Report to Stapledon Memorial Trust Vacation Studentship Bursary

Host Institute: Rothamsted Research, North Wyke, Devon

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Student: Bríd Hanrahan **Email:** b.hanrahan5@universityof galway.ie

Supervisor: Alison Carswell **Email:** alison.carswell@rothamsted.ac.uk

Abstract

The use of sieved versus intact soils is often debated for their use in controlled environment experiments. While intact soils might give a better representation of the "in-field environment" sieving homogenises soils and removes spatial heterogeneity, soil flora and fauna and stones. There have been few studies comparing the effects of sieved and intact cores specifically for controlled environment experiments and this study aimed to investigate how soil preparation methods effected crop growth and herbage yield.

The effect of air-drying and sieving soil to < 2 mm and < 9 mm on perennial ryegrass and white clover mix yield, under controlled environment conditions, from packed soil columns was examined relative to intact soil columns.

Herbage yields were significantly greater from the sieved soil relative to that of the intact soil. Dry weight herbage yields were 1.16 ± 0.313, 4.99 ± 0.919 and 5.32 ± 0.315 t ha^{-1} from the intact soil, < 9 mm sieved soil and < 2 mm sieved soil columns respectively.

Sieving and air-drying soil creates a finer seedbed, creating a more favourable environment for seed germination and seed development, allowing initially greater yields from sieved soil columns relative to intact soil columns under controlled environment conditions.

Keywords

Lolium perenne
Controlled environment
Intact soil
Sieved soil
Soil preparation

Background

Both sieved and intact soils are used in controlled environment pot and column experiments. However, differences in soil properties between sieved and intact soils, linked to the processing of soils which have been sieved and typically air-dried may have the potential to affect crop establishment, growth and yield. While intact soils might give a better representation of the “in field” environment, sieved soils are often used to combine soil cores across multiple field locations and create a more homogenous set of samples.

Sieving changes the soil structure by disrupting aggregates. Soil structure influences water retention, carbon (C) sequestration and nutrient transformations (Blaud, et al., 2017). Soil aggregates are hotspots for bacterial activity and different sizes of aggregates have been shown to hold different bacterial diversity, abundance and biomass (Helgason et al., 2010). The disturbance effect of sieving is thought to result in an increase in organic C mineralisation especially in the days immediately after sieving (0 – 3 days). Sieving breaks up macro-aggregates into micro-aggregates increasing surface area and exposing organic material to biotic and abiotic transformation processes. Some authors have also shown an increase in soil microbial biomass after sieving, however, this does begin to subside with time (Franzluebbers, 1999). Although, other studies showed no difference in mineralisation of organic C or N between sieved and intact soils unless the soil was sieved to <3 mm, in which case mineralisation was 25 – 50% greater in soils sieved to <=1 mm compared to soils sieved to >=4mm (Curtin et al., 2014).

Materials and Methods

Soil was collected from 4 separate locations in a bare fallow plot from the North Wyke research site of Rothamsted Research (50.767197 N, -3.903286 W). Four intact soil cores (to 30 cm depth) and 4 replicates of approximately 10 kg of loose soil (from the plough layer, at approximately 20 cm depth) were collected in a 2 × 2 m square shaped sampling strategy. The soil was a clayey, non-calcareous soil from the Hallsworth soil series (Harrod and Hogan, 2010) that had recently been ploughed.

Photo 1. Left to right, Dr Alison Carswell, Bríd Hanrahan, Dr William Roberts and Jake Le Grice collecting soil cores from Little Burrows for my experiment. (Photo source: Griffiths, 2024)

Any vegetation was removed from the top of the intact cores and the cores were stored in gas permeable bags at 4°C in the dark to reduce biological activity prior to sowing. The loose soil was maintained at ambient air temperature in gas permeable bags prior to air drying and sieving. The individual replicates of loose soil were passed through a 9 mm sieve and half of each rep was then passed through a 2 mm sieve. The sieved soil was left to air dry at < 30 °C before being packed into columns the same size and depth as the intact cores. Phosphorus (P) and potassium (K) fertiliser and lime was mixed with the soil and prior to packing. Nitrogen (N) fertiliser was dissolved in solution and applied to the top of the intact cores 15 days after seed germination. Fertiliser additions were made according to the soil pH, P, K and Mg status and for clover containing swards for the N application, in accordance with AHDB recommendations (ADHB, 2023) on an individual experimental replicate basis. Soil pH varied between 5.6 and 6.0, available P varied between 6.6 and 12 mg dm^{-3} and available K varied between 56 and 79 mg dm^{-3}.

