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Reducing crop disease risk through residue management*

Jacqueline Stroud[1], Dasuni Jayaweera[2] and Rumiana Ray[2]

[1]SRUC, Craibstone Estate, Aberdeen, AB21 9YA

[2]Division of Plant and Crop Sciences, School of Biosciences, University of Nottingham

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CONTENTS

1. Abstract

The ambitions of the sector for net zero include the sustainable management of pests, weeds and diseases. The latest science has informed a business case for the effective biocontrol of the plant pathogen *Fusarium culmorum* and the mycotoxin, deoxynivalenol, by *Lumbricus terrestris* earthworms. The estimated biocontrol value is £59 ha^{-1} through reducing tillage (fuel) and fungicide inputs.

This project demonstrated the potential for applying Artificial Intelligence (AI) to process images of the soil surface to detect *Lumbricus terrestris* middens. *Lumbricus terrestris* earthworms live in a permanent vertical burrow, emerging at night to collect plant residues from a 30 cm radius of their burrow entrance to form the midden (food resource and burrow microclimate control). Middens are biodegradation hotspots which can cover up to 30 % of the soil surface. Practically, small robots collect image data for plant assessments, incidentally collecting soil surface information. This created an opportunity to process this type of image towards the integration of soil biology and plant health information at field scales.

Research attention has been directed towards residue-dwelling crop pathogens causing Fusarium head blight (*Fusarium* spp.). This is the first study to include *Zymoseptoria tritici*, *Oculimacula acuformis* and *O. yallundae*, and field collected plant residues (wheat, OSR and maize) containing mixtures of pathogenic fungi. Both the field and laboratory conditions identified that pathogen-infected materials were an attractive food source for *Lumbricus terrestris* earthworms which rapidly removed infected materials from the soil surface. However, there were indications of fitness penalties in terms of worm survival with straw containing *F. graminearium* and *Oculimacula* spp. Further research is needed to determine the agroecological interactions between worm activity and individual pathogens in wheat. Two major gaps in understanding were identified as scientific barriers for biocontrol implementation: (1) pathogen dispersal risks from earthworm activities, and (2) linking earthworm activities and fitness to plant disease incidence.

We used an audience response system at a farming conference (111 people) to inform development needs for biocontrol strategies. Earthworm ecology knowledge was poor, but 90 % people routinely monitor or observe earthworms, and an opportunity to develop an experiential learning activity in October 2022 was identified. Midden mapping activities were well received, and there was potential for co-development (providing images to accelerate AI development). Information seeking behaviours were characterised as browsing style, and preferences were towards animated social media posts, in a format for mobile phone viewing, disseminated via Twitter, by the active scientist. This highlights that network building between scientists-farmers is an important component of change management in agriculture.

2. Introduction

Lumbricus terrestris earthworms are tillage sensitive, midden building earthworms that selectively feed on plant residues at the soil surface. These plant residues are heavily colonised by fungi. Earthworms prefer feeding on pioneer saprotrophic fungi and pathogenic fungi, probably linked to their cellulolytic activities, meaning the capacity to breakdown cellulose (plant cell walls) (Bonkowski et al., 2000). The abundance of these fungi may be indicators of food quality to earthworms, signalling that organic matter is at an early stage of decomposition (Bonkowski et al., 2000).

The trajectory of agricultural soil management practices is a reduction in tillage intensity for reasons including reducing fuel use (costs of crop establishment), reducing soil erosion risks and conservation of biotic activities. However, by decreasing soil tillage, farmers are faced with increased risks to food production caused by infestations of phytopathogenic fungi. For example, *Fusarium graminearum* can survive as a saprotroph in crop residues, and serves as a primary inoculum impacting host plants when conditions facilitate perithecium formation, fruiting bodies discharging ascospores (Abid et al., 2021). Hence, farmers and consultants are focussing on intrinsic biocontrol options to prevent and control infections to keep plants and soils healthy (van Capelle et al., 2021).

Research into *Lumbricus terrestris* activities is a frontier of biology linked to the potential biocontrol of phytopathogenic fungi. These earthworms have been shown to use *Fusarium*-infected residue as a food source, thus promoting the degradation of associated mycotoxins, and are abundant in unploughed arable fields (van Capelle et al., 2021). It has been estimated that the value of *Lumbricus terrestris* for *Fusarium culmorum* reduction is £59 per hectare in fuel and pesticide savings (Plaas et al., 2019).

We identified three priorities to support the net zero ambitions of the agricultural sector: (1) strategies to map *Lumbricus terrestris* middens abundance at field scales, (2) measure *Lumbricus terrestris* feeding behaviours towards additional residue-dwelling crop pathogens causing Eyespot (*Oculimacula* spp.) or Septoria tritici blotch (*Zymoseptoria tritici*), field straw expected to be densely populated with mixtures of pathogenic fungi including *Fusarium* spp, and (3) assess ecological literacy and social learning network communications to enhance the accessibility of frontier research.

The 3 objectives/KPIs for this research project were (1st January 2022 – 23rd March 2022):
1) Analyse soil surface images to detect earthworm midden abundance
2) Rank breakdown efficiency of plant residues infected (by *Fusarium* spp., *Oculimacula* spp. and *Zymoseptoria tritici*) by *Lumbricus terrestris* earthworms
3) Identify pathway(s) for easy access to research for farmers and growers

3. Materials and methods

3.1. Field surface residue cover and pathogen populations

We worked with the AHDB knowledge exchange team to connect to farmers and growers using reduced tillage soil management practices. No personal identifying information was requested or recorded (i.e. participation was anonymous) and farms in Buckinghamshire, Gloucestershire, Wiltshire and Nottinghamshire were visited. The sampling strategy required: the current crop to be winter wheat, earthworm middens to be present, and the previous crop (returned residues) could be wheat, oilseed rape or maize only. From the 16 potential fields, 7 fields were suitable and used to collect photographic midden data and surface straw for laboratory analyses. The photographic image data (360 images) were used for the object detection model. The field straw was dried, and analysed in the laboratory for presence and quantity of targeted pathogens using real-time qPCR assays as previously described (Brown et al., 2020, Ajigboye et al., 2021).

3.2. Object detection model to classify middens in images

Two types of data image sets were collected to develop the object detection model. The first set of training images of middens were taken at varying sizes, angles and distance to the ground. This set was used to develop manual identification skills (image annotator). The images were annotated using CVAT, an annotation tool creating a bounding box surrounding the worm middens located in an image. The annotated images were then used to train the Artificial Intelligence (AI) model. The AI was designed to locate where in an image the worm midden is and representations in that image that allow it to classify worm middens. The second set of training images were collected using a uniform image size, angle and distance to the ground (matching the soil surface image data collected by small robots) from one field in Nottinghamshire. Images were collected using a digital camera pointing face down from a 1.5 m height over a 10 x 10 m transect. These images were collected in pairs, the second image contained coloured pegs as midden markers to aid annotation. The training set for the model comprised of all images from set 1 and 50 % of images from set 2 (260 images). The validation test was run on 50 % of images from set 2 (49 images).

