# ORIGINAL ARTICLE



# Functional root trait-based classification of cover crops to improve soil physical properties

Csilla Hudek<sup>1</sup> | Cristinel Putinica<sup>1</sup> | Wilfred Otten<sup>1</sup> | Sarah De Baets<sup>2</sup>

<sup>1</sup>School of Water, Energy and Environment, Cranfield Soil and Agrifood Institute, Cranfield University, Cranfield, UK

<sup>2</sup>Research Coordination Office, KU Leuven, Leuven, Belgium

#### Correspondence

Csilla Hudek, School of Water, Energy and Environment, Cranfield Soil and Agrifood Institute, Cranfield University, College Road, Cranfield, MK43 0AL, Bedfordshire, UK. Email: csilla.hudek@cranfield.ac.uk

#### Abstract

Cover crop use is a well-established soil conservation technique and has been proven effective for erosion control and soil remediation in many arable systems. Whereas the obvious protection mechanism of cover crops occurs through the canopy, plant roots perform multiple functions. It is important to consider the soil functions delivered by different root systems in order to increase the uptake of cover crops for sustainable soil and water management. A classification of cover crop root systems up to 0.6 m deep based on functional traits will allow us to better study their potential role in soil bio-engineering, soil structural improvements for hydrological services and soil resource protection. This was a glasshouse experiment, using large 1-m<sup>3</sup> containers filled with loam soil, loose topsoil and compacted subsoil, in which seven cover crop species (oat, rye, buckwheat, vetch, radish, mustard, phacelia) were grown for 90 days. Root cores were taken at the end of the experiment, washed and imaged to determine root traits (total root length density, average root diameter, root specific length and root surface area) for both the topsoil and subsoil layers. Root identity was determined from a distinctive combination of single root traits and related to three soil functional variables, representing soil structural improvement, runoff mitigation and erosion control. The results showed that total root length and root surface area correlate well with aggregate stability and soil macroporosity. Buckwheat, mustard and rye had significantly greater aggregate stability, as well as 10, 8 and 7% greater microporosity, respectively, at the interface with the compacted layer when compared to the control bare soil. Furthermore, average root diameter negatively correlated with soil macroporosity, indicating that cover crops with a fine root system are more beneficial for creating pore-space than those with thicker taproots. Selecting cover crop species with the right root traits is therefore crucial to improve soil health.

#### Highlights

- · Roots of cover crops are a largely unexplored frontier for bio-engineering of agricultural soils.
- · Combinations of root traits were identified that most improve soil characteristics.

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- Cover crops with finer root systems were better at enhancing porosity.
- Root length and surface area were most important for enhancing soil structure.

#### KEYWORDS

aggregate stability, cover crops, erosion mitigation, root morphology, soil compaction, soil conservation

# **1** | INTRODUCTION

Agriculture is often accountable for soil erosion, water pollution and land degradation. One-third of the earth's soils are suffering from degradation, and much of this is due to a decline in the physical soil structure. Goal 15 of the UN's Sustainable Development Goals is specifically addressing these problems. In the UK, topsoil loss is estimated to reach 2.2 million tonnes per year, affecting nearly 50% of all arable lands (Pimentel & Burgess, 2013). Quantifiable soil degradation costs range between £0.9 bn and £1.4 bn per year, with a central estimate of £1.2 bn, mainly linked to loss of organic content of soils (47% of total cost), compaction (39%) and erosion (12%) (Graves et al., 2015).

Devising strategies to prevent land degradation are urgently needed to preserve the essential ecosystem services provided by soils. This can be done by ensuring the soil surface is covered, to increase soil strength (to resist erosion) and improve soil structure (for hydrological services and optimal crop growth). Soils have been manipulated for many years to reduce soil compaction and optimize crop growth, to improve drainage, to provide slope stability and to manage nutrients, pests and contaminants. In modern agriculture, this is mainly carried out using machinery or engineered amendments such as fertilizers. These options often have a limited effect and are costly and not sustainable, so alternatives need to be sought.

Plant roots are important improvers of soil structure, enhancing aggregate formation and stability (Haynes & Beare, 1997; Kavdir & Smucker, 2005) and improving soil cohesion (de Baets, Torri, Poesen, Salvador, & Meersmans, 2008). Root-induced macropores are of particular importance for runoff mitigation due to their large diameters and high connectivity, enhancing rapid rainfall infiltration and percolation to deeper soil layers (Cresswell & Kirkegaard, 1995; Ghestem, Sidle, & Stokes, 2011), and improving soil aeration. The mechanisms by which this engineering occurs include the physical penetration of the soil matrix by roots through vertical and lateral expansion (physical creation of macropores, also called bio-drilling) and the subsequent triggering of microbial activity arising from rhizodeposition of root exudates, cells and debris, which serve as energy sources for

the rhizosphere microbiota. These biota improve the properties of the soil through adhesion, kinetic restructuring and filamentous binding (Miransari, 2014). In turn, the resulting soil structure subsequently promotes future root growth, creating a sustainable positive feedback loop.

In arable systems, increasing the capacity of soils to resist erosion and receive, retain and release water through structural rejuvenation is most feasible via the use of appropriate cover crops in the rotation as a best management practice. Cover crops are fast-growing annuals or perennials which, planted sequentially between two cash crops, have the ability to boost soil health and reduce the negative impact of agromanagement on the environment. They are usually planted immediately after harvest (Abdalla et al., 2019). Left to grow all winter, they cover and protect the soil surface against erosion and die off or are removed (mechanically or chemically) in early spring to make way for the cash crop.

There are a number of studies on the effect of specific cover crop species, as recently reviewed by Blanco-Canqui and Ruis (2020); examples include their impact on tackling soil compaction (Chen & Weil, 2010; Williams & Weil, 2004), improving infiltration (Kahimba, Sri Ranjan, Froese, Entz, & Nason, 2008; Yu et al., 2016), enhancing aggregate stability (Liu, Ma, & Bomke, 2005; Walsh, MacKenzie, Salmins, & Buszard, 1996) and reducing erosion (de Baets, Poesen, Meersmans, & Serlet, 2011; Kasper, Radke, & Laflen, 2001). However, there is not much focus on the effect of the traits of their root systems on soil physical properties. Research shows that soils planted with fit-for-purpose roots are better adapted to disturbances such as nutrient or water shortage (Barkaoui, Roumet, & Volaire, 2016) or soil erosion (Yu et al., 2016). Cover crops are able to create important macropores in the soil by shifting the soil during the growth of their taproots (e.g., mustard), or by granulation of soil particles into aggregates in terms of sod-forming plants (e.g., ryegrass) (McGourty & Reganold, 2005). Despite its importance for plant productivity, the study of cover crop species' root systems at multiple depths is a largely unexplored frontier in bio-engineering of agricultural soils. Root responses to the combination of soil physical stresses (e.g., mechanical impedance and water stress) depend on the communication and coordination

of the different regions of the same root system (Bengough et al., 2006). No research has yet focused on matching root traits that are beneficial for soil functions, such as soil physical stability and water infiltration capacity, even though simultaneous consideration of more than one trait is required to understand adaptation and functioning (e.g. root response to soil compaction (Bengough et al., 2006). Bacq-Labreuil, Crawford, Mooney, Neal, and Ritz (2019) showed that the diversity of root morphology and interactions between roots and soil biota impact soil structural formation and dynamics. As root morphology can have different effects upon soil structure, the choice of cover crop with specific root traits can have both practical and ecological implications. It remains, however, unclear how combinations of root traits affect multiple soil properties such as infiltration, aggregate stability, porosity and compaction. Therefore, to increase the uptake of cover cropping for soil bioengineering in agriculture, a classification of cover crop root systems based on functional traits is needed to study their potential for soil structural improvements, hydrological services and soil resource protection. Such knowledge of root-soil interactions would allow for screening of the right cover crop species for a specific problem.

