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Soil Biology and Soil Health Partnership Project 11: Developing UK-relevant benchmarks for the soil health indicators: Potentially Mineralisable Nitrogen and Solvita respiration burst

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This is the final report of a 12 month project (Project 11) within the Soil Biology and Soil Health Partnership (AHDB: 91140002) which started in April 2019. The work was funded by a contract for £9,734 from AHDB and BBRO.

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1. Abstract

This project is part of a suite of integrated projects (Soil Biology and Soil Health Partnership) specifically aimed at addressing the AHDB and BBRO Soils Programme call - "Management for Soil Biology and Soil Health". This project was one of a number of activities funded through the Innovation Fund, designed to address knowledge gaps that arose over the 5-year duration of the programme (see Figure 1).

The size and activity of soil microbial biomass is considered to be a key indicator of soil biological health. However, the ‘standard’ method of assessment which uses a chloroform extraction is not currently offered by any of the main commercial labs in the UK due to the hazardous reagents required. Two alternative methods can be used to infer the size and activity of the microbial community: (i) potentially mineralisable nitrogen (PMN) which measures the amount of N readily decomposed under controlled (anaerobic) conditions, and (ii) CO_2-C burst which measures the amount of C released as CO_2 when a dried soil is rewetted. These processes are both dependent on the size and activity of the soil microbial biomass. The methods are currently delivered by commercial laboratories in the UK. The interpretation frameworks (or guideline values) developed so the methods can be used as indicators, have largely been based on data from the United States; this project has used data to derive guideline values that are relevant for UK agro-climatic conditions.

The project used results from UK agricultural soils (restricted to mineral soils at <20% SOM) analysed by Hill Court Farm Research (PMN; n=625; arable soils) and NRM laboratories (CO_2-C burst; n=1803 arable and 310 grassland soils). Rather than deriving critical thresholds, the project developed ‘typical’ values based on the median and lower quartile values for each of the datasets. These values can be used as a comparative tool to understand whether levels of microbial activity within the soil are typical or if there is scope for improvement.

The project found:

- Both PMN and CO_2-C burst levels were negligible in the subsoil confirming that these methodologies provide integrated indicators of both the size and activity of the soil microbial biomass.
- There was no relationship between soil texture and CO_2-C burst or PMN value. A single set of guideline values were therefore determined for all mineral soil textures (i.e. those containing <20% SOM).
- Separate guidelines were developed (and justified) for grass and cropped (all annual crop types) land for CO_2-C burst, but there are currently insufficient PMN results from grassland systems to develop guidelines for PMN in grassland.

- Evaluation of the guideline values using data from sampling at the long-term experimental sites within Project 4 of the Soil Biology and Soil Health partnership clearly demonstrated that the revised values were more sensitive at identifying treatment differences (due to variations in organic matter content and pH) than the existing guidelines from the US.
- Further evaluation of these benchmark values will be undertaken using results obtained from the final year of sampling at the long-term experimental sites, as part of the reporting for Soil Biology and Soil Health Project 4.

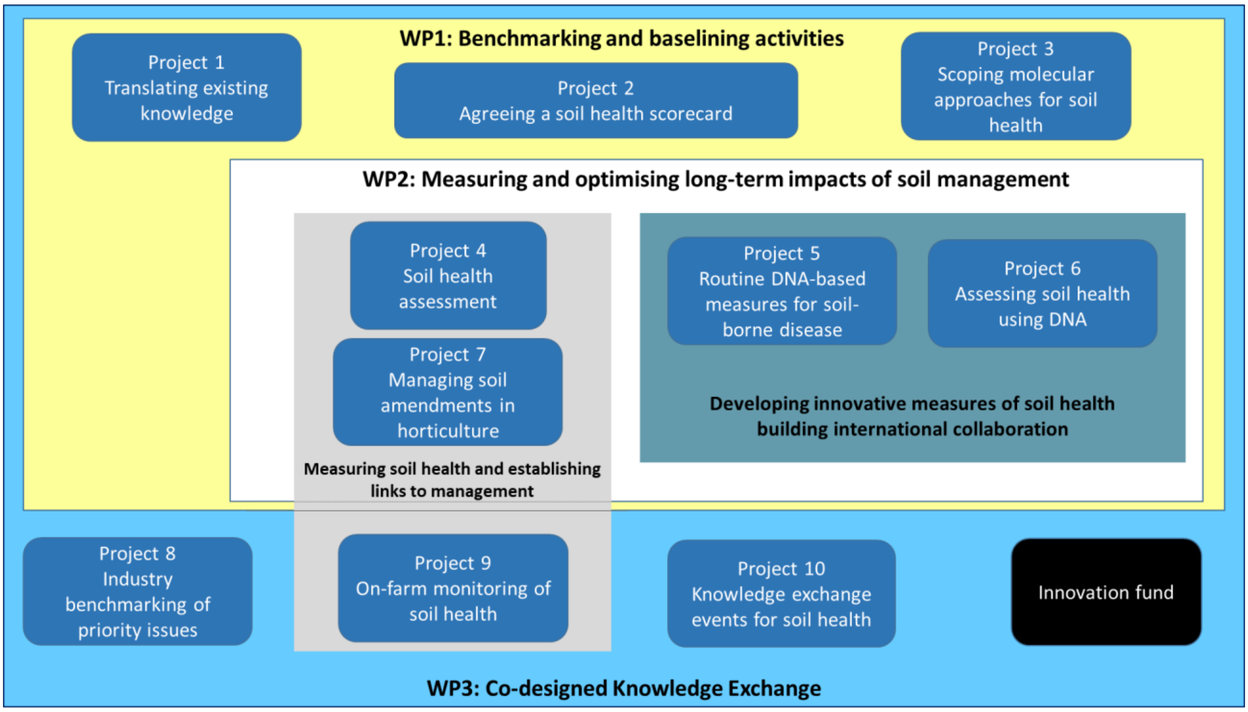

Figure 1. Diagram to show how this project fits into the organisation of the Soil Biology and Soil Health Partnership: Project 11 is one of the projects funded under the “Innovation fund”