The model was trained using the RetinaNet framework, which is used to deal with small objects in an image. Object detection provides two outputs: one is the localisation of the midden in the image, and the other output is classifying the object as a midden or not. The mean average precision (mAP) metric was used to evaluate the performance of the model.

Intersection over Union (IoU) calculation to describe how well the object was located in the image (Figure 1).

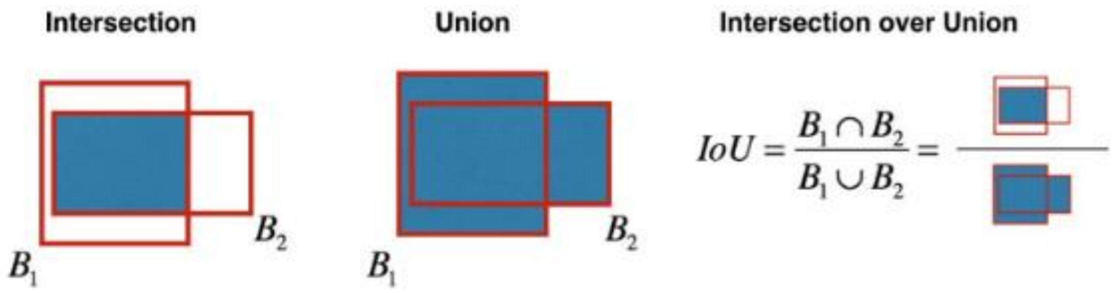

Figure 1: Description of Intersection of Union calculations. Image credit: Small Robot Company.

Classification:
True Positive: A midden is correctly identified as a midden,
False Positive: An object which is not a midden is incorrectly identified as a midden,
True Negative: An object which is not a midden is correctly not identified as a midden
False Negative: A midden is incorrectly not identified as a midden.
Precision: How many of the results are relevant?
True Positives / (True Positives + False Positives)
Recall: How many of the relevant results are correct?
True Positive / (True Positive + False Negative)

The precision and recall of the model are calculated using the validation test set over a range of different Intersection over Union (IoU) thresholds to plot a graph of precision vs recall. The resulting area is the mean average precision (mAP) or model performance.

3.3. Straw biodegradation efficiency by earthworms

3.3.1. Laboratory infected straw

Sterilised straw (20g) was inoculated with fungal pathogens *Fusarium graminearum* (100ml of spore concentration of 10^6 ml^{-1}), 10 mycelial (5mm diameter) plugs of *Oculimacula acuformis* or *O. yallundae,* and *Zymoseptoria tritici* (100ml of spore concentration of 10^6 ml^{-1}) (Brown et al., 2021, Ajigboye et al., 2021). Inoculated straw was incubated for 2 weeks at room temperature (~20ºC) prior to use. Wheat plant leaves exhibiting Septoria tritici blotch (STB) disease symptoms (lesions and pycnidia) were also used for this experiment. The food choice arena design has been previously described (Doube et al., 1997; Bonkowski et al., 2000). Briefly, the arena was an aluminium plate with four equally spaced, black microfuge tubes penetrating the rim of the plates at the bottom. All the plant material was cut to the length of the microfuge tube (2.5 cm deep) and positioned at random within each plate. Each plate was lined with three layers of moist filter paper to prevent earthworm desiccation and was closed with a cardboard lid. One *Lumbricus terrestris* was added to a chamber (n = 40) at 6pm and straw preference was determined at 9am the following morning. The experiment was conducted in a polytunnel with an overnight temperature of 6 – 9 °C. Choice was measured by plant piece(s) moved from the microfuge tube into the arena. This was used to inform a ranking preference for pathogen infected plant residues.

These results were explored further using a binary choice chamber experiment (Carolina™ Large choice chamber). Each choice chamber was filled with topsoil, with one side containing surface straw inoculated with *Fusarium graminearum* and the other side containing material inoculated with *Oculimacula acuformis*. One *Lumbricus terrestris* earthworm was added to the chamber pathway at 6pm and preference was determined at 9am the following morning. Choice was measured as the side of the chamber the earthworm was located. Student's t-test was used to compare treatment effects.

The surface straw biodegradation experiment design has been previously described (Oldenburg et al., 2008). The *Lumbricus terrestris* earthworms were purchased from wormsdirect.co.uk and incubated in topsoil for 1 week prior to use. A 1 litre tub was filled with 700 g topsoil (pH 7.3, organic matter (LOI) 6.09 %, sandy texture) and moisture content was adjusted to 20 %. A starter burrow in the centre was made using a 5 mm diameter rod and three clitellate individuals (3 – 4 g) were added to each microcosm. The following day, 4 g crop residue was applied to the soil surface including controls: uninfected straw and no earthworms, uninfected straw. Pathogen treatments included straw infected with *Fusarium graminearum (Fusarium g)*, *Oculimacula acuformis* (Eyespot R) or *O. yallundae* (Eyespot W), *Zymoseptoria tritici* (Septoria straw), wheat plant leaves exhibiting Septoria tritici blotch symptoms (Septoria leaves) and control straws from natural infection in fields Malin and C6. The experiment was conducted in a controlled environment room (15 – 17 °C), and each of the replicates were arranged as a randomized factorial block design (n = 4). Photographs of the surface were collected at 0 d and 7 days. Image processing to determine surface area of straw was performed using ImageJ 1.44 (http://rsbweb.nih.gov/ij/). Student's t-test were used to compare treatment effects. At the end of the experiment (28 days) the number of surviving earthworms was recorded.

3.3.2. Field collected straw

Field straw experiments used the choice arena as described previously, with three modifications. As 13 % earthworms had escaped the choice chamber by dragging a microfuge tube into the centre of the arena and escaping through the hole, a different tool was used to make the hole in the aluminium plate to create a very tight fit for the microfuge tube and replication was increased (n = 56). Seven equally spaced microfuge tubes were used so that all the field straw (n = 7) could be assessed at the same time.

The surface straw biodegradation experiment design used the same experimental design as described previously with two modifications. A total of 8 g crop residue was applied to the soil surface and a lower controlled environment room temperature was used (12 – 15 °C).

3.4. Agricultural knowledge and information system preferences

Audience responsive software (CLiKAPAD) was used to explore ecological literacy, change readiness and communication preferences at a farming conference in February 2022. Participants were verbally informed that their participation was entirely voluntary and anonymous, and the collective results would be used to shape communication and research directions. That is, by using the keypads they were providing informed consent. A 90-minute PowerPoint presentation included voting slides with questions ranging from earthworm identification to likert scales to explore attitudes. There were six question sets with 1 – 5 questions per set within the presentation, and participants had 10 seconds to vote for each question. The results were immediately presented back to the audience in an appropriate format e.g. answers to earthworm identification questions or bar charts of preferences, with opportunities for questions and clarifications. After the event, the likert scale results were processed to calculate a net activity score (-100 to +100 %) to aid interpretations.

The likert responses e.g. "how confident are you in knowing what epigeic, endogeic and anecic earthworms are?" were used to categorise participants into one of three groups:

Active: 5 (e.g. "very confident")

Prepared: 4

Passive: 1 – 3 (e.g. "not at all confident")

The calculation is:

Active (%) – passive (%) = net activity score (%). This scoring technique facilitates interpretations, for example, a negative score in earthworm ecology knowledge indicates poor ecological literacy.