The soil columns were saturated and left for 24 hrs before sowing with an 85:15 perennial ryegrass (*Lolium perenne*, cv. Abermagic) and white clover (Trifolium repens cv. Aberdai) mix, after which they were laid out in the controlled environment room in a randomised control block design, allowing for differences in lighting and air flow. The controlled environment room settings were 16:8 hours day: night with respective temperatures of at 20:15 °C. Whilst seeds were germinating and seedlings becoming established the soil columns were watered lightly using a spray bottle daily for the first six weeks and then watering was increased once the plants were established.

Table 1. Experimental design

	Column 1	Column 2	Column 3
Row 1	9mm	2mm	Intact
Row 2	9mm	2mm	Intact
Row 3	2mm	Intact	9mm
Row 4	Intact	2mm	9mm

Where 9 mm is soil that was air dried and passed through a 9 mm sieve.
Where 2 mm is soil that was air dried and passed through a 2mm sieve.
Where Intact is soil that was not dried or processed and remained in core from collection.

White clover germination was poor in two of the intact soil columns, with no clover germination in one and only one seed germinated in another despite a germination test showing a germination rate of 80%. These two columns were resown with 0.004 g of white clover seed in the seventh week after the original sowing date. Eight weeks after the initial sowing date the crop was cut to a height of 2 cm. Fresh herbage from each column was weighed and then oven-dried for 48 h at 80 °C. After drying the dried herbage weight was recorded and the dried herbage ground to <0.5 mm. Following which the herbage sample was analysed for total N content, using a Carlo Erba NA 2000 linked to a Sercon 20/22 isotope ratio mass spectrometer (Sercon, Crewe, UK; Carlo Erba, CE instruments, Wigan, UK).