The objectives of this study are therefore (a) to define the root system morphology of seven typical cover crops grown in loam soil vulnerable to soil degradation, (b) to use a set of experimental assays to measure the potential of these cover crops for improving multiple soil functions related to erosion control, runoff mitigation and soil structure and (c) to identify root traits that link to erosion control, runoff mitigation and improved soil structure.

# 2 | MATERIALS AND METHODS

#### 2.1 | Experimental set-up

A greenhouse experiment was carried out, growing seven different cover crop species as monocultures in large 1 m × 1.2 m × 0.8 m containers (S1). The 24 plant containers were placed in a randomized block design, including three replicates per species and three control bare soils, without plants. The soil was purchased from a specialized topsoil supplier (Boughton loam, UK) and sieved through a 4-mm sieve. Soil was characterized as having a loam soil texture (52% sand, 20% clay, 28% silt) following the USDA soil classification system, containing 0.17% of total nitrogen, 48 mg kg<sup>-1</sup>of total phosphorus and 237 mg kg<sup>-1</sup>of potassium. The bottom 50 cm of the containers were filled with the soil compacted at a bulk density of 1.5 g cm<sup>-3</sup> and the top 30 cm of soil was loosely packed at a bulk density of 1.2 g cm<sup>-3</sup>. Each of the Soil Science—WILEY

24 containers was placed on a weighing system with a capacity of 4,000 kg and accuracy of 100 g (Gravicon -C3-4000; Transicon Ltd, Shropshire, UK). The balances are connected to a control computer, and weights were recorded sequentially at 10-s intervals. A control system (LemnaTec) was programmed to top up the water content four times a day through a series of soaker hoses up to a reference gravimetric soil water content of 0.18 g  $g^{-1}$ . Air temperature was continuously monitored. Moisture sensors (SM150T, Delta T) were installed into each container at 60 and 70 cm depth and recorded moisture content hourly. In addition, soil moisture profile probes with an HH2 reader were used (Delta-T) to measure at 10, 20, 30 and 40 cm from the surface, weekly. The cover crops were sown in rows in June 2018 and grown for 90 days. Table 1 lists the cover crop species used, their seeding densities (ranging from 10 to 45 kg  $ha^{-1}$ ) and their plant densities (ranging from 12 to 144 plant  $m^{-2}$ ). The selected seeding rates correspond with the recommended lowest seeding densities used in agriculture.

# 2.2 | Plant phenotyping, sampling and analysis

After 90 days of growth, samples were collected to measure and calculate the following above- and belowground plant traits: aboveground biomass (AB;  $g m^{-2}$ ), root biomass (RB;  $g m^{-3}$ ), root length density (RLD;  $cm cm^{-3}$ ) root diameter (D; mm), specific root length (SRL; m  $g^{-1}$ ), root surface area (RSA; m<sup>2</sup>), deep root length fraction (DRLF; -), calculated as the ratio of total length of roots in the deep soil (30-60 cm) over total root length in the entire profile, and root-to-shoot ratio (R:S) calculated as ratio of total root mass and total shoot biomass. The root traits were determined from cored samples with a volume of 754 cm<sup>3</sup> collected at the following depths: 0–15 cm, 15-30 cm, 30-45 cm and 45-60 cm. Four replicated root cores were taken from each container using a bipartite Eijkelkamp root auger (80 mm in diameter). Roots were separated from the soil using the wet sieving method (Smit et al., 2000) and a 500-µm sieve. Washed roots were scanned with a flatbed scanner and the images were analysed with WinRHIZO 2018a software. The scanned roots and the harvested aboveground plant material were oven-dried at 70°C for 48 h and weighed with an analytical scale.

# 2.3 | Soil tests

All soil samples were taken prior to sampling for root traits. Post-harvest soil samples were taken at 10 cm

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TABLE 1 The seven cover crop species, their common names, ID, seeding densities and number of plants per m<sup>2</sup>

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Species	Family	Common name	ID	Seeding density (kg $ha^{-1}$ )	Plant density (plants m <sup>-2</sup> )
Avene sativa L	Poaceae	Oat	OT	25	41
Secale cereale L.	Poaceae	Rye	RY	40	30
Fagopyrum esculentum Moench	Polygonaceae	Buckwheat	BW	45	35
Vicia villosa L.	Fabaceae	Vetch	VC	25	12
Raphanus sativus L.	Brassicaceae	Tillage radish	RD	12	15
Sinapsis alba L.	Brassicaceae	White mustard	MS	10	33
Phacelia tanacetifolia Benth	Boraginaceae	Phacelia	PH	12	144

(topsoil) and 30 cm (subsoil) depth, air dried, mixed and sieved to 2 and 0.5 mm.

The following soil physical properties were determined post-harvest (Table 2): bulk density samples were taken at 15, 30 and 50-cm depth using rings of 5 cm in diameter. Penetration resistance (MPa) was measured with a penetrologger (Eijkelkamp Penetrologger SN, cone  $1.2 \text{ cm}^2$  and  $30^\circ$ , speed 2 cm s<sup>-1</sup>) recording resistance up to 80-cm depth in 1-cm increments with three replicated profiles per box.

The following soil tests to quantify soil functioning regarding runoff mitigation, erosion control and soil structural improvements were performed post-harvest: topsoil macropore infiltration rate, topsoil aggregate stability and soil porosity. Water infiltration rate (cm  $h^{-1}$ ) was measured using single ring infiltrometers (Burt & Soil Survey Staff, 2014). Core samples were collected with the bipartite root auger from the topsoil layer (0-15 cm)to test and calculate the effects of the roots of the cover crops on soil aggregate stability by using a modified wet sieving method (Frei, 2009) with a sieve size of 20 mm. In this method a value of 1 means that roots hold all soil in place and a value of 0 means that the root system does not help in holding the soil together, meaning that all soil is eroded away. Soil rings (PVC, 3 cm in diameter, 3 cm height) were extracted for determination of soil porosity at 15, 30 and 50-cm soil depth. These soil samples were scanned with Phoenix V|TOME|X M 240 high-resolution X-ray computer tomography (CT) (GE Sensing and Inspection Technologies, Wunstorf, Germany), with a resolution of 28 µm, exposure time of 200 ms, number of projections of 2,998, voltage of 170 kV and a current of 150 µA. Each scan was reconstructed using DatosRec software (GE Sensing and Inspection Technologies, Wunstorf, Germany) then manually combined in VG Studio MAX v2.2 (Volume Graphics GmbH, Heidelberg, Germany). Samples were thresholded to separate solid and pores using in-house developed software (Hapca, Houston, Otten, & Baveye, 2013) and geometric properties of the pore space were calculated following Houston et al. (2017).