4. Results

4.1. Object detection model to classify middens in images.

Training images (set 2) were collected from a field which contained an average of 2 middens per m^2 (Figure 2).

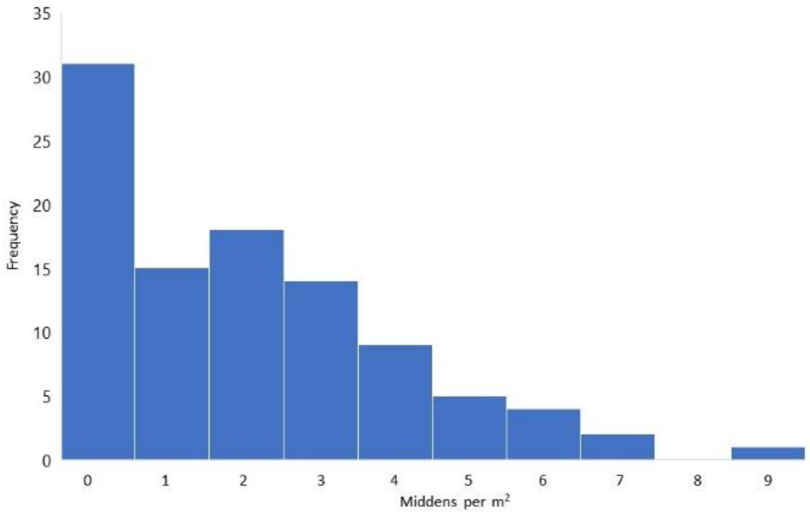

Figure 2: Histogram of midden abundance used to train the model

The model performance (mAP) score was 25 % (Figure 3) and the validation results demonstrated that the model is learning the correct representations of middens (Figure 4).

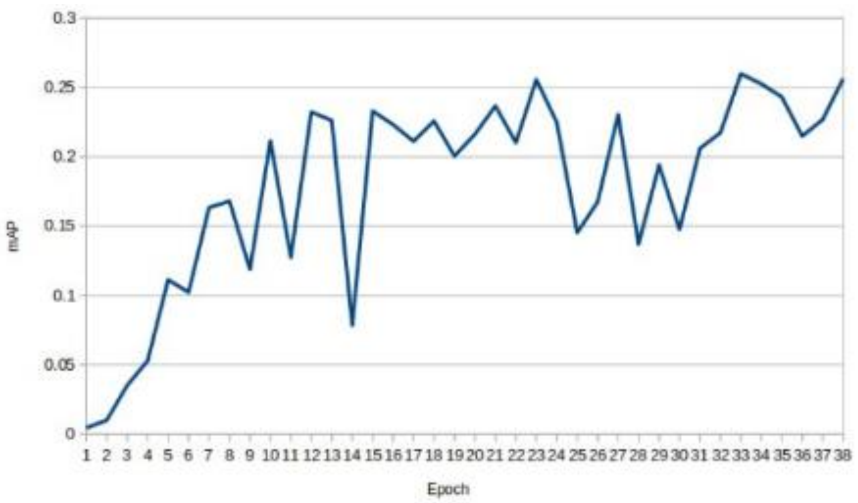

Figure 3: Mean average precision (mAP) score of the object detection model used to detect middens in images

Figure 4: Example model output, the predicted box is red and ground truth box is green.

4.2. Straw biodegradation efficiency by earthworms

4.2.1. Laboratory infected straw

The choice arena experiment resulted in a total of 83 % of the earthworms (n = 35, 5 escaped) selecting at least one infected plant material, with 17 % earthworms making no selection. The maximum number of straw types selected was all four types (of the four choices) by one earthworm, and the average selection was 1.4 types (of the four choices) by each earthworm. The trend in straw pieces taken are shown in Figure 5 below. The preference ranking is *Zymoseptoria tritici* infected leaf (Septoria leaf), *Fusarium graminearum* inoculated straw (Fusarium g), *Zymoseptoria tritici* inoculated straw (Septoria Straw) and *Oculimacula acuformis* inoculated straw (Eyespot R).

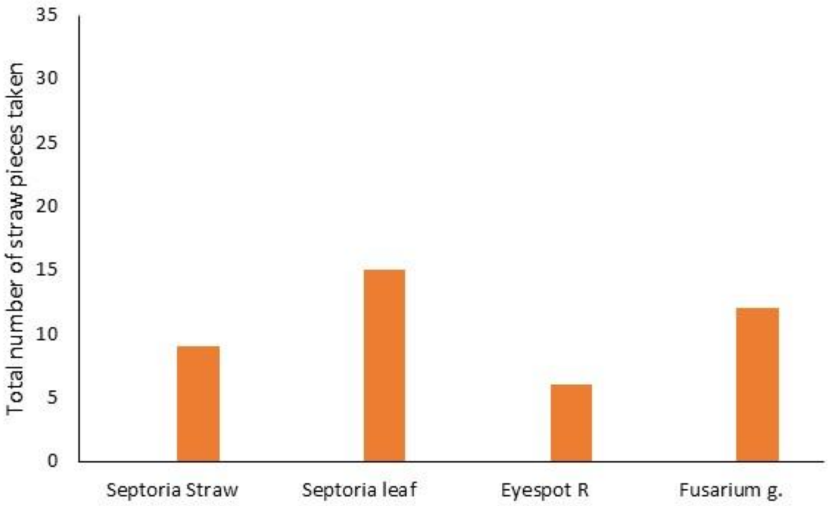

Figure 5: Preferences shown by *Lumbricus terrestris* for straw infected with pathogenic fungal species and wheat leaves infected with septoria.

The trend indicated that *Fusarium graminearum* was twice as popular as *Oculumacula acuformis* (Eyespot R), so the binary choice chamber was used to investigate whether *Lumbricus terrestris* earthworms moved towards straw infected with *F. graminearum* or *O. acuformis*. A total of 92 % of the earthworms (n = 23, 1 escaped) selected a chamber, with 8 % earthworms making no selection (found in the introduction compartment). There was no significant difference (P> 0.05) in choice.

The surface straw biodegradation experiment demonstrated that in the presence of *Lumbricus terrestris*, the surface residue significantly (P< 0.05) decreased within 7 days (Figure 6). The non-inoculated straw was least efficiently incorporated into the soil (5 %) compared to laboratory infected (14 – 35 %) and field collected (C6, Malin wheat: 8 – 26 %) straw. There was no indication that *F. graminearum* was more popular than other plant pathogens.

