatments trafficked at conventional pressure. RLD is an important parameter  
548 for characterizing root growth (Doussan et al., 2006), and has been used in previous studies as a key root parameter  
549 for modelling water uptake (Tinker and Nye, 2000; Javaux et al., 2013). Munos-Romero et al., (2010) and  
550 Chakraborty et al., (2008) results indicate that RLD is a positive predictor of crop yield. Although RLD had a  
551 positive correlation with crop yield in this study, root depth (using X-ray) displayed a much stronger relationship  
552 with crop yield (Figure S5).

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555 4.2.1 Soil physical responses to traffic

556 In line with this papers hypothesis, trafficking effects were more influential on crop and root performance than  
557 tillage system. The presence of wheeled areas in both zero and deep cultivation treatments increased soil bulk  
558 density significantly in deep tillage treatments (Table 2 Figure 4.1). Our data shows similar findings with zero and  
559 deep tillage significantly reduced bulk density values in untrafficked zones. However, in trafficked treatments,  
560 high tyre pressure combined with deep tillage treatments resulted in higher bulk density values due to the loss of  
561 inherent strength by tilled soil, resulting in compression of soil particles (Raper, 2005; Soane, Godwin and Spoor,  
562 1986). Chan et al., (2006) observed that trafficking after deep tillage increased bulk density values from 1.27 Mg  
563 m<sup>-3</sup> to 1.54 Mg m<sup>-3</sup>, emphasizing the effect of trafficking on the reduced bearing capacity of the deep tilled soil.  
564 The optimum soil density has been reported to differ between soil types in previous studies. Indeed, Czyż, (2004)  
565 established a soil type interaction between crop yield, bulk density and root mass concluding with sandy loam  
566 soils (similar to this study) having an optimum bulk density value of 1.54-1.66 Mg m<sup>-3</sup>. Yet, in this study, root  
567 biomass was significantly reduced with treatments displaying similar soil density values to that reported optimum.  
568 Although conventional pressure tyres significantly affected zero tillage in the 100 – 200 mm layer, trafficking  
569 affected the 0 – 200 mm later under deep tillage. In shallow tillage treatments, the top 0- 100 mm layer was  
570 considerably impacted by high tyre pressure.

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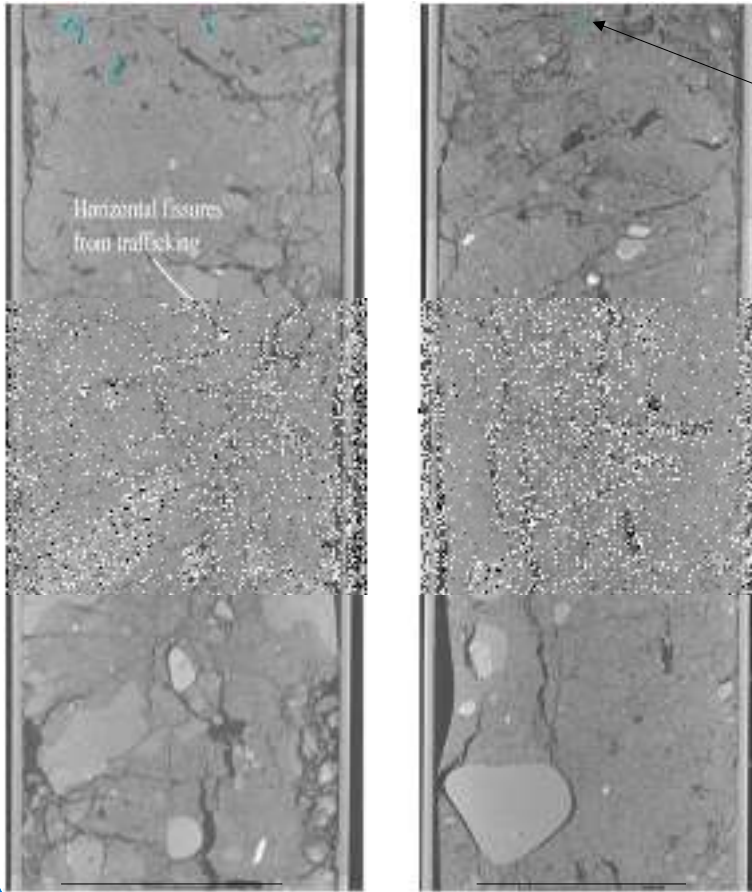
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Region grower root "seed points"

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573

574 **Figure 54.7:** Vertical view of X-ray CT images through centre of soil core using VGStudioMax® software for  
 575 (a) Shallow tillage conventional pressure (b) Zero tillage low tyre pressure. Scale bar = 50 mm.

