



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

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9           **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, root length  
15          density and deeper seminal rooting analysed using X-ray Computed Tomography (CT). In general, conventional  
16          pressure trafficking had a significant negative influence on crop yield, root development, bulk density and total  
17          soil porosity of deep and shallow tillage conventional pressure systems compared no traffic zero and deep tillage  
18          systems. Visual improvements in soil structure under zero tillage may have improved crop rooting in zero tillage  
19          treatments through vertical pore fissures (biopores), enhancing water uptake during the crop flowering period.  
20          This study highlights the implications of soil structural damage on root system architecture created by compaction  
21          in crop production. The constricted root systems found in conventional pressure shallow tillage, zero and deep  
22          tillage trafficked regimes emphasizes the importance of using technology to improve soil management and reduce  
23          the trafficked areas of agricultural fields.

24

25

26           **1. 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          affecting 33 million hectares in Europe alone (Akker and Canarache, 2001). Soil compaction is a form of physical  
34          degradation caused by short crop rotations and heavy farm machinery working on low organic matter soils in wet  
35          conditions resulting in the loss of pore space due to an externally applied load, forcing soil aggregates together



36 (Defossez and Richard, 2002). The resulting anaerobic high density soils have significantly reduced capacity to  
37 store water and nutrients required by growing crops (Hamza and Anderson, 2005) and severely compacted soils  
38 prevent soil exploration from root growth (Tracy et al., 2012).

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

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

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

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



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

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

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

96

## 97 **2. Materials and Methods**

### 98 2.1 Site and soils

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

107 In the year prior to this study, it was necessary to plant a break crop (2017/18) as part of a standard crop rotation  
108 to improve soil conditions and reduce diseases such as take all (*Gaeumannomyces graminis* var. tritici). A field  
109 bean (*Vicia Faba*) break crop was planted, and yields were assessed to ensure the trial site was uniform with no  
110 underlying issues. Since the trial site began, the crop rotation has been first winter wheat (*Triticum aestivum* L.)



111 harvest in 2012 followed winter wheat in 2013, winter barley (*Hordeum vulgare L.*) 2014, winter barley 2015,  
112 followed by a cover crop “TerraLife-N-Fixx” (DSV United Kingdom Ltd, 2015); Spring oats 2016, spring wheat  
113 2017 and winter beans 2018. For this trial, winter wheat (*Triticum aestivum L. cv. Graham*) was drilled early  
114 October 2018 when the soil was dry, friable and soil temperatures  $>6$  °C. The seeding rate was 250 seeds per m<sup>2</sup>  
115 and drilling took place on the 5<sup>th</sup> of October. This is in line with local normal farming practice.

116

117 **Table 1.** Description of the topsoil (0-300 mm) properties for Harper Adams University trial site, Shropshire, 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

118 \*Landis Soil guide (Cranfield University, 2021).

119 LOI, Loss of Ignition.

120

121

## 122 2.2 Experiment design

123

124 The experiment was a randomised 3 x 3 factorial arrangement of 9 treatments in four complete replicate blocks.  
125 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.  
126 Tramlines were at a 90° angle to plots with 24 m spacing for fertilising and spraying operations throughout the  
127 growing season. A split-plot design was used, half the plot (30 m) designated for sampling and the other half was  
128 undisturbed for yield data collection. The half plot for sampling was sub-divided for the two sampling stages,  
129 ensuring sampling did not occur near the same location as the previous sample. Cultivation for spring beans in  
130 2017 was performed at three depths, 250 mm for deep tillage, 100 mm for shallow tillage and direct into stubble  
131 for zero tillage. In the winter wheat trial, soil cores were collected at tillering (Growth stage (GS) 25) and the  
132 flowering stage (GS 61-69) (Zadoks, Chang and Konzak, 1974) in July 2019.

133 Three commercial crop establishment systems were used consisting of three different tillage depths. The following  
134 tillage treatments are denoted as: Treatment 1 = Deep tine cultivator at 250 mm (DT) for deep tillage similar to  
135 (Ren *et al.*, 2019), treatment 2 = shallow disc cultivation at 100 mm (ST) and treatment 3 = zero tillage using a  
136 direct seed drill (ZT). In combination with the different tillage depths, three traffic regimes were used in this study



137 no traffic (NT), conventional tyre pressure (CP) and low tyre pressure (LP). Tillage depths were combined with  
138 traffic management practices for the 9 treatments (DTNT, DTCP, DTLP, STNT, STCP, STLP, ZTNT, ZTCP &  
139 ZTLP).

140

#### 141 2.2.2 Tillage equipment and tyres

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

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

155

156

157

#### 158 2.3.1 Soil physical properties

159 Soil bulk density samples were also collected within the trafficked and non-trafficked area of the plot, to represent  
160 the bulk density of the tillage treatments. Samples were replicated three times. Each core sample was 50 mm in  
161 width and 300mm in length. An Eijkelkamp® soil corer was used to take bulk densities samples. Each bulk density  
162 sample was taken within 0.5 m of the location of the soil cores taken for X-ray CT. The objective was to represent  
163 the physical constraints (or lack of) for root growth in each plot examined. The method used in this study involved  
164 splitting the bulk density sample into three 100 mm sections (0-100 mm, 100-200 mm and 200 – 300 mm) similar  
165 to (Smith, 2016). The corer was opened in the field and split using a knife and ruler.

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

170 
$$\text{Moisture \%} = \frac{\text{fresh weight(g)} - \text{dry weight (g)}}{\text{dry weight(g)}} * 100 \quad \text{Equation 1}$$



171 Dry bulk density ( $\text{Mg m}^{-3}$ ) = dry soil weight ( $\text{Mg}$ )/ soil volume ( $\text{m}^{-3}$ )

172

Equation 2.

173

174

175 2.3.2 Penetration resistance (PR)

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

183

184 2.3.3 Soil porosity analysis

185 Before soil porosity analysis on ImageJ software (version 1.52) (Schneider et al., 2012) could commence, an  
186 image stack was created in VG Studio Max<sup>®</sup> for each scan. The contrast was adjusted to improve the uniformity  
187 and visibility of the soil pores. The register object tool corrected scan discrepancies for soil core angle.  
188 Straightening the scan allowed a cylindrical shape to be cropped and the tube edges and air space outside of the  
189 soil core removed. This enabled soil data to be captured throughout the soil core. A new volume was selected and  
190 extracted from the original. This created a separate cropped image volume to work from. The surface  
191 determination tool in VG Studio Max<sup>®</sup> was used to threshold pore spaces within the solid matrix. The tool defines  
192 the contour of objects, separating 3D data into regions, providing meaningful soil data (Borges de Oliveira et al.,  
193 2016). The image was then inverted to remove the extracted variables from the image and highlighting the pore  
194 spaces in the soil core. The processed image was exported as an \*.TIFF image stack for further analysis using  
195 ImageJ software.

196 Soil pore characteristics were measured using X-ray CT to establish information about the 3D soil environment  
197 for root growth without disrupting the structural integrity of the soil core. The original grey-scale X-ray CT images  
198 were analysed using ImageJ software. The scale was set for each dataset to define to spatial scale of the active  
199 image. The unit of length was set in millimeters and the known distance was 0.045mm (45 $\mu\text{m}$ ). Each scanned  
200 core was cropped to remove the area outside of the soil column. The action of soil coring during sampling had the  
201 effect of loosening the bottom 20 mm of the core, therefore 415 slices at the bottom of each scan were discarded  
202 to remove the loosening effect due to the sampling process. The downward movement of the PVC pipe also caused  
203 a smearing effect on the soil at the outside edge of the core and this area was also removed by cropping.