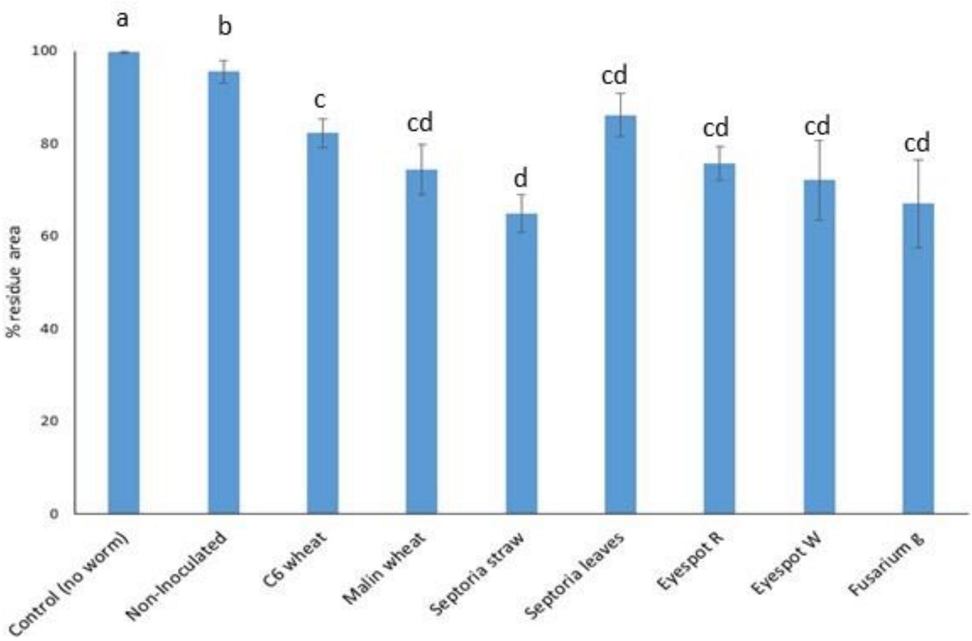

Figure 6: Surface residue cover after 7 days incubation due to incorporation activities by *Lumbricus terrestris*. Letters (a, b, c, d) above the columns indicate significant differences (P <0.05). Pathogen treatments: straw infected with *Fusarium graminearum (Fusarium g)*, *Oculimacula acuformis* (Eyespot R) or *O. yallundae* (Eyespot W), *Zymoseptoria tritici* (Septoria straw), wheat plant leaves exhibiting Septoria tritici blotch symptoms (Septoria leaves) and naturally infected wheat straws from fields Malin and C6.

No worms survived following feeding on straw inoculated with *O. yallunadae* (Eyespot W) and the general trend was reduction in survival on artificially inoculated straw (Figure 7).

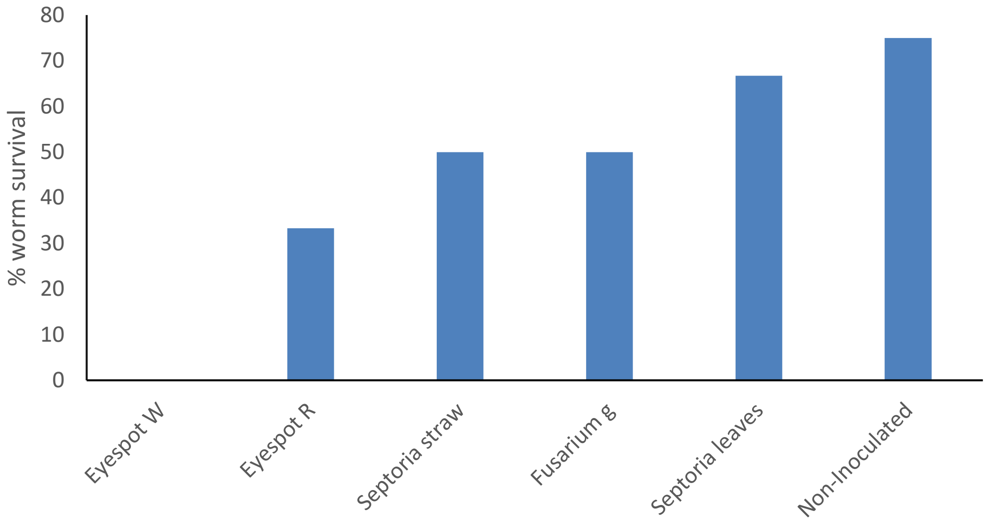

Figure 7. Survival (%) of *Lumbricus terrestris* following 28 days feeding on inoculated straw.

4.2.2. Field collected straw

DNA from the collected straw was used to quantify, using real-time PCR, *F. culmorum, F. graminearum, Microdochium nivale, M. majus*, *Z. tritici*, *Rhizoctonia solani* (AG2-1), *R. cerealis* and *Leptosphaeria maculans* or *L. biglabosa*. The material contained mixtures of plant pathogenic fungi, with the exception of maize where no pathogens were detected. The most prevalent species were *F. graminearum, M. nivale and L. maculans. Leptosphaeria biglabosa, R. solani, R. cerealis* and *Oculimacula* species were not detected.

Table 1: Pathogen suite associated with the field straws

			DNA (pg/ng)										
Field	Field location	Previous crop	*F. graminearum*	*M. nivale*	*F. culmorum*	*Z. tritici*	*L. maculans*	*M. majus*	*L. biglobosa*	AG2-1	*R. cerealis*	*O. yallundae*	*O. acuformis*
C5 Wheat	Nottinghamshire	Spring Barley	1.93781	0.07735		0.00023	0.00004						
B1 Wheat	Nottinghamshire	Maize	0.28487	0.34705	0.00998		0.00010						
C6 Wheat	Nottinghamshire	Maize		1.51641	0.04032	0.00248	0.00022	0.02012					
C3 Wheat	Nottinghamshire	Maize	0.27268				0.00005						
Malin wheat	High Wycombe	Wheat	0.58478	0.15636				0.00012					
OSR	Malmesbury	Barley	0.02167	0.04128		0.00078	0.00489						
Maize	Salisbury	Barley											

The choice arena experiment resulted in a total of 77 % of the earthworms (n = 53, 3 escaped) selecting at least one field straw type, with 23 % earthworms making no selection. The maximum number of straw types selected was five types (of the seven choices) by one earthworm, and the average selection was two types (of the seven choices) by each earthworm. The trend in straw pieces taken are shown in Figure 8 below.

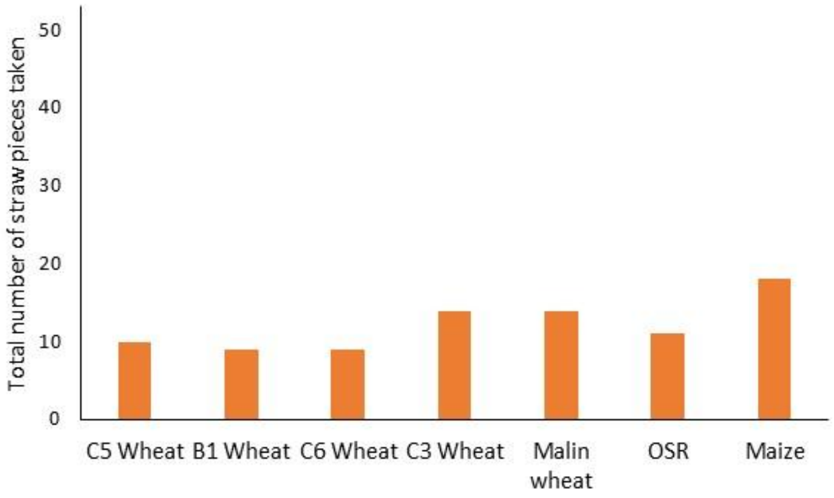

Figure 8: Preferences shown by *Lumbricus terrestris* for field collected straw, with wheat and OSR straw infected with a mixture of pathogens (see Table 1).