2. Introduction

Soil physics, chemistry and biology are interlinked and all play a role in maintaining productive agricultural and horticultural systems. However, our understanding of biological indicators of soil health are less developed than indicators of soil physical and chemical properties and improving this is a key aim of the AHDB-BBRO Soil Biology and Soil Health Partnership (SBSH). In an earlier SBSH partnership project, Griffiths *et al.,* (2018) identified a number of biological indicators that have the potential for inclusion within a scorecard approach for soil health testing, which is currently being evaluated as part of the partnership programme. In particular, assessment of the size and activity of the microbial biomass was considered to be a key indicator of soil biological health, demonstrating a soils ability to store and recycle nutrients. In general, higher soil microbial biomass/activity are linked with 'better' soil quality (Dick, 1992). There are a number of methods for measuring this, including:

- *Chloroform fumigation-extraction* measures the carbon content of the microbial community in soils and is an indicator of its size/biomass (often termed 'microbial biomass carbon' or 'MBC' (and equivalents for N); Brookes *et al.,* 1985, Vance *et al.,* 1987, Wu *et al.,* 1990). It is considered to be the 'reference' method, described by ISO standard 14240-2, and often used to provide 'correction factors' for other methods of measuring MBC such as substrate induced respiration (Anderson & Domsch, 1978; ISO-1420-1). This technique involves measuring the difference in carbon content between split samples of soil, one of which has been fumigated with chloroform to lyse all living cells and release their contents as organic carbon ('MBC') and one of which is untreated. It is a method predominantly used by research organisations and is not currently offered by any of the main commercial laboratories in the UK, principally as it involves the use of chloroform which is a known carcinogen and a flammable/explosive organic compound.
- *Potentially mineralisable N* (PMN) measures the release of ammonium-N from soils incubated for 7 days under anaerobic conditions (Keeney, 1982). The test is provided by commercial laboratories in the UK and is currently used to determine 'Additional Available Nitrogen – AAN). PMN originates mainly from the microbial biomass and recent residues (plant material, organic manures etc) and has been used to indicate the medium-term capacity of the microbial community to decompose (mineralise) organic N into plant available forms.
- *CO_2 respiration burst* measures the release of CO_2 following the addition of water to an air-dried soil sample. The test was developed for rapid use and is offered by commercial laboratories in the UK. It has the advantage that it uses dried soil samples, so can be performed alongside other routine soil tests, such as nutrient analysis and organic matter content, whereas both PMN and MBC require fresh soil samples (which should be stored in refrigerators at <4°C before analysis). CO_2-burst measures the activity of microbes rather than the size of the microbial community (www.solvita.com). The method has been used to indicate the capacity of the microbial community to mineralise carbon and nitrogen (Franzluebbers *et al.,* 2000; Haney *et al.,* 2008 a,b).

Griffiths et al. (2018) proposed MBC indicator values for potential use in the AHDB-BBRO Soil Health scorecard based on a global meta-analysis by Kallenbach & Grandy (2011), which included data from 41 studies where the response of MBC (as measured by either chloroform extraction or substrate induced respiration) to organic amendments was measured. The authors proposed a MBC threshold > 200 mg C/kg to represent a 'good' level of microbial biomass that required no improvement (i.e. given a 'green' traffic light). Whilst MBC is being tested within Project 4 ('Soil health assessment') of the partnership programme using the chloroform incubation method, an alternative method is required if soil microbial biomass is to be used as a routine indicator of soil health as the extraction is not an option for commercial laboratories.

Benchmark/indicator values have been developed for both PMN and CO_2-burst, but only using data from the United States ('The Cornell Framework', Moebius-Clune *et al.,* 2017 and www.solvita.com), where soils are very different as a result of differences in parent material, climate and management, and tend to be lower in organic matter than those in the UK. As a result, the 'traffic lights' that are currently proposed for these indicators may not be appropriate for UK soils and cropping conditions. Analysis of UK soils has shown PMN and CO_2 burst values much higher than those typically measured in the USA (with most UK soils scoring highly for these indicators and given a 'green' traffic light). For example, early results from Project 4 of the SBSH partnership at Harper Adams University, showed a significant effect of repeated FYM addition on PMN levels in the topsoil, linked to a higher (but not significant) MBC (Table 2.1). Results from the qPCR analysis also showed high bacterial (16sRNA) and fungal (18sRNA) populations on the repeated FYM treatment (Elphinstone; pers. comm. SBSH Project 5); although these differences were not detected using CO_2 burst method.

Table 2.1 Soil biological indicators measured at the long-term experimental site at Harper Adams University evaluating the effect of repeated addition of a range of organic materials; 'traffic light' scoring based on those proposed by Griffiths et al. (2018) and www.solvita.com using international datasets ('green' = no action required).

Attribute	Control	FYM (23 years)	Slurry (23 years)	Green compost (13 years)	Green/food compost (6 years)	Food-based digestate (9 years)
MBC (mg/kg)	350	393	315	317	334	345
PMN (mg/kg)*	23[a]	90 [b]	24 [a]	43 [a]	38 [a]	43 [a]
CO_2-C burst (mg/kg)	198	228	247	222	219	228

**Statistically significant treatment effect at P<0.05, with values across a row with a different letter significantly different from each other.*

However, when the results from Harper Adams were evaluated in the light of the Cornell Framework thresholds for PMN (Moebius-Clune *et al.,* 2017) and Solvita thresholds for CO_2 burst (www.solvita.com), all treatments (including the inorganic fertiliser control) scored the same, despite the statistically significant differences between treatments ('green' traffic light – good soil biological health; Table 2.1). There was no difference in CO_2 burst activity with treatment, with all scoring a 'green' traffic light for this metric (Table 2.1). This lack of sensitivity suggests there is a need to revise the scorecard benchmark values to ensure they are appropriate for UK soils, crops and climatic conditions. This will provide greater resolution in the benchmarking values to better distinguish differences in the biological activity of UK soils.

2.1. Objectives

This project aimed to update the soil health scorecard benchmark values for PMN and CO_2 burst to more accurately reflect UK soils and climatic conditions.

3. Materials and methods

3.1. Source data

3.1.1. Potentially mineralisable N (PMN)

Hill Court Farm Research analyse a large number of soil samples for PMN and hold a database of *c*.400 soils (all arable) where PMN, soil organic matter (SOM) and texture (by hand) have been measured. The database is largely comprised of medium and heavy textured soils, with less than 10% of samples being classed as light textured (e.g. loamy sand and sandy loam). In order to increase the number of light soils tested an 50 additional sites were sampled in spring or autumn 2019 as part of planned soil mineral N sampling undertaken by the partners in existing studies, as well as samples from the farmer groups working with the programme as part of Project 9. The majority of samples were taken from the top 0-15cm or 0-30cm, however in order to understand the 'extremes' of microbial biomass/activity possible within UK soils, samples were also taken from the subsoil (60-90cm) at four sites. Soil samples were analysed for texture (by hand), organic matter (loss on ignition) and PMN (anaerobic incubation, Keeney et al., 1982). Data gathered as part of previous experimental work and other funded projects (e.g. sampling of the AHDB Strategic and Monitor Farms) was also added to the database where appropriate; this included results from SAC Laboratories 'Soil Health Test' suite (including PMN, SOM, nutrients and pH), comprising *c*.40 soils from Scotland (arable and grassland).