# 2.4 | Statistical analysis

Differences in soil physical and chemical properties, as well as in plant traits between the different species and the bare control soil, were evaluated using one-way ANOVA and to identify the differences the Tukey HSD test was applied. Penetration resistance data were analysed using repeated measures ANOVA. Principal component analysis was performed with all root traits measured, to identify which root traits cluster and to synthetize the root information for each species. Species root traits (SRL, RSA, D and DRLF) were summarized in a common principal component analysis (PCA) retaining two principal components. The PCA coordinates of each species on the two main principal axes (PC1 and PC2) were used to calculate root identity. The PC1 and PC2 components are further called root factor 1 and root factor 2, respectively. Root identity was calculated for each species as the mean of species PC1 (or PC2) coordinates. The objective was to classify the root systems of the species grown in monocultures using their root traits only. The mean PCA coordinates of each species on the two main principal axes (i.e., root identity) were then used to find correlations with the soil variables: topsoil aggregate stability, infiltration rate and soil porosity. This was done by performing linear or non-linear regressions using standard deviation on the mean values as a WLS estimator. For porosity, the effect of the compacted layer and the interaction of compaction with the PC1 and PC2 values were tested as well. All statistical tests were carried out using the statistical computing software SPSS (IBM SPSS Statistics 26).

### 3 | RESULTS

#### 3.1 | Soil parameters

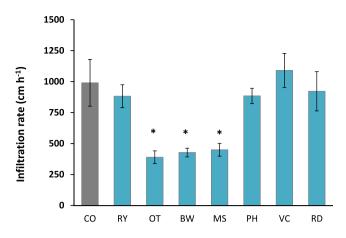
No significant difference was found in bulk density among the species at any of the three soil depths (15–20, 30–35 and 50–55 cm) but in all three depths, the treatments with cover crops had less bulk density compared to the control bare

TABLE 2 N	Number of soil and plant samples taken per soil layer	plant samples t	aken per soil lay	ver for the different soil tests or plant traits per container	l tests or plant traits	s per containe	er				
	Soil					Plant					
Test	water infiltration (saturated hydraulic Penetratio conductivity) resistance (K; cm h <sup>-1</sup> ) (MPa cm <sup>-1</sup>	Penetration resistance (MPa cm <sup>-1</sup> )	Bulk density Soil (g cm <sup>-3</sup> ) ma	Root-induced Soil aggregate macroporosity (%) stability (g g <sup>-1</sup> )	Root-induced aggregate stability (g g <sup>-1</sup> )	Root biomass (g m <sup>-3</sup> )	Root length Root density diam (cm cm <sup>-3</sup> ) (mm)	Root diameter (mm)	Root Specific surface root len area $(m^{-2})$ $(m g^{-1})$	gth	Deep root- length fraction
Method used	Single ring infiltrometer	Penetrologger	Core method Image based (X-ray CT and thresholdii	Image based (X-ray CT and thresholding)	Modified wet sieving method	Core + oven dry	Core + WinRHIZO	Core + WinRHIZO			
Soil layer (cm) 00	00	00-80	15-20 30-35 50-55	15-20 30-35 50-55	00-15	0–15 15–30 30–45 45–60	0–15 15–30 30–45 45–60	0–15 15–30 30–45 45–60	0-15 15-30 30-45 45-60	0–15 15–30 30–45 45–60	30-45 45-60
N° of samples/ 3 layer/ container	/ 3	ω	£	2	1	ε	ŝ	ω	£		6
Total number of replicates/ species/soil layer	0	6	6	٩	ς	6	6	6	6	6	6
Note: Each species	Note: Each species treatment had three replicate containers. CT, computer tomography.	eplicate containe	ers. CT, computer	tomography.							

soil treatment  $(1.34 \pm 0.1, 1.58 \pm 0.05, 1.6 \pm 0.03 \text{ g cm}^{-3})$ . In the top 15–20 cm, buckwheat had the least and rye and radish the greatest bulk density  $(1.23 \pm 0.04 \text{ and } 1.31 \pm 0.08 \text{ g cm}^{-3})$ , respectively). At the interface with the compacted subsoil (30–35 cm) mustard and vetch had the least and rye, radish and phacelia the greatest mean values  $(1.43 \pm 0.04 \text{ and } 1.49 \pm 0.11 \text{ g cm}^{-3})$ , respectively). In the sub-50–55 cm, radish gave the least bulk density values and mustard the greatest  $(1.4 \pm 0.03 \text{ and } 1.55 \pm 0.07 \text{ g cm}^{-3})$ , respectively).

The soil moisture content of the different treatments is presented in S2. There was no significant difference in the water content of the different treatments in the top 0-30 cm. As the topsoil moisture content did not vary between treatments the infiltration measurements could not be affected by this. At 40 cm, significantly less water content (p < 0.02) was measured for the bare control soil compared to phacelia, vetch, mustard, radish and buckwheat, as well as for oat (p < 0.05) compared to phacelia and buckwheat, and for rye (p < 0.03) compared to phacelia. At 60 cm, vetch had significantly greater water content (p < 0.01) compared to phacelia, radish, oat, buckwheat and the control bare soil treatments and significantly lower values were measured for the control bare soil compared (p < 0.03) to rye and mustard. At the bottom of the container only the control bare soil had significantly less (p < 0.02) water content compared to all other cover crop treatments.

Penetration resistance at the end of the experiment showed less soil strength for all species in the top 0-25 cm (from  $0.1 \pm 0.03$  to  $0.9 \pm 0.3$  MPa), similarly to the control bare soil (0.1–0.8 MPa). At the interface with the compacted subsoil, the mean penetration resistance values significantly increased (p < 0.05) for all species treatments  $(2.2 \pm 0.4 \text{ MPa})$  compared to the control bare soil ( $1.0 \pm 0.1$  MPa), and reached the greatest mean value of  $3.1 \pm 0.3$  MPa at 50-cm depth, compared to 1.7  $\pm$  0.2 MPa for the control bare soil. At the top 0–25 cm vetch, phacelia and buckwheat had greater penetration resistance values (p < 0.01) compared to all other treatments, and rye and radish when compared to oat and the control bare soil. At 25-50 cm, mustard and the bare soil had less penetration resistance (p < 0.001) compared to all other treatments, as well as buckwheat, with the exception of phacelia. Additionally, vetch also showed greater penetration resistance (p < 0.0001) when compared to phacelia. At the subsoil (50-80 cm), radish had greater penetration resistance (p < 0.001) and the control bare soil less penetration resistance (p < 0.0001) compared to the other treatments. Oat and vetch also showed significantly greater penetration resistance (p < 0.0001)when they were compared to rye, buckwheat and mustard.