576

577 4.25.2 Soil porosity in response to trafficking & tillage

578 ~~Sandy soils due to their adhesive and coarse grain nature, have reduced porosity, including lower levels of~~  
 579 ~~micropores compared to loamy soils (Arvidsson, 1998). The aggregation potential in this sandy loam soil is low.~~  
 580 ~~In the presence of plants, porosity and pore connectivity as shown to reduce further compared to clay cohesive~~  
 581 ~~soils which tend to increase in porosity through flocculation and aggregation (Baeq Labreuil et al., 2018). Here,~~  
 582 ~~we found soil porosity to be low in general across all treatments. When comparing cultivation systems, we found~~  
 583 ~~that shallow tillage in the 0-100 mm layer had significantly lower porosity (10.58%) compared to deep tillage~~  
 584 ~~(22.72%). Although zero tillage recorded low porosity values also (10.72%), it was not significantly different to~~

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585 ~~the other two systems.~~ Compared to non-trafficked treatments, trafficked soil in general caused a sharp decline in  
586 soil porosity in the top 0-100 mm layer. Tyre inflation pressure is one of the key contributors to soil stress in the  
587 100 to 1000 mm layer (Botta et al., 2008). The effect of re-compaction from trafficking after cultivation was often  
588 worse in deep tillage treatments, with a lower percentage porosity than in zero and shallow tillage (Table 42 for  
589 DTLP and DTCP treatments). In deeply cultivated soils, water infiltration rates can be reduced by up to 82% after  
590 a single wheeling's (Chyba, 2012), which has agronomic implications such as reduced water and nutrient use  
591 efficiency by up to 22% thus, potentially resulting in crop yield penalties of up to 38% (Ishaq et al., 2001). Yield  
592 effects by trafficking were modest in our study due to low soil moisture conditions during sowing in autumn 2018  
593 (Met office, 2019). Dry soil has increased soil strength, reducing the effects of soil compaction as the soil load  
594 support capacity would have increased thus, increasing permissible ground pressure (Hamza and Anderson, 2005).

595 A key characteristic of zero tilled soils is a change in soil pore architecture with vertically orientated  
596 fissures connected down through the soil profile created by biopores (Figure 4.7). Similar findings have resulted  
597 in reduced CO<sub>2</sub> fluxes and increased saturated hydraulic conductivity by surface connected porosity (Cooper et  
598 al., 2021). The same study found similar soil porosity levels between conventional and zero tillage with zero  
599 tillage total porosity ranging from <5%, 10% and 12% on average over 1-5, 6-10 and 11-15 years respectively.  
600 The significant increase in deep tillage soil porosity substantially increases soil respiration, resulting in up to 13.8  
601 times higher CO<sub>2</sub> emissions through increased oxidation and carbon breakdown (Reicosky et al., 1999). The lower  
602 porosities in zero and shallow tilled soils reduces space for gas exchange, reducing soil respiration and supporting  
603 carbon sequestration, thus increasing recalcitrant levels of carbon in soil. Mangalassery et al., (2014) found similar  
604 porosity results using X-ray CT methods to measure the effect of tillage method on greenhouse gas emissions,  
605 finding significantly higher porosity in tilled soil (13.6%) compared to zero tilled soil (9.6%) in the top 0-100 mm  
606 layer. However, in deeper soil horizons, no difference could be found between tillage system. The findings in this  
607 experiment agree with that study, showing both tillage methods did not differ significantly in the 100-200 mm  
608 layer with lower soil porosities recorded.