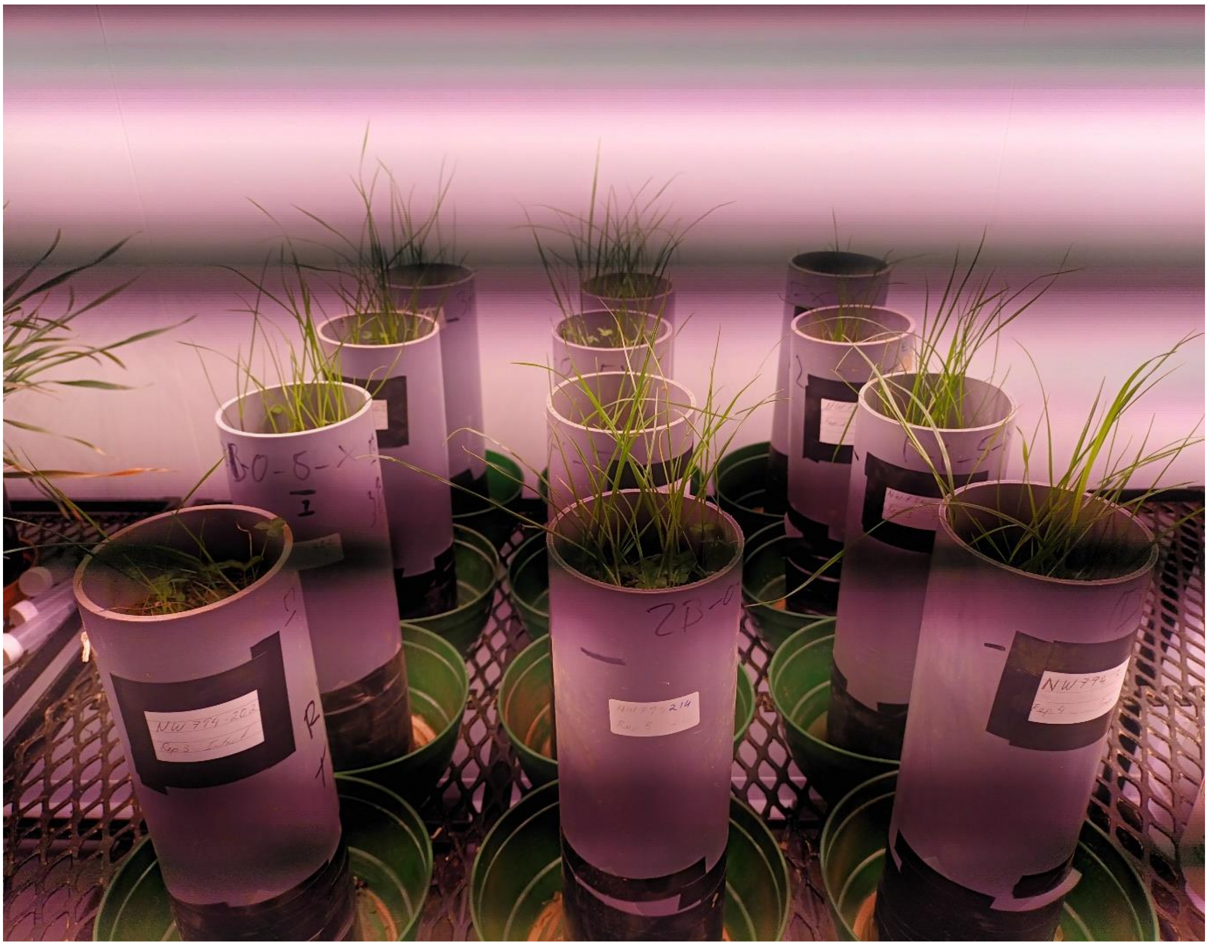

Photo 2. Showing soil column layout in the controlled environment room (Photo source: Hanrahan, 2024)

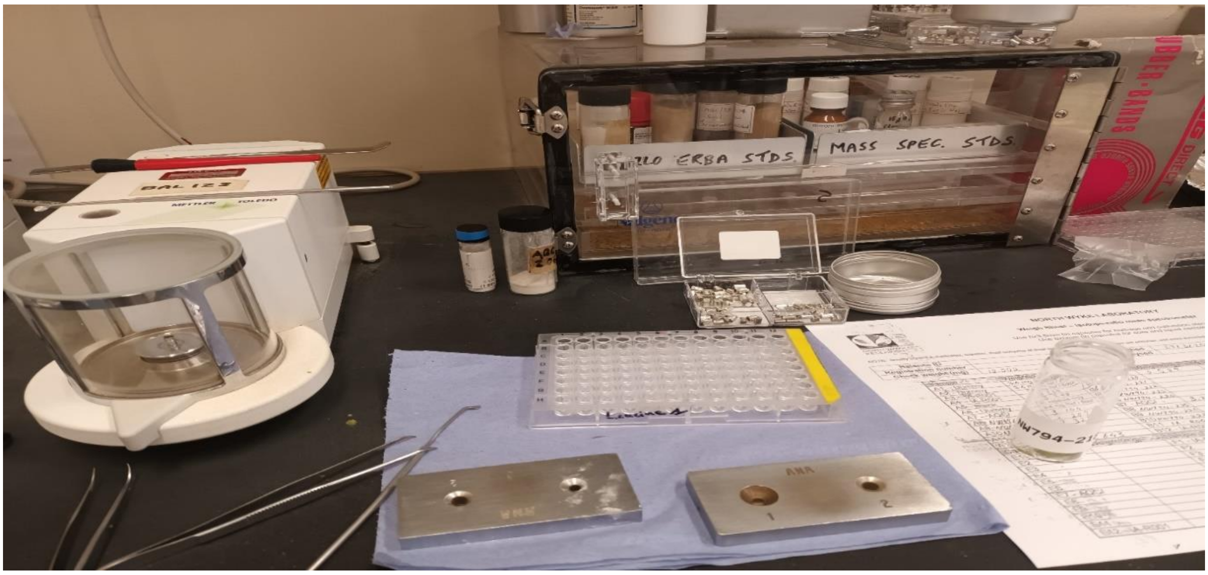

Photo 3: Preparing herbage samples for nitrogen analysis. (Photo source: Hanrahan, 2024)