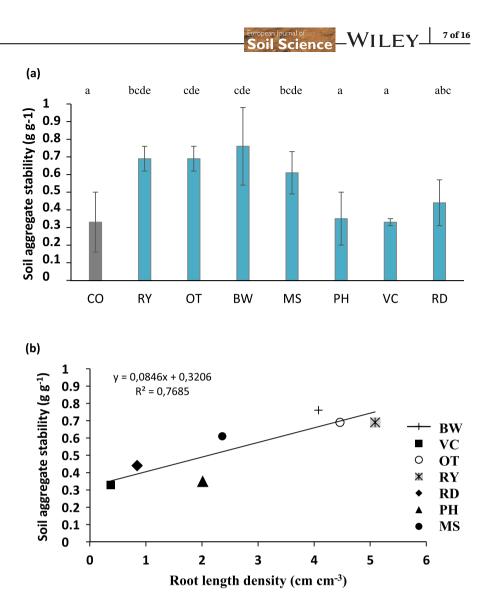
**FIGURE 1** Infiltration rate (cm  $h^{-1}$ ) of the seven cover crop species (buckwheat (BW), vetch (VC), oat (OT), rye (RY), radish (RD), phacelia (PH), mustard (MS)) and control bare soil (CO) measured with a single ring infiltrometer. Error bars indicate the variability of the data and \* indicates significant differences

There was a significant difference between the way cover crops affected the infiltration of water. The infiltration rates ranged between  $389 \pm 51$  and  $1,090 \pm 137$  cm h<sup>-1</sup> for the different cover crop treatments, compared to 990  $\pm$  188 cm h<sup>-1</sup> for the control bare soil. Significantly slower infiltration rates (p < 0.05) were obtained for buckwheat, mustard and oat when compared to vetch, radish, phacelia, rye and the control bare soil (Figure 1).

The greatest increase of topsoil aggregate stability provided by the roots was for soil permeated with buckwheat roots (0.76 g g<sup>-1</sup>) and the least value was for vetch (0.33 g g<sup>-1</sup>). Aggregates in the topsoil (0–15 cm) for mustard, rye, oat and buckwheat were more stable than those for vetch, phacelia, radish and the control bare soil (Figure 2a). Aggregate stability increased with root length density ( $r^2 = 0.75$ ) (Figure 2b).

Cover crops had only an insignificant effect (p < 0.24) on the detectable macroporosity (16.1-21.1%) as measured by X-ray CT on the top 15-cm soil layer. However, buckwheat  $(16.8 \pm 0.8\%)$ , rye  $(13.4 \pm 2.1\%)$  and mustard (14.5) $\pm$  2.1%) increased the soil's macroporosity compared to the control bare soil  $(6.5 \pm 3.6\%)$  at 30-cm soil depth (p < 0.05). At 50 cm (Figure 3), radish  $(16.2 \pm 2.6\%)$ increased soil porosity (p < 0.004) compared to the control bare soil ( $8.0 \pm 3.8\%$ ) and phacelia ( $7.3 \pm 3.1\%$ ), and mus- $(13.9 \pm 2.2\%)$ had greater porosities tard than phacelia (p < 0.02).

Mustard had a greater (p < 0.01) pore surface area in all three soil layers compared to the control bare soil, and at 15 and 30 cm compared to oat (p < 0.04). At 30 cm, vetch, radish, buckwheat and rye showed greater (p < 0.042) results compared to the control bare soil. At the subsoil layer (50 cm), buckwheat and **FIGURE 2** (a) Root-induced topsoil aggregate stability (0–15 cm) of the seven cover crops (vetch (VC), phacelia (PH), radish (RD), mustard (MS), rye (RY), oat (OT) and buckwheat (BW)) and the control bare soil (CO) and (b) the linear relationship between aggregate stability and root length density. Lowercase letters indicate significant differences between the treatments



the control bare soil had less pore surface area (p < 0.035) when compared to radish, and phacelia had less pore surface area when compared to mustard and radish (p < 0.001).

# 3.2 | Plant traits

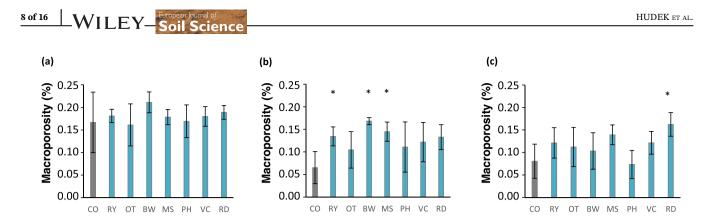
# 3.2.1 | Root architecture

When fully excavated at the individual plant level, the root systems of the studied cover crops show an architecture as displayed in Figure 4. With the exceptions of vetch, all the studied species showed a well-developed, extensive root system. Both cereals (oat and rye), being monocots, developed a fibrous root system with a bulk mass in the top 20–30-cm soil layers that gradually decreased and reached a maximum rooting depth of 40–50 cm. The remaining species, being dicots, had a tap root system penetrating to deeper layers (40–80 cm). Mustard and buckwheat developed strong, well-branched lateral roots, arising at all depths. Near the soil surface, phacelia's fine, net-like lateral roots branched out. On the other hand, vetch had scarcely developed lateral roots, with a shallow tap root system reaching no deeper than 30 cm. The radish tuber tap root penetrated through the entire soil profile, reaching the bottom of the containers (80 cm). Its short, profuse lateral roots run spirally downward all the way to the tip.

# 3.2.2 | Plant functional traits

The aboveground biomass substantially varied among the studied species. Mustard had a significantly (p < 0.01) greater aboveground biomass compared to all other species, and the aboveground biomass of radish was greater than that of vetch (p < 0.01).