204 The processed image was 1220 x 1220 pixels in size. Applying the contrast enhancement filter helped normalize  
205 all slices. The filter reduces the differences in pixel grey-level between slices known as beam hardening  
206 (Wildenschild et al., 2002). The ImageJ Huang automatic threshold algorithms were used for each scan to create  
207 binarized images and separate the air-filled pores from the background region. The binarized scans were de-  
208 speckled twice to remove unwanted noise within each scanned image, improving analysis and accuracy of the



209 investigated pores. The Look Up Table (LUT) was inverted to change the white pores to black, ensuring analysis  
210 calculated the air-filled pores and not the soil matrix. The resulting binary images were analysed using the Analyze  
211 Particles tool which provided information for average pore size, total area and percentage porosity for each  
212 individual image.

213

#### 214 2.4.1 *Soil core sampling*

215 Field soil core size was chosen to capture as much root material growing in the field as possible while minimizing  
216 the trade-off that exists with the X-ray CT technology between image resolution and core size (Mooney et al.,  
217 2012; Zhou et al., 2021). The core dimensions were consistently 70 x 300 mm (diameter x depth) for each sample.  
218 Soil cores were extracted from the field sites at GS 25 in February and again at GS 61 in June. Sampling was  
219 carried out at GS 61 during wheat anthesis, when root growth is at its peak (Gregory et al., 1978). Due to high  
220 moisture deficits in HAU (43 mm) during sampling at GS 61 in early July, the soil sample area was wetted with  
221 2.5 L of water and allowed to infiltrate. This lubricated the soil, reduced soil fracturing, and allowed tube insertion  
222 and soil core extraction to take place as smoothly as possible. Polyvinyl chloride (PVC) drainage pipes were cut  
223 to size (70 x 300 mm) and these tubes were used to collect soil cores (as per Millington et al, 2017).

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

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#### 240 2.5.1 *X-ray computed tomography (CT) – Root analysis*

241

242 Soil cores were transferred to the University College Dublin (UCD) X-ray CT facility at the Rosemount  
243 Experimental Research Station at Belfield Campus, UCD, Ireland. The soil cores were scanned using a Phoenix®  
244 v|tome|x M 240 kV scanner (GE Measurement and Control solution, Wunstorf, Germany). The v|tome|x M was



245 set at a voltage of 90 kV and current of 400  $\mu$ A to optimize contrast between background soil and root material.  
246 A voxel resolution of 45  $\mu$ m was achieved by using the ‘Multi Scan option’ to scan in 4 segments. A total of 1800  
247 projection images per section were taken at 200 m/s per image using the ‘Fast Scan option’, which has the default  
248 values of an image averaging of 1 and 0 skip. No filters were used during scanning. The total scan time per core  
249 was 24 minutes or 6 minutes per section. Once scanning was complete, the images were reconstructed using  
250 Phoenix datos|x2 rec reconstruction software, the four scans were assembled into one 3D volume for the whole  
251 core. Core samples were scanned within a week of the sampling date, the scanned core was 300 mm in length and  
252 70 mm diameter. The software corrected movements during the scanning process and removed noise from scanned  
253 images.

#### 254 2.5.2 X-ray CT root segmentation

255  
256 Image analysis for X-ray CT images was performed using the software VGStudioMax<sup>®</sup>, version 3.2 (Volume  
257 Graphics GmbH, Heidelberg, Germany) to segment roots and soil porosity. Roots were segmented by setting seed  
258 points and using selected threshold values in the “*Region grower*” that enabled fast and accurate selection of grey-  
259 scale voxels (3D pixels) pertaining to root materials. The root system was extracted from the greyscale CT image  
260 of soil using the VGStudioMax<sup>®</sup> semi-automated local adaptive thresholding “*Region Growing*” selection tool,  
261 similar to (Tracy et al., 2013). Root volumes were calculated by segmenting the root region of interest (ROI).  
262 Once the roots were segmented from the image, erosion and dilation tool was selected at 1 pixel using the *Region*  
263 *Growing* tool. Root system architecture parameters such as root vertical depth, root volume and root surface area  
264 were measured from the segmented root systems. Root vertical depth was calculated on the Z axis in  
265 VGStudioMax<sup>®</sup> from the length of a complete root from the base seed point.

266

#### 267 2.5.3 Destructive 2D root analysis

268 After the soil cores were scanned, the soil and root material were separated by root washing gently with a water  
269 jet hose. Two sets of sieves with a mesh size of 2 mm and 1 mm collected root material. Roots were washed and  
270 soil material removed before the roots were placed into a sealed and labelled bag filled with water. The washed  
271 root samples were placed into a freezer until scanning and analysis with WinRHIZO<sup>™</sup> scanning and software  
272 (version 2016a Regent Instruments, Canada) commenced. The root samples were thawed before scanning with  
273 the WinRHIZO<sup>™</sup> software. Large root stumps were removed from the sample prior to placing it inside the tray to  
274 reduce root misrepresentation (Wang and Zhang, 2009). Roots were placed onto a clear transparent tray (30 cm x  
275 20 cm) with water. A pair of plastic forceps were used to spread out root seminal and lateral roots. Images were  
276 scanned at a resolution of 600 dpi (42  $\mu$ m pixel size) with an Epson Perfection V800 scanning system. Root  
277 images were measured for root length, root surface area, average root diameter and root volume for the total soil  
278 core. This output was used to verify the 3D root outputs from VGStudioMax<sup>®</sup> (Flavel et al., 2017; Tracy et al.,  
279 2012). The WinRHIZO<sup>™</sup> software enabled rapid assessment of root parameters. It calculated the root volume by  
280 determining the average root diameter and root length by pixel counting the 2D root image and then assuming the  
281 root shape was cylindrical. The WinRHIZO<sup>™</sup> used a skeletonization method for characterizing root systems  
282 (Himmelbauer, Loiskandl and Kastanek, 2004). The software uses greyscale values in \*.TIFF file format. The



283 output of the images was distinguished by global thresholding analyses for root diameter while root length was  
284 validated by skeleton images. After WinRHIZO™ scanning, the roots were removed from the scanning tray using  
285 forceps. The root samples were dried at 70°C for 24 hours and the root biomass samples were weighed.

## 286 2.6 Soil Moisture Deficit Model

287

288 Soil Moisture Deficit (SMD) was calculated based on the SMD hybrid model for Irish grassland (Schulte et al.,  
289 2005). Rainfall, wind speed (m/s), sunshine hours, maximum and minimum temperature data were taken from the  
290 nearest weather station located in Newport, Shropshire 6km from the site (Met office, 2019).

291

## 292 2.7 Statistics

293

294 Data from the scanned (destructive and non-destructive) images and root biomass were not normally distributed.  
295 Non-normal data do not meet the assumptions underpinning ANOVA (Analysis of Variance); therefore, all data  
296 underwent log transformation (in Microsoft Excel) before being exported to Minitab 18® where analysis of  
297 variance (ANOVA) was performed to homogenize the variances of the compared means (Poorter and Garnier,  
298 1996). For linear regression analysis, residuals of data were made to ensure that the assumptions of the analysis  
299 were met (normal distribution, constant variance, etc). Normality was tested using the Anderson-Darling test in  
300 Minitab 18®.

301

## 302 3. Results

### 303 3.1 Growing conditions during crop season

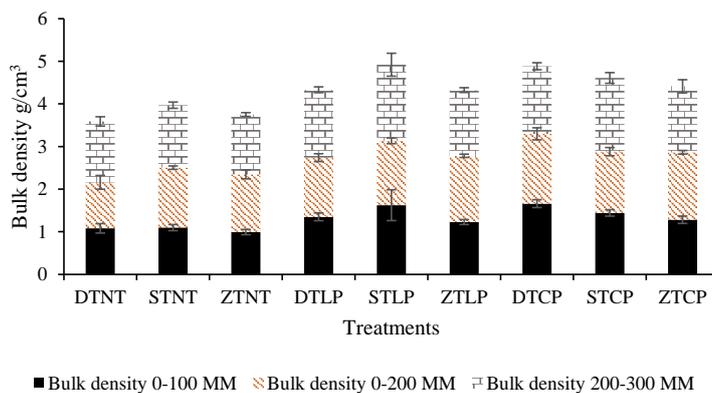
304 In 2018, crops were established at low soil moisture levels, which may have reduced soil compaction caused by  
305 tillage operations across all site locations. From January to August (2019), 418.6 mm of rainfall was recorded at  
306 HAU, 68 mm in total for January and February. Soil moisture deficits reached 66.2 mm in HAU (Supplementary  
307 fig. S1) by early June 2019. High soil moisture deficits were recorded from early April to June, causing drought  
308 stress during rapid growth periods (Met office, 2019).