609

610

611

#### 612 4.25.3 Penetrometer responses to ~~tillage traffic and traffic~~

613 Penetrometer resistance (PR) is a useful parameter for evaluation of soil physical resistance to root growth (Otto  
614 et al., 2011). In general, trafficking had a considerable influence on soil PR in this study as depicted in Figure 6-  
615 8. The greatest contrast in soil penetration resistance was between trafficked and un-trafficked soil with zero  
616 tillage showing the highest resistance under conventional tyre pressure. Recent studies have shown that roots can  
617 exploit pores and bypass layers of strong soil (Atkinson et al., 2020). Axial pressure from repeated trafficking in  
618 ZTCP resulted in the highest PR values. However, root depth was less affected in contrast to STCP and DTCP.  
619 This might explain why roots could exploit existing pore networks in undisturbed soils compared to tillage  
620 treatments. In the middle layer examined, shallow till conventional pressure treatments suffered from a tillage pan  
621 effect shown in Figure 64.7. In fact, all trafficked zero and shallow tillage systems resulted in PR values beyond

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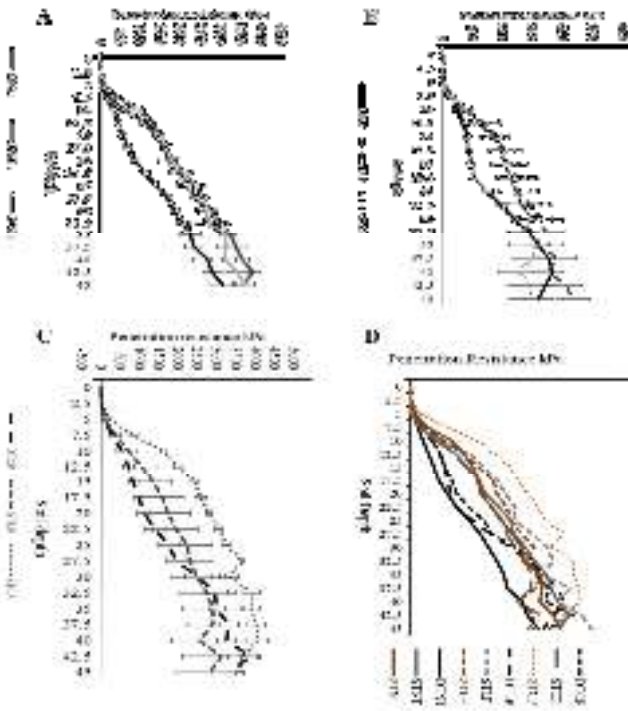
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622 2,000 kPa, a threshold level which several studies show there is a reduction in root growth (da Silva et al., 1994;  
623 Lapen et al., 2004; Tormena, da Silva and Libardi, 1999). A compact zone at shallow depths is detrimental to  
624 plant growth and crop yield in rainfed temperate climates when short term droughts occur (Campbell, Reicosky  
625 and Doty, 1974).

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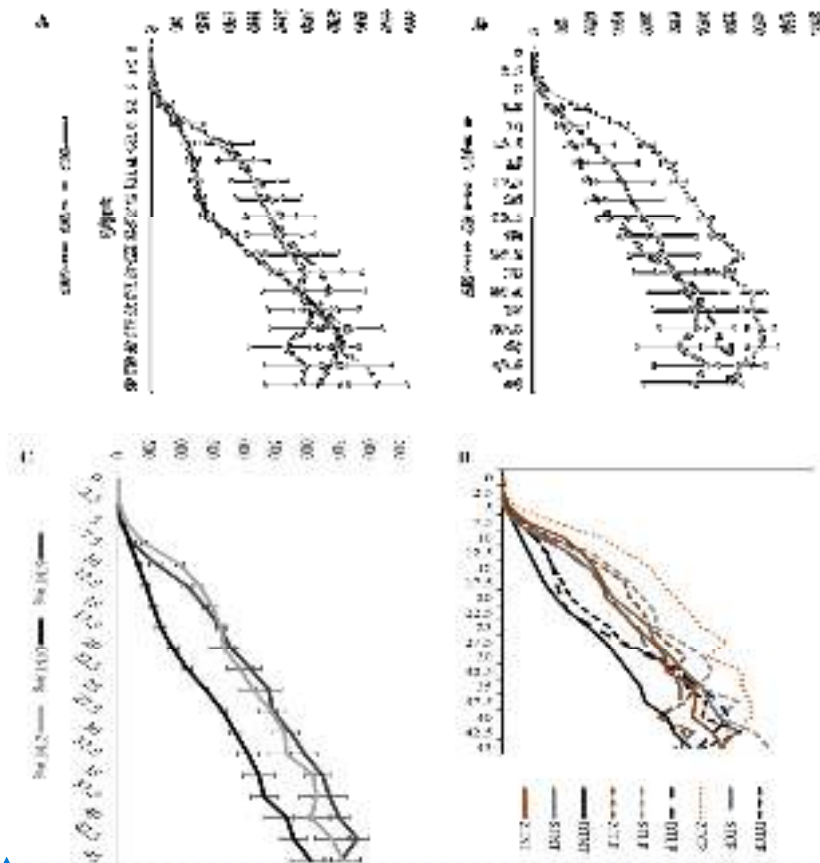
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628  
 629 **Figure 64.8:** Penetration resistance (kPa) for tillage and traffic treatments at soil depths of 0 - 450 mm. X axis  
 630 depicts soil depth. Y axis depicts Soil penetration resistance (kPa). Treatments represented by initials (Tillage: D  
 631 = Deep, S = Shallow, Z = Zero), (Traffic: NT = No traffic, LP = Low pressure tyre, CP = Conventional pressure  
 632 tyre). **A** low tyre pressure no traffic, **B** conventional low tyre pressure, **C** conventional tyre pressure no traffic and  
 633 **D** traffic x tillage treatments combined.