The highest root biomass was found in the top 0-15 cm, and with increasing soil depth root biomass



**FIGURE 3** Detectable soil macroporosity (resolution >28 µm) of the soil with cover crop species (vetch (VC), radish (RD), oat (OT), buckwheat (BW), phacelia (PH), rye (RY), mustard (MS)), and the control bare soil (CO) as measured by X-ray computer tomography (CT) on samples collected at (a) 15, (b) 30 and (c) 50 cm soil depth. \* Indicates significant differences compared to bare soil

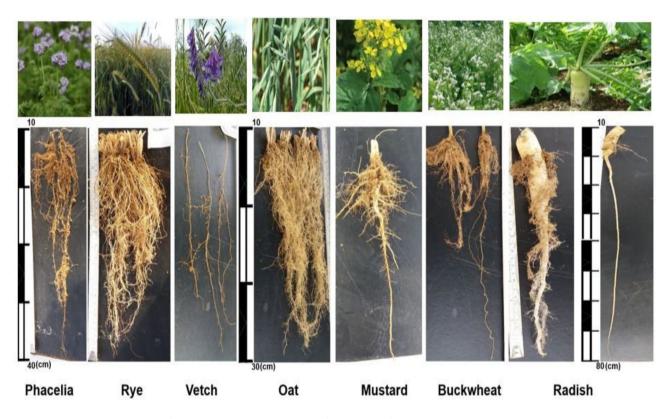


FIGURE 4 Root architecture of the studied cover crop species after 90 days of growth

decreased. A significantly higher (p < 0.01) total root biomass was measured for radish and mustard compared to buckwheat, vetch, oat, rye and phacelia. Due to their thick taproot, mustard and radish had the highest root biomass throughout the soil profile (0–60 cm) and vetch the lowest.

A significantly higher root-to-shoot ratio was measured for radish compared to buckwheat, phacelia and vetch, as well as for rye compared to phacelia (p < 0.05). Root length density (RLD) significantly varied among the different cover crop species in the top 0–15-cm soil layer (p < 0.05) (Table 3). At 15–30 cm oat had significantly greater (p < 0.05) RLD compared to all other studied species and at 30–45 cm the density was greater than that for phacelia, buckwheat, vetch and radish (p < 0.01). At 45–60-cm soil depth, only mustard showed a greater RLD value when compared to phacelia (p < 0.05).

Eighty percent of the root systems of all studied species had very fine roots (i.e., < 0.5 mm in diameter), both

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**TABLE 3** Plant trait results of the seven cover crop species at the topsoil (0–15 cm and 15–30 cm) and subsoil (30–45 cm and 45–60 cm) layers

	DIV	VC	07	DV	<b>ND</b>	DII	MG
	BW	VC	ОТ	RY	RD	PH	MS
0–15-cm depth							
$AB (g m^{-2})$	7,463 ± 2,406	784 ± 363	$6,100 \pm 4,190$	$2,373 \pm 695$	8,711 ± 2,611	5,134 ± 2,777	$16,587 \pm 1755$
$RB (g m^{-3})$	$0.92 \pm 0.49$	$0.04\pm0.01$	$1.02\pm084$	$0.75\pm0.22$	$2.72 \pm 2.04$	$0.24 \pm 0.09$	$3.13 \pm 1.47$
R:S	$0.029 \pm 0.003$	$0.034 \pm 0.007$	$0.042 \pm 0.006$	$0.081 \pm 0.01$	$0.097 \pm 0.03$	$0.017 \pm 0.005$	$0.041 \pm 0.002$
D (mm)	$0.46 \pm 0.27$	$0.42 \pm 0.06$	$0.41\pm0.04$	$0.36\pm0.07$	$107.00 \pm 36.34$	$0.28 \pm 0.03$	$14.09 \pm 15.25$
$RLD (cm cm^{-3})$	$4.09 \pm 1.49$	$0.37 \pm 0.08$	$4.45 \pm 2.17$	$5.08 \pm 1.45$	$0.84 \pm 0.50$	$2.06 \pm 0.72$	$2.65 \pm 1.47$
$SRL (m g^{-1})$	$44.3 \pm 14.3$	$62.5 \pm 13.8$	38.7 ± 9.8	$54.8 \pm 22.2$	$2.5 \pm 1.8$	79.9 ± 45.5	$6.5 \pm 3.0$
$RSA(m^2)$	338.7 ± 123	$36.1 \pm 6.1$	$411.1 \pm 200.3$	404 ± 93.9	$113.4 \pm 183$	$120.7 \pm 54.5$	$240.5\pm118.7$
15–30-cm depth							
$RB (g m^{-3})$	$0.03 \pm 0.02$	$0.02\pm0.01$	$0.07 \pm 0.22$	$0.04 \pm 0.02$	$0.16\pm0.09$	$0.03 \pm 0.01$	$0.04\pm0.02$
D (mm)	$0.26 \pm 0.05$	$0.39 \pm 0.05$	$0.25 \pm 0.03$	$0.25 \pm 0.03$	4.93 ± 12.87	$0.29 \pm 0.05$	$0.26 \pm 0.05$
$RLD (cm cm^{-3})$	$0.57 \pm 0.41$	$0.17 \pm 0.09$	$2.04 \pm 1.91$	$0.87 \pm 0.43$	$0.22 \pm 0.14$	$0.44 \pm 0.29$	$0.75 \pm 0.35$
SRL (m $g^{-1}$ )	108.4 ± 34.8	69.7 <u>+</u> 18.1	204.1 ± 135.4	142.3 ± 31.5	$10.3 \pm 7.9$	96.0 ± 34.1	$128.9 \pm 60.6$
$RSA(m^2)$	32.6 ± 19.4	$16.1 \pm 8.9$	124.7 ± 127.4	52.3 ± 26.0	$28.7 \pm 14.6$	27.6 ± 14.7	42.6 ± 14.9
30–45-cm depth							
$RB (g m^{-2})$	$0.01 \pm 0$	$0.01 \pm 0.01$	$0.03 \pm 0.03$	$0.02\pm0.01$	$0.07 \pm 0.05$	$0.01 \pm 0$	$0.06 \pm 0.06$
D (mm)	$0.28 \pm 0.04$	$0.37 \pm 0.05$	$0.28 \pm 0.05$	$0.30 \pm 0.04$	$0.61 \pm 0.32$	$0.31 \pm 0.04$	$0.27 \pm 0.06$
RLD (cm cm $^{-3}$ )	$0.22 \pm 0.1$	$0.16 \pm 0.06$	$0.73 \pm 0.66$	$0.41 \pm 0.32$	$0.12 \pm 0.07$	$0.15 \pm 0.08$	$0.47 \pm 0.21$
SRL (m $g^{-1}$ )	105.6 ± 31.8	115.5 ± 52.0	159.9 <u>+</u> 69.8	159.9 ± 42.2	26.3 ± 35.5	104.4 ± 45.7	$116.1 \pm 48.8$
$RSA(m^2)$	$15.0 \pm 6.7$	$14.0 \pm 6.6$	43.9 ± 31.9	28.9 ± 18.1	16.7 ± 9.5	11.7 ± 6.6	32.3 ± 17.1
45–60-cm depth							
$RB (g m^{-2})$	$0.02 \pm 0.02$	$0.02 \pm 0.02$	$0.04 \pm 0.05$	$0.01 \pm 0$	$0.05 \pm 0.03$	$0.01 \pm 0.01$	$0.03 \pm 0.02$
D (mm)	$0.29 \pm 0.03$	$0.37 \pm 0.07$	$0.36 \pm 0.09$	$0.30 \pm 0.07$	$0.47 \pm 0.09$	$0.27 \pm 0.05$	$0.26 \pm 0.02$
RLD (cm cm $^{-3}$ )	$0.29 \pm 0.22$	$0.16 \pm 0.1$	$0.26 \pm 0.21$	$0.23 \pm 0.15$	$0.14 \pm 0.06$	$0.12 \pm 0.1$	$0.39 \pm 0.21$
SRL (m $g^{-1}$ )	136.7 ± 48.6	132.4 ± 68.1	151.1 ± 55.5	141.7 ± 45.3	$35.3 \pm 23.0$	142.2 ± 94.4	$124.2 \pm 58.0$
$RSA(m^2)$	$21.0 \pm 17.6$	$14.1 \pm 10.4$	19.9 ± 14.0	$16.2 \pm 8.9$	$15.9 \pm 7.8$	$8.3 \pm 8.2$	$24.0 \pm 8.2$
DRMF	$0.10 \pm 0.04$	$0.35 \pm 0.06$	$0.13 \pm 0.06$	$0.10 \pm 0.03$	$0.24 \pm 0.16$	$0.08 \pm 0.02$	$0.22 \pm 0.11$