3.1.2. CO_2-burst

NRM Laboratories hold a large database (> 5,500 samples) from their Soil Health Index testing service, which includes texture, organic matter (loss on ignition) and CO_2 burst. The results from 4,600 samples taken from 2017 onwards were included within the data analysis. Given the size of this database, no additional sampling was required. The database was carefully screened to remove subsoils (> 30cm depth), non-UK data, non-agricultural land (sports turf, land restoration), incomplete data sets and organic soils (>20% SOM – benchmarking restricted to mineral soils only).

The remaining database was then split into cropping categories: tillage land (arable and horticultural), permanent crops (miscanthus, orchards, top fruit etc), grassland and ‘unknown’. The ‘unknown’ cropping comprised samples that had been submitted without information on land use, these were screened and cropping added where other identifiers provided an indication of the type of land use (e.g. samples from the Yield Testing Network – YEN, AHDB Monitor farms and Strategic farms etc.). Table 3.1 gives details of the final database that was used in the benchmarking exercise; note there were 1,936 samples that could not be reliably allocated to a crop class, so these were excluded from the analysis.

Table 3.1 Land-uses covered by the NRM soil health database

Land use	Number of samples
Cropped[a]	1803
Grassland[b]	310
Permanent crops[c]	142

[a]Cropped land included cereals, oilseeds, potatoes, field vegetables (brassicas, onions, carrots), salad crops, peas and beans, maize, set aside;
[b]Grassland included samples labelled: ‘permanent pasture’, ‘grazing’, ‘grassland’, ‘silage’, ‘reseeds’ and ‘leys’;
[c]Permanent crops included orchards, topfruit, miscanthus, also glasshouse crops; these samples have been removed from the data analysis.

3.2. Data analysis

The databases collated in 3.1.1 and 3.1.2 were scrutinized using a similar approach to that developed for the SOM typical ranges (Griffiths *et al.,* 2018, Defra 2000 and 2004), taking into account soil texture (clay content: light, medium and heavy) and land use (for PMN the database comprised mainly arable land). Due to lack of locational data it was not possible to evaluate the effect of climate (low, medium and high annual rainfall) on PMN and CO_2-burst, as used in the SOM benchmarking exercise (Griffiths *et al.,* 2018). Descriptive statistics were used to generate ‘typical ranges’ of PMN and CO_2-burst for each soil grouping and land use according to the median and interquartile range with traffic lights allocated as follows:

≥ median: no action required (green) – results are typical for the soil type and cropping

<median & ≥ lower quartile: monitor (amber) – results are below average for the soil type and cropping

< lower quartile: investigate (red) – results are very low for the soil type and cropping, with considerable scope for improvement.

The provisional ranges and traffic lights proposed were compared to those previously suggested by Griffiths *et al*. (2018) and evaluated using results from the long-term experimental sites sampled as part of Project 4 where MBC has also been measured using the traditional chloroform extraction methodology.

4. Results

4.1. Potentially mineralisable N 'typical ranges'

The final PMN database of samples analysed by Hill Court Farm Research comprised 598 samples from arable topsoils in England and Wales (sampling depths predominantly 0-15cm or 0-30cm), with PMN values ranging from 2 to 174 mg/kg. A total of 27 arable topsoils were analysed by SAC laboratories from sites in Scotland, with PMN ranging from 22 to 42 mg/kg. These results were added to the Hill Court Farm Research database. There were only 26 samples from grassland sites, 14 of which were in Scotland and analysed by SAC laboratories. PMN on these grassland soils ranged from 15 to 197 mg/kg, but given the sample number it was not possible to determine whether separate benchmarks are required for grassland soils and so these samples were removed from the analysis. The benchmarks derived for arable soils could be used, with caution, to help interpret PMN in grassland systems, but further sampling and a larger database of PMN values in grassland soils is required to provide a more robust interpretation framework for this land-use.

For the arable sites, the data was positively skewed (Skew = 1.3; Figure 4-1), with 50% of values falling between 27 and 64 mg/kg PMN, but the upper quartile of values much greater (62 - 174 mg/kg) than the lower quartile (2 - 27 mg/kg). Comparison with the Cornell Soil Health Framework for benchmarking PMN showed how inappropriate this benchmark system is for UK soils, with over 90% of soils falling into the 'green' category at >18 mg/kg (Table 4.1).

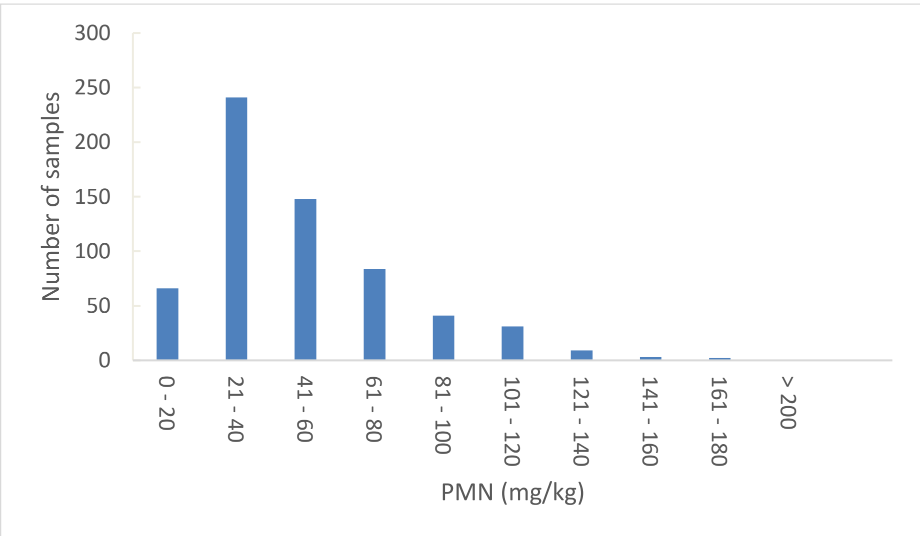

Figure 4-1 Frequency distribution for the PMN content of arable topsoils in Great Britain.