634

635 4.5.4 Root system architecture responses to tillage and traffic

636 The 'hidden half' (i.e. roots) of plants are difficult to interpret in field studies (Lynch and Brown, 2001). A large  
 637 root system is characterized by large biomass, root length and root length density (Ehdaie et al., 2010; Hamblin  
 638 and Tennant, 1987). Root biomass was an important indicator of root size, showing treatment effect at anthesis  
 639 compared to the tillering stage. In general, root biomass had a positive relationship with grain yield. Zero tillage  
 640 treatments both untrafficked and trafficked at low pressure had greater root biomass over all shallow tillage  
 641 treatments and deep till trafficked at conventional pressure. Although deep tillage treatments with no traffic had  
 642 the highest root biomass by GS61, it did not achieve the highest yield. No significant difference in root biomass  
 643 was found between tillage treatments in untrafficked samples, confirming that roots are more sensitive to

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644 trafficking than tillage method. The compaction effects of trafficking on soil structure exacerbated the impact on  
645 rooting in general. Typically, studies report shallower rooting, increases in root diameter and decreased axial and  
646 lateral rooting (Grzesiak et al., 2014). Due to the high moisture deficits depicted in (Figure 4.10) experienced  
647 during April and May 2019, it is likely that the deeper vertical rooting in zero tillage treatments retained more  
648 moisture at depth compared to other establishment methods.

649 4.2.4 Root system architecture responses to traffic

650 Traffic significantly affected root volume, root surface area, root length and RLD in shallow tillage treatments  
651 and zero tilled treatments trafficked at conventional pressure. RLD is an important parameter for characterizing  
652 root growth (Doussan et al., 2006) and has been used in previous studies as a key root parameter for modelling  
653 water uptake (Finker and Nye, 2000; Javaux et al., 2013). Munos-Romero et al., (2010) and Chakraborty et al.,  
654 (2008) results indicate that RLD is a positive predictor of crop yield. Although RLD had a positive correlation  
655 with crop yield in this study, root depth (using X-ray) displayed a much stronger relationship with crop yield  
656 (Figure 4.12). When comparing the highest root biomass (under deep tillage with no traffic) and bulk density  
657 results in the 100-200 mm layer, we found a reduction in root biomass when trafficked under conventional  
658 pressure by 28% in deep tillage under conventional pressure (BD = 1.66 g cm<sup>-3</sup>), 37% in shallow till conventional  
659 pressure (1.437 g cm<sup>-3</sup>) and 39% in zero tillage conventional pressure (1.583 g cm<sup>-3</sup>) treatments. The compaction  
660 effects of trafficking on soil structure exacerbated the impact on rooting in general. Typically, studies report  
661 shallower rooting, increases in root diameter and decreased axial and lateral rooting (Grzesiak et al., 2014).  
662 Colombi and Walter, (2017) observed decreased shoot dry weights in pot studies by 19 and 82% under moderate  
663 (1.45 g cm<sup>-3</sup>) and high (1.6 g cm<sup>-3</sup>) soil strength conditions. In the same study root dry weight was also reduced  
664 by 36 and 87% under the same soil strength conditions. Shallow tillage had the lowest root biomass in both  
665 trafficked and untrafficked treatments. Shallow tillage treatments suffered from visible horizontal fissures or  
666 "tillage pan" in Figure 5.10, causing significantly reduced rooting compared to deep tillage treatments. Moreover,  
667 a combination of <10% porosity and PR reaching >2,000 kPa in the 100-200 mm layer, it is likely that roots may  
668 also have suffered from anaerobic conditions due to poor infiltration rates through the tillage pan during heavy  
669 rainfall events. Conversely, root impedance may have occurred during drought periods through May and June  
670 (Batey, 2009). Alameda, Anten and Villar, (2012) proposed that axial growth suffers more than radial root growth.  
671 These effects of increased PR and soil bulk density were observed underin the current study. However, the increase  
672 in root diameter reported by several authors was not detected here (Chen et al., 2014; Lipiec et al., 2012; Tracy  
673 et al., 2012a; Alameda et al., 2012).