The surface straw biodegradation experiment demonstrated that in the presence of *Lumbricus terrestris*, the surface residue significantly (P< 0.05) decreased within 7 days (Figure 9). The C5 and Malin wheat field straw were least efficiently incorporated into the soil (5 – 7 %) compared to other field collected straws (C6, C3 wheat, OSR and Maize: 22 – 27 %).

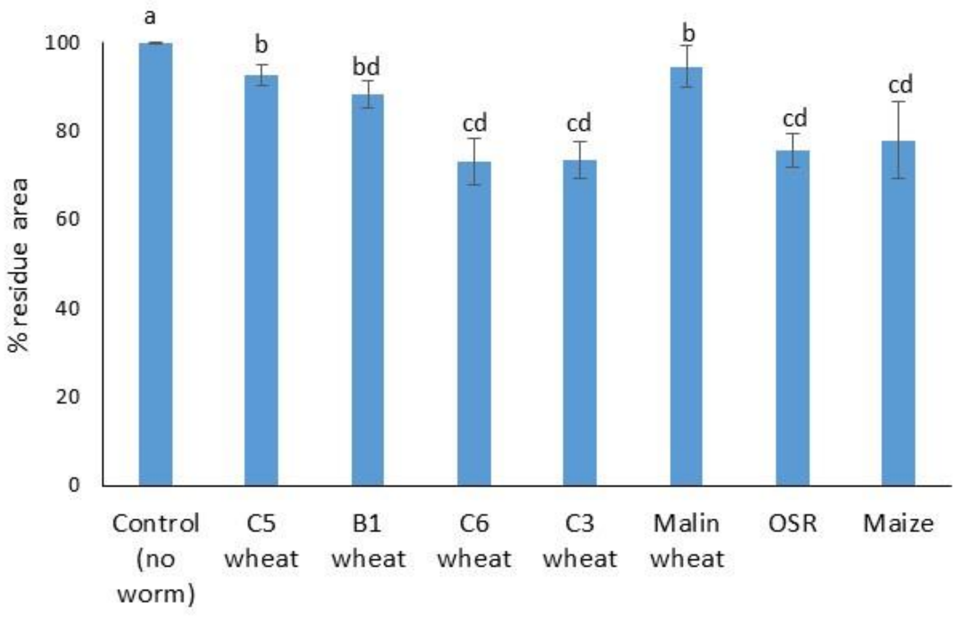

Figure 9: Surface residue cover after 7 days incubation due to incorporation activities by *Lumbricus terrestris*. Letters (a, b, c, d) above the columns indicate significant differences (P <0.05).

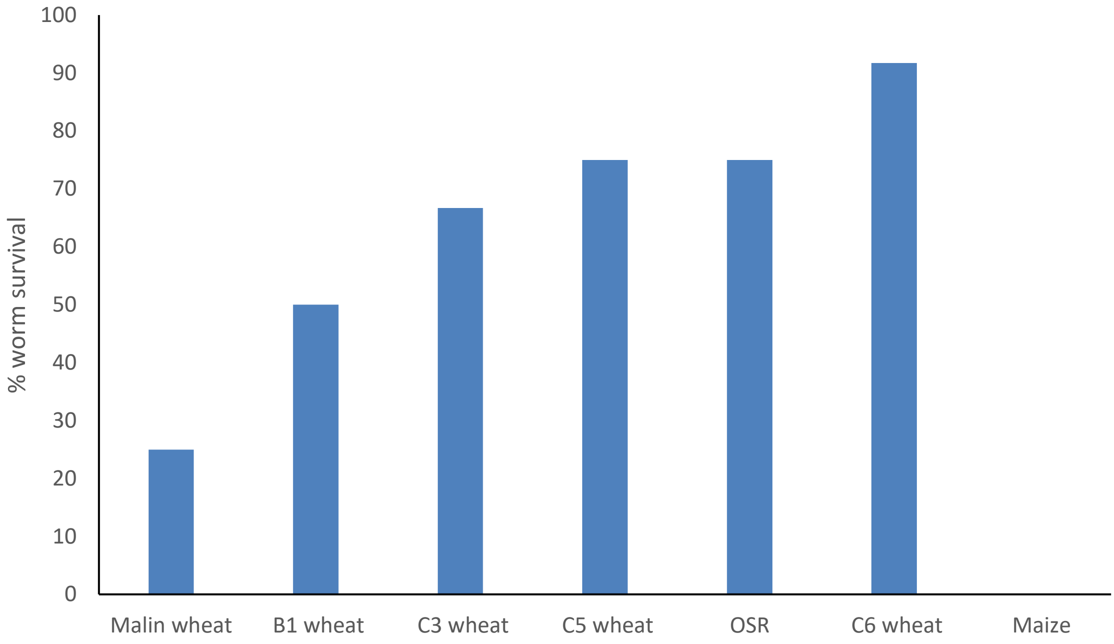

Figure 10. Survival (%) of *Lumbricus terrestris* following 28 days feeding on naturally infected straw.

No worms survived feeding on maize residues (Figure 10). Greatest mortality was observed in Malin wheat (75%) compared to C6 wheat (8%). There were no clear associations between pathogen DNA and worm survival.

4.3. Agricultural knowledge and information system preferences

There were 100 registered attendees for the event but a larger number of people attended, meaning the average CLiKAPAD response rate was 111 people (ranging between 102 – 122 people) per question. Participants described their roles in land management principally as farmers (76 %), with agronomists (12%), policy (1 %), research and teaching (6%) and conservationists (5 %).

Table 2: Net activity scores calculated from the likert scale questions

Attitude questions:	Active (%)	Passive (%)	**Net activity score (%)**
How confident are you in knowing what epigeic, endogeic and anecic earthworms are?	8.5	83	**-74.5**
How confident are you in knowing what an earthworm midden is?	55.4	28.1	**27.3**
How would you rate your expertise in crop residue management?	27.2	47.4	**-20.2**
How interested are you in mapping middens in your field(s)?	23.9	39.4	**-15.5**
Are you interested in technology assisted earthworm mapping?	33.3	30.5	**2.8**
Would you contribute photos to accelerate midden AI identification development?	58	15	**43**
How likely would you be to recommend viewing the animation (earthworm biology) to others?	49.2	24.6	**24.6**

The audience generated a very negative activity score (Table 2) in terms of their confidence in earthworm ecology knowledge (-74.5 %). To explore ecological knowledge, an illustration showing an anecic earthworm was correctly identified by 25 % of the audience, with 45 % selecting “no, do not know”. In contrast, a photograph of an anecic earthworm was correctly identified by 45 % of the audience, with just 19 % selecting “no, do not know”. In terms of earthworm monitoring, 18 % reported they deliberately measure and record earthworm populations, 72 % observe earthworms, and 10 % do not monitor earthworms (“not at all”). The cohort that deliberately measures earthworms (n = 20) had a confidence score of 2.5/5 in earthworm ecology, with 35 % correctly identifying the anecic earthworm from the illustration and 30 % from the photograph. In contrast, those who do not monitor earthworms (n = 11) had a confidence score of 1.5/5 in earthworm ecology, with 9 % correctly identifying the anecic earthworm from the illustration and 45% from the photograph.