#### 309 3.2.1 Soil properties – Bulk density & Penetrometer resistance

310 The calculated probability (*P*-value) and standard error of the mean (SEM) from one-way ANOVA analysis is  
311 given in Fig. 1 for bulk density presented for 0-100 mm, 100-200 mm, and 200-300 mm measurements. In the top  
312 0-100 mm, bulk density was significantly higher in DTCP (1.66 Mg m<sup>-3</sup>) and STCP (1.44 Mg m<sup>-3</sup>) treatments  
313 compared to ZTNT (0.994 Mg m<sup>-3</sup>) and DTNT (0.97 Mg m<sup>-3</sup>) (*P*<0.01). STNT (1.09 Mg m<sup>-3</sup>) was significantly  
314 higher than ZTNT and DTNT and only significantly lower than DTCP. In the middle horizon (100-200 mm), a  
315 significant interaction between trafficking treatment was found. Bulk density was significantly lower in DTNT



316 (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>) treatments (P<0.05). In the bottom  
317 200-300 mm layer measured, no significant tillage x traffic interaction was found (P>0.05).

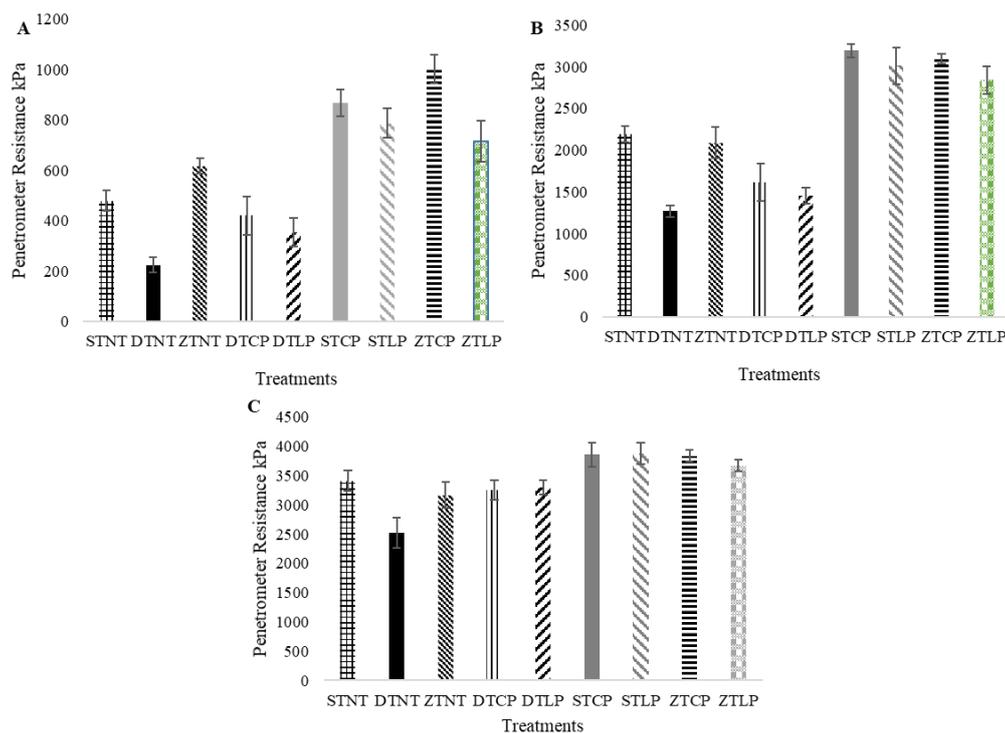


318

319 **Figure 1.** Soil bulk density g/cm<sup>3</sup> for tillage x traffic treatments for three depth layers.

320 Penetration resistance (PR) was recorded in February 2019 when the soil was at field capacity. Measurements  
321 were grouped into three groups, 0-150 mm, 150-300 mm, and 300-450 mm depth layers. Figure 2 depicts the  
322 combined three layers grouped into one 0-450 mm graph. The ANOVA analysis revealed highly significant  
323 differences for each layer. In the 0-150 mm layer, DTNT recorded the lowest kPa (kilopascals) readings and was  
324 significantly lower than ZTCP, STCP, STLP, ZTLP and ZTNT (P< 0.000). DTCP and DTLP were significantly  
325 lower kPa than ZTLP, STLP, STCP and ZTCP. ZTCP recorded the highest kPa reading and was significantly  
326 higher than ZTLP, ZTNT, STNT, DTLP, DTCP and DTNT. In the second layer (150-300 mm), similar trends  
327 were found and highly significant (P<0.000). STCP showed the highest kPa (3193.5 kPa) and was significantly  
328 higher than STNT, ZTNT, DTNT, DTLP and DTCP. In contrast, DTNT recorded the lowest reading (1268.4 kPa)  
329 and was significantly lower than ZTNT, STNT, ZTLP, ZTCP, STCP and STLP. STNT revealed significantly  
330 lower kPa than STLP, ZTCP and STCP. ZTNT penetrometer readings were significantly lower than all trafficked  
331 ZT and ST treatments. In the lower depth (300-450 mm), DTNT was significantly lower than STLP, STCP, ZTCP,  
332 ZTLP and STNT (P<0.000).

333



334

335 **Figure 2.** Penetration resistance for three layers (a) 0-150 mm ( $P < 0.000$ ), (b) 150-300 mm ( $P < 0.000$ ) and (c) 300-  
 336 450 mm ( $P < 0.000$ ) during wheat tillering (GS25). Soil moisture conditions were at field capacity during sampling.

337

### 338 3.2.2 Soil porosity

339 The results of the ANOVA analysis of the CT-measured porosity (0-220 mm) are presented in Table 2. Soil  
 340 porosity results were split into two soil layers of 0-100 mm and 100-200 mm respectively. In the top 0-100 mm  
 341 layer, DTNT showed significantly higher total pore space ( $P < 0.01$ ) compared to all other treatments except ZTNT.  
 342 Tillage had a significant effect on soil porosity in the no traffic samples in the 0-100 mm layer ( $P < 0.05$ ). Deep  
 343 tillage with no traffic had higher soil porosity (22.72%) than in shallow tillage (no traffic) (10.58%). There was  
 344 no significant difference between soil porosity under zero tillage and shallow tillage in the no traffic samples.  
 345 Trafficking had a significant effect on overall porosity. In deep tillage treatments, overall porosity 22.72% (no  
 346 traffic) was reduced to 8.08% (under low tyre pressure) and 6.50% under conventional tyre pressure. Traffic had  
 347 little effect on shallow and zero tillage porosity in the top 0-100 mm when compared to the no traffic samples  
 348 with small reductions in porosity. In the second examined layer, 100-200 mm zone, tillage and traffic were not  
 349 significantly different ( $P < 0.487$ ). The percentage porosity shown in Table 2, indicate a sharp decline in the lower  
 350 depth with only 9.02% in DTNT. DTCP treatments recorded the lowest porosity (3.96%).

351



352

353

354 **Table 2.** Soil porosity for tillage x traffic for two soil layers.