674 4.2.5 Tillage and trafficking effects on rooting and crop yield

675 In the present study, it was found that long term zero tillage plots under low tyre pressure increased yield by up  
676 to 0.772 Mt ha<sup>-1</sup> compared to the deep tillage conventional tyre pressure treatments. All zero tillage treatments  
677 yielded over 11 Mt ha<sup>-1</sup> compared to deep and shallow tillage treatments (10.71 Mt ha<sup>-1</sup>mean). Evidence using  
678 data collected from the X-ray CT scans showed deeper vertical rooting in zero tillage plots compared to shallow  
679 and deep tillage treatments (Figure 7). Coupled with deeper rooting, zero tillage no traffic treatments had  
680 significantly lower bulk density than deep tillage conventional pressure plots. Munoz-Romero et al., (2010)  
681 reported a yield increase of 0.5 Mt ha<sup>-1</sup> in zero tillage compared to conventional tillage which was associated with

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682 greater water use and increased water use efficiency, similar to (Chakraborty et al., 2008). Improvements in  
683 moisture retention, soil pore structures and reduced soil compaction under zero-tillage, may also have contributed  
684 to a yield increase over conventionally tilled treatments.

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685 It is possible that the lower levels of porosity found in zero tillage aided with water retention during drought  
686 periods on the highly sandy soil in this trial. Coupled with the development of vertically oriented soil structural  
687 characteristics attributed to earthworm activity and old root channels (Figure 5), the zero tillage treatments may  
688 also have had increased access to water by roots at lower soil horizons. Indeed, biopores benefit root growth by  
689 altering the surrounding chemical, physical and biological properties of soil (Stroud et al., 2017; Banfield et al.,  
690 2017). Thus providing macropore pathways with lower mechanical resistance in which deeper rooting  
691 preferentially grow towards (Zhou et al., 2021). In contrast, deep cultivation created a porous structure which has  
692 shown to increase respiration of aerobic microorganisms, improving the flow of air and water thus increasing  
693 CO<sub>2</sub> emissions (Mangalassery et al., 2014a). Crop yield was influenced less in zero tillage treatments by  
694 trafficking than the other tillage treatments. The lower sensitivity to compaction in zero tillage is attributed to an  
695 elastic behavior or increase in bearing capacity, with soil acquiring similar structural properties to grassland soil  
696 (Ehlers and Claupein, 1994).

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#### 699 4.35.5 2D & 3D imaging for studying root-soil relationships

700 Due to the complexity of measuring root systems, two methods were conducted to provide comprehensive  
701 analysis. Important topology (root networks) and geometrical (physical positions) characteristics of wheat rooting  
702 using X-ray CT were found in this study. A strong significant relationship between RLD (WinRHIZO™) and root  
703 depth (X-ray CT) was found (Figure S44.11) validating the suitability of image analysis methods in field studies.  
704 Further, root depth showed the strongest correlation with crop yield compared to root biomass and RLD (Figure  
705 S54.12). Moreover, the large environmental variance (low r number) in root relationships may have been caused  
706 by spatial effects reported in previous studies (Guo et al., 2020; Zhou et al., 2021). Compared to traditional 2D  
707 WinRHIZO™ analyses, the significant difference found with *in situ* root depth between treatments using X ray  
708 CT was not detected by destructive WinRHIZO™ analysis (i.e., it involves the washing of soil from root material,  
709 thus losing important architectural data). Destructive root analysis showed evidence of superior rooting properties  
710 under deep tillage treatments (e.g., root length density and root volume). Visualizing important behaviors of wheat  
711 rooting in field scale trials, highlights the importance-significance of root depth to sustain high yields in drought  
712 conditions. Figure 74-9 depicts significantly longer root length in zero tillage treatments compared to trafficked  
713 deep and shallow tillage, with trafficked treatments roots were generally confined to the top 0-50 mm of soil. In  
714 general, root length rarely surpassed 100 mm in depth. This was partly due to insufficient resolution available  
715 with the X-ray CT scanner to capture finer root materials (Pfeifer et al., 2015).