Table 4.1 Use of the Cornell Framework to benchmark UK PMN values (n=625). Very low values indicate a constraint to proper soil functioning; low and medium values do not necessarily represent a major constraint but suggest sub-optimal functioning; high and very high values indicate near optimal or optimal functioning and management goals should aim to maintain such conditions.

Cornell 'traffic lights'	PMN (mg/kg)	% of UK Soils in each category
Red – very low	< 9	1
Orange - low	9.1 - 13	2
Yellow - medium	13.1 - 18	4
Light Green - high	18.1 - 21	5
Dark Green – very high	> 21.1	89

PMN and SOM were positively related, albeit with only 23% of the variation in PMN considered to be due to variation in organic matter content (Figure 4-2).

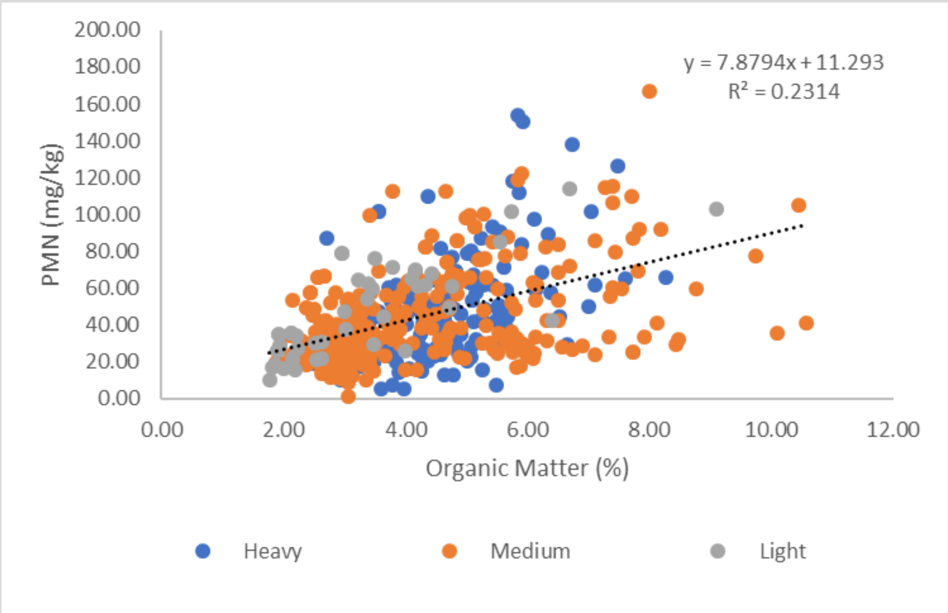

Figure 4-2 Relationship between PMN and organic matter content of arable topsoils (light < 18% clay; medium 18-35% clay and heavy > 35% clay).

Samples were divided into 3 textural groups: light (loamy sand, sandy loam or silt loams; n=68), medium (sandy clay loam or silty clay loams; n=366) and heavy (clays, clay loams, silty clays and sandy clays; n=191), and descriptive statistics calculated for each group (Table 4.2). The PMN content of the light-textured soil group was more variable (high standard error and 95% confidence interval) compared to the other textural groups; the SOM content of these soils also varied in a similar manner (Table 4.3). However, the 95% confidence intervals of the PMN content of all three textural groups over-lapped, suggesting that the average PMN of all these soil types was very similar. Guideline values (or benchmarks) for UK arable soils were therefore estimated from the median and lower quartile PMN values of the whole dataset, regardless of soil type (Table 4.4).

Table 4.2 PMN summary statistics for UK arable topsoils

Soil type:	Light	Medium	Heavy	All data
Mean	51	48	48	48
Standard error	3.9	1.5	2.1	1.2
Median	40	39	42	41
Skew	1.4	1.2	1.4	1.3
95% CI	7.6	2.9	4.0	2.3
Minimum	10	1.4	5.2	1.4
Maximum	174	167	154	174
Lower quartile	27	27	28	27
Upper quartile	68	62	60	62
Count	68	366	191	625

Table 4.3 SOM (%LOI) summary statistics for UK arable topsoils

Soil type:	Light	Medium	Heavy	All data
Mean	3.3	4.2	4.5	4.2
Standard error	0.22	0.10	0.09	0.07
Median	3.0	3.5	4.5	3.9
Skew	1.7	1.2	0.6	1.2
95% CI	0.44	0.19	0.17	0.13
Minimum	1.8	1.9	2.3	1.8
Maximum	9.1	10.6	8.3	13.3
Lower quartile	0	2.6	3.3	3.0
Upper quartile	3.5	4.8	5.1	5.1
Count	47	314	170	531

Table 4.4 Guideline values for the 'typical' PMN content of UK agricultural soils

Category	PMN (mg/kg)	Description
Investigate	<27	Very low
Monitor	28-40	Low (below average)
No action required	>40	Typical

4.1.1. Effect of sampling depth on PMN

At four of the sites sampled in 2019 covering the range of textures (light, medium and heavy), the PMN and SOM content of the topsoil (0-30cm) was compared with that in the subsoil (60-90cm). There was a marked decline in both properties in the subsoil (Figure 4-3), with subsoil PMN <10% of topsoil PMN, and subsoil SOM *c*. 50% of topsoil SOM. Topsoil PMN ranged from 20 – 27 mg/kg and subsoil concentrations from 0.4 – 3.0 mg/kg. Soil organic matter varied more with soil type ranging from 2.1 % (sandy loam) to 7.7% (silty clay loam) in the topsoil, and from 0.65% (loamy sand) to 3.8% (silty clay) in the subsoil.