ImageJ soil porosity % 0-100mm	<i>n</i>	No traffic	low tyre pressure	Conventional tyre 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	<i>n</i>			
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				

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

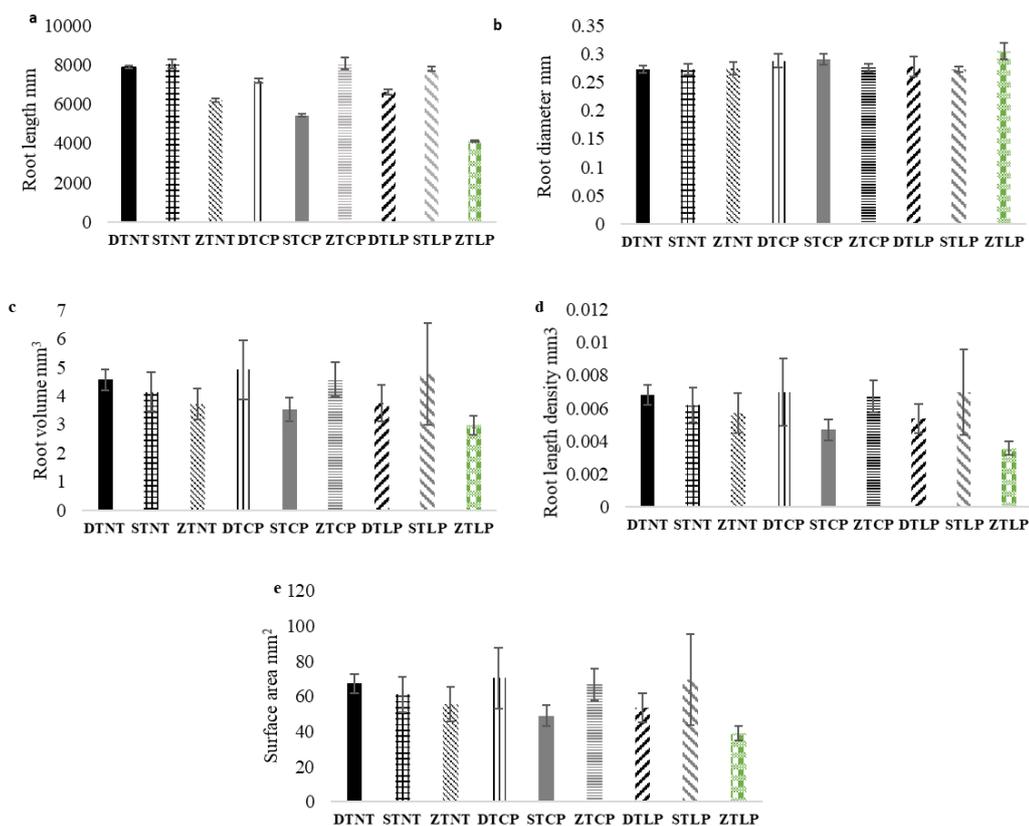
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357 3.3.1 *Destructive 2D root analysis*

358 The interaction between tillage system and trafficking protocols using destructive root measuring methods  
 359 (WinRHIZO™) are shown in fig 3 for GS 25 and fig 4 for GS 61. At GS25, no significant differences were found  
 360 between traffic and tillage treatments. However, the WinRHIZO™ analysis revealed a tendency towards increased  
 361 root growth in no traffic treatments. At the later growth stage (GS61), Figure 3 depicts the results showing highly  
 362 significant interactions between trafficking systems on root length density (RLD) (P<0.001) and root length (P<  
 363 0.001), root surface area (P<0.002) and root volume (P< 0.05). DTNT showed significantly higher RLD, root  
 364 surface area and root length compared to ZTCP, STCP and STLP. Root volume was significantly higher in DTNT  
 365 over ZTCP and STCP. DTNT produced nearly double the root length compared to ZRCP. In contrast to DTCP,  
 366 root surface area reduced by 36% compared to untrafficked areas (no traffic samples). In shallow and zero tillage,  
 367 root surface area was reduced by 32% and 63.6% respectively in conventional pressure samples compared to  
 368 untrafficked samples. There was no significant difference for root diameter and between all tillage and trafficking  
 369 regimes. The results demonstrate that there was no significant difference in RLD at the tillering stage, nor could  
 370 trends be found as roots were undeveloped. However, at anthesis, the RLD was significantly higher under non-  
 371 trafficked tillage treatments when compared to DTCP, STCP and ZTCP (Fig 3b).

372

373



374

375 **Figure 3.** Tillering (GS25) root system architecture using destructive root method. (a) Root length (mm), (b) Root  
 376 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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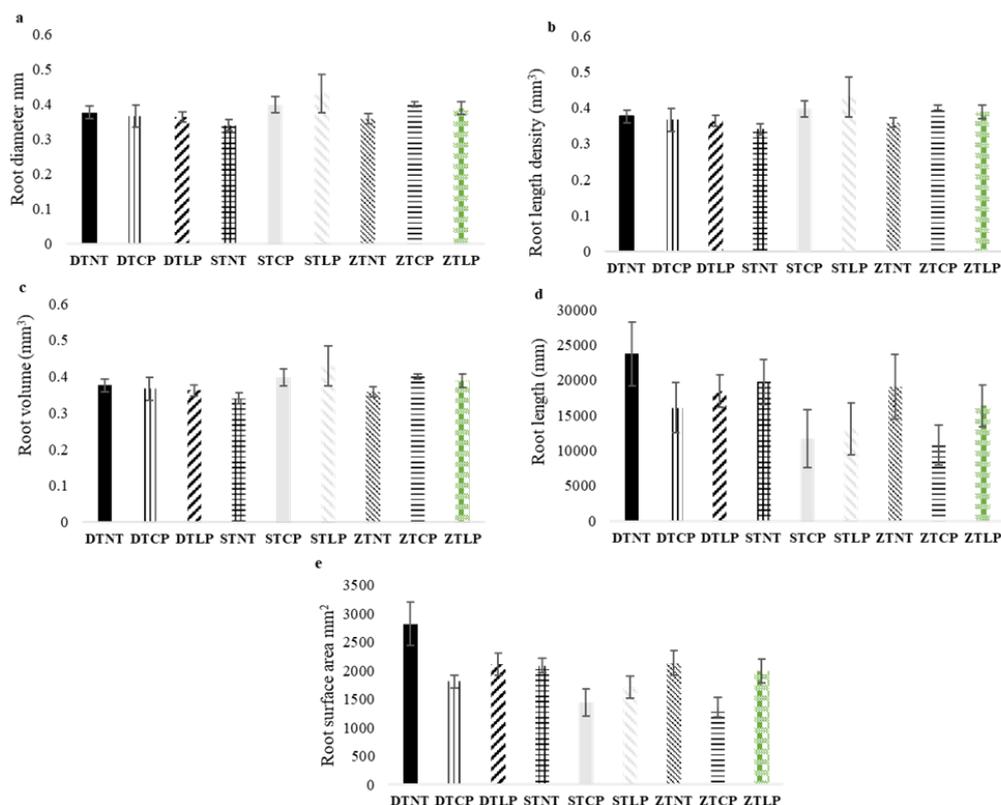
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388 **Figure 4.** Flowering growth stage 61 root system architecture using destructive root method. (a) Root diameter,  
 389 (b) Root length density (mm<sup>3</sup>), (c) Root volume (mm<sup>3</sup>), (d) root length (mm), (e) Root surface area (mm<sup>2</sup>)

390

391 3.3.2 X-ray CT root analysis results

392 Significant differences were found between trafficking treatments at GS61 for RLD and vertical root depth using  
 393 non-destructive VGStudioMax 3.2 (Table 3). The X-ray CT scans revealed significantly longer vertical rooting  
 394 (measured via the Z axis in VGStudioMax®) in ZTNT (112.7 mm) compared to DTCP (60.44 mm), DTLP (66.96  
 395 mm), STLP (65.39 mm) treatments ( $P < 0.001$ ). ZTNT showed significantly greater RLD (0.000098 mm<sup>3</sup>/m<sup>3</sup>) over  
 396 DTCP (0.000052 mm<sup>3</sup>/m<sup>3</sup>), DTLP (0.000058 mm<sup>3</sup>/m<sup>3</sup>), STLP (0.000058 mm<sup>3</sup>/m<sup>3</sup>) and ZTCP (0.000060 mm<sup>3</sup>/m<sup>3</sup>)  
 397 treatments ( $P < 0.001$ ). Root volume and surface area showed no significant difference using X-ray CT. However,  
 398 similar trends were found to the conventional WinRHIZO™ method. Trafficking had more of an influence on  
 399 rooting than tillage method which did not have any significant effect on root parameters. As RLD is an important  
 400 root trait commonly measured to estimate water uptake (White, Sylvester-Bradley and Berry, 2015), linear  
 401 regression was used to verify the relationship between root depth and RLD. A significant relationship ( $P < 0.001$ )  
 402 was found with a coefficient of determination  $R^2 = 0.54$  (Supplementary Fig. S2).