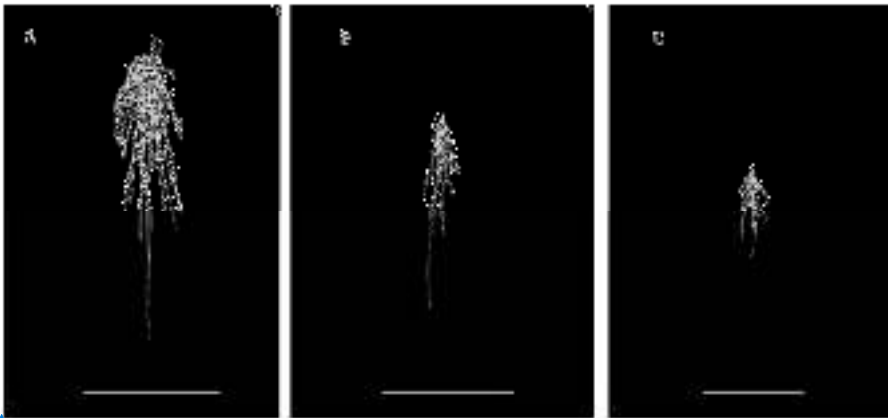
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717 In general, both root analysis methods showed agreement in the results. Zero tillage treatments had significantly  
718 deeper rooting over shallow tillage and deep tillage trafficked treatments. Using the WinRhizo™ method,  
719 untrafficked deep tillage treatments showed superior root length. Similar disagreements in findings between  
720 methods could be explained by the difference in methodology between the two imaging approaches as X-ray CT  
721 is 3D and scans roots in soil whilst, WinRhizo™ is 2D and scans washed roots (Tracy et al., 2012a). Root volume  
722 and surface area were also examined using X-ray CT. In contrast to the WinRhizo™ analysis, no significant  
723 differences could be detected between treatments. The root volumes obtained by the WinRhizo™ were much  
724 greater than the volumes attained from the X-ray CT scan. The difference can be attributed by much clearer  
725 contrasts between air and root material with the destructive method compared to limitations with resolution and  
726 density differences between soil, root and organic materials (Mooney et al., 2012) in the X-ray CT scan-images.

727



728

729 **Figure 74.9:** Root system architecture of winter wheat during anthesis for (a) Deep tillage no traffic, (b) Zero  
730 tillage low tyre pressure and (c) deep tillage conventional tyre pressure. (a) and (b) showed significantly longer  
731 root length on the primary axis compared to (c) deep tillage trafficked treatments. Scale bar = 70 mm.

#### 732 4.5.6 Traffic and tillage effects on rooting and crop yield

733

734 In the present study, it was found that long term zero tillage plots under low tyre pressure increased yield by up  
735 to 0.772 Mt ha<sup>-1</sup> compared to the deep tillage conventional tyre pressure treatments. All zero tillage treatments  
736 yielded over 11 Mt ha<sup>-1</sup> compared to deep and shallow tillage treatments (10.71 Mt ha<sup>-1</sup> mean). Evidence using  
737 data collected from the X-ray CT scans showed deeper vertical rooting in zero tillage plots compared to shallow  
738 and deep tillage treatments (Figure 4.9). Coupled with deeper rooting, zero tillage no traffic treatments had  
739 significantly lower bulk density than deep tillage conventional pressure plots. Munoz-Romero et al., (2010)  
740 reported a yield increase of 0.5 Mt ha<sup>-1</sup> in zero tillage compared to conventional tillage which was associated with  
741 greater water use and increased water use efficiency, similar to (Chakraborty et al., 2008). Improvements in

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742 moisture retention, soil pore structures and reduced soil compaction under zero tillage, may also have contributed  
743 to a yield increase over conventionally tilled treatments.