*Note:* Mustard (MS), phacelia (PH), radish (RD), rye (RY), oat (OT), vetch (VC) and buckwheat (BW). Values are averages and standard error out of nine replicates. Aboveground biomass (AB; g), root biomass (RB; g  $m^{-2}$ ), root diameter (D; mm), root length density (RLD; m  $m^{-2}$ ), specific root length (SRL; m  $g^{-1}$ ), root surface area (RSA;  $m^2$ ), deep root length fraction (DRLF, -) and root-to-shoot ratio (R:S), calculated as the ratio of total root mass and total shoot mass.

in the topsoil and subsoil layers. Species with taproots (buckwheat, radish, phacelia and mustard) had thicker roots (i.e., > 2 mm in diameter), but these only made up 1 to 5% of roots in the top (0–30 cm), and 7% (radish) in the subsoil layers (30–60 cm) (Figure 5).

The root surface area (RSA) results showed that in the top 0–15 cm of soil, oat, rye and buckwheat had significantly greater values compared to vetch, radish and phacelia (p < 0.001), as well as mustard when compared to vetch (p < 0.05). In the 15–30-cm soil layer, oat had greater RSA compared to buckwheat, mustard, phacelia, radish and vetch (p < 0.05). At 30–45 cm depth, oat had greater (p < 0.01) RSA compared to buckwheat, phacelia, radish and vetch, and at 45–60 cm, there were no significant differences in RSA between species.

The mean specific root length (SRL) among the studied species ranged from  $2.5 \pm 1.8 \text{ m g}^{-1}$  (radish in top 0–15 cm) to  $204.1 \pm 135.4 \text{ m g}^{-1}$  (oat at 15–30 cm). In the top 0–15 cm, the SRL values of radish and mustard were significantly less compared to buckwheat, vetch, oat, rye and phacelia (p < 0.05), and phacelia had a significantly greater topsoil SRL compared to buckwheat, oat, radish and mustard (p < 0.05). At 15–30 cm, oat had greater SRL compared to buckwheat, vetch, and phacelia (p < 0.05), and at 30–60 cm, radish and phacelia (p < 0.05).

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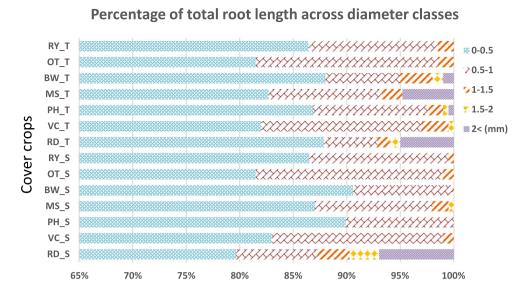


FIGURE 5 Root length distribution in different diameter classes of cover crop species measured in the topsoil (0– 30 cm) and subsoil (30–60 cm) layers of subsamples (mustard (MS), phacelia (PH), radish (RD), rye (RY), oat (OT), vetch (VC), buckwheat (BW) top soil layer (\_T), subsoil layer (\_S))

We found significantly greater deep root length fraction (DRLF, -) for radish when compared to buckwheat, rye and phacelia (p < 0.05) and for vetch when compared to buckwheat, oat, rye and phacelia (p < 0.001). These results show that radish and vetch distributed more of their root lengths in the deeper soil layers than the other cover crops.

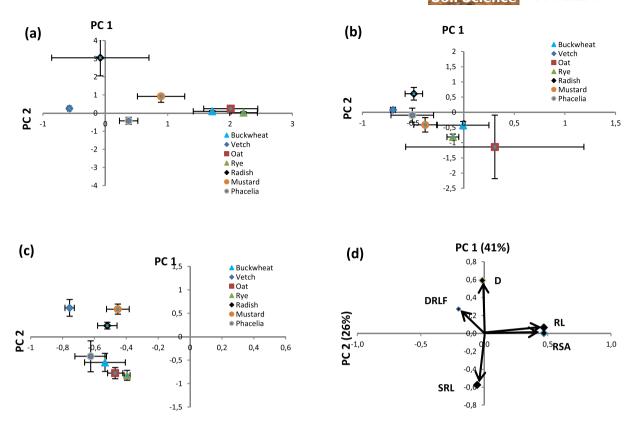
# 3.3 | Principal component analysis

To summarize all the root information, a PCA was performed. Two principal components are retained and explain 67% of the variance in the root data (Figure 6). PC 1, or root factor 1, is strongly correlated with root length density (0.967) and with root surface area (0.967). PC 2, or root factor 2, is negatively correlated with specific root length (-0.757) and positively with diameter (0.781). Table 4 presents the correlations of all root variables with PC 1 and 2, respectively. The scores of all root variables on PC 1 and PC 2 axes are shown in Figure 6d.

PC 1 and PC 2 coordinates were averaged per species and soil layer and plotted in Figure 6 to visualize the root identity profiles for all the studied species. Error bars indicate standard errors on the average species values. Figure 6a indicates that both cereals (oat and rye) and buckwheat have a very similar root profile with high root lengths and high root surface area values. The two Brassicaceae species (mustard and radish) are characterized by the highest PC 2 values, indicating they have the thickest mean root diameters. The legume vetch, showing negative PC 1 and positive PC 2 values (Figure 6a), is characterized by very few, thin roots, but has a high deep root length fraction value. Phacelia has few roots but has one of the highest root specific length values, indicating many long fine roots per unit mass. Figure 6b indicates that at the interface of the loose topsoil with the compacted subsoil, root profiles start to overlap more. Total root length drops significantly with increasing soil depth for most species. Oat, however, seems to have the highest PC 1 scores, indicating greater root lengths at this depth, whereas radish still has the greatest average root diameter at this depth (15–30 cm) (Figure 6b). In the compacted subsoil the two cereals, buckwheat and phacelia, have the greatest specific root length values, whereas radish, mustard and vetch have the greatest deep root length fraction values (Figure 6c).