403



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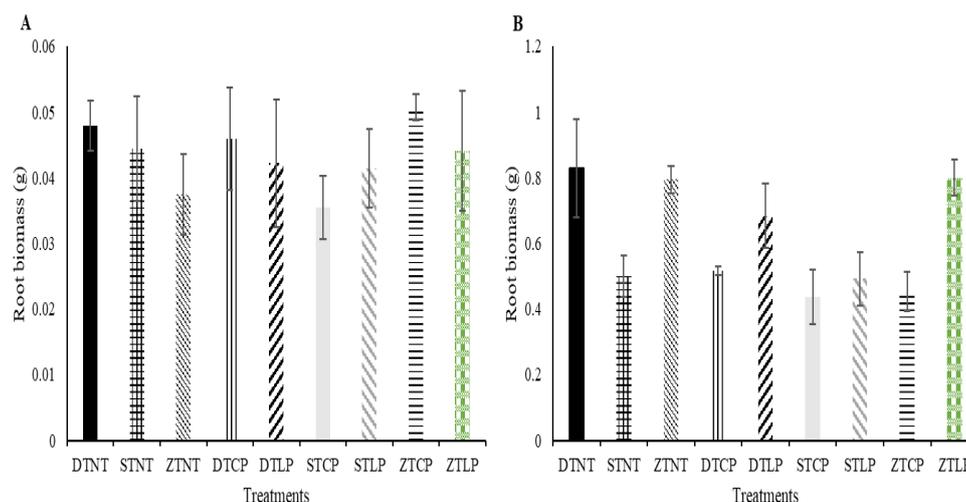
406

407 **Table 3.** Root system architecture using non-destructive method.

Tillage x traffic	Root system Architecture flowering growth stage			
	Root surface area		Length (Z) axis (mm <sup>3</sup> )	Root length density (mm/m <sup>3</sup> )
	Root volume mm <sup>3</sup>	mm <sup>2</sup>		
DTNT	3900.00	23448	96.1 ab	0.000083 ab
STNT	2648.00	17350	88.4 abc	0.000077 ab
ZTNT	3048.00	17907	112.7 a	0.000098 a
DTCP	2276.00	12114	60.44 c	0.000052 b
DTLP	3525.00	20269	66.96 bc	0.000058 b
STCP	2900.00	18052	67 abc	0.000058 ab
STLP	2358.00	14211	65.39 bc	0.000057 b
ZTCP	2533.00	15040	69.43 abc	0.000060 b
ZTLP	4480.00	25104	97.89 ab	0.000085 ab
<b>P value</b>	NS	NS	0.001	0.001

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

409 Figure 5 shows root biomass results for GS25 and GS61. No significant differences between treatments at GS25  
 410 ( $P < 0.848$ ) were found. However, root biomass was significantly different for tillage x traffic with high confidence  
 411 level ( $P < 0.001$ ) at GS61. DTNT (0.829 g) showed significantly ( $P < 0.001$ ) greater root biomass, than STCP (0.437  
 412 g) and ZTCP (0.4530 g) treatments. DTNT did not significantly differ from ZTLP (0.7992 g), ZTNT (0.7939 g),  
 413 DTLP (0.6837 g), STNT (0.4991 g) and STLP (0.4923 g). The results show that, DTNT, ZTLP and ZTNT resulted  
 414 in nearly 50% greater root biomass over STCP and ZTCP treatments. Tillage treatments (center line where there  
 415 was no traffic effect) did not differ significantly with respect to root biomass.



416

417 **Figure 5.** Root biomass at tillering (GS25) and flowering (GS61) for traffic and tillage treatments. Treatments  
418 represented by initials (Tillage: D = Deep, S = Shallow, Z = Zero), (Traffic: NT = No traffic, LP = Low pressure  
419 tyre, CP = Conventional pressure tyre).

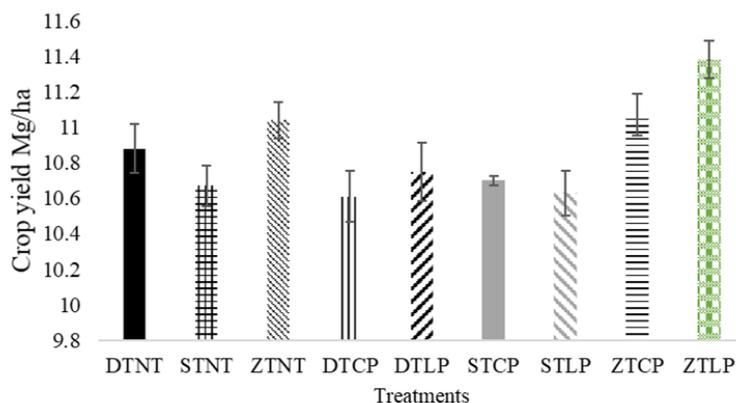
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#### 421 3.4 Crop yield

422 Crop yield was highly significant between trafficking treatments and tillage ( $P < 0.01$ ) shown in Fig. 6. ZTLP had  
423 the highest yield ( $11,385 \text{ kg ha}^{-1}$ ) and was significantly greater than DTLP ( $10,757 \text{ kg ha}^{-1}$ ), STCP ( $10,700 \text{ kg ha}^{-1}$ )  
424  $^1$ ), STNT ( $10,678 \text{ kg ha}^{-1}$ ), STLP ( $10,638 \text{ kg ha}^{-1}$ ) and DTCP ( $10,613 \text{ kg ha}^{-1}$ ). All three zero tillage treatments  
425 trended higher than deep tillage and shallow tillage treatments. ZTLP showed a  $500 \text{ kg ha}^{-1}$  yield advantage over  
426 DTNT (NS) and between  $628 - 772 \text{ kg ha}^{-1}$  over trafficked treatments and STNT with high significance. In general,  
427 this study did not show a trend in yield between conventional and low tyre pressure treatments. For deep tillage,  
428 conventional tyre pressure reduced crop yield compared to low tyre pressure by  $144 \text{ kg ha}^{-1}$  (1.34%). When  
429 compared to the no traffic sample, conventional tyre pressure consistently reduced yield by  $272 \text{ kg ha}^{-1}$  (2.5%)  
430 in deep tillage. Although not significant, trafficking trended towards improving yield by  $30 \text{ kg ha}^{-1}$  (0.03%) using  
431 conventional tyre pressure and  $340 \text{ kg ha}^{-1}$  (3.07%) using low tyre pressure. No trends were found in shallow  
432 tillage treatments. Linear regression of root depth using X-ray CT showed a significant relationship to crop yield  
433 ( $P < 0.001$ ) and positive correlation ( $r = 0.54$ ). However, the coefficient of determination was low  $R^2 = 0.3094$   
434 (Fig. S3). Moreover, regression analysis also showed a significant relationship between root biomass and crop  
435 yield ( $P < 0.01$ ). However, the correlation between the two variables was weaker ( $r = 0.43$ ) (coefficient of variance  
436  $R^2 = 0.1859$ ). This indicates that root depth is a stronger predictor of crop yield.

437

438



439

440 **Figure 6.** Crop yield in Mg/ha for traffic x tillage treatments.