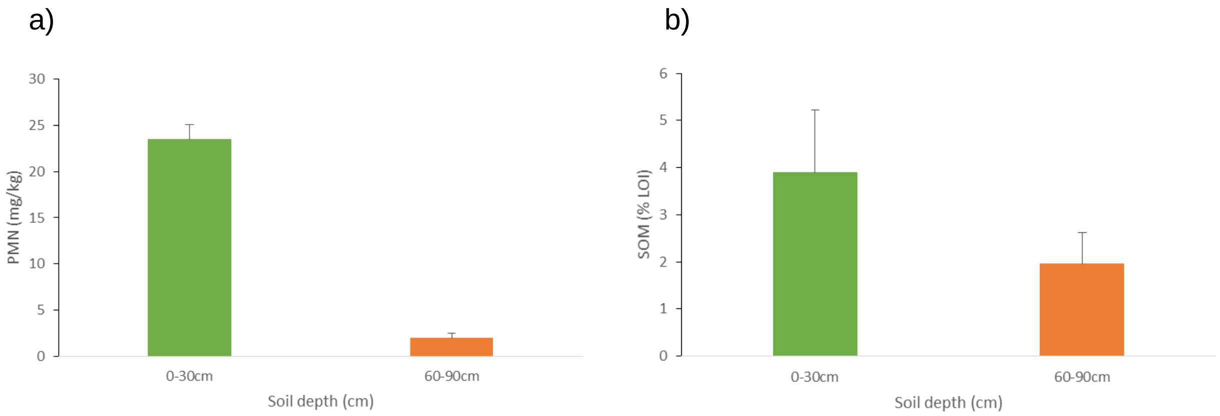

Figure 4-3 Change in a) PMN and b) SOM with sampling depth

4.2. CO_2 burst

4.2.1. Cropland CO_2-C benchmarks

The CO_2-C burst of cropped soils was normally distributed, with a slight positive skew (skew = 0.56; Figure 4-4). Using the current interpretation framework, 8% of values were considered to have low microbial activity (red traffic light; <25 mg/kg CO2-C), 12% moderate activity (amber traffic light; 25-74 mg/kg CO_2-C), with the majority of samples considered to have high microbial activity (green traffic light; ≥75 mg/kg CO_2-C).

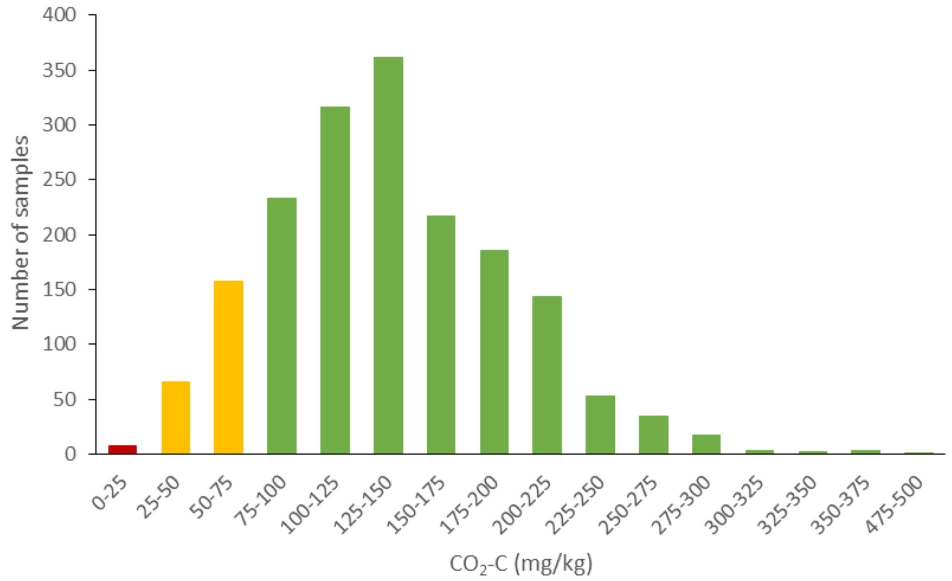

Figure 4-4 Frequency distribution of CO_2-C burst data for mineral (< 20% OM) topsoils in the UK under cropping (n=1803); bars coloured according to current guidance: red = low; amber = moderate; green = high/very high activity

There was a weak relationship between CO_2-C burst and soil organic matter content (%LOI; Figure 4-5a), but no relationship with clay content (Figure 4-5b) or topsoil pH (Figure 4-5c), although CO_2-C tended to be lower at higher pH levels. Consequently, there was no justification for generating different benchmarks according to soil type. Descriptive statistics were generated for the whole dataset (Table 4.5), from which guideline values (or benchmarks) for UK arable soils were estimated using the median and lower quartile CO_2-C values (Table 4.6). It was assumed that soils with CO_2-C levels less than the lower quartile (*c.* 100 mg/kg) had 'low' levels of microbial activity (red traffic light), soils with CO_2-C levels falling between the lower quartile and median value for UK soils (100-135 mg/kg) had 'moderate' levels (amber traffic light) and those greater the median (> 135 mg/kg) had 'good' or high (i.e. above average) levels of activity (green traffic light); Table 4.6. Adding the 1,936 samples with unknown land use to the cropland database gave almost identical results and class boundaries, with a lower quartile of 98.9 mg/kg and a median of 128 mg/kg.