#### 3.4 | Root traits predicting soil functions

The PC 1 root factor has a significant effect on rootinduced topsoil aggregate stability (Table 5), but there is no significant effect of the root factors on infiltration rate (Table 5). Both PC1 and PC2 had a significant effect on the macroporosity of the topsoil (Table 5). PC 1 correlates positively with macroporosity, whereas PC 2 correlates negatively with soil pore space, indicating that total root length and root surface area increase soil macroporosity, whereas soil macroporosity decreases with increasing mean diameter and a reduced specific root length. For the interface soil layer (15-30 cm) and the deep subsoil (45-60 cm) separately, no significant correlations were found between soil macroporosity and root factors. However, there is a significant effect of the interaction between root factor 1 (which correlates with total root length and root surface area) and the compaction level (corresponding to a mean bulk density of  $1.2 \text{ g cm}^{-3}$  for topsoil and 1.5 g cm<sup>-3</sup> for subsoil) on soil macroporosity (Table 6).



**FIGURE 6** (a) Root identity profiles in loose topsoil (0–15 cm), (b) at the interface with the compaction layer (15–30 cm) and (c) in compacted subsoil (45–60 cm), expressed as their average coordinates  $\pm$  standard error (*SE*) on the two main principal component analysis (PCA) components, for the seven studied cover crops species grown as monocultures. (d) Scores of five root traits (root length (RL, m), specific root length (SRL, m g<sup>-1</sup>), root surface area (RSA, m<sup>2</sup>), mean root diameter (D, mm) and deep root-mass fraction (DRMF, –)) on two main PCA factors

<b>TABLE 4</b> Loading of root traits (specific root length (SRL;
m g <sup>-1</sup> ), root length (RL; m), root diameter (D; mm), root surface
area (RSA; $m^2$ ) and deep root length fraction (DRLF, $-$ )) on the
two PCA components

	PC 1	PC 2
SRL	-0.114	<b>-0.757</b>
RL	0.967	0.003
D	-0.029	0.781
RSA	0.967	0.086
DRLF	-0.415	0.359

Note: Values in bold are correlations with PC1 and 2 higher than 0.7.

# 4 | DISCUSSION

The integration of cover crops in land management is a well-recognized practice in conservation agriculture (Lal, 2015). However, increasing the uptake of cover cropping in conventional agriculture, where soil degradation is a common problem, still needs to be encouraged. Our work and results provide information on how the root system of the studied cover crops plays an important role **TABLE 5** The *p*-values indicating the significance of PC1 and PC 2 root factors for the soil variables topsoil (0–15 cm) aggregate stability, topsoil macroporosity (%) (resolution 28  $\mu$ m) and infiltration rate (mm h<sup>-1</sup>)

	Topsoil aggregate stability	Infiltration rate	Topsoil macroporosity
PC1	0.003	0.092	0.034
PC2	0.588	0.353	0.001

in improving the physical quality of the soil and preventing soil degradation, which should support farmers in decision making. The differences in root morphology among the different cover crop species highlighted the functional differences they provide in mitigating soil degradation. To assess the studied cover crops' potential in improving soil functions associated with erosion control and runoff mitigation, we measured the most informative root traits (root biomass, root-to-shoot ratio, root length density, root diameter, root surface area, specific root length and deep root length fraction) and linked these to specific soil quality indicators (compaction, porosity, infiltration and aggregate stability).

**TABLE 6** Impact of the compacted layer and the root factors on soil macroporosity (resolution  $28 \mu m$ )

	Macroporosity
PC1	0.002
PC2	0.063
Compacted layer	0.025
PC1*compacted layer	0.012
PC2*compacted layer	0.703

Bulk density and penetration resistance are important indicators of soil compaction which have direct implications on runoff and soil erosion (Nawaz, Bourrié, & Trolard, 2013). A number of studies highlight the effect of bulk density and penetration resistance on the development of the root system (Bécel, Vercambre, & Pagès, 2011), often linking compaction to restricted root growth (Tardieu, 1994). However, plants respond differently in root development when tackling compaction (Grzesiak, Grzesiak, Hura, Marcinska, & Rzepka, 2013). Burr-Hersey, Mooney, Bengough, Mairhofer, and Ritz (2017) found three different development responses in cover crops to compaction. In their study, tillage radish and vetch showed altered root growth, while the root system of black oat did not show any deviation penetrating through the compacted layer. In our experiment we also hypothesized that some cover crop species would be better adapted to grow in the compacted soil, whereas others would not be able to produce a significant amount of root biomass in the compacted soil after 90 days of plant growth. We could not detect significant differences in bulk density but the penetration resistance and the results from the root surface area values indicate the species with the most penetrative roots. Our findings show that oat had the greatest root surface area values in the top 45 cm of soil (i.e., a significantly greater root soil surface contact compared to buckwheat, phacelia, radish and vetch). Mustard and rye also grew lots of fine roots in the compacted soil, which indicates that among the studied species, oat, mustard and rye overcame compaction significantly better than other species, which would most likely result in having better access to nutrients and water. This finding for oat is in line with Burr-Hersey et al. (2017), who observed the same undisturbed root development in their study. In terms of different tillage systems, Materechera and Mloza-Banda (1987) found a negative correlation between root length density and penetration resistance both in the conventional and minimum tillage systems. Root response to compacted layers could also be linked to the result of root system architecture or soil depth. Correa, Postma, Watt, and Wojciechowski (2019) highlighted the influence of increased compaction on the decrease of total root length. Root plasticity allows the plant to overcome unfavourable soil conditions and allocate roots to layers where nutrient and water availability is more favourable.

Root traits such as the deep root length fraction provide information on the distribution of root length to the deeper soil layers (30-60 cm). Our study found that radish and vetch had a significantly greater deep root length fraction compared to the other cover crops. In general, most roots are present in the upper soil layers and with increasing soil depth root presence decreases. All of our studied cover crops follow this pattern, but in the case of radish and vetch the decrease with depth is significantly less, resulting in greater deep root length fraction results. However, the total root length density of these species in the 30-45-cm layer is low (Table 3) compared to oat, rye and mustard. Additionally, radish's storage organ penetrates through the entire soil profile, suggesting an increased soil aeration and water infiltration in the deeper soil layers, but further testing needs to be carried out to verify this. However, being able to produce a significant mass of roots in compacted soil does not mean that these roots affect soil structure in a positive way. Several studies show that roots with increased diameter can penetrate better through compacted soil and are able to alleviate soil compaction (Correa et al., 2019; Materechera, Alston, Kirby, & Dexter, 1992). On the other hand, increased root diameter could increase soil densification (Kolb, Legué, & Bogeat-Triboulot, 2017), negatively impacting on porosity, which decreases the water conductivity and water holding capacity of the soil (Tubeileh, Groleau-Renaud, Plantureux, & Guckert, 2003).