The audience generated a positive activity score (Table 2) in terms of their confidence in identifying earthworm middens (27.3 %). To explore this knowledge, a photograph of worm casts and a worm midden was provided, with the midden correctly identified by 83 % of the audience. Further, 86 % of the audience correctly identified that anecic earthworms are responsible to residue redistribution on fields (making middens). The cohort who deliberately monitor earthworms had a confidence score of 4/5 in knowing midden identification was and 78 % correctly identified the midden from the photograph. In contrast, those who do not monitor earthworms had a confidence score of 2.9/5 in knowing what a midden was and 80 % correctly identified the midden from the photograph.

Enabling the agricultural transition to net zero is a change of system, so change readiness was explored. The audience generated a moderately negative activity score (-20.2 %) in terms of their expertise in crop residue management (Table 2). Asked if they were interested in (a) midden mapping their field(s) and (b) technology assisted midden mapping generated different responses. Personal midden mapping generated a net negative activity score (-15.5 %) compared to a net positive activity score (2.8 %) for technology assisted midden mapping. It was highlighted the technology assisted midden mapping could be accelerated with contributions of photos, and 58 % indicated that “yes, definitely contribute” generating a net positive activity score of 43 % (Table 2).

Communication styles were explored in terms of engagement in technical report outcomes. This highlighted that information seeking behaviours were primarily (61 %) browsing activities (passive, indirect) noting that 21 % of the audience reported “none” in terms of reading technical reports. Focussing on the cohort who identified as ‘farmers’: 10 % would actively seek out the report, 57 % may come across the report via browsing activities and 33 % selected “none” for report engagement. In terms of presentation styles within the technical report, graphical information was preferred (61%)

over visual/experimental outcomes (photographs showing the change in surface straw area) informing presentation style in this report.

To raise awareness of the online publication of the technical report, the audience was asked which type of social media post might ‘ignite their curiosity’ and the majority (56 %) selected an animation (over the static graph, experiment photograph or text based content) (Figure 11a). A 1-minute animation demonstration was presented to the audience and they were asked if they would recommend viewing to others, and this generated a positive activity score (Table 2). In terms of communication formats and channels, the information would be principally viewed via mobile phone (79 %) (Figure 11b) via Twitter (42 %) (Figure 11c). In terms of the source of digital information: 25 % had no preference and 5 % do not use social media, but in terms of the 70 % who thought the source of information mattered (Figure 11d); 50 % had a preference towards the active scientist(s). This choice was 6–times more popular than the host (institution or funding source) and twice as popular as the soils community hub.

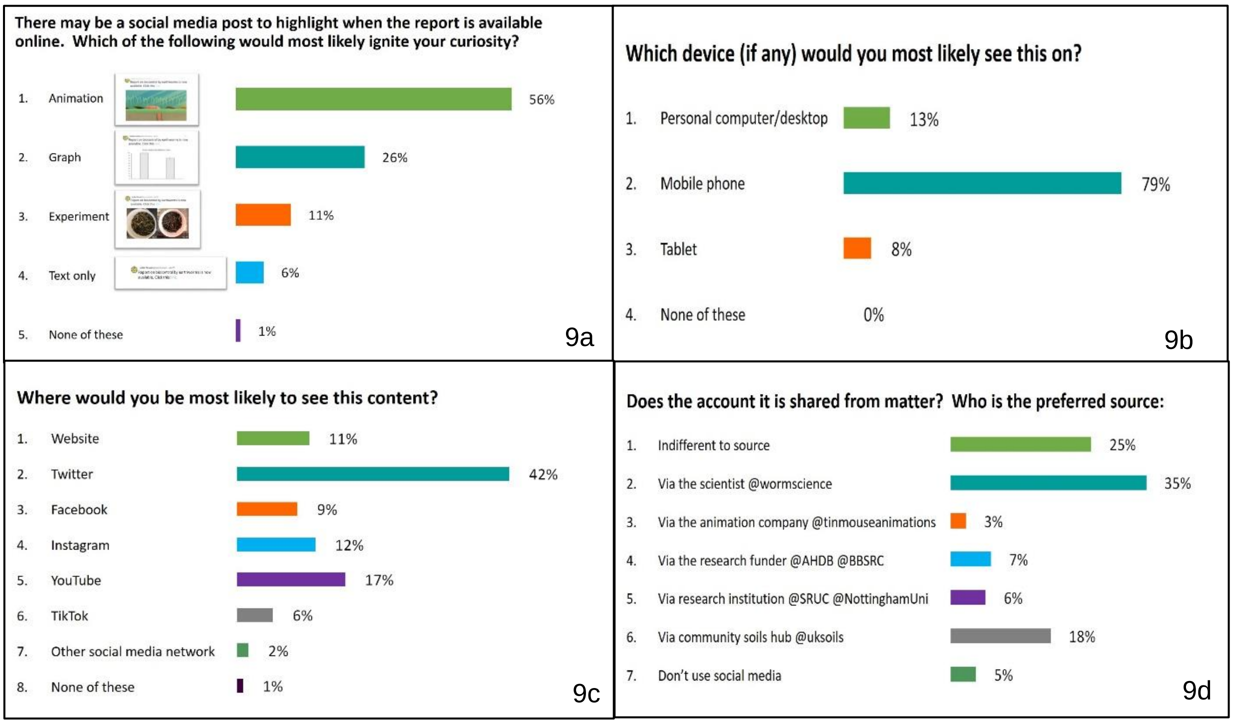

Figure 11: (a) Social media communication preferences, (b) device used for viewing digital content, (c) digital network use and (d) source of information.

An animation was informed by audience responses with a storyboard including: context (field setting), connection to prior knowledge (earthworms biodegrade crop residues), and linking new learning to own knowledge (epigeic and anecic earthworms are the litter feeding earthworm types) (Figure 12). It is available as a .mov file, ready for dissemination with the publication of this report.