To examine the variance around the mean herbage yields and the total N offtake by the herbage for each soil processing treatment a one-way Analysis of Variance was conducted, with a square root transformation applied to the yield and N offtake data to ensure it met model assumptions of data normality. Where significant differences between treatments were observed Fisher's protected least significant differences test was used to make multiple comparisons.

Results and Discussion

Significant differences were observed in the dry herbage yields between soil processing treatments ($p < 0.001$; Figure 1). The intact cores had a significantly lower herbage yield of 1.16 t ha^{-1} relative to both sieved soil treatments which had average yields of 4.99 and 5.32 t ha^{-1} for the <9 mm and <2 mm sieved soils respectively. The total N harvested in the herbage from each treatment also varied with significantly less taken up and removed by the lower yielding intact soil column treatment, relative to the sieved soil treatments (p <0.001; Figure 2). Total N concentration of the herbage was 1.43, 2.57 and 2.30 for the intact soil columns, the < 9 mm sieved soil columns and the < 2 mm sieved soil columns respectively (Table 2). The total N is mainly determined by the clover crop in the soil however the soil preparation method influenced the germination rate of the seeds.

Table 2: Dry matter and nitrogen offtake.

Treatment	Dry Matter Yield (t ha^{-1})	Standard error	Total N offtake kg N ha^{-1}	Standard error of mean
Intact soil	1.16	0.313	16.35	4.13
< 9 mm sieved soil	4.99	0.919	122.66	16.35
< 2mm sieved soil	5.32	0.315	121.41	4.55