441

#### 442 4. Discussion

##### 443 4.1.1 Soil physical responses to tillage & trafficking

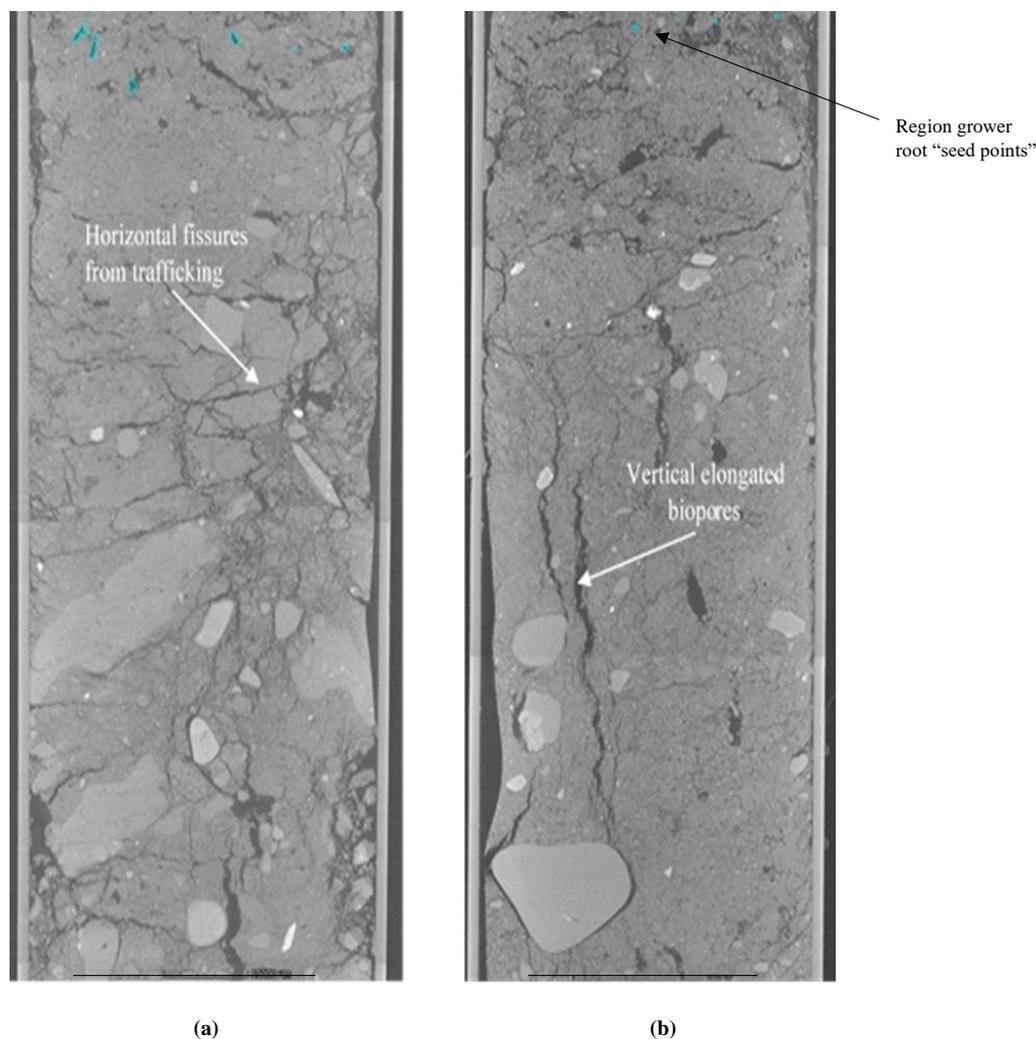
444 In line with this paper's hypothesis, trafficking effects were more influential on crop and root performance than  
445 tillage system. The presence of wheeled areas in both zero and deep cultivation treatments increased soil bulk  
446 density significantly in deep tillage treatments (Fig. 1). Previous studies have shown that zero tillage systems  
447 increase in bulk density, penetration resistance and reduce in porosity in the early years of adoption from  
448 conventional tillage systems (Christian and Ball, 1994; Six et al., 2004; Mangalassery et al., 2014a; Smith, 2016).  
449 Vogeler et al., (2009) showed that bulk density is higher under conservation tillage methods in the top 100 mm  
450 layer during the first five years of adoption from conventional systems. Indeed, Soane et al., (2012) reported that  
451 significant regeneration of soil structure requires a three-year period from tillage depending on previous historic  
452 land management practice. Moreover, values decrease in the long term with multiple benefits including improved  
453 saturated conductivity, soil organic matter and air permeability in lower soil horizons. Arvidsson, 1998 showed  
454 that soils with  $<30 \text{ g kg}^{-1}$  of organic matter were likely to suffer 11% higher crop yield loss due to compaction  
455 using uniaxial compression tests. It is plausible that the actions of soil fauna such as earthworms and old root  
456 channels could have reduced bulk density over time (Fig. 7) as identified by (Angers and Caron, 1998). Roots  
457 promote soil structural formation through increasing soil aggregation. Root mucilage production, root hair  
458 formation, and localised wetting and drying cycles encourage a reduction in soil bulk density (Bengough, 2012).

459 Our data shows similar findings with zero and deep tillage significantly reduced bulk density values in  
460 untrafficked zones. However, in trafficked treatments, high tyre pressure combined with deep tillage treatments  
461 resulted in higher bulk density values due to the loss of inherent strength by tilled soil, resulting in compression  
462 of soil particles (Raper, 2005; Soane, Godwin and Spoor, 1986). Chan et al., (2006) observed that trafficking after  
463 deep tillage increased bulk density values from  $1.27 \text{ Mg m}^{-3}$  to  $1.54 \text{ Mg m}^{-3}$ , emphasizing the effect of trafficking  
464 on the reduced bearing capacity of the deep tilled soil. The optimum soil density has been reported to differ



465 between soil types in previous studies. Indeed, Czyż, (2004) established a soil type interaction between crop yield,  
466 bulk density and root mass concluding with sandy loam soils (similar to this study) having an optimum bulk  
467 density value of 1.54-1.66 Mg m<sup>-3</sup>. Yet, in this study, root biomass was significantly reduced with treatments  
468 displaying similar soil density values to that reported optimum. Although conventional pressure tyres significantly  
469 affected zero tillage in the 100 – 200 mm layer, trafficking affected the 0 – 200 mm later under deep tillage. In  
470 shallow tillage treatments, the top 0- 100 mm layer was considerably impacted by high tyre pressure.

471



**Figure 7.** Vertical view of X-ray CT images through centre of soil core using VGStudioMax® software for (a) Shallow tillage conventional pressure (b) Zero tillage low tyre pressure. Scale bar = 50 mm.



477

478 4.1.2 *Soil porosity in response to trafficking & tillage*

479

480 Sandy soils due to their adhesive and coarse grain nature, have reduced porosity, including lower levels of  
481 micropores compared to loamy soils (Arvidsson, 1998). The aggregation potential in this sandy loam soil is low.  
482 In the presence of plants, porosity and pore connectivity as shown to reduce further compared to clay cohesive  
483 soils which tend to increase in porosity through flocculation and aggregation (Bacq-Labreuil et al., 2018). Here,  
484 we found soil porosity to be low in general across all treatments. When comparing cultivation systems, we found  
485 that shallow tillage in the 0-100 mm layer had significantly lower porosity (10.58%) compared to deep tillage  
486 (22.72%). Although zero tillage recorded low porosity values also (10.72%), it was not significantly different to  
487 the other two systems. Compared to non-trafficked treatments, trafficked soil in general caused a sharp decline in  
488 soil porosity in the top 0-100 mm layer. Tyre inflation pressure is one of the key contributors to soil stress in the  
489 100 to 1000 mm layer (Botta et al., 2008). The effect of re-compaction from trafficking after cultivation was often  
490 worse in deep tillage treatments, with a lower percentage porosity than in zero and shallow tillage (Table 2 for  
491 DTLP and DTCP treatments). In deeply cultivated soils, water infiltration rates can be reduced by up to 82% after  
492 a single wheelings (Chyba, 2012), which has agronomic implications such as reduced water and nutrient use  
493 efficiency by up to 22% thus, potentially resulting in crop yield penalties of up to 38% (Ishaq et al., 2001). Yield  
494 effects by trafficking were modest in our study due to low soil moisture conditions during sowing in autumn 2018  
495 (Met office, 2019). Dry soil has increased soil strength, reducing the effects of soil compaction as the soil load  
496 support capacity would have increased thus, increasing permissible ground pressure (Hamza and Anderson, 2005).