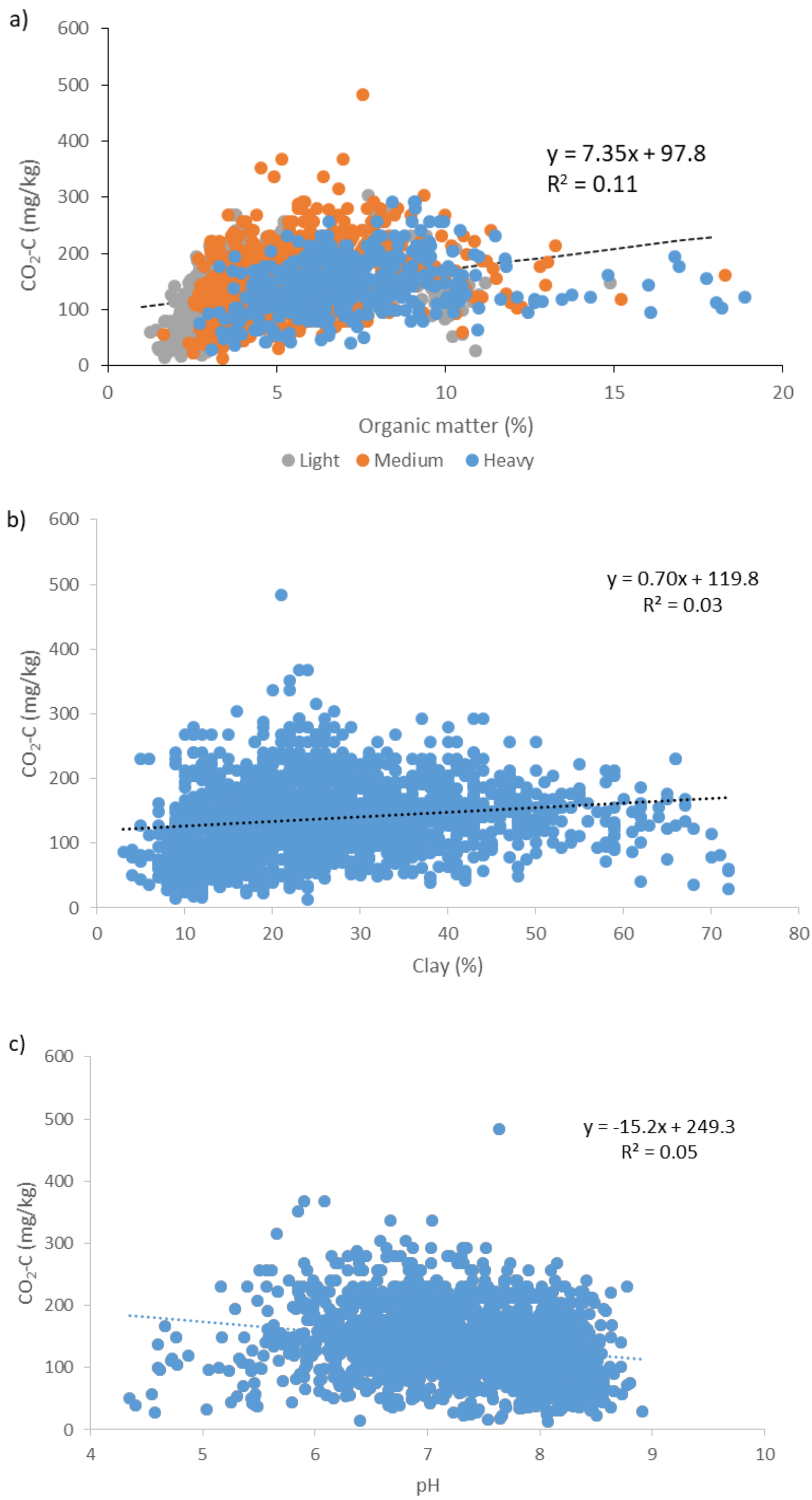

Figure 4-5 Relationship between CO_2-C burst and topsoil a) organic matter content (% LOI), b) clay content, and c) pH, for cropped soils in the UK (light soils < 18% clay; medium soils 18-35% clay; heavy soils > 35% clay).

Table 4.5 Descriptive statistics for topsoil organic matter and CO_2-C burst in mineral soils under cropland (n=1803).

Statistic	SOM (% LOI)	CO_2-C (mg/kg)
Mean	5.5	138
Standard error	0.06	1.32
Median	5.0	134
Minimum	1.3	13
Maximum	19	483
lower quartile	3.6	99
upper quartile	6.8	172
Skewness	1.2	0.6
95 % Confidence interval	0.11	2.6

Table 4.6 CO_2-C benchmarks for UK mineral soils under cropland

CO_2-C class	Traffic light colour	Description
< 100 mg/kg		Low activity
100-135 mg/kg		Moderate activity
>135 mg/kg		Good activity (above average)

4.2.2. Grassland CO_2-C burst benchmarks

The grassland category included samples labelled: 'permanent pasture', 'grazing', 'grassland', 'silage', 'reseeds' and 'leys'. For the SOM benchmarking (Griffiths et al., 2018) ley grassland was included within the arable category. However, as there were only 27 samples that had the identifier as 'ley grassland' and no details were provided on the length of the ley, these samples were included with the grassland analysis. Despite fewer samples than the 'cropped' database, CO_2-C in grassland soils was normally distributed (Figure 4-6). Using the current interpretation framework, as reported by NRM, very few values were allocated to the low (1%) and moderate (3%) categories, with the majority of samples considered to have high/very high microbial activity (green traffic light; ≥75 mg/kg CO_2-C).

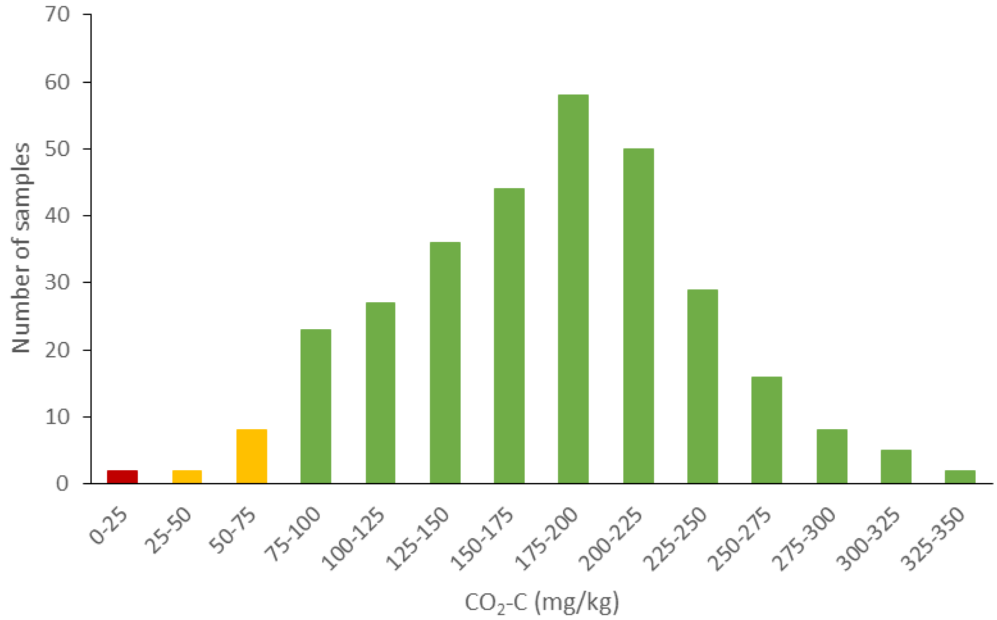

Figure 4-6 Frequency distribution of CO_2-C burst data for mineral (< 20% OM) topsoils in the UK under grassland (n=310); bars coloured according to current guidance: red = low; amber = moderate; green = high/very high activity

A greater proportion of the variation in CO_2-C measured from the grassland soils could be explained by variation in topsoil OM content (R^2 = 0.25; Figure 4-7). This probably reflects the more even distribution of values across the full range of organic matter contents compared to the cropland soils (which had a greater proportion of soils with low SOM and low CO_2-C). The relationship with clay content (R^2 = 0.03) and pH (R^2 = 0.02) was again poor, and there was no justification for generating different benchmarks according to soil type.