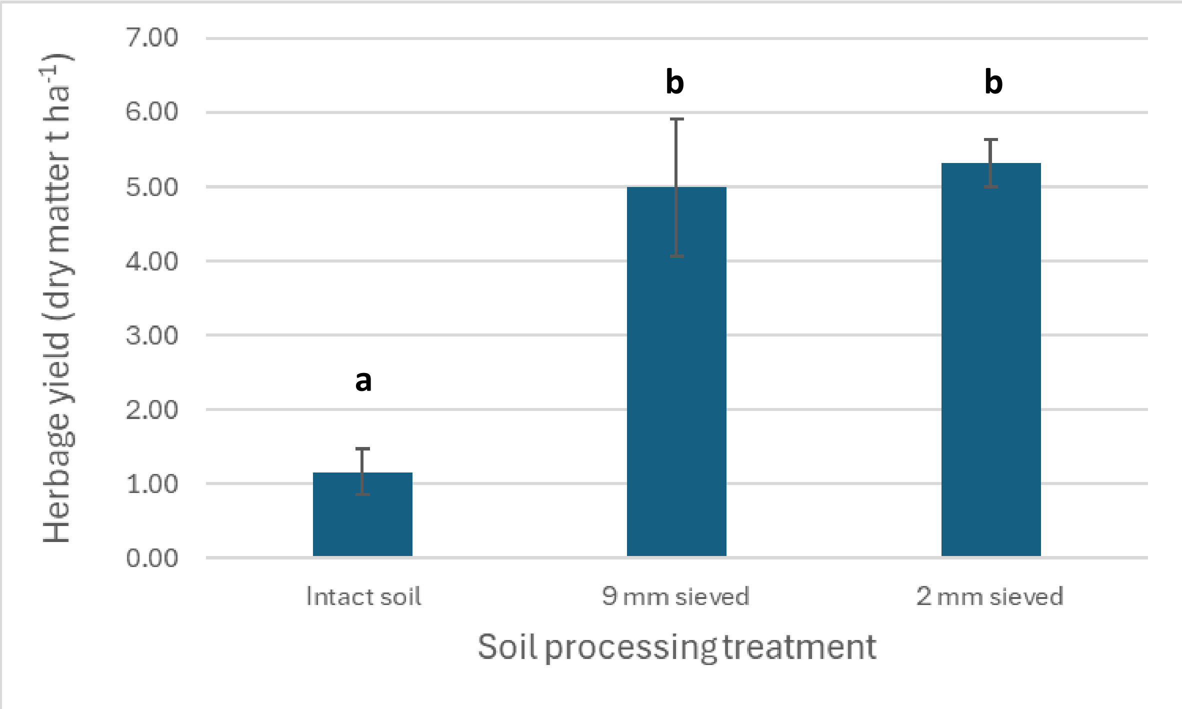

Figure 1. Herbage yield for differing soil processing treatments. Bars represent mean values (*n* = 4), error bars represent standard error of mean and the letters indicate significant differences between treatments according to Analysis of Variance with Fisher's LSD test.

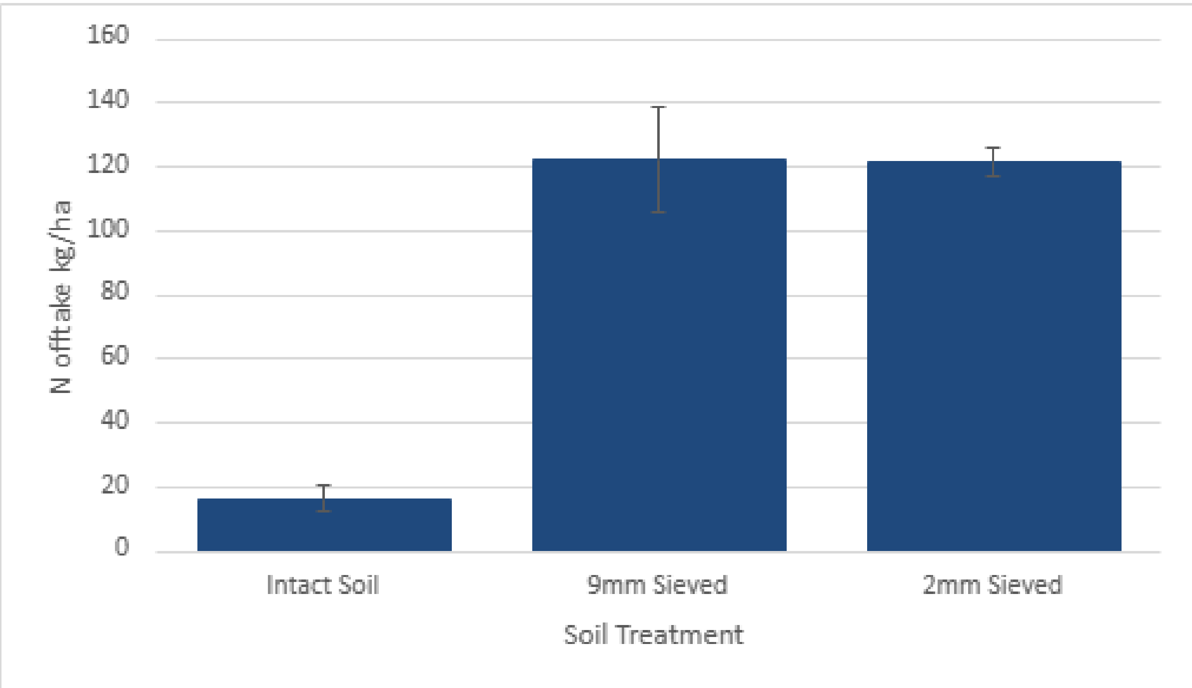

Figure 2. Nitrogen offtake in herbage grown under different soil processing treatments. Bars represent mean values (*n* = 4) and error bars represent standard error of mean.

The experiment will be continued for a further three cuts allowing for establishment issues to be resolved and providing more information. The clover germinated poorly in two of the intact cores so this will also allow the resown clover time to geminate and grow. As the rate of N fertiliser was applied with consideration that clover would contribute N via biological N fixation this could have influenced the lower yield in the intact cores. However, visual inspections showed that one intact core replicate had clover germination and growth levels similar to sieved cores and the yield was still low.