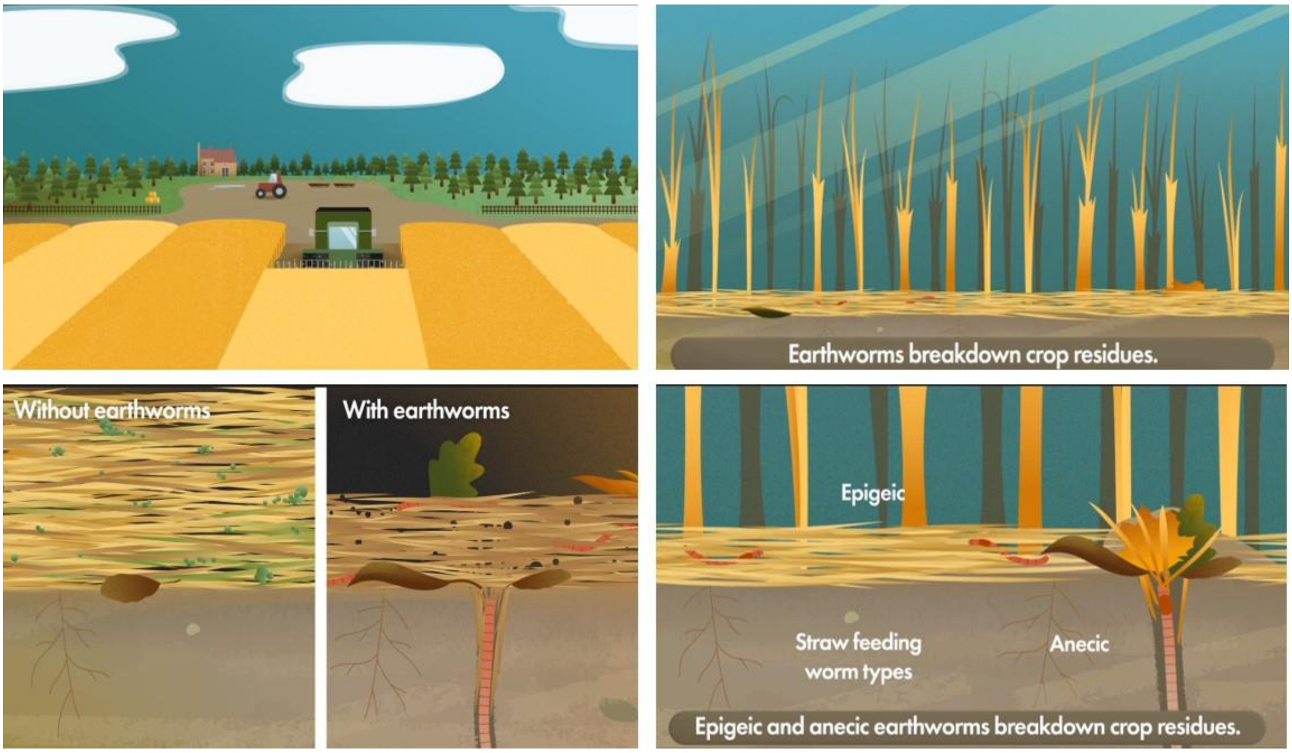

Figure 12: Storyboard for 1-minute animation based on farmer feedback to link earthworm behaviours to their ecological groups and build ecological literacy in the community.

Audience responses included positive feedback about this approach making it easy to communicate with a "lab coat". Questions included using midden mapping as a measure of soil health, the impact of using a stripper header for crop residue management (implications of standing straw for earthworm food source and disease risk), the best length to chop straw to enhance earthworm activities and if there was any research into the impact of *Lumbricus terrestris* straw biodegradation on the biocontrol of bean seed fly larvae. A farming magazine editor approached the scientist to propose producing 4000 earthworm photography mats for distribution in October 2022, requesting a quote for production (£12k, or £3 each) so that they could explore sponsorship options with companies trying to engage farmers in carbon markets.

5. Discussion

The object detection model to classify worm middens in images was a successful proof of concept. Training images developed annotation skills within the industry (set 1) that were applied to develop the object detection model to classify worm middens in images (set 2). The model performance was good (25 % mAP), indicating the potential for real world implementation (Figures 3, 4). Small robots collect field surface images for plant counts, weed and disease assessments and middens could be added to this image processing suite. Discussions identified that the pathway to develop model performance would be via image collection method used for set 2 (uniform image sizes, angles and distance to the ground) across different field settings, with a training set between 1000 – 9999 images needed to optimise model performance. Two applications were highlighted: integrating soil biological properties into crop health, disease and performance monitoring at field scales (Figure 13) and soil health assessment, based on research which correlated midden counts to soil physical and microbial properties (Jemisen et al., 2019). The company reported receiving several enquiries by customers about midden mapping their fields following the presentation.

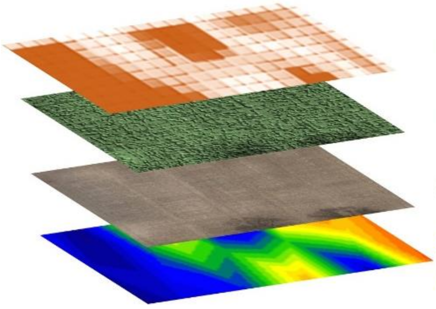

Figure 13: Potential application of midden monitoring: integration of soil biology into field monitoring

The complementary laboratory studies measuring *Lumbricus terrestris* earthworm behaviours demonstrated that they select pathogen infected plant residues. A total of 77 – 83 % *Lumbricus terrestris* earthworms chose plant and straw material infected with plant pathogens (Figures 5, 8). There was no indication of preferential behaviours towards *F. graminearum* with all the straw types - septoria (*Zymoseptoria tritici*), Eyespot-R (*Oculimacula acuformis)* and mixtures of pathogens (Table 1) - being chosen by at least one individual earthworm. A binary chamber experiment indicated that *Lumbricus terrestris* does not specifically move towards soil patches with *Fusarium graminearum* surface straw type, indicating infected materials do not induce chemotaxis-like behaviours. Both septoria leaf and maize strips were the most frequently selected plant materials, but this finding could be confounded by their physical form (leaf shape) in comparison to the straw, rather than fungal composition. *Lumbricus terrestris* earthworms have a preference for leaves of specific shapes (Darwin, 1882).

The wheat straw infected with pathogens was more attractive as a food source compared to non-infected wheat straw (Figure 6), with up to 6-fold higher surface removal rates by *Lumbricus terrestris* earthworms. This is in agreement with other research indicating that *Lumbricus terrestris* is a selective feeder, with a preference towards pathogens and early successional fungal species (Doube et al., 1997; Bonkowski et al., 2000; Oldenburg et al, 2008). However, worm mortality increased on artificially infected material (Figure 7) with straws including *Oculimacula* spp. being most detrimental to their survival. There were no clear associations between worm behaviour, activity, survival and pathogen load or diversity on field straw, which may be because cellulolytic activity which enhances the nutrient quality of the straw was not assessed (Oldenburg et al, 2008). The behaviours measured, for example, comparing Malin field wheat straw (*Fusarium graminearum*) to C6 field straw (*Fusarium culmorum)*, suggested that although both were similarly attractive food sources in the first biodegradation experiment (Figure 6) in a repeated experiment, Malin was the least attractive food source (Figure 8) and resulted in the lowest survival (Figure 10) suggesting that *F. graminearum* may be negatively impacting on the fitness of the worms. Further research is required to identify if these differences are related to the intrinsic ability of the individual *Fusarium* spp. to produce mycotoxins. The least attractive food source were straws inoculated with *O. acuformis* and whilst the biodegradation experiment showed increased incorporation activity, *O. yallundae* was associated with 100% mortality of the worms. Field straw was collected from fields where midden abundances ranged between 2 – 15 middens per m^2 and differences detected in the laboratory may help to explain the patchy distribution of middens within fields. Research attention has been principally directed towards *Fusarium* species, but this is the first study (to our knowledge) to include feeding responses to pathogen DNA-characterised field straw or artificially infected straw with *Z. tritici* or *Oculimacula* species. In summary, in both the field and laboratory conditions, *Lumbricus terrestris* earthworms select pathogen infected plant residues and rapidly remove infected materials from the soil surface. However, it remains unclear if these benefits are partially offset by reduced worm fitness and increased mortality depending on the pathogenic species present.