Bodner, Leitner, and Kaul (2014) found that greater root density significantly increased the micropore volume of cover crops but in the meantime reduces the volume of larger pores. We observed that species with either greater root length density or greater root surface area, such as buckwheat, rye and mustard, show significantly greater soil macroporosity at 30 cm (Figure 3b). In our result, root factor 1, correlated with total root length and root surface area, had a significant effect on soil macroporosity. Oat, however, did not significantly increase soil macroporosity at 30 cm compared to bare soil, even though it had the greatest root length density. This could be explained by the fact that fine oat roots are more likely to fill up the existing soil pores during their growth and an increase in pore space was therefore not detected at the time of scanning. Hao, Wei, Cao, Guo, and Shi (2020) also reported increased effects on porosity from grass species. Our study shows that not only grass species and cereals, in our case rye, are capable of increasing pore space, but species with a greater root length density and root surface area, such as

mustard and buckwheat, can also increase soil porosity in compacted soil layers.

The greatest aggregate stability of the soil permeated by roots, was measured for buckwheat, followed by the two cereals, oat and rye. In general, a high number of fine roots can provide greater aggregate stability (Erktan et al., 2016); however, our study cannot provide clear evidence for it because 80% of the roots of all studied species had very fine roots (<0.5 mm in diameter). It has been reported by a number of authors (e.g., Hudek, Stanchi, D'Amico, & Freppaz, 2017; Vergani & Graf, 2015) that greater root length density results in an increased aggregate stability. Our results support this observation; with increasing root length density, the aggregate stability is increased, with the exception of buckwheat. The greatest aggregate stability was measured on buckwheat samples; however, the greatest root length density results were measured for the two cereals (oat and rye). The difference was not significant. Many complex processes can contribute to aggregate stability. Root-induced soil aggregate stability not only works through its physical reinforcement, but other mechanisms are likely to contribute to it (e.g., the chemical properties of the root exudates and the interactions between these exudates and the soil biota). The role of exudates was beyond the scope of the present study and warrants further investigation. Decomposed roots and root exudates are chemical and biological binding agents (Tisdall & Oades, 1982). The rate of root turnover and the composition of root exudates can have significant effects on aggregate stability, which can explain our results.

Plant and ecosystem functioning responses to environmental changes have been shown to be indicated by changes in specific root length values (Wright et al., 2004). In general, species with a resourceacquisitive strategy show greater specific root length and lower root dry matter values and plants with a resource-conservation strategy show lower specific root length and greater root dry matter values (Wright et al., 2004). Our results support these findings and show that mustard and radish, with their significantly lower specific root length and high root biomass values, have a resource-conservation strategy. The significantly greater aboveground biomass values for mustard and radish also reinforce this theory. At the interface (30-45-cm soil layer) oat and rye had greater specific root length and root length density values, as well as greater root biomass values. This indicates that the cereals were capable of investing more into producing roots in the compacted layer, with the purpose of gaining access to an abundant water and nutrient supply. These roots possibly have a shorter root lifespan (Pérez-Harguindeguy et al., 2013).

The role the different root systems play in improving soil functions related to soil structural improvements, erosion control and runoff mitigation is based on increasing the resistance of the soil by modifying its hydrological and mechanical properties (Gray & Sotir, 1996). Some of the chosen experimental assays were directly able to link the changes to specific cover crop species and root systems; others did this with less certainty. The effects of root functional identity on soil porosity and soil aggregate stability were clear, with root length density and root surface area affecting soil porosity and soil aggregate stability in a positive way. Root identity, however, did not affect infiltration rate. The infiltration assay provided direct information on the changes in the hydrological properties of the soil after 90 days of growth. To be able to link root identity to the changes caused in infiltration capacity, the time of sampling and measurements was probably wrong as the measurements should have been performed after the decay of the root system. Storr et al. (2020) did not find a significant difference in yearly soil moisture variability between cover crop fields and their corresponding bare control fields, indicating that differences in soil moisture, infiltration rate and evapotranspiration between cover crop and bare control plots are small and probably need to be measured with more precise equipment such as Microtensiometer probes.

# 5 | CONCLUSION

Our study provides evidence, by comparing the functional traits of seven cover crops and linking them to erosion control, runoff mitigation and soil structural improvement, of the benefits of using cover crops in agriculture.

The results indicate that species with a similar root identity in the topsoil, showing greater root length and root surface area, such as oat, rye and buckwheat, can significantly increase topsoil (0-15 cm) aggregate stability and soil porosity in compacted soil at 30-cm depth. Also, mustard was effective at increasing aggregate stability in the topsoil and creating soil pores at 30 cm, which can also be linked to its greater fine root length in these layers. Deeper in the compacted subsoil, at 50-cm depth, only the species mustard and radish produced enough roots to impact on soil porosity compared to the unplanted soil. Interestingly, this study showed that it is the fine roots of these species that are effective at creating pore space. The roots of the studied cover crops did not have an effect on infiltration rates. However, our topsoil was loosely packed, and it will require further investigation if an absence of an effect would be detected in case a higher bulk density was present at the surface.

Cover cropping is a successful soil conservation technique, but it has limitations. It can only be effective if it is recognized as part of a well-planned, integrated farming system. There are a number of factors (e.g., environmental conditions, soil degradation status, cash crop type, method of tillage) that should be taken into consideration before building cover crops into the farming system to sustain both the ecological and economic benefits of cover cropping. The present work provided a new "toolbox" for both farmers and soil conservationists by presenting correlations between root traits and soil physical properties showing how root traits can help alleviate soil compaction or increase aggregate stability. The monoculture results provide a valuable starting point for the selection and combination of different cover crop mixtures. They can inform further studies on how different mixtures of cover crops and their root systems can affect and enhance multiple soil functions. This would, however, also require the development of tools to distinguish between the different cover crops' root biomass in a multispecies mixture.

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#### **CONFLICT OF INTEREST**

The authors have no conflicting interests that might be perceived to influence the results and/or discussion reported in this paper.

#### AUTHOR CONTRIBUTIONS

Study concept and design: Sarah De Baets and Csilla Hudek. Data collection and laboratory analysis: Cristinel Putinica, Csilla Hudek and Sarah De Baets. Analysis and interpretation of data: Csilla Hudek and Sarah De Baets. Drafting of the manuscript: Csilla Hudek, Sarah De Baets and Wilfred Otten. Statistical analysis: Csilla Hudek and Sarah De Baets. Obtained funding: Sarah De Baets. Review and editing: Csilla Hudek, Sarah De Baets, Wilfred Otten and Cristinel Putinica.

#### DATA AVAILABILITY STATEMENT

The underlying data can be accessed through https://doi. org/10.17862/cranfield.rd.13573907 [Correction added on 17 August 2021, after first online publication: The Data Availability Statement has been updated in this version.]

#### ORCID

Csilla Hudek b https://orcid.org/0000-0002-7527-8862

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#### SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

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# Functional root trait based classification of cover crops to improve soil physical properties

Hudek, Csilla

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