Conclusion

Soil preparation methods are shown to have an impact on crop yield with significantly greater herbage yields observed for both the sieved soil treatments. Sieve size did not have a significant impact on herbage yield. The germination of clover seed might have an important effect on herbage yield as it provides N via biological N fixation and the slow establishment may influence ryegrass growth. The N offtake is relevant to agricultural production as it directly correlates to the protein content of the crop.

It is advisable to consider the effects of soil preparation when carrying out a controlled environment experiment, as sieving will result in faster germination and increased yield in comparison to intact cores.

My Experience

During my time at Rothamsted I had the opportunity to get involved with other projects that were happening. I travelled to Rothamsted's other sites, Harpenden and Broom's Barn, to collect soil cores as part of the Resilient Farming Futures Strategic Program, led by Professor Adie Collins and Dr Martin Blackwell. We collected 60 cores from each site, and they were taken from plots of wheat, oats and ryegrass clover. I got involved marking out the random sampling design where the cores were being taken from and labelling and packing the cores. These cores were brought back to North Wyke where they will be used in a drought and saturation experiment. While in Harpenden I got to see the Broadbalk and Park Grass experiments, the oldest and longest running agricultural experiments in the world. I also got to see the sample archive which had over 300,000 samples of grass, grains, soil and forages. It was a really interesting trip and I learned a lot from it.

The North Wyke farm is part of the Environmental Change Network of farms that are being monitored long-term for changes in soil water and air quality as well as biodiversity. I assisted Helena Taylor on the weekly Environmental Change Network field work. We emptied moth traps, checked ponds for frog spawn and collected water samples. I got to follow the water samples through the labs as we first tested for electrical conductivity and pH before filtering them in readiness for the analytical lab, where they were analysed using the Aquachem for total oxidised nitrogen, phosphorus and ammonium.

Photo 4 and 5: Left photo shows lunch outside while carrying out Farm Platform field surveys with Maria Resendez and right photo shows the sample archive at Rothamsted research, Harpenden (Photo source: Hanrahan, 2024).

Another project I worked on during my time here was the measurement of soil pH buffer power. This was done on soils from the UK (North Wyke) and Nigeria. For the Nigerian soils I worked in the quarantine laboratory and followed quarantine procedures as stipulated by the Department of Environment, Food and Rural affairs license. Increasing concentrations of NaOH were added to the soils, after which I measured the pH and performed plant available P analysis using a malachite green reagent and plate reader which works on the basis of colourimetry.