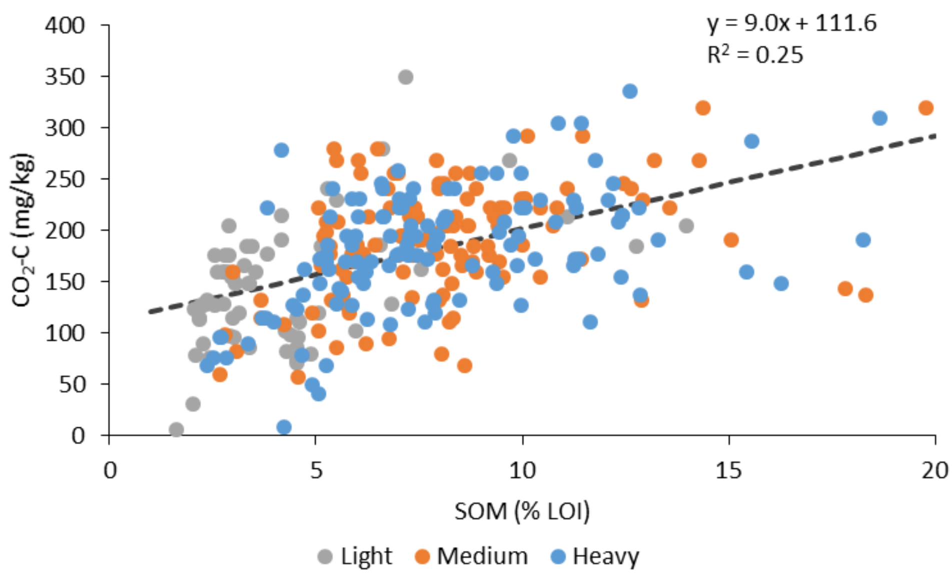

Figure 4-7 Relationship between topsoil organic matter content and CO_2-C burst in UK mineral soils under grass (light soils < 18% clay; medium soils 18-35% clay; heavy soils > 35% clay).

Descriptive statistics were therefore generated for the whole of the grassland dataset (Table 4.7), from which guideline values (or benchmarks) for UK grassland soils were derived using the median and lower quartile CO_2-C values (Table 4.8). It was assumed that soils with CO_2-C levels less than the lower quartile (*c.* 130 mg/kg) had 'low' levels of microbial activity (red traffic light), soils with CO_2-C levels falling between the lower quartile and median value for UK soils (130-180 mg/kg) had 'moderate' levels (amber traffic light) and those greater the median (> 180 mg/kg) had 'good' or high (i.e. above average) levels of activity (green traffic light). These benchmarks are *c.* 30 mg/kg higher than for cropped soils; comparison of the mean +/- 95% confidence intervals suggest that a split between grass (95% CI: 170 – 184 mg/kg) and arable (95% CI: 135 - 141 mg/kg) is justified and given SOM contents tend also to be higher under grassland soils, is also to be expected.

Table 4.7 Descriptive statistics for topsoil organic matter and CO_2-C burst in mineral soils under grassland (n=310).

Statistic	SOM (% LOI)	CO_2-C (mg/kg)
Mean	7.3	177
Standard error	0.2	3.4
Median	7.0	177
Minimum	1.6	6.2
Maximum	20	349
lower quartile	5.1	132
upper quartile	9.2	215
Skewness	0.8	-0.1
95 % Confidence interval	0.4	6.7

Table 4.8 CO_2-C benchmarks for UK mineral soils under grassland

CO_2-C class	Traffic light colour	Description
< 130 mg/kg		Low activity
130-180 mg/kg		Moderate activity
>180 mg/kg		Good activity (above average)

4.2.3. Effect of sampling depth on CO_2-C burst

Within the NRM database there were 9 sets of samples that compared topsoil (0-15 cm) and subsoil (60-90 cm) CO_2-C burst levels. These clearly demonstrated a significant decrease (P<0.05) in microbial activity (CO_2-C burst) in the subsoil (Figure 4-8); there was also a significant decrease in soil organic matter levels (topsoil mean = 4.7%; subsoil mean = 1.9%).

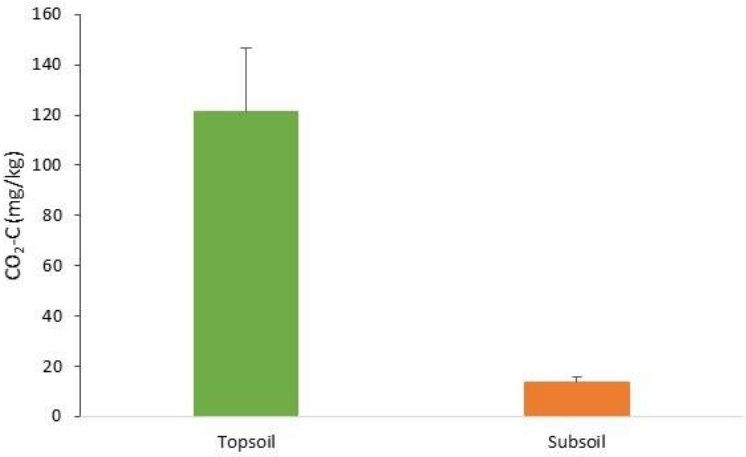

Figure 4-8 Change in CO_2-C burst with sampling depth

4.3. Testing the benchmarks at the long-term experimental sites

Results obtained from the four long-term experimental sites undertaken as part of Project 4 of the partnership programme between 2017 and 2019 were evaluated in the light of the proposed benchmark values (Table 4.9). All the results would have been given a green traffic light (‘no action required’) using the non-UK specific benchmarks (‘Old’ Benchmarks; Table 4.9), whereas with the new benchmarks this was reduced to *c.* 50% of values for both PMN and CO_2-burst.