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

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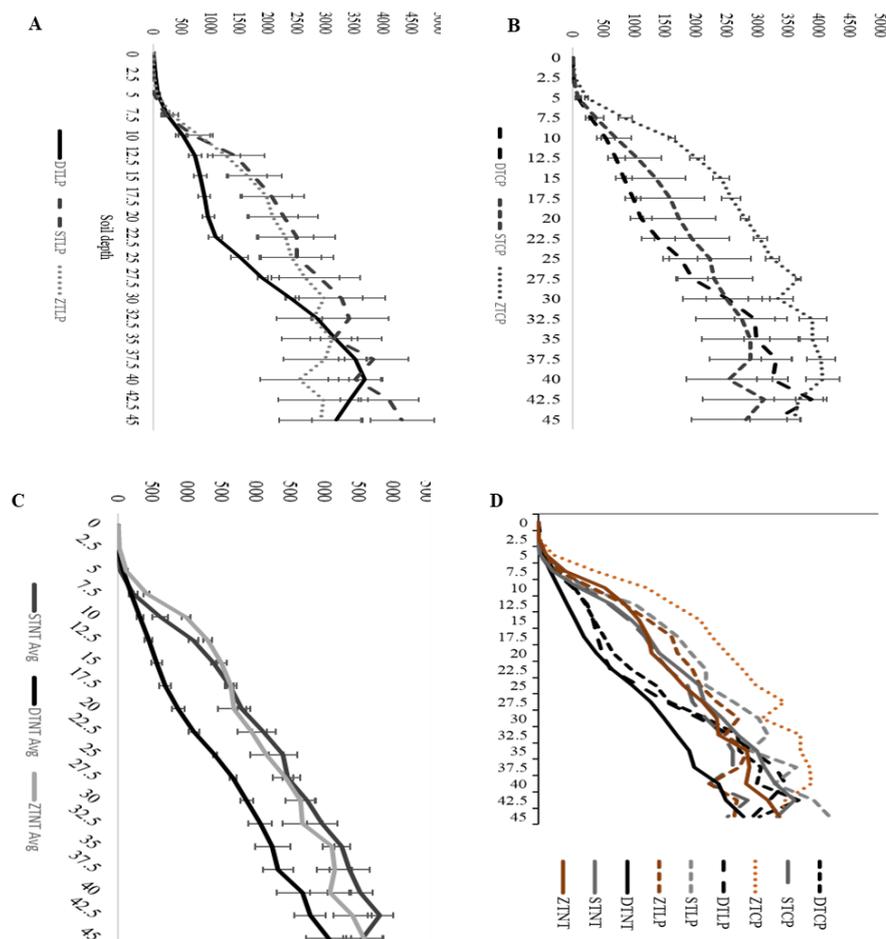
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514 4.1.3 *Penetrometer responses to tillage and traffic*

515

516 Penetrometer resistance (PR) is a useful parameter for evaluation of soil physical resistance to root growth (Otto  
517 *et al.*, 2011). In general, trafficking had a considerable influence on soil PR in this study as depicted in fig. 8. The  
518 greatest contrast in soil penetration resistance was between trafficked and un-trafficked soil with zero tillage  
519 showing the highest resistance under conventional tyre pressure. Recent studies have shown that roots can exploit  
520 pores and bypass layers of strong soil (Atkinson *et al.*, 2020). Axial pressure from repeated trafficking in ZTCP  
521 resulted in the highest PR values. However, root depth was less affected in contrast to STCP and DTCP. This  
522 might explain why roots could exploit existing pore networks in undisturbed soils compared to tillage treatments.  
523 In the middle layer examined, shallow till conventional pressure treatments suffered from a tillage pan effect  
524 shown in Fig. 7. In fact, all trafficked zero and shallow tillage systems resulted in PR values beyond 2,000 kPa, a  
525 threshold level which several studies show there is a reduction in root growth (da Silva, Kay and Perfect, 1994;  
526 Lapen *et al.*, 2004; Tormena, da Silva and Libardi, 1999). A compact zone at shallow depths is detrimental to  
527 plant growth and crop yield in rainfed temperate climates when short term droughts occur (Campbell, Reicosky  
528 and Doty, 1974).



529

530 **Figure 8.** Penetration resistance (kPa) for tillage and traffic treatments at soil depths of 0 - 450 mm. X axis depicts  
 531 soil depth. Y axis depicts Soil penetration resistance (kPa). Treatments represented by initials (Tillage: D = Deep,  
 532 S = Shallow, Z = Zero), (Traffic: NT = No traffic, LP = Low pressure tyre, CP = Conventional pressure tyre). **A**  
 533 low tyre pressure, **B** conventional tyre pressure, **C** no traffic and **D** traffic x tillage treatments combined.

534

535

536 **4.2 Root system architecture responses to tillage and traffic**

537 The ‘hidden half’ (i.e. roots) of plants are difficult to interpret in field studies (Lynch and Brown, 2001).  
 538 A large root system is characterized by large biomass, root length and root length density (Ehdaie et al., 2010;  
 539 Hamblin and Tennant, 1987). Root biomass was an important indicator of root size, showing treatment effect at  
 540 anthesis compared to the tillering stage. In general, root biomass had a positive relationship with grain yield. Zero  
 541 tillage treatments both untrafficked and trafficked at low pressure had greater root biomass over all shallow tillage



542 treatments and deep till trafficked at conventional pressure. Although deep tillage treatments with no traffic had  
543 the highest root biomass by GS61, it did not achieve the highest yield. No significant difference in root biomass  
544 was found between tillage treatments in untrafficked samples, confirming that roots are more sensitive to  
545 trafficking than tillage method. The compaction effects of trafficking on soil structure exacerbated the impact on  
546 rooting in general. Typically, studies report shallower rooting, increases in root diameter and decreased axial and  
547 lateral rooting (Grzesiak et al., 2014). Due to the high moisture deficits depicted in (Fig S1) experienced during  
548 April and May 2019, it is likely that the deeper vertical rooting in zero tillage treatments retained more moisture  
549 at depth compared to other establishment methods.

550

551 Traffic significantly affected root volume, root surface area, root length and RLD in shallow tillage  
552 treatments and zero tilled treatments trafficked at conventional pressure. RLD is an important parameter for  
553 characterizing root growth (Doussan et al., 2006) and has been used in previous studies as a key root parameter  
554 for modelling water uptake (Tinker and Nye, 2000; Javaux et al., 2013). Munos-Romero et al., (2010) and  
555 Chakraborty et al., (2008) results indicate that RLD is a positive predictor of crop yield. Although RLD had a  
556 positive correlation with crop yield in this study, root depth (using X-ray) displayed a much stronger relationship  
557 with crop yield (fig. S3). When comparing the highest root biomass (under deep tillage with no traffic) and bulk  
558 density results in the 100-200 mm layer, we found a reduction in root biomass when trafficked under conventional  
559 pressure by 28% in deep tillage under conventional pressure ( $BD = 1.66 \text{ g cm}^{-3}$ ), 37% in shallow till conventional  
560 pressure ( $1.437 \text{ g cm}^{-3}$ ) and 39% in zero tillage conventional pressure ( $1.583 \text{ g cm}^{-3}$ ) treatments. Colombi and  
561 Walter, (2017) observed decreased shoot dry weights in pot studies by 19 and 82% under moderate ( $1.45 \text{ g cm}^{-3}$ )  
562 and high ( $1.6 \text{ g cm}^{-3}$ ) soil strength conditions. In the same study root dry weight was also reduced by 36 and 87%  
563 under the same soil strength conditions. Shallow tillage had the lowest root biomass in both trafficked and  
564 untrafficked treatments. Shallow tillage treatments suffered from visible horizontal fissures or “tillage pan” in Fig  
565 10, causing significantly reduced rooting compared to deep tillage treatments. Moreover, a combination of <10%  
566 porosity and PR reaching >2,000 kPa in the 100-200 mm layer, it is likely that roots may also have suffered from  
567 anaerobic conditions due to poor infiltration rates through the tillage pan during heavy rainfall events. Conversely,  
568 root impedance may have occurred during drought periods through May and June (Batey, 2009). Alameda, Anten  
569 and Villar, (2012) proposed that axial growth suffers more than radial root growth. These effects of increased PR  
570 and soil bulk density were observed underin the current study. However, the increase in root diameter reported by  
571 several authors was not detected here (Chen et al., 2014; Lipiec et al., 2012; Tracy et al., 2012; Alameda, Anten  
572 and Villar, 2012).