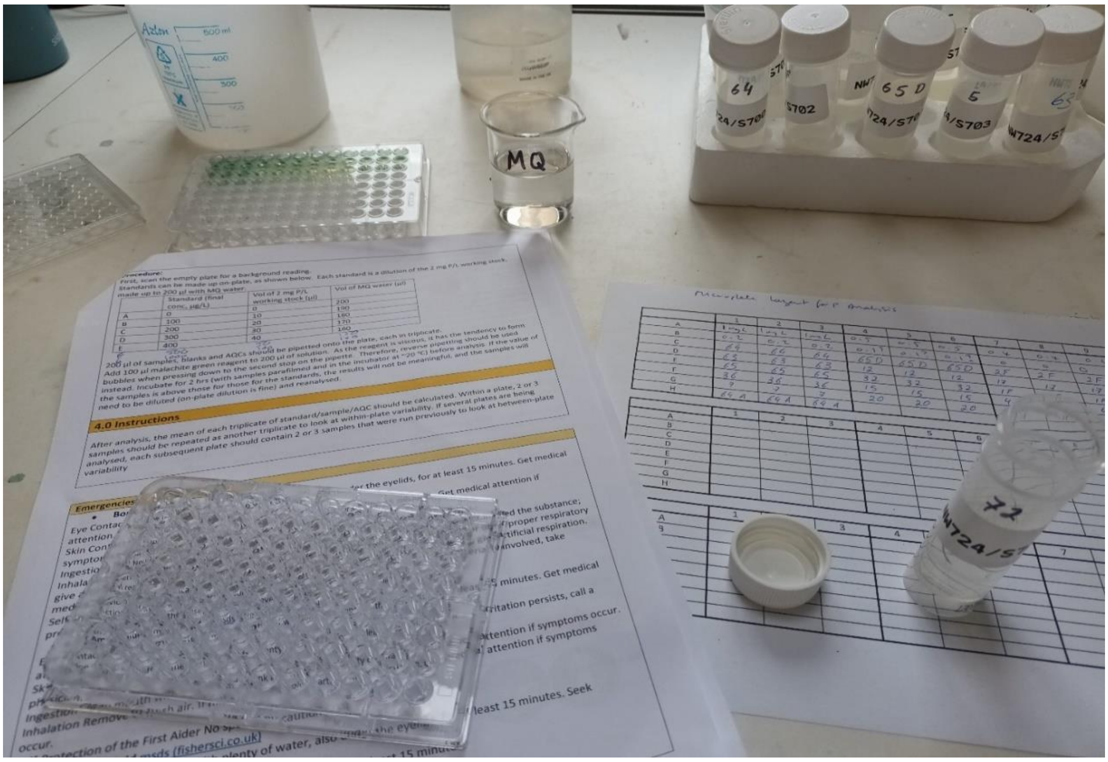

Photo 5: Phosphorous analyses (Photo source: Hanrahan, 2024)

Summary:

I thoroughly enjoyed my twelve weeks at Rothamsted Research, North Wyke. I gained a great insight into the world of scientific research and in particular soil and grassland science. Not only did I get to work on my own experiment I also had the chance to assist on other projects and as a result of my experience I gained new and valuable skills that I am sure will stand me in my future career and life. Coming from Ireland I got to see the differences and similarities within the agricultural industry as well and had many enjoyable weekends exploring the Devon countryside on my bike. I am incredibly grateful to my supervisor Dr Alison Carswell for all her support and guidance throughout the placement. Thanks also to Dr Martin Blackwell for showing me and explaining the other experiments happening at Harpenden and Brooms barn. I also wish to acknowledge the wider staff (from research, facilities to farm) at North Wyke who made me feel so welcome and were always willing to answer my questions. I feel very lucky to have such fond memories of my placement. Overall, it has been a really great opportunity and without the support of the Stapleton this opportunity may not have been possible for me, so I am very thankful for its support.

Photo 5: Cows due to calve enjoying some hay. (Photo source: Hanrahan, 2024)

References

Blaud, A. *et al.* (2017) 'Effects of dry and wet sieving of soil on identification and interpretation of microbial community composition', *Advances in Agronomy*, pp. 119–142. doi:10.1016/bs.agron.2016.10.006.

AHDB (2023) 'Nutrient Management Guide (RB209)', Section 3 Grass and Forage crops https://projectblue.blob.core.windows.net/media/Default/Imported%20Publication%20Docs/Nut%20Man%20Guide%20RB209%20-%20SECTION%203%20UPDATE%20(2023)%20_230821_WEB.pdf.

Curtin, D. *et al.* (2014) 'Mineralization of soil carbon and nitrogen following physical disturbance: A laboratory assessment', *Soil Science Society of America Journal*, 78(3), pp. 925–935. doi:10.2136/sssaj2013.12.0510.

Franzluebbers, A. (1999) 'Potential C and N mineralization and microbial biomass from intact and increasingly disturbed soils of varying texture', *Soil Biology and Biochemistry*, 31(8), pp. 1083–1090. doi:10.1016/s0038-0717(99)00022-x.

Helgason, B.L., Walley, F.L. and Germida, J.J. (2010) 'No-till soil management increases microbial biomass and alters community profiles in soil aggregates', *Applied Soil Ecology*, 46(3), pp. 390–397. doi:10.1016/j.apsoil.2010.10.002.

Harrod, T. and Hogan, D. (2010) The soils of North Wyke and rowden. *Unpublished report to North Wyke Research, revised edition of original report by TR Harrod, Soil Survey of England and Wales. Available at;* http://resources.rothamsted.ac.uk/sites/default/files/SoilsNWRowden.pdf (*Accessed:*15 February 2024)