During this project, two major gaps in understanding were identified as the scientific barriers for implementation. Firstly, it is a priority to research pathogen dispersal risks from earthworm activities. Traditionally, it was thought that earthworms generate adverse populations of plant pathogen outcomes due to dispersal mechanisms (Edwards and Fletcher, 1988). This interpretation is based on one publication where *Fusaria* were isolated from the digestive tracts of earthworms collected in India (Khambata and Batt, 1957), referencing observations made in 1917 and 1918 that earthworms distribute fungal spores in soils. This mechanism has received little research attention. To date, it is known that some *Fusarium* species are digested in the gut of *Lumbricus terrestris*, including *F. culmorum* (Schrader et al., 2009) and *F. laieritium* spores (Moody et al., 1996), but the fate of *F. graminearum* and other phytopathogens are unknown. Middens are biodegradation hotspots made by *Lumbricus terrestris,* but the ecology has been overlooked: midden patches have elevated populations of other earthworm types. Specifically, earthworm types characterised by their horizontal mobility at the soil surface (epigeic) and within the topsoil (endogeic). If pathogenic species can survive the passage through these earthworm guts, then there is the potential for dispersal which may increase the incidence of disease. It would be reasonably simple to use ecologically intact earthworm populations (epigeic, endogeic and anecic earthworms) and characterise pathogen communities associated with midden soil patches to improve our understanding of pathogen dispersal risks from earthworm activities.

The second major gap in understanding which forms a scientific barrier for implementation is linking earthworm activities to plant disease incidence and severity and their own fitness. A rapid improvement in understanding could be achieved with mechanisms to support soil science – plant pathology collaborative research. For example, greenhouse experiments with eyespot - W

(*Oculimacula yallundae*) detected a lower disease incidence (44 – 70 %, cultivar effects) in the presence of *Lumbricus terrestris*, indicating that these earthworms are potentially effective biocontrol agents for this pathogen (Bertrand et al., 2015). In agreement, field research from 1991 – 1995 in Switzerland reported reduced tillage increased earthworm populations and reduced eyespot infection in wheat (Anken et al., 2004). However, effects of the eyespot pathogens on worm fitness were not assessed. In this study, both straws infected with eyespot pathogens fed to the earthworms resulted in reduced survival. As this is only one experiment further work is required to confirm the observations. Real world implementation is dependent on linking earthworm activities and fitness to plant disease occurrence.

In terms of real-world implementation, ecological literacy is linked to pesticide dependencies and uptake of biocontrol strategies (Wyckhuys et al., 2019). Further, facilitating the transformation to a net zero system is change management, so it is important to determine the level of readiness in order to prepare appropriate resources. Readiness ranges from denial (no need for change), vague awareness (limited knowledge), pre-planning (no action), initiation (training), implementation (routine) to professionalization (sophisticated knowledge, dynamic improvements). We explored these key properties of the agricultural knowledge and information system with the people expected to benefit from this research activity.

The audience reported low confidence levels in their knowledge of earthworm ecology (Table 2), and this was reflected in the widespread inability to identify an earthworm to its ecological group via an illustration or photograph format. It could be assumed that people who deliberately measure earthworms would be confident in earthworm ecology and correctly identify earthworms, but they were neither confident nor correct in their earthworm ecological group identification. These results indicate poor soil ecological literacy rates and a readiness stage of 'vague awareness'. This requires attention towards preparing appropriate resources because there is potential for this situation to be improved. In total, 90 % of the audience reported that they routinely monitor or observe earthworms. Earthworm ecological groups are a type of knowledge which are an abstract representation of earthworm population characteristics. For simple information to be encoded and widely known, it needs to be related to personal experience (e.g. experiential learning with earthworm identification success) (Gigerenzer & Hoffrage, 1995). This suggests promoting experiential learning resources provided by AHDB (e.g. 'How to count worms'), updating this guide by replacing illustrations with photographs (as earthworm ecological group identification success rates were 44 % higher for photographic images compared to illustrations) and co-ordinating feedback (e.g. photographic images for verification checks) would likely build ecological knowledge in the community to support change management in agriculture.

In contrast to earthworm ecology, knowledge about anecic earthworm behaviours was good. There was a positive net activity score in confidence in earthworm midden identification (Table 2) and 83 % of the audience correctly identified this surface feature from a photograph. These results indicate a pre-planning stage of readiness, particularly as people had negative activity score when rating their personal expertise in crop residue management (Table 2). In terms of implementation, it is unlikely that people will invest time to walk fields to monitor earthworm middens (net negative activity score), the interest was in technology assisted midden mapping (net positive activity score). This was corroborated by people indicating they would contribute photographs to develop the midden identification model (58 %) resulting in a high net positive activity score of 43 %. This complements the technical development of the object detection model which is at the stage of requiring images from a range of field situations to improve AI performance.

In terms of communications, the customer of science is the funding organisation, and it is essential for scientists to produce written outcomes (reports, articles, publications for REF assessments) and webinars for academics. This communication strategy can be problematic because farmers developing reduced tillage management practices state that: “farmer to farmer learning is a powerful tool whilst there is a whole lot of science paperwork out there, but it is on a shelf somewhere” (Skaalsveen et al., 2020).

Exploring interactions with technical reports highlighted that 21 % people selected “none”, with 1 in 3 farmers indicating that they do not read technical reports. Information seeking behaviours were aligned with routine digital behaviours, that is 61 % people reported a browsing style (passive, indirectly come across the report) rather than traditional information seeking behaviours e.g. library book style (active, directly seek out information). That is the: “on a shelf somewhere” problem in communication networks is metaphorical (not literal), as information seeking behaviours are principally passive and indirect. This highlights the importance of planning appropriate digital resources to better integrate the two knowledge systems together.

Here we explored the opportunities improving the accessibility of the report using social media approaches that would ‘ignite curiosity’, and the majority (56 %) selected an animation style (over graphical, photographic or written content, Figure 9a), in a format suitable for mobile phone screens (Figure 9b), with attention towards Twitter (Figure 9b). Importantly, the animation had potential for social learning network spread with a positive net activity score (Table 2) relating to people personally recommending it to be viewed by others. This behavioural response indicates the animation was perceived to have salience, was consistent with public reputations and egos to engage the messenger effect (Dolan et al., 2012). The messenger effect can be easily overlooked, but responses to content are linked to trust, credibility, authority, accountability, reputation and/or likeability of the messenger (Dolan et al., 2012). Therefore, it is important to know if there is a preferred messenger for digital content. The majority (70 %) considered the messenger mattered, with a clear preference towards the active scientist(s) over organisation (funding source or university) or community groups (Figure 9d). This indicates that network building via direct communications with active scientists is an important component of change management in agriculture.

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