744 It is possible that the lower levels of porosity found in zero tillage aided with water retention during  
745 drought periods on the highly sandy soil in this trial. Coupled with the development of vertically oriented soil  
746 structural characteristics attributed to earthworm activity and old root channels (Figure 4.7), the zero tillage  
747 treatments may also have had increased access to water by roots at lower soil horizons. Indeed, biopores benefit  
748 root growth by altering the surrounding chemical, physical and biological properties of soil (Stroud et al., 2017;  
749 Banfield et al., 2017). Thus providing macropore pathways with lower mechanical resistance in which deeper  
750 rooting preferentially grow towards (Zhou et al., 2021). In contrast, deep cultivation created a porous structure  
751 which has shown to increase respiration of aerobic microorganisms, improving the flow of air and water thus  
752 increasing CO<sub>2</sub> emissions (Mangalassery et al., 2014a). Crop yield was influenced less in zero tillage treatments  
753 by trafficking than the other tillage treatments. The lower sensitivity to compaction in zero tillage is attributed to  
754 an elastic behavior or increase in bearing capacity, with soil acquiring similar structural properties to grassland  
755 soil (Ehlers and Claupein, 1994).

#### 756 4.5. Conclusion

757 The results from this research highlight the importance of traffic management for improving crop productivity.  
758 Physical and visual implications of soil compaction on the soil profile were demonstrated in this study, signifying  
759 the implications of tyre pressure on root growth. High tyre pressure Traffic significantly reduced root development  
760 in all tillage treatments with tyre pressure having no significant effect on mitigating compaction effects on soil  
761 and roots. However, Moreover, deep, and shallow tillage systems were more influenced by compaction than zero  
762 tillage with roots confined to the top 0-60 mm thus, reducing primary vertical rooting and inhibiting roots access  
763 to deeper soil moisture reserves. The highly significant impact on crop yield was highlighted by the strong  
764 relationship between root depth and crop yield. The visible effects of trafficking on the soil profile depicted  
765 through X-ray CT, provides evidence of the damage modern farm machinery can cause for root resource capture,  
766 leading to potential increased drought stress and yield loss in crop production. This long-term trial site has shown  
767 that zero tillage does not affect root growth, in fact, reduced bulk density, improved grain yield and rooting depth  
768 significantly through deeply connected vertical soil pore fissures created by earthworms and old root channels,  
769 allowing roots to access deeper soil moisture reserves. These findings suggest that scientists and farmers should  
770 focus on designing improved zero tillage cropping systems, managing field trafficking protocols with controlled  
771 traffic farming. Moreover, further investigation on tracks and dual radial tyres are required to quantify practical  
772 compaction mitigation measures. Furthermore, this research shows that the combination of X-ray CT scanning  
773 along with traditional destructive methods provide a robust method for assessing in field rooting for future crop  
774 breeding initiatives and soil management practice. This research concludes that trafficking has more profound  
775 effects for root growth and crop yield than tillage method little differences were found between deep tillage and  
776 zero tillage methods in the absence of traffic in terms of overall physical root growth.

777 However, in abundance of biopores and increased soil bearing capacity to withstand machinery traffic in in zero  
778 tillage systems increased rooting depth and moisture retention during the growing season.

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[Supplement](#). The supplement related to this article is available in a separate word file as per submission.

4.7 Supplementary tables and figures

HAU	Deep	Shallow	Zero	Mean
<b>Traffic/Tillage 0—100 mm</b>				
No traffic	0.971	1.099	0.994	1.058 <sub>b</sub>
LP	1.351	1.625	1.230	1.401 <sub>a</sub>
CP	1.661	1.444	1.282	1.462 <sub>a</sub>
$P < 0.01$				
<b>Traffic/Tillage 100—200 mm</b>				
No traffic	1.079	1.406	1.353	1.279 <sub>b</sub>
LP	1.389	1.509	1.552	1.483 <sub>a</sub>
CP	1.637	1.437	1.583	1.553 <sub>a</sub>
$P < 0.01$				
<b>Traffic/Tillage 200—300 mm</b>				
No traffic	1.429	1.466	1.404	1.433
LP	1.593	1.787	1.619	1.666
CP	1.537	1.548	1.548	1.544

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**Table 4.5:** Analysis of variance table for penetration resistance x tillage, traffic and tillage x traffic interactions between 0—150 mm, 150—300 mm and 300—450 mm.