For both properties, the light textured soils at the Gleadthorpe long-term experimental site (7% clay) were classed as moderate (amber traffic light) or low (red traffic light), with the control (no organic material additions) having the lowest PMN and CO_2-C values. This was supported by both the measurements of microbial biomass undertaken using the chloroform extraction methodology (note benchmarks for this parameter are not UK-specific) and by the soil organic matter content (%LOI). The absence of any ‘green’ traffic lights at this site for the microbial parameters, despite higher values where organic materials had been applied (albeit, not statistically significant) and significant increases in SOM ($P<0.05$) with organic material addition, suggests that separate benchmarks for light textured soils may be required. This is contrary to the findings in sections 4.1 and 4.2 – further analysis on a larger number of light-textured soils is required in order to determine whether separate benchmarks are required for this soil type.

Table 4.9 Evaluation of PMN and CO_2-burst measurements at the long-term experimental sites from project 4 of the SBSH partnership in the light of the newly proposed, UK-specific benchmarks.

Site & texture (date sampled)	Treatment	'Old' Benchmarks PMN (mg/kg)	'Old' Benchmarks CO_2-C (mg/kg)	'New' Benchmarks PMN (mg/kg)	'New' Benchmarks CO_2-C (mg/kg)	Biomass C (mg/kg)[a]	SOM (%LOI)[b]
Harper Adams[c] (2017)	Control	23	198	23	198	350	3.0
	FYM	90	228	90	228	393	4.1
	Slurry	24	247	24	247	315	3.6
	Green compost	43	222	43	222	317	4.0
	Green/food compost	38	219	38	219	334	3.7
	Food-based digestate	43	228	43	228	345	3.4
Loddington (2018)	Plough	71	143	71	143	401	7.1
	Direct drill	74	139	74	139	394	7.2
Craibstone (2018)[d]	pH 4.5	62	99	62	99	97	10.3
	pH 6.0	67	124	67	124	157	10.1
	pH 6.5	77	140	77	140	231	10.3
	pH 7.0	75	101	75	101	162	10.3
Gleadthorpe[c] (2019) Loamy sand	Control	12	68	12	68	96	1.9
	Broiler litter	26	85	26	85	129	2.1
	FYM	31	80	31	80	164	2.7
	Slurry	36	82	36	82	181	2.2
	Green compost	35	77	35	77	153	2.8

[a]Microbial biomass measured using the chloroform extraction methodology and interpreted using benchmark values proposed by Griffiths *et al.* (2018) from an international meta-analysis (Kallenbach & Grandy, 2011).
[b]SOM benchmarks dependent on soil texture (light/medium/heavy) and rainfall region (low, medium, high); Griffiths *et al. (*2018).
[c]Organic materials applied on an annual basis for 7-23 years depending on treatment, with N fertiliser rates adjusted accordingly; control treatment = recommended rates of fertiliser only.
[d]Average values across 4 crop types: winter wheat stubble, potatoes, 2nd year ley and oat stubble (undersown with grass).

At Craibstone and to a lesser extent, Harper Adams, there were differences in the allocation of results to benchmarks between the different methodologies. At Craibstone, although pH significantly affected PMN (P<0.05), the overall amount of PMN in these soils was above average at all pH levels (a green traffic light), whereas the CO_2-burst benchmarks suggested low activity at pH 4.5, moderate activity at pH 6 and 7.5, with above average activity at pH 6.5; this was mirrored by the variation (and benchmark allocation) in microbial biomass C. These differences in interpretation, suggest that the two methodologies (PMN and CO_2 burst) are measuring different soil properties/processes. The fact that SOM content did not vary with pH, and was high (i.e. within typical ranges for this soil type; Table 4.9) across all treatments at this site (similar to PMN) also suggests this. PMN measures a specific soil organic matter 'pool' - one that is readily decomposable by the microbial biomass (including the microbial biomass killed by the anaerobic incubation process) - whereas CO_2-burst measures the activity of the organisms involved in that decomposition (including decomposition of microbial biomass killed during the air drying process).

5. Overall evaluation

It is well established that the soil microbial community is fundamental to the regulation of organic matter transformations in soils and the cycling of nutrients, as well as the stabilisation of soil structure (e.g. via fungal hyphae and the release of polysaccharide 'glues'). This activity is, predominantly found in the topsoil, linked to the supply of fresh organic matter (residues, roots etc), oxygen and water. The fact that both PMN and CO_2-C burst levels were negligible in the subsoils within the respective databases demonstrates that these methodologies are integrated indicators of both the size and activity of the microbial community.

It is important that soil health indicators are relevant and clearly demonstrate the ability of a soil to deliver a specific function. This is relatively simple for some properties, such as soil nutrient status, where crop yields are reduced when nutrients drop below certain threshold concentrations (e.g. using the Nutrient Management Guide – RB209 index system for extractable P, K and Mg), or when roots cannot penetrate soil layers with resistances > 2.0MPa (Valentine *et al.,* 2012). For other properties, such as SOM, absolute thresholds are difficult to establish (Loveland *et al.,* 2001, 2003) and instead, 'envelopes of normality' (Verheijen *et al.,* 2005) or typical ranges can be defined based on the soil type and location. The draft soil health scorecard uses this approach for interpreting SOM values (Griffiths *et al.,* 2018), providing guidance to farmers and growers on how their soil compares to others in their locality, and the potential scope for improvement. For soil microbial biomass, there have been some studies showing positive correlations between the size of the microbial community and crop yield (e.g. Lupwayi *et al.,* 2015, Kiani *et al.,* 2017), but guidance on threshold values is limited, and not appropriate for UK cropping conditions. The databases provide by NRM and Hill Court Farm Research have provided the opportunity to quantify typical ranges for PMN and CO_2 burst for UK agroclimatic conditions.

The results from the long-term sites suggest that the proposed UK specific guidelines for both PMN and CO_2-C burst are more sensitive than the previously published values. Both parameters picked up differences in microbial activity due to the effect of organic material additions at Gleadthorpe (Table 4.9), but only the PMN guidelines suggested differences in function at Harper Adams (even though CO_2-C burst tended to be higher where organic materials were applied at this site; albeit not statistically significant). By contrast, for pH (i.e. at Craibstone), it was CO_2-C burst guideline values that appeared more sensitive. Clearly both parameters are an indicator of how the microbial community is functioning (both in terms of size and activity), but they measure different properties/processes. PMN is largely a measure of the readily decomposable N pool in soils (which will include microbial biomass N), whereas CO_2-C burst gives an indicator of the activity of the microbial community. At Craibstone, the latter measurement guidelines appeared more closely aligned to the fumigation extraction method for determining microbial biomass.

Further evaluation of these benchmark values will be undertaken using results obtained from the final year of sampling at the long-term experimental sites, as part of Project 4 reporting.

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