573

#### 574 4.3 2D & 3D imaging for studying root-soil relationships

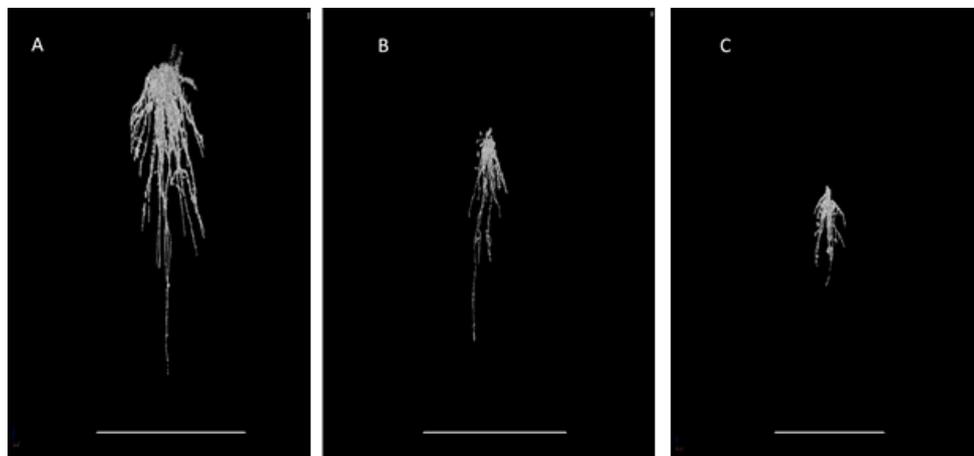
575 Due to the complexity of measuring root systems, two methods were conducted to provide comprehensive  
576 analysis. Important topology (root networks) and geometrical (physical positions) characteristics of wheat rooting  
577 using X-ray CT were found in this study. A strong significant relationship between RLD (WinRHIZO™) and root  
578 depth (X-ray CT) was found (fig. S2) validating the suitability of image analysis methods in field studies. Further,  
579 root depth showed the strongest correlation with crop yield compared to root biomass and RLD (fig. S3).



580 Moreover, the large environmental variance (low  $r$  number) in root relationships may have been caused by spatial  
581 effects reported in previous studies ( Guo et al., 2020; Zhou et al., 2021). Compared to traditional 2D  
582 WinRHIZO™ analyses, the significant difference found with *in-situ* root depth between treatments using X-ray  
583 CT was not detected by destructive WinRHIZO™ analysis (i.e., it involves the washing of soil from root material,  
584 thus losing important architectural data). Destructive root analysis showed evidence of superior rooting properties  
585 under deep tillage treatments (e.g., root length density and root volume). Visualizing important behaviors of wheat  
586 rooting in field scale trials, highlights the importance of root depth to sustain high yields in drought conditions.  
587 Figure 9 depicts significantly longer root length in zero tillage treatments compared to trafficked deep and shallow  
588 tillage, with trafficked treatments roots were generally confined to the top 0-50 mm of soil. In general, root length  
589 rarely surpassed 100 mm in depth. This was partly due to insufficient resolution available with the X-ray CT  
590 scanner to capture finer root materials (Pfeifer et al., 2015).

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

601



602

603 **Figure 9.** Root system architecture of winter wheat during anthesis for (a) Deep tillage no traffic, (b) Zero tillage  
604 low tyre pressure and (c) deep tillage conventional tyre pressure. (a) and (b) showed significantly longer root  
605 length on the primary axis compared to (c) deep tillage trafficked treatments. Scale bar = 70 mm.



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#### 612 *4.4 Traffic and tillage effects on rooting and crop yield*

613 In the present study, it was found that long term zero tillage plots under low tyre pressure increased yield by up  
614 to 0.772 Mt ha<sup>-1</sup> compared to the deep tillage conventional tyre pressure treatments. All zero tillage treatments  
615 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  
616 data collected from the X-ray CT scans showed deeper vertical rooting in zero tillage plots compared to shallow  
617 and deep tillage treatments (Fig. 9). Coupled with deeper rooting, zero tillage no traffic treatments had  
618 significantly lower bulk density than deep tillage conventional pressure plots. Munoz-Romero et al., (2010)  
619 reported a yield increase of 0.5 Mt ha<sup>-1</sup> in zero tillage compared to conventional tillage which was associated with  
620 greater water use and increased water use efficiency, similar to (Chakraborty et al., 2008). Improvements in  
621 moisture retention, soil pore structures and reduced soil compaction under zero-tillage, may also have contributed  
622 to a yield increase over conventionally tilled treatments.

623 It is possible that the lower levels of porosity found in zero tillage aided with water retention during drought  
624 periods on the highly sandy soil in this trial. Coupled with the development of vertically oriented soil structural  
625 characteristics attributed to earthworm activity and old root channels (Fig 7), the zero tillage treatments may also  
626 have had increased access to water by roots at lower soil horizons. Indeed, biopores benefit root growth by altering  
627 the surrounding chemical, physical and biological properties of soil ( Stroud et al., 2017; Banfield et al., 2017).  
628 Thus providing macropore pathways with lower mechanical resistance in which deeper rooting preferentially  
629 grow towards (Zhou et al., 2021). In contrast, deep cultivation created a porous structure which has shown to  
630 increase respiration of aerobic microorganisms, improving the flow of air and water thus increasing CO<sub>2</sub>  
631 emissions ( Mangalassery et al., 2014). Crop yield was influenced less in zero tillage treatments by trafficking  
632 than the other tillage treatments. The lower sensitivity to compaction in zero tillage is attributed to an elastic  
633 behavior or increase in bearing capacity, with soil acquiring similar structural properties to grassland soil (Ehlers  
634 and Claupein, 1994).

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640 **5 Conclusion**

641 The results from this research highlight the importance of traffic management for improving crop productivity.  
642 Physical and visual implications of soil compaction on the soil profile were demonstrated in this study, signifying  
643 the implications of tyre pressure on root growth. High tyre pressure significantly reduced root development in all  
644 tillage treatments. However, deep, and shallow tillage systems were more influenced by compaction with roots  
645 confined to the top 0-60 mm thus, reducing primary vertical rooting and inhibiting roots access to deeper soil  
646 moisture reserves. The highly significant impact on crop yield was highlighted by the strong relationship between  
647 root depth and crop yield. The visible effects of trafficking on the soil profile depicted through X-ray CT, provides  
648 evidence of the damage modern farm machinery can cause for root resource capture, leading to potential increased  
649 drought stress and yield loss in crop production. This long-term trial site has shown that zero tillage does not affect  
650 root growth, in fact, reduced bulk density, improved grain yield and rooting depth significantly through deeply  
651 connected vertical soil pore fissures created by earthworms and old root channels. These findings suggest that  
652 scientists and farmers should focus on designing improved zero tillage cropping systems, managing field  
653 trafficking protocols. Furthermore, this research shows that the combination of X-ray CT scanning along with  
654 traditional destructive methods provide a robust method for assessing in field rooting for future crop breeding  
655 initiatives and soil management practice. This research concludes that little differences were found between deep  
656 tillage and zero tillage methods in the absence of traffic in terms of overall physical root growth. However, in  
657 abundance of biopores and increased soil bearing capacity to withstand machinery traffic in in zero tillage systems  
658 increased rooting depth and moisture retention during the growing season.

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662

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

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

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