	0 – 150 mm 2019	150 – 300 mm 2019	300 – 450 mm 2019
<b>Traffic</b>			
No traffic	432.3 <sub>b</sub>	1848 <sub>b</sub>	3028.5 <sub>b</sub>
CP	538.8 <sub>a</sub>	2614 <sub>a</sub>	3753.6 <sub>a</sub>
LP	626.7 <sub>a</sub>	2422 <sub>a</sub>	3655.7 <sub>a</sub>
P value	< 0.001	< 0.000	< 0.000
<b>Tillage</b>			
Deep	240 <sub>c</sub>	1366.3 <sub>b</sub>	3135.2 <sub>b</sub>
Shallow	488.4 <sub>b</sub>	2811.4 <sub>a</sub>	3800 <sub>a</sub>
Zero	869.4 <sub>a</sub>	2706.7 <sub>a</sub>	3502.7 <sub>a</sub>
P value	< 0.000	< 0.000	< 0.000
Tillage x traffic	0.001	0.067	NS

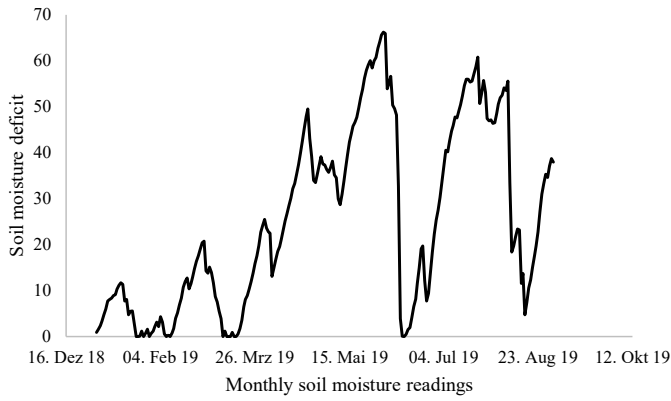
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\*Significant difference between means is represented by different letters





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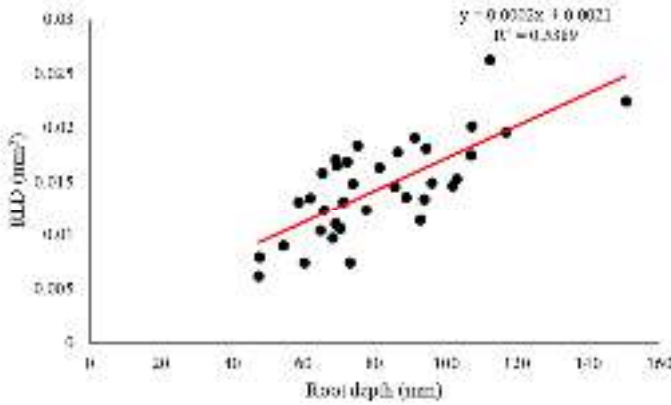
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856 **Figure 4.10:** Soil moisture deficit model during the growth period (January–August) in Harper Adams University.

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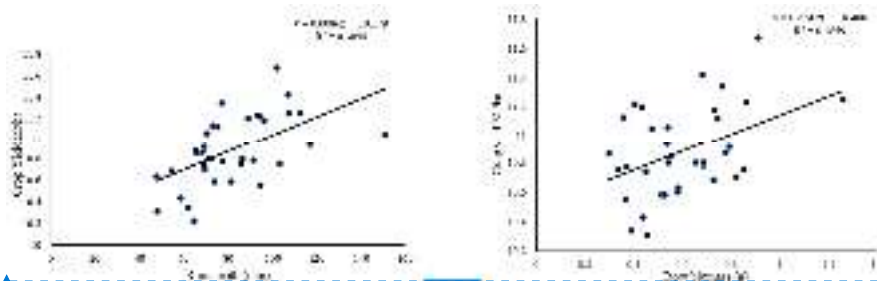
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859 **Figure 4.11:** Linear regression measuring the relationship between RLD (mm<sup>3</sup>) (destructive analysis) and root depth (mm) (X-ray CT).

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 863 **Figure 4.12:** Linear regression between crop yield (Mt/ha) and (a) Root depth and (b) root biomass as predictors  
 864 of crop yield.

865  
 866 **Author contributions.** KMc and ST conceived the experiment. DH & MH carried out sampling and soil analysis.  
 867 DH processed and analysed all samples. DH analysed and interpreted the data and wrote the manuscript. All  
 868 authors contributed to the data, providing interpretation and comments to the manuscript.

869  
 870 **Competing interests.** The authors declare that they have no conflict of interest

871  
 872 **Disclaimer.**

873  
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 875 Adams University. Long term crop rotation treatments at the Large Marsh site on the University grounds are  
 876 managed and maintained by the agricultural staff at